

# Journal of Animal Science

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*Journal of Animal Science* (ISSN 0021-8812) is published 12 times per year (monthly) by the American Society of Animal Science. Periodicals postage paid at 201 W. Springfield, Ste 1202, Champaign, IL 61820 and at additional mailing offices. Form 3579 to be returned to the ASAS Business Office. Postmaster: Send change of address to American Society of Animal Science, PO Box 7410, Champaign, IL 61826.

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# Triennial Reproduction Symposium: Developmental programming of fertility

## 1 The importance of understanding the impacts of developmental programming on fertility: An overview. Lawrence Reynolds\*, *North Dakota State University, Fargo, ND.*

Increasing evidence supports the idea that reproductive efficiency may be “programmed” during prenatal or early postnatal life. Developmental programming is the concept that an “insult” during any developmental stage can result in programming of organ/organismal function in the short or long term. Insults that result in developmental programming include external factors such as (1) malnutrition pre-mating, during pregnancy, or during infancy; and (2) exposure to environmental factors such as social stress, high temperature-humidity, smoke, herbicides, pesticides, or phytosteroids. Intrinsic factors that result in developmental programming include (1) maternal age, (2) multiple fetuses, and (3) maternal and embryonic genetic background. Based on large epidemiological studies primarily in humans, as well as controlled studies in animal models including livestock, developmental programming affects the function of various organ systems in the offspring, resulting in altered growth, body composition, metabolism, and behavior. The organ systems that have been shown to be affected include, among others, adipose tissue, brain, cardiovascular system, endocrine system, gastrointestinal tract, kidney, muscle, and reproductive system, including the ovary, utero-placenta, testis, and hypothalamic-pituitary-gonadal axis. In addition, the effects of developmental programming have been shown to be transmissible across generations. The 2015 Triennial Reproduction Symposium will focus on how developmental processes and systems can affect reproductive success in males and females in livestock and other species.

**Key Words:** Triennial Reproduction Symposium, developmental programming of fertility, overview

## 2 Beef heifer development systems and lifetime productivity.

Andrew J. Roberts\*<sup>1</sup>, Rick N. Funston<sup>2</sup>, Elaine E. Grings<sup>3</sup>, and Mark K. Petersen<sup>1</sup>, <sup>1</sup>*USDA-ARS, Fort Keogh LARRL, Miles City, MT,* <sup>2</sup>*University of Nebraska West Central Research and Extension Center, North Platte, NE,* <sup>3</sup>*Department of Animal Science, South Dakota State University, Brookings, SD.*

Research continues to amass proving nutrition and other environmental factors experienced during pregnancy affect the fetus, resulting in changes in traits exhibited through life. In livestock, small differences in nutritional profiles during gestation may result in fetal programming that alters lifetime production efficiency. Thus, the potential of fetal programming should be considered when determining supplemental feeding strategies during gestation. For example, evaluation of offspring born to cows grazing dormant pasture supplemented with either 1.1 or 1.8 kg/d of alfalfa hay during the last 3rd of gestation demonstrated females born to cows provided the lower supplementation level were 10 kg heavier and had greater BCS at 5 yr of age. These differences appeared beneficial for maintaining reproductive performance in offspring managed with less harvested feed inputs. Comparison of offspring from cows wintered on native range (low quality) or improved pasture (greater quality) for 30 to 45 d during the 5th to 6th mo of gestation resulted in a trend for longer productivity in daughters from cows wintered on improved pasture. In recent studies comparing offspring from cows with or without protein supplementation while grazing dormant winter range during late gestation, heifers from protein-supplemented dams

had greater BW at weaning. This increase in BW persisted through pregnancy diagnosis and subsequent calving, and pregnancy rates were greater in heifers from protein-supplemented dams. Heifers from protein-supplemented dams appeared less feed efficient compared with heifers from un-supplemented dams. Thus, in utero exposure to nutritionally restricted environments (non-supplemented dams) may promote greater feed efficiency later in life. Nutrition during post-weaning development may also affect lifetime productivity. Heifers developed on low quality native range with RUP supplementation had greater retention beyond 3 yr of age than cohorts developed in a feed lot with higher quality feed and greater ADG. Collectively, these examples show long-term effects of nutritional management strategies used during gestation and development in given production environments need to be evaluated.

**Key Words:** fetal programming, lifetime productivity, nutritional supplementation

## 3 Effects of nutrition on reproductive development in bulls.

Leonardo F. C. Brito\*, *ABS Global Inc., DeForest, WI.*

A series of experiments were conducted to evaluate the effects of nutrition during calfhod (defined as the period from 10 to 26–30 wk of age) and peripubertal period (27–31 to 70–74 wk of age) on reproductive development in bulls. The overall objective of these experiments was to evaluate the effects of nutrition on endogenous metabolic hormones (leptin, insulin, GH, and IGF-I), gonadotropins and testosterone concentrations, sexual development, sperm production, and semen quality in bulls. Results of these experiments demonstrated that nutrition affected GnRH secretion and sexual development in bulls. Increased nutrition during calfhod resulted in a more sustained increase in LH pulse frequency during the early gonadotropin rise and greater testicular development at maturity. On the other hand, low nutrition during calfhod suppressed LH secretion during the early gonadotropin rise and resulted in delayed puberty and reduced testicular development at maturity. When low nutrition was accomplished by restricted feed intake, hypothalamic and pituitary function were compromised and LH secretion was more severely affected. Temporal associations between LH secretion patterns and circulating IGF-I concentrations implied that IGF-I is a possible signal to the central “metabolic sensor” involved in translating body nutritional status to the GnRH pulse generator. Nutrition also affected testicular steroidogenesis (testosterone concentrations), indicating effects on the number or function of Leydig cells, or both. Age-related increases in physiological and GnRH-stimulated circulating testosterone concentrations were hastened in bulls receiving high nutrition and delayed in bulls receiving low nutrition; these effects were probably mediated by both LH secretion and IGF-I concentrations. Circulating leptin and insulin may have only permissive roles on GnRH secretion, but may enhance testicular development. Growth hormone concentrations decreased concomitantly with increasing IGF-I concentrations during sexual development in bulls, suggesting that the testes could contribute considerable amounts of circulating IGF-I. In conclusion, management strategies to optimize reproductive function in bulls should focus on increasing nutrition during calfhod.

**Key Words:** bull, nutrition, sexual development

**4 Nutritional programming of puberty in heifers.** Gary L. Williams\*<sup>1,2</sup>, Rodolfo C. Cardoso<sup>1,2</sup>, Bruna R. C. Alves<sup>2</sup>, and Marcel Amstalden<sup>2</sup>, <sup>1</sup>Texas A&M AgriLife Research, Beeville, TX, <sup>2</sup>Texas A&M University, College Station, TX.

Because multiple estrous cycles are required for heifers to express full fertility before first breeding, ensuring that puberty is achieved in advance of 14 mo of age is critical. This allows heifers to calve as 2 year olds and optimizes lifetime efficiency. Feeding diets that promote elevated BW gain during the prepubertal period has been the primary strategy for achieving these end-points. Based on research conducted in our laboratories and others, it appears that the interval between approximately 4 and 7 mo of age (juvenile period) is a critical window for metabolically programming the neuroendocrine axis to achieve targeted reproductive maturation in heifers. By focusing on nutritionally mediated changes in the hypothalamus, we have been able to characterize several structural and functional modifications in hypothalamic pathways that respond to high-energy diets during this period. These include the differential expression of several genes (NPY, AGRP, POMC, and GHR) within the arcuate nucleus, a reduction in NPY innervation of GnRH neurons, decreased concentrations of central NPY, and increased innervation of kisspeptin neurons by  $\alpha$ MSH. These alterations are consistent with the hypothesis that accelerated growth during the juvenile period, resulting in increased circulating leptin, leads to decreased inhibition and accelerated excitation of GnRH neurons. Novel nutritional and managerial approaches for optimizing growth during sensitive periods of infantile and juvenile development are required in order for fundamental biology to reach practical translation. These approaches must facilitate nutritional imprinting of the brain during early calthood, while minimizing feeding costs, optimizing the consistent establishment of estrous cycles by 11 to 12 mo of age, and avoiding precocious puberty or disrupted mammary gland development. Our recent studies indicate that this may be achieved by exposing heifers to a stair-step nutritional regimen. However, a role for the maternal metabolic environment in heifer offspring responses to postnatal diets may also be operative. If confirmed, successful managerial strategies will also include the pregnant dam (USDA-NIFA grants 2009-65203-05678 and 2013-67015-20960).

**Key Words:** heifer, programming, puberty

**5 Future reproduction in gilts and boars is affected prenatally by sow management and early in life by management conditions to which the developing swine are exposed.** Mark J. Estienne\*, Virginia Tech, Blacksburg, VA.

Applied research at Virginia Tech has focused on effects of pre- or post-natal conditions on reproduction in adults. In utero, swine fetuses can be programmed as a consequence of intrauterine growth retardation (IUGR) caused by insufficient uterine capacity. Although negative effects of IUGR on postnatal growth are well documented, possible reproductive effects have been less studied. Preliminary work from our laboratory, however, showed that low BW boars had poorer libido and semen quality than high BW boars; in gilts, age at puberty was negatively correlated with pig BW. It is becoming apparent that fetuses can also be programmed by management of the gestating sow. For example, we compared growth and reproductive characteristics of gilts farrowed by sows that were kept in individual crates throughout gestation, group pens throughout gestation, or individual crates for the first 30 d post-mating and then group pens for the remainder of pregnancy. During the last 4 wk of the grow-finish period, BW of gilts farrowed by females housed in crates throughout gestation was greater than BW of gilts in the other 2 groups. Also, the efficiency of feed conversion was greatest and last-rib

backfat thickness the least, in gilts farrowed by females housed in crates throughout gestation. Interestingly, fewer gilts farrowed by females kept in crates throughout gestation reached puberty by 165 d of age compared with the other 2 groups. In intensively managed swine operations, there are many potential environmental stressors to which postnatal pigs are exposed early in life and this may affect future reproduction as well. For example, we determined the effects of crowding in the nursery on future reproduction and longevity in gilts. Reproductive performance during the second parity, including the number of pigs born alive was significantly greater for gilts each allowed 0.25 m<sup>2</sup> of floor space in the nursery compared with gilts restricted to 0.17 m<sup>2</sup>. Research will continue to identify prenatal or early-in-life stressors and to develop management strategies for mitigating adverse effects on swine reproduction.

**Key Words:** gilts, boars, development

**6 Our stolen figures: Using the process of sexual differentiation to think about endocrine-disrupting compounds and their effects on energy balance.** Jill E. Schneider\*, Lehigh University, Bethlehem, PA.

Reproductive processes (gametogenesis, hypothalamic-pituitary-gonadal function, and behavior) are masculinized or feminized during fetal development. At least some of these effects are determined during fetal development by androgens, estrogens, and glucocorticoids acting on cognate steroid receptors. These receptors, however, render reproductive development sensitive to endocrine-disrupting compounds that also act on the same receptors. Less well known is the fact that endocrine disruptors can alter energy balance (energy intake, storage, and expenditure). Recent evidence indicates that endocrine disruptors affect many individual processes known to contribute to obesity, including the gut microbiome, adipocyte differentiation, energy metabolism, ingestive behavior, and the tendency to accumulate adipose tissue in response to certain diets. Understanding effects of endocrine disruptors on energy balance will be aided by attention to the processes involved in sexual differentiation. This is because many energy-balancing traits are sexually dimorphic with the masculine phenotype most closely linked to metabolic diseases such as type II diabetes and heart disease. So far, it is clear that at least some endocrine disruptors have masculinizing effects via classical organizational effects on sexually dimorphic energy balancing traits during fetal development. In addition, we should expect endocrine disruptors to affect other defined mechanisms of sexual differentiation (e.g., sex chromosome action, aromatization, active feminization, and organizing actions at later periods of development, such as puberty). Investigators interested in effects of endocrine disruptors on peripheral metabolism often work in isolation from those interested in the effects of endocrine disruptors on ingestive behavior. In fact, changes in peripheral metabolism have organizational and activational effects on the neural circuitry that controls ingestive behavior. Together, these considerations demand a concerted multidisciplinary and integrative approach to the study of endocrine disruptors.

**7 Environmentally induced epigenetic transgenerational inheritance of disease: Ancestral ghosts in your genome.** Michael K. Skinner\*, Center for Reproductive Biology, School of Biological Sciences, Washington State University, Pullman, WA.

Transgenerational effects of environmental toxicants significantly amplify the effect and health hazards of these compounds. One of the most sensitive periods to exposure is during embryonic gonadal sex determination when the germ line is undergoing epigenetic programming and DNA re-methylation. Previous studies have shown that endocrine

disruptors can cause an increase in adult onset disease such as infertility, prostate, ovary and kidney disease, cancers and obesity. Interestingly, this effect is transgenerational (F<sub>1</sub>, F<sub>2</sub>, F<sub>3</sub>, and F<sub>4</sub> generations) and hypothesized to be due to a permanent (imprinted) altered DNA methylation of the germ-line. The transgenerational epigenetic mechanism appears to involve the actions of an environmental compound at the time of sex determination to permanently alter the epigenetic (i.e., DNA methylation) programming of the germ line that then alters the transcriptomes of developing organs to induce disease susceptibility and development transgenerationally. A variety of different environmental compounds have been shown to induce this epigenetic transgenerational inheritance of disease including: fungicide vinclozolin, plastics BPA and phthalates, pesticides, DDT, dioxin and hydrocarbons. The suggestion that environmental factors can reprogram the germ line to induce epigenetic transgenerational inheritance of disease and phenotypic variation is a new paradigm in disease etiology that is also relevant to other areas of biology such as evolution.

### **8 Environmental effects on programming of reproductive behavior.** Frederick vom Saal\*, *University of Missouri-Columbia, Columbia, MO.*

Fetal development is a period of heightened sensitivity to hormones that regulate the differentiation of tissues. An example showing that very small differences in testosterone (T) and estradiol (E<sub>2</sub>) during fetal life can lead to changes in the life history of males and females is the intrauterine position phenomenon or IUP. Developing between fetuses of the same or opposite sex in species in which there are multiple fetuses (polytocous species) results in very small differences in fetal serum T and E<sub>2</sub> and differences in the development of tissues, including the brain, responsive to these sex steroids. For example, in mice, 2F males (located in utero between 2 females) have elevated serum E<sub>2</sub> during fetal life and in adulthood show an increase in sexual behaviors relative to 2M males (located between 2 males); 2M males have elevated serum T during fetal life and in adulthood are more aggressive than 2F males. Similarly, 2F female mice are more sexually attractive to males and more sexually receptive, but less aggressive, than their 2M female siblings. There are now numerous environmental chemicals that have been found to bind to estrogen receptors and disrupt normal estrogen signaling. The best studied of these estrogenic endocrine-disrupting chemicals is bisphenol A or BPA. Developmental exposure to BPA has been related to numerous changes in brain structure, function, and behavior in both males and females. Of great interest is the finding that the magnitude of the sex differences in some behaviors observed in untreated rats and mice is reduced or eliminated as a result of exposure to doses of BPA that are relevant to exposures experienced by humans based on biomonitoring studies. While there is less information regarding the effects of endocrine disrupting chemicals such as BPA in farm animals relative to rodents or humans, there is evidence for effects of fetal exposure to BPA on neuroendocrine function in sheep. There is also evidence for transgenerational transmission of altered phenotype, including behavior,

caused by exposure to endocrine disrupting chemicals during the period of germ cell epigenetic programming.

**Key Words:** endocrine disruptor, bisphenol A, fetal programming

### **9 Potential effects of real life exposure to environmental contaminants on reproductive health.** Neil P. Evans\*<sup>1</sup>, Michelle Bellingham<sup>1</sup>, Corinne Cotinot<sup>2</sup>, Stewart M. Rhind<sup>3</sup>, Richard Sharpe<sup>4</sup>, and Paul A. Fowler<sup>5</sup>, <sup>1</sup>*College Medical Veterinary and Life Sciences, Institute of Biodiversity Animal Health & Comparative Medicine, University of Glasgow, Glasgow, UK*, <sup>2</sup>*INRA, 1198 Biologie du Developpement et Reproduction, Jouy en Josas, France*, <sup>3</sup>*James Hutton Institute, Aberdeen, UK*, <sup>4</sup>*Queens Medical Research Institute, MRC Centre for Reproductive Health, University of Edinburgh, Edinburgh, UK*, <sup>5</sup>*Institute of Medical Sciences, Division of Applied Medicine, University of Aberdeen, Aberdeen, UK.*

While much research has focused on the effects of individual chemical exposures on animal health, far less is known about the effects of exposure to the mixtures of chemicals often found within our environment, even though this is a more typical exposure pattern. Biosolids (processed human sewage sludge) contain low individual concentrations of an array of contaminants including heavy metals and organic pollutants [e.g., polycyclic aromatic hydrocarbons, polychlorinated biphenyls and polychlorinated dibenzo(p)dioxin and furan] and form the basis of our model with which to study the effects of exposure to mixtures of environmentally relevant concentrations of pollutants in a domestic animal, the sheep. Studies using this model have investigated the effects of developmental exposure to biosolids on a variety of reproductive endpoints, including GnRH, kisspeptin, and estradiol receptor expression within the hypothalamus, LH and estradiol receptor expression within the pituitary gland, and protein, mRNA, and gamete production within the gonads of male and female sheep. The studies suggest that exposure to biosolids in utero, via maternal exposure, has detrimental effects on the fetal hypothalamo-pituitary-gonadal axis that could affect subsequent fertility. Studies with adult animals, also exposed during fetal life, suggest long-term effects of environmental chemical exposure on the reproductive axis. Investigation of lambs born to ewes grazed on biosolid-treated pastures (1) throughout life, (2) up until gestation, or (3) only during gestation have shown that some effects of environmental chemicals (relative to unexposed controls) may be more pronounced when exposure is acute, or that physiological compensation may occur when exposure is prolonged. Overall, the results of this study question the reliance on no observed adverse effects levels, with regard to chemical safety, when chemical exposure normally occurs as complex mixtures. The results suggest that developmental chemical exposure may affect the hypothalamo-pituitary-gonadal axis at a variety of levels, although whether the effects are driven by central effects or occur at each organ studied remains to be determined. [Wellcome Trust grant 080338].

**Key Words:** endocrine-disrupting chemicals, environmental contamination, ruminant



# Breeding and Genetics Symposium: Joint Interbull/JAM Session: Milk spectral data—Cost-effective information to improve expensive and limited traits in dairy cattle breeding

**10 Capitalizing on fine milk composition for breeding and management of dairy cows.** Nicolas Gengler<sup>\*1</sup>, H el ene Soyeurt<sup>1</sup>, Fr ed eric Dehareng<sup>2</sup>, Catherine Bastin<sup>1</sup>, Fr ed eric Colinet<sup>1</sup>, Hedi Hammami<sup>1</sup>, and Pierre Dardenne<sup>2</sup>, <sup>1</sup>*University of Li ge-GxABT, Gembloux, Belgium*, <sup>2</sup>*Walloon Agricultural Research Center, Gembloux, Belgium*.

Management and breeding of dairy cows face the challenge of permanently adapting to changing production circumstances under socio-economic constraints. If management and breeding addresses different timeframes of action, both need relevant phenotypes that allow for precise monitoring of the status of the cows, their products (i.e., milk and subsequently dairy products), their behavior and their environmental impact. Milk composition has been identified as an important source of information since it could reflect, at least partially, all these elements. Major milk components such as fat, protein, urea, and lactose contents are routinely predicted by mid-infrared (MIR) spectrometry and have been widely used for these purposes. But, milk composition is much more complex and other components might be informative. Such new milk-based phenotypes should be considered given that they are cheap, rapidly obtained, usable on a large scale, robust and reliable. In a first approach, new phenotypes can be predicted from MIR spectra using classical prediction equation based techniques. This method was used successfully for many novel traits (e.g., fatty acids, lactoferrin, minerals, milk technological properties, citrate), that can then be useful for management and breeding purposes. An innovation was to consider the longitudinal nature of the relationship between the trait of interest and the MIR spectra (e.g., to predict methane from MIR). By avoiding intermediate steps, prediction errors can be minimized when traits of interest (e.g., ketosis) are predicted directly from MIR spectra. In a second approach, in an innovative manner, patterns detected by comparing observed from expected MIR spectra can be used directly. All these traits can then be used to define best practices, adjust feeding and health management, improve animal welfare, improve milk quality and limit environmental impact. Under the condition that MIR data are available on a large scale, phenotypes for these traits will allow genetic and genomic evaluations. Introduction of novel traits into the breeding objectives will need additional research to clarify socio-economic weights and genetic correlation with other traits of interest.

**Key Words:** milk, spectra, novel traits

**11 Applications of milk spectroscopy data to select for feed efficiency and reduced methane emissions in dairy breeding programs.** Sinead McParland<sup>\*1</sup> and Frederic Dehareng<sup>2</sup>, <sup>1</sup>*Animal and Grassland Research & Innovation Centre, Teagasc Moorepark, Fermoy, Co. Cork, Ireland*, <sup>2</sup>*Walloon Agricultural Research Centre, Gembloux, Belgium*.

Feed efficiency (FE), the difference between energy intake and the energy demanded and supplied by various reservoirs and sinks, is of increasing importance. One such energy sink is methane emissions (ME); ME also have repercussions on climate change. Collection of the necessary data for the computation of genetic evaluations for both FE and ME are, however, laborious and expensive. Mid-infrared spectrometry (MIRS) offers a solution to routinely collect data on milk recorded

animals at no additional cost to milk recording. The use of MIRS to accurately predict animal and milk traits, including energy balance (EB) and milk fatty acids (FA) has recently been shown and validated. When changes in body mass are zero, EB is mathematically identical to residual feed intake (RFI), a measure commonly used to depict FE. The relationship between ME and milk FA has also been reported. Therefore, it was recently hypothesized that both RFI and ME could also be predicted from milk. Two separate studies were conducted using 1,270 records of RFI from Irish Holstein-Friesians, and 77 records of ME from Belgian Holstein cows. Accuracy (i.e., correlation) of predicting RFI and ME using partial least squares regression on the MIR spectral data was 0.62 and 0.87, respectively, in cross validation. Both studies were limited by small numbers, and have yet to be independently validated. The heritability of MIR-predicted RFI was 0.06, and was lower than the heritability of measured RFI using the same data set (0.10), however the genetic correlation between MIR-predicted and measured RFI was 0.78 using the limited data set. Furthermore, when prediction equations were applied to larger data sets of MIR spectral data, the lactation profile for RFI was in line with expectations based on profiles estimated from measured RFI. The shape of the lactation profile of predicted ME was, however, dependent on stage of lactation. The major limitation precluding FE and ME from breeding programs in the past can be resolved through the use of MIRS on milk recorded animals.

**Key Words:** feed efficiency, methane, mid infrared spectrum

**12 Using milk spectroscopy phenotypes in genetic selection programs to improve the nutraceutical value of milk in dairy cows.** Henk Bovenhuis<sup>\*</sup>, *Animal Breeding and Genomics Centre, Wageningen University, Wageningen, the Netherlands*.

Milk is a unique product that is specifically suited to nourish the offspring and because initially milk is the young's only food it has to contain all essential ingredients. However, several studies have indicated that in modern human diets, a high intake of saturated fatty acids is a risk factor for cardiovascular diseases. Milk and dairy products are major sources of these fatty acids. Milk fat composition is strongly affected by genetic factors, which suggests that there are opportunities to change milk fat composition by means of selective breeding. A crucial factor in successful selection strategies is the availability of phenotypes. Milk fat composition can be accurately determined based on gas chromatography (GC); however, this analytical method is expensive and relatively time consuming. Alternatively, fatty acids can be predicted based on infrared spectra. Infrared prediction of unsaturated fatty acids might be based on indirect relations or on a direct relationship between infrared frequencies and the double bond within the fatty acid chain. Distinguishing direct and indirect relations is relevant as it might have consequences for the range of conditions under which prediction equations are valid. Bouwman (2014) compared genome-wide associations for GC-based fatty acids with infrared-predicted fatty acids and found considerable differences. A typical example was the region on BTA26 containing the SCD1 polymorphism. More recently, we studied the effect of the SCD1 polymorphism on all 1,060 individual infrared wavelengths (Wang et al., 2015). No wavelengths were significantly affected by the SCD1 polymorphism whereas the SCD1 polymorphism has been shown to have significant effects on the content of C10:0, C14:0, C18:0, C10:1, C12:1,

C14:1, and C16:1 (Duchemin et al., 2012). This suggests that infrared spectra contain little direct information on the content of these fatty acids. This confirms results by Eskildsen et al. (2014), who concluded that prediction of individual fatty acids relies on correlations with fat content rather than on direct relations with specific Infrared frequencies.

**Key Words:** infrared, prediction, milk fatty acids

**13 Do milk spectroscopy phenotypes have a role to play in dairy fertility and health breeding programs?** Catherine Bastin\*<sup>1</sup>, Léonard Théron<sup>2</sup>, Aurélie Lainé<sup>1</sup>, and Nicolas Gengler<sup>1</sup>, <sup>1</sup>*University of Liège, Gembloux Agro-Bio Tech, Animal Science Unit, Gembloux, Belgium*, <sup>2</sup>*Faculty of Veterinary Medicine, Clinical Department of Production Animals, University of Liège, Liège, Belgium*.

Genetic selection allows for permanent improvement of dairy cow fertility and health. However, these traits are generally complex, difficult to record, and lowly heritable (<0.10). Hence, indicators have been proven as useful in the prediction of genetic merit for direct fertility and health traits as long as they are easier to measure, heritable, and genetically correlated. Considering that changes in (fine) milk composition over the lactation reflects the physiological status of the cow, the mid-infrared (MIR) analysis of milk opens the door to a whole new range of potential indicator traits of fertility and health. Previous studies investigated the phenotypic and genetic relationships between fertility and MIR-predicted phenotypes, most of them being related to negative postpartum energy balance and body fat mobilization (e.g., fat to protein ratio, urea, fatty acids profile). Results showed that a combination of various fatty acids traits (e.g., C18:1 *cis*-9 and C10:0) could be used to improve fertility. Furthermore, occurrence of (sub-)clinical ketosis has been related to milk-based phenotypes such as fatty acids and ketone bodies. Hence, MIR-predicted acetone and  $\beta$ -hydroxybutyrate contents in milk were demonstrated as useful for breeding cows less susceptible to ketosis. Although studies investigating the genetic association among mastitis and MIR-predicted phenotypes are scarce, a wide range of traits, potentially predicted by MIR spectrometry, are worth considering. It includes traits related to the disease response of the cow (e.g., lactoferrin), to the reduced secretory activity (e.g., lactose) and to the alteration of blood-milk barrier (e.g., minerals, citrate). Moreover, direct

MIR-prediction of fertility and health traits should be further considered. To conclude, MIR-predicted phenotypes have a role to play in the improvement of dairy cow fertility and health. However, further studies are warranted to (1) grasp underlying associations among MIR-predicted indicator and fitness traits, (2) estimate the genetic parameters, and (3) include these traits in broader breeding strategies.

**Key Words:** health, fertility, mid-infrared spectrometry

**14 Implementation of mid-infrared calculated tools to select for novel traits in dairy cattle breeding.** Eileen Wall\*<sup>1</sup>, Stephanie Smith<sup>1</sup>, Sinead McParland<sup>2</sup>, Vicky Hicks<sup>3</sup>, and Mike Coffey<sup>1</sup>, <sup>1</sup>*Scotland's Rural College, Edinburgh, UK*, <sup>2</sup>*Teagasc, Cork, Ireland*, <sup>3</sup>*National Milk Records, Chippenham, UK*.

Currently, mid infrared (MIR) spectroscopy of milk samples is used to predict milk fat, protein, and lactose. While the technology has been utilized for milk recording for numerous years, the application of mid-infrared to measure other parameters in milk is a recent development. More recent studies have shown that MIR can predict different milk quality attributes (e.g., Soyeurt et al., 2011, 2012) as well as other performance traits such as energy balance (McParland et al., 2011, 2014), methane emissions (Dehareng et al., 2012) and health and fertility traits. Here we extend the predictions of novel traits from milk MIR to implementation in the industry as part of routine milk recording. We will discuss the system established to routinely capture and used milk spectra in an operational milk recording business. The system established has allowed us to apply prediction equations for a range of traits to national spectra on almost 117,000 animals (over 1 million test-dates) from 355 farms across the UK (from Feb 2013 to Jan 2015). Systems for routinely predicting novel traits have been established and routine extraction of data for the estimation of genetic parameters and breeding values developed. For example, heritability estimates for cow energy balance, body energy content and energy intake were 0.12, 0.23, and 0.15 respectively. Mid-infrared-based traits predictions from routinely collected data at a national level have potential use in genetic improvement of livestock for previously difficult and expensive to record traits.

**Key Words:** milk mid-infrared, genetic improvement, dairy cows



# Breeding and Genetics Symposium: Joint Interbull/JAM Session: Use of genomics to improve limited and novel phenotypes in animal breeding

**15 The effect of genomic technologies in the selection of novel phenotypes in dairy crossbreeding programs.** Richard J. Spelman\*, Mathew D. Littlejohn, Ric G. Sherlock, and Steve Davis, *Livestock Improvement Corporation, Hamilton, New Zealand.*

Genomic selection in dairy cattle has been successfully applied for milk production traits in many countries around the world. This has been enabled through the routine collection of phenotypes over several years and the large body of animals that have been genotyped. The application of genomics for non-routinely collected phenotypes has been less successful due to the cost of phenotyping and the smaller number of animals available for genotyping. Genomic estimates for Johnes susceptibility and residual feed intake have been recently commercialized in the New Zealand dairy population. Johnes susceptibility has been estimated in a case control experimental setting. Over 1,500 animals that have been identified to be affected by Johnes have been genotyped and compared with a control derived from the general population of animals that have been genotyped in the LIC genomic selection data set. The accuracy of evaluation for Johnes susceptibility is approximately 30% with a heritability of 18%. Residual feed intake was measured over 2000 growing Holstein-Friesian heifers in New Zealand and Australia, which were phenotyped over 42 d for feed intake and live weight gain. Genomic estimates for residual feed intake have a reliability of 10%.

**Key Words:** dairy, genomics, crossbreeding

**16 Using genomic technologies to improve broiler health and production.** Rachel J. Hawken\*, *Cobb-Vantress, Siloam Springs, AR.*

Health and welfare traits have always been a critical factor toward the genetic improvement of broiler breeding stock. Current trends toward antibiotic free broiler production have emphasized the need to engage new technologies toward the improvement of these traits. Genomic technologies such as genome selection, genome sequencing, and mutation detection all offer great opportunities to advance these traits. For example, with the aid of genomics, genetic information from birds in disease challenged environments (on pure line breeders or crossbred product birds) can be utilized for the selection of superior breeding stock at the pedigree level, in non-diseased environments. Furthermore, the genetic basis of genetic diseases can be identified and deleterious alleles eliminated from breeding programs. This presentation will illustrate various opportunities for improving health and production traits using genomics at Cobb.

**Key Words:** poultry, genomics

**17 Application of genomic technologies to improve feed efficiency traits in swine.** Kent A. Gray\*, *Smithfield Premium Genetics, Rose Hill, NC.*

One challenge that faces the US pork industry is the efficient use of feed resources as feed costs continue to be the largest variable expenditure. Typically, feed efficiency is measured as feed conversion ratio (FCR) or calculated through residual feed intake (RFI). Regardless of the measure defined as feed efficiency, average daily gain (ADG) and average daily feed intake (ADFI) remain the 2 main components for estimating nutrient utilization traits. Several studies demonstrate improvement of feed efficiency through selection programs focused on improving either

FCR or RFI. However, these programs require recording individual feed intake, which is labor intensive, time consuming, and requires specialized equipment. Additionally, data from feed intake recording equipment (FIRE) systems contain several data errors that require careful editing before the data can be used. With the introduction of advanced genomic technologies in livestock species there is an opportunity to improve selection of feed efficiency. Genomic tools have helped us identify genomic regions associated with feed efficiency and other production traits through whole-genome association studies. One region in particular located on SSC1 ranging from 166 to 170 Mb was significantly associated with ADFI, ADG, and backfat (BF), where *SOCS6* and *DOK6* are proposed as the most likely candidate genes. The use of whole-genome selection methodologies can also be used to improve response to selection by increasing accuracy. Variance components were estimated with AIREML and (G)EBV were computed with either BLUP or single-step genomic BLUP (SSGBLUP). Comparisons of reliabilities between traditional BLUP estimates and SSGBLUP estimates indicate that GEV estimates for FCR and component traits BF, ADFI, ADG, and off-test weight (WT) have increased reliabilities by 9% to 60% when compared with traditional EBV estimates. Because of this increased accuracy, we can conclude that application of genomic technologies will help us improve feed efficiency traits in swine.

**Key Words:** feed efficiency, swine, genomic selection

**18 The role of genomics in the development of novel phenotype-based products in the beef industry.** Matthew A. Cleveland\*, *Genus plc, DeForest, WI.*

The use of genomics in beef cattle evaluation has experienced relatively widespread adoption. Breed associations and improvement programs have used genomic information to varying degrees, particularly for well-characterized traits. The opportunity for transformational change from genomics, however, lies with novel traits that are typically more difficult to capture or describe during normal production processes. Genomic associations have been reported for novel traits such as those related to feed efficiency, animal health, meat quality and consumer preference, reproduction, healthfulness of beef and effects on human health, the environment and animal welfare. While several interesting genomic regions have been identified across traits, there is scant evidence of application in structured breeding programs for beef cattle. The collection of phenotypes for some novel traits can have near-term direct genetic and economic impact. For example, improvements in individual feed intake or even disease resistance can generate measurable increases in beef system profitability. Other novel traits of interest, such as fatty acid composition or methane production, have large perceived consumer impact, but remain aspirational in terms of providing a direct economic benefit or even a logical selection objective. Regardless of the trait, the optimal use of genomics in a beef improvement program requires a structured data collection system, or large project-based phenotyping efforts, to realize the benefits and deliver a truly differentiated product. Ultimately, the value derived from genomics-enabled differentiation must at a minimum cover the cost of the investment. The value of genomics in the beef industry is difficult to elucidate without some measure of actual or "virtual" integration of the value chain, which is often lacking. Commercial breeding programs, such as Genus ABS, have developed the structure to deliver well-defined genetically improved products to

the beef supply chain. At the core of this improvement is the ability to make rapid progress for economically relevant traits, which is often optimally achieved with the aid of genomic information.

**Key Words:** beef cattle, genomics, novel phenotype

### 19 Using genomics to enhance selection of novel traits in North

**American dairy cattle.** Jacques P. Chesnais\*<sup>1</sup>, Mehdi Sargolzaei<sup>1,3</sup>, Filippo Miglior<sup>2,3</sup>, and Jennie E. Pryce<sup>4</sup>, <sup>1</sup>*The Semex Alliance, Guelph, Ontario, Canada*, <sup>2</sup>*Canadian Dairy Network, Guelph, Ontario, Canada*, <sup>3</sup>*CGIL, University of Guelph, Guelph, Ontario, Canada*, <sup>4</sup>*Department of Economic Development, Jobs, Transport and Resources, Bundoora, Victoria, Australia.*

Genomics offers new opportunities for the effective selection of novel traits. For traits such as mastitis resistance, hoof health, or milk composition records based on medium infrared (MIR) data, for example, enough records are usually available to carry out genomic evaluations based on sire genotypes and the phenotypes of their daughters. For traits that are more novel or expensive to collect, such as individual feed intake or immune response, the development of a cow reference population is the most effective approach. The reliability of the resulting genomic

predictions depends primarily on the size of the reference population and on trait heritability, as shown by Daetwyler et al. (2008). To provide an empirical check of these theoretical estimates of reliability, the reliability of genomic selection was estimated for various traits using a reference population of 1,000 to 10,000 Canadian-born Holstein cows that had been genotyped with a panel of 6,000 SNP or more. All genotypes were imputed to 50K. The effects of SNP were estimated from cow records only, after excluding the dams of validation bulls. Bulls first proven in 2013 and 2014 were then used to carry out a validation and estimate the accuracy of genomic selection based on these SNP effects. Differences between accuracies obtained this way and using the Daetwyler formula are reported for traits of varying heritability and degree of indirect selection. Results confirm that large reference populations are usually required to achieve adequate accuracy. In many instances, the accuracy of genomic selection for novel traits can be increased through the use of indicator traits. Cow size and MIR data are used as examples to show how they can increase the accuracy of genomic selection for feed efficiency. Expected rates of genetic progress are calculated for each scenario, using the selection intensities and generation intervals currently realized in North American dairy cattle.

**Key Words:** novel trait, genomic evaluation, dairy cattle

# ADSA-SAD (Student Affiliate Division) Undergraduate Competition: ADSA-SAD Undergraduate Student Poster Competition

**M1 An analysis of cow cooling methods in lactating Jersey cows, while simultaneously assessing the accuracy of infrared thermography in characterizing heat stress.** Amy P. McBirney\*<sup>1</sup>, Stan L. Henderson<sup>1</sup>, and Luis A. Rodriguez<sup>2</sup>, <sup>1</sup>*California Polytechnic State University, San Luis Obispo, CA*, <sup>2</sup>*Innovative Liquids LLC, El Dorado Hills, CA*.

Cow comfort is a crucial component to maximizing dairy cattle health and productivity. Combating high temperatures is of particular importance to dairy producers located in California's Central Valley who must ensure their herd remains cool and comfortable to maintain animal wellbeing and productivity. The purpose of this project was to determine differences in heat stress based on cow cooling methods on a California Jersey dairy while also using infrared technology to measure and predict heat stress. Infrared rear udder surface temperatures, digital rectal temperatures and respiration rates were collected on 30 Jersey cows. Fifteen cows from each of 2 different high-producing mature cow pens were selected to obtain measurements during 3 different times: morning in freestalls (8–9:15 AM), mid-morning in the milk barn (9:30–11 AM), and afternoon in freestalls (1:30–2:45 PM). Management and freestall barn design were similar between pens, with the exception of differing cow cooling methods. One pen was provided with soakers only, while the other pen was provided with soakers and fans. Low to high heat stress was observed in the sample of 30 cows throughout the day. The lowest respiration rates, rectal temperatures, and udder temperatures were observed in the milking parlor during the mid-morning, particularly as cows stood in the holding pen for greater lengths of time. Rectal temperatures for cows in Pen 6 (no fans) differed significantly ( $P = 0.003$ ) from rectal temperatures measured on the same group of cows when in the milk barn. The positive response to the more frequent soaking times of the milk barn cooling system suggests this method may be more effective in reducing the effect of heat stress than either of the freestall cooling systems. Infrared body surface temperature is currently being explored as a potentially dependable and accurate means of quantifying heat stress in cattle (Jones, 1999; Berg et al., 2013). Data indicated a significant correlation ( $P = 3 \times 10^{-4}$ ) between infrared udder temperatures and digital rectal temperatures, suggesting that infrared thermography is valuable in identifying heat stress.

**Key Words:** heat stress, infrared thermography, cow comfort

**M2 Comparison of dry matter measurements between a hand-held near infrared unit and 48 hour-60°C oven drying with corn silage and alfalfa silage.** Derek M. Donnelly\*, Robb W. Bender, and David K. Combs, *University of Wisconsin-Madison, Madison, WI*.

Calculating accurate dry matter of forages is important for adjusting rations on a day-to-day basis. This study evaluated the accuracy of a hand-held near infrared (NIR) unit (Moisture Tracker, Digistar Inc., Fort Atkinson, WI) compared with conventional oven drying for 48 h at 60°C using 2 different forages: alfalfa silage and corn silage. Twenty frozen samples each of alfalfa silage and corn silage were obtained from a commercial feed testing laboratory (Rock River Labs Inc., Watertown, WI). The NIR unit was calibrated for each different forage type. Approximately 105 g of each sample was spread over an 8 cm × 8 cm area on brown paper and scanned with the NIR unit 30 times, each scan was in a different location on the sample. Upon the thirtieth measurement, the NIR unit would show the average dry matter content on its display. The

average of the 30 scans was recorded as the dry matter content of the sample. Two 100-g subsamples of each forage sample were weighed into aluminum pans. The samples were then oven-dried for 48 h at 60°C and weighed back to calculate oven dry matter. Dry matter determined by NIR and oven were compared by *t*-test within each forage type. Dry matter determined for corn silage via the NIR (39.59%) were similar ( $P = 0.557$ ) to those obtained from conventional oven drying (39.92%). Dry matter estimated from the NIR for alfalfa (48.6%) were greater ( $P < 0.0001$ ) than obtained from oven drying (44.17%). The results of this experiment showed that dry matter values were similar with oven dry matter when analyzing corn silage, but were not similar when analyzing alfalfa silage.

**Key Words:** dry matter, near infrared, dairy cattle

**M3 Evaluation of pH test strips for accuracy in determining pH of cow urine.** Colleen N. Curtiss\*<sup>1</sup>, Kayleigh G. Gratz<sup>1</sup>, David P. Chamberlin<sup>1</sup>, Timothy F. Brown<sup>2</sup>, and Elizabeth L. Karcher<sup>1</sup>, <sup>1</sup>*Michigan State University, East Lansing, MI*, <sup>2</sup>*West Central, Ralston, IA*.

Hypocalcemia is a periparturient metabolic disorder that is costly to the US dairy industry. One strategy to reduce incidences is to feed anionic supplements during the close-up period. The effectiveness of this strategy may be monitored by evaluating urine pH of cows, with an effective preventative pH in the range of 6.0 to 7.0. There are multiple commercial urine pH strips available to test the acidity of urine. The objective of this study was to evaluate several pH test strips for accuracy in determining pH of cow urine. Urine was collected from 4 mid-lactation Holstein dairy cows, composited, and distributed into ten 250-mL samples. The pH of each sample was then adjusted, using 12 N HCl and a laboratory pH meter, to 1 of 10 pH values: 5.5, 6.0, 6.25, 6.5, 6.75, 7.0, 7.25, 7.50, 7.75, and 8.0. The pH of each standard was recorded at the end of sampling and the average of the 2 values (5.68, 6.10, 6.33, 6.63, 6.81, 7.06, 7.33, 7.57, 7.76, and 8) was used to compare the accuracy of the strips. Seven evaluators assessed each pH sample with 9 commercially available pH strips to determine equivalence between the pH strip reading and the pH of the prepared standard. Confidence intervals (90%) were calculated for each pH strip and sample combination and compared with the standard. Seven of the 9 strips fell within the 90% confidence interval ranges for 1 to 3 urine samples and 1 strip was within the range for 5 samples. Of the pH strips tested, only 1 was able to identify a correct pH for 7 of the 10 urine samples. Based on our results, this strip could be used to identify urine pH in a range from 5.7 to 7.75. These results depict the variation in the accuracy of the test strips to detect urine pH. Producers must be aware of these variations as they utilize urine pH to measure the effect of feeding anionic supplements in a transition cow program.

**Key Words:** hypocalcemia, pH, urine

**M4 Rumen epithelial gene expression in response to oral NaHCO<sub>3</sub> treatment in Holstein bull calves.** Emily R. Meese\*<sup>1</sup>, Taylor T. Yohe<sup>1,2</sup>, Hannah L. M. Tucker<sup>1,2</sup>, and Kristy M. Daniels<sup>1,2</sup>, <sup>1</sup>*The Ohio State University, OARDC, Wooster, OH*, <sup>2</sup>*Virginia Tech, Blacksburg, VA*.

While common, concern exists that low rumen pH in periruminant calves should be avoided. Oral NaHCO<sub>3</sub> can increase rumen pH. The objective

was to study effects of oral administration of  $\text{NaHCO}_3$  on expression of select genes in the rumen epithelium at 49 d of age. Genes included in the investigation encode for proteins that are used for volatile fatty acid (VFA) absorption, pH regulation, and  $\text{NaHCO}_3$  transport. Neonatal Holstein bulls ( $n = 12$ ) were randomly assigned to one of 4 treatments, arranged in a  $2 \times 2$  factorial design. Factors were treatment [control (con) or  $\text{NaHCO}_3$  (bicarb)] and oral route of delivery (water-based drench or gelatin-filled capsule). Administration of  $\text{NaHCO}_3$  dose was adjusted weekly and reached a maximum of 48 g/d. Treatments were administered before each of 2 daily feedings. Calves were fed 543 g DM/d of a 22% CP, 20% milk fat replacer, had access to a 20% CP starter and water. At wk 8, rumen tissue was obtained from the cranial ventral region of each calf. Total RNA was extracted from the epithelial portion of rumen tissue, reverse transcribed into cDNA, and used in quantitative reverse transcription PCR assays. Relative abundance of 10 VFA transporters, pH regulators, anion exchangers and cotransporters were quantified. Rumen pH was not increased by  $\text{NaHCO}_3$ , (measured 2, 4, and 6 h post-dosing) but urine pH was. This observation went against past findings regarding ruminal pH with oral  $\text{NaHCO}_3$  administration but generally supports the notion that post-ruminal, rather than ruminal,  $\text{NaHCO}_3$  absorption and metabolism occurs in young calves. In support of this,  $\text{NaHCO}_3$  had no effect on expression of any ruminal gene measured here. Findings suggest that in young calves, low rumen pH is part of the developmental process and regulatory mechanisms associated with non-diffusional VFA transport, rumen pH, and  $\text{NaHCO}_3$  transport are not affected by supplemental  $\text{NaHCO}_3$ .

**Key Words:** dairy calf, rumen, gene expression

**M5 Effect of overstocking at the feed bunk on indicators of cow temperament.** Danielle N. Coleman\*, Maurice L. Eastridge, Jessica A. Pempek, and Kathryn L. Proudfoot, *The Ohio State University, Columbus, OH.*

Our objective was to investigate the effect of overstocking the feed bunk on dairy cow behavioral responses to human approach and reactivity to blood sampling. One hundred and 20 dry Holstein cows were allocated to 1 of 2 treatment groups with different stocking densities [overstocked (OS): 0.88 headlocks/cow; understocked (US): 1.17 headlocks/cow]. Over 2 testing periods (7 d apart), flight response was assessed using a human-approach test with a 5-point ordinal scale defining the distance at which the cow stepped away from the approaching experimenter (0 = not approachable from 3 m to 4 = cow moves away when experimenter is 0 m from the cow). A qualitative assessment was also made of the cow's response to the experimenter using a visual analog scale (VAS) that included the terms: relaxed, nervous, alert, shy, aggressive, social, and curious. Reactivity to blood sampling via the coccygeal vein was assessed in the pen using a 4-point scale (0 = least reactive to 3 = most reactive). Data were analyzed through a mixed model analysis, using treatment, time, and their interaction. The relationship between qualitative measures was assessed using a Pearson correlation. Treatment did not affect the cow's flight response; however, there was a significant treatment by time interaction whereby flight response scores decreased with time in OS cows and increased with time in US cows (OS: 1.65 to 1.47, US: 1.33 to 1.68;  $P = 0.02$ ). Reactivity to blood sampling did not differ by treatment (OS: 1.11, US: 0.98;  $P = 0.47$ ), and there was also no treatment by time interaction (OS: 1.17 to 1.11, US: 1.01 to 0.98;  $P = 0.88$ ). The overall correlation between qualitative terms was low. However, the terms 'relaxed' and 'nervous' showed a significant negative correlation (OS:  $r = -0.71$ ;  $P < 0.0001$ ; US:  $r = -0.61$ ;  $P < 0.0001$ ). In conclusion, overstocking the feed bunk affected the animal's response to an approaching human. Cows in the OS treatment

became less approachable over time, which may indicate fear, stress, or an increase in arousal. Future research should investigate the effect overstocking may have on cow temperament for a longer duration, as this may further decrease approachability.

**Key Words:** stocking density, feed bunk, temperament

**M6 Relationship between cow lying behavior and freestall barn design.** Jennifer Callanan\* and Amber L. Adams-Progar, *Washington State University, Pullman, WA.*

There is concern that barn design may cause underutilization of isolated freestalls and, in turn, lead to overcrowding and decreased cow comfort. The objective of this study was to observe and describe cow utilization of isolated stalls in one pen of lactating dairy cattle. One pen of 67 Holstein cows, with lactations ranging from one to 7, was selected for observations because the pen was designed to include one row of 8 stalls (isolated stalls = IS) out of a total of 72 stalls that faced the northwest concrete wall of the barn. This design allowed cows within the IS to face a wall rather than other cows. All 67 cows were randomly assigned a study number and identified with fluorescent animal marking paint. Cow attendance in the IS was observed via 2 time-lapse cameras mounted on the northeast and southwest ceilings of the barn. Time-lapse images were captured at 20-s intervals over the course of 5 d. The scan sampling method was implemented to record the number of cows within the IS, cow study numbers, and cow behavior (standing or lying). Descriptive statistics and correlations were applied to analyze the data. A correlation was detected between time of day and the percentage of cows utilizing the IS ( $r = -0.53$ ,  $P = 0.007$ ), where maximum attendance ( $4.78 \pm 0.19\%$  of cows) occurred at 0300 h and 0500 h, and minimum attendance ( $0.30 \pm 0.08\%$  of cows) occurred at 1000 h. Throughout the study, most the cows within the IS preferred to lie down ( $95.25 \pm 2.27\%$  of cows) rather than stand ( $4.75 \pm 2.27\%$  of cows). Thirty-seven percent of the cows in the pen attended the IS at least once during the study, with 7.5% of the cows accounting for 64% of the visits to the IS. No correlation was detected between cow lactation and IS utilization ( $r = 0.007$ ,  $P = 0.95$ ). These results indicate that the IS in this study were utilized primarily for lying purposes by a small percentage of cows; however, additional research is needed to determine whether cow health or social status may explain the small number of cows that visited the isolated stalls throughout this study.

**Key Words:** cow comfort, lying behavior, barn design

**M7 Effect of carbon dioxide on oxidative stability of raw milk.** Jayendra Kumar Amamcharla and Jia Xin Tang\*, *Kansas State University, Manhattan, KS.*

Sensory attributes play a critical role in the consumer acceptability of pasteurized milk. Any off-flavor present in raw milk is carried onto the pasteurized milk. One such off-flavor is oxidized flavor (OF). This can have a huge economic impact on dairy farmers as well as on milk processors. Raw milk invariably undergoes oxidative changes as soon as the milk leaves the udder. Still, the rate of OF development mainly depends on nature, concentration, and distribution of pro- and anti-oxidants in the milk system. Antioxidants in raw milk play an important role by donating a hydrogen ion to terminate auto-oxidation. In this process, the antioxidant capacity raw milk diminishes during storage. Recent studies showed that the total antioxidant capacity (TAC) of raw milk decreases by 10% after 24 h and 27% after 48 h of storage. The objective of the present work was to mitigate the reduction in TAC of raw milk by using carbon dioxide ( $\text{CO}_2$ ). It has been established that



CO<sub>2</sub> inhibits growth of psychrotrophic bacteria and extends the shelf-life of raw milk but the effect of CO<sub>2</sub> treatment on TAC of raw milk has not been established. Approximately one gallon of raw milk was divided into 4 sub-samples and 3 of the sub-samples were injected with CO<sub>2</sub> to 3 pH levels (pH 6.65, 6.55, and 6.45) and the remaining fourth subsample was used as a control. Subsequently, each of these 4 sub-samples were divided into 25 equal samples and stored at 4°C. On each experimental day, 5 equal samples were withdrawn and analyzed using a modified ferric reducing ability of plasma (FRAP) assay. Modified FRAP assay was used to determine the TAC of samples on Day 0, 1, 2, 3, and 4. The data were analyzed as repeated measures design using the MIXED procedure in SAS. The presence of CO<sub>2</sub>, storage time, and their interaction significantly ( $P < 0.05$ ) influenced the FRAP value of raw milk. It was observed that the TAC of all raw milk samples was gradually reduced during refrigerated storage. However, the drop in TAC was found to be significantly ( $P < 0.05$ ) lower in CO<sub>2</sub> treated milk. Overall, CO<sub>2</sub> treatment of raw milk shows potential in reducing TAC loss during storage of raw milk.

**Key Words:** modified FRAP assay, total antioxidant capacity, raw milk

**M8 Hepatic patatin-like phospholipase domain-containing protein 3 protein is regulated during the transition to lactation period in dairy cows.** Ryan S. Pralle\*, Rafael C. Oliveira, Tawny L. Chandler, Sandra J. Bertics, and Heather M. White, *University of Wisconsin-Madison, Madison, WI.*

Patatin-like phospholipase domain-containing protein 3 (PNPLA3) is a membrane bound triglyceride lipase that contributes to lipid metabolism in the liver and is responsive to feed restriction. PNPLA3 mRNA expression is dynamic in dairy cows with lower expression during negative energy balance. The objective of this study was to characterize PNPLA3 protein abundance in transition cows subjected to fatty liver induction. Multiparous cows were blocked by expected calving date and randomly assigned to a control ( $n = 3$ ) ad libitum intake group, or a fatty liver induction ( $n = 6$ ) group that was overfed during the dry period, and feed restricted (80% of dry matter intake) at +14 d relative to calving (DRTC) until onset of clinical ketosis. Liver biopsies were taken in the prepartum (-28, -14 DRTC), lipid accumulation (+1, +14, +28 DRTC), and recovery (+42, +56 DRTC) periods and at the time of clinical ketosis. Liver PNPLA3 protein abundance was determined through Western blot analysis and normalized to total protein. Data were analyzed using PROC MIXED of SAS 9.3. Abundance of PNPLA3 was analyzed with main effects of treatment, period, and treatment x period, and random effect of cow. Protein abundance was also analyzed by PROC MIXED and PROC CORR based on total lipid accumulation diagnosed as high (>15%, dry matter) or low (<15%, dry matter). All fatty liver induction cows became clinically ketotic and developed fatty liver. Abundance of PNPLA3 was greater ( $P = 0.02$ ) during the recovery period compared with the accumulation period (1.12 vs. 0.79  $\pm$  0.09, arbitrary units). Cows with high liver lipids had decreased ( $P < 0.01$ ) PNPLA3 abundance compared with cows with low liver lipids (0.69 vs. 1.0  $\pm$  0.08, arbitrary units). There was a negative correlation ( $P < 0.01$ ;  $r = -0.427$ ) between liver lipid concentration and PNPLA3 abundance. These data indicate that regulation of PNPLA3 may influence liver lipid accumulation and potential recovery from fatty liver disease.

**Key Words:** patatin-like phospholipase domain-containing protein 3, adiponutrin, fatty liver

**M9 Effect of management practices and housing type on somatic cell counts in Kentucky dairy farms.** Mickayla A. Myers\*, Amanda E. Stone, and Jeffrey M. Bewley, *University of Kentucky, Lexington, KY.*

The objective of this research was to compare the effects of management practices and housing types on SCC. Data were acquired for 34 Kentucky dairy herds, participating in the Southeast Quality Milk Initiative project. Herds were grouped into SCC categories where low = 2013 mean bulk tank SCC  $\leq 250,000$  cells/mL ( $n = 16$ ), and high =  $>250,000$  cells/mL ( $n = 18$ ). Herd size ranged from 42 to 660 lactating cows. Thirty-eight percent ( $n = 6$ ) and 50% ( $n = 9$ ) of low and high herds, respectively, were housed in freestalls. Lactating cow bedding of sawdust or wood shavings was most commonly used for low (47%,  $n = 7$ ) and high (53%,  $n = 9$ ) herds. Fans were provided in the parlor and housing for low (75%,  $n = 12$ ) and high (67%,  $n = 12$ ) herds. Mastitis-treated cows were not commonly housed separately from lactating cows for low (75%,  $n = 12$ ) and high (67%,  $n = 12$ ) herds. Lactating cow housing was split into 3 respective groups: total confinement, pasture access for  $<4$  h a day, and pasture access for  $\geq 4$  h a day. Total confinement was used in 50% ( $n = 8$ ) and 33% ( $n = 6$ ) of low and high herds, respectively. Pasture access for  $<4$  h a day was 6% ( $n = 1$ ) and 17% ( $n = 3$ ) for low and high, respectively. Finally, pasture access for  $\geq 4$  h a day was 38% ( $n = 6$ ) and 50% ( $n = 9$ ) for low and high, respectively. Of the herds using freestalls, manure was removed from stalls twice daily in 67% ( $n = 8$ ) and 77% ( $n = 10$ ) for low and high herds, respectively. Soakers were used in 31% ( $n = 5$ ) and 41% ( $n = 7$ ) of low and high herds, respectively. Udder hair was singed or clipped in 25% ( $n = 4$ ) and 39% ( $n = 7$ ) of low and high herds, respectively. Overall system checks of the parlor were performed annually in 31% ( $n = 5$ ) and 53% ( $n = 9$ ) of low and high herds, respectively. Results of this research may be used to help producers improve their dairy operations by understanding the management practices low and high SCC herds implement. This work was supported by a grant award from USDA-NIFA-AFRI (2013-68004-20424).

**Key Words:** low SCC, mastitis management practices, milk quality

**M10 Effect of milking procedures and mastitis detection methods on somatic cell counts for Kentucky dairy herds.** Kerri A. Bochantin\*, Amanda E. Stone, and Jeffrey M. Bewley, *University of Kentucky, Lexington, KY.*

The objective of this research was to compare the effects of milking procedures and mastitis detection methods on SCC. Data were acquired for 34 Kentucky dairy herds participating in the Southeast Quality Milk Initiative project. Herds were grouped into SCC categories where low = 2013 mean bulk tank SCC  $\leq 250,000$  cells/mL ( $n = 16$ ; L), and high  $\geq 250,000$  cells/mL ( $n = 18$ ; H). Herd size ranged from 42 to 660 cows. Fifty six percent ( $n = 10$ ) of H and 38% ( $n = 6$ ) of L herds used the California Mastitis Test (CMT) to detect clinical mastitis. Similarly, 33% of H ( $n = 6$ ) and 31% ( $n = 5$ ) of L herds used CMT to detect subclinical mastitis. All herds pre- and post-dipped ( $n = 34$ ). Pre-dip application methods included pre-dip with a non-return dip cup with commercial disinfectant (H = 94%,  $n = 17$ ; L = 80%,  $n = 12$ ), non-return dip cup with homemade disinfectant (H = 6%,  $n = 1$ ), and applied by sprayer with commercial disinfectant (L = 20%,  $n = 3$ ). Teat drying methods included multiple-use paper towels (H = 6%,  $n = 1$ ; L = 13%,  $n = 2$ ), single-use paper towels (H = 47%,  $n = 8$ ; L = 27%,  $n = 4$ ), multiple-use cloth towels (L = 20%,  $n = 3$ ), and single-use cloth towels (H = 47%,  $n = 8$ ; L = 40%,  $n = 6$ ). Pre-stripping was used in 72% of H ( $n = 13$ ) and 75% of L ( $n = 12$ ) herds. Post-dip application methods included



dip cups with commercial disinfectant (H = 94%, n = 17; L = 94%, n = 15) or homemade disinfectant (H = 6%, n = 1). Wearing latex or nitrile gloves “always” during milking was practiced in 33% of H (n = 6) and 81% of L (n = 13) herds. Producers were asked to estimate the cost of clinical mastitis and answers ranged from \$0 to \$250 (26%, n = 9), \$251 to \$500 (35%, n = 12), \$501 to \$750 (3%, n = 1), \$751 or more (6%, n = 2), or unknown (24%, n = 8). When asked to rate the pain experienced in a mild case of clinical mastitis, responses ranged from 0 to 2 (24%, n = 8), 3 to 5 (62%, n = 21), and 6 to 8 (15%, n = 5), where 1 is no pain and 10 is severe pain. Responses for rating severe clinical mastitis pain included 5 to 8 (32%, n = 11) and 9 to 10 (68%, n = 23). Results of this survey may be used to improve mastitis management in dairy cattle herds in Kentucky.

**Key Words:** mastitis, milking procedure, Kentucky

**M11 Effect of a probiotic on dairy calf diarrhea and growth rate.** Alexis C. Thompson\*, Donna M. Amaral-Phillips, and Jeffrey M. Bewley, *University of Kentucky, Lexington, KY.*

Probiotics may prevent or reduce illness occurrence. The objective of this study was to observe the effects of a probiotic (Protect, Provita Animal Health, Omagh, Northern Ireland) on calf diarrhea incidence and average daily gain. This study was conducted at the University of Kentucky Coldstream Dairy during the winter of 2014 and 2015. All female and male Holstein calves were enrolled at birth, fed a colostrum replacer (Bovine IgG Colostrum Replacer, Land O’Lakes, Shoreview, MN), and did not receive any vaccinations. Calves were housed in individual sawdust-bedded hutches with ad libitum access to water and 18% CP starter until weaning at 8 weeks of age. Calves were fed a 20:20 non-medicated milk replacer (total 0.71 kg powder daily) twice daily. Calves were randomly assigned to one of 3 treatments: 10 g liquid oral probiotics at birth (O, n = 10), 10 g liquid oral probiotics at birth plus 1 g powder probiotic once daily incorporated in morning milk from d 2 to 14 (O+P, n = 9), and a negative control (CON, n = 9). Calves were weighed once weekly using a scale (Cardinal Digital HSDC Series, Cardinal Scale Mfg. Co, Webb City, MO). Feces were scored once weekly, where 1 represented firm feces which settled once hitting the ground, 2 feces that did not hold form well, 3 feces that readily spread, and 4 feces that was liquid and splattered (Larson et al., 1977). A fecal score of 4 indicated diarrhea. The GLM and FREQ procedures of SAS Version 9.3 (SAS Institute Inc., Cary, NC) were used to analyze data. The incidence of an episode of diarrhea was 80% for O, 75% for O+P, and 88% for CON ( $P = 0.79$ ). Mean fecal scores from birth to weaning were  $2.18 \pm 0.55$  for O,  $2.18 \pm 0.35$  for O+P, and  $2.28 \pm 0.52$  for CON ( $P = 0.73$ ). Calves were confirmed with *Rotavirus* and *Coronavirus* through PCR in all treatment groups. Average daily gain during the study was  $0.47 \pm 0.10$  kg/d for O,  $0.52 \pm 0.10$  kg/d for O+P, and  $0.56 \pm 0.10$  kg/d for CON ( $P = 0.42$ ). No significant differences were observed among treatments for average daily gain, fecal score, or incidence of calf diarrhea.

**Key Words:** calf diarrhea, probiotic, growth rate

**M12 Genomic testing of female Holsteins: A resource for selection and improvement.** Kimberly M. Davenport\*, Joshua J. Peak, and Brenda M. Murdoch, *University of Idaho, Moscow, ID.*

Genomic testing is providing a powerful tool for use in dairy management decisions across the United States. While genetic testing has been more broadly implemented for sire selection, its use in selection of

females is only now being applied. The identification of females with the most promising genetic potential can influence selection decisions including replacement heifers and cull cows, as well as identify female candidates for embryo donations. Identifying the genetic potential in both male and female contributors to the herd will accelerate genetic improvement and identify potential problems of disease carrier matings before they occur. The purpose of this study was to (1) confirm pedigree information with genetic parentage, (2) identify females that may carry a disease-associated allele, and (3) compare recorded herd data of cows that have been phenotypically selected for reproduction performance to genomic predictions of the Clarifide test. Purebred Holstein cows (n = 162) were sampled by ear punch. Tissue samples were sent to Zoetis and a low density Clarifide genomic test was used to correct pedigree parentage and evaluate the genomes for several markers associated with common selection parameters. Several traits economically important to dairy herds such as milk yield, milk fat component percentage, and traits related to reproductive efficiency and productive life, were compared with results from the Clarifide genomic tests for each animal. The accuracy of parentage records utilizing the parentage correction tool was used to eliminate animal misidentification. The relative similarity of the genomic prediction to phenotypic data and parentage corrections for these traits suggest all be incorporated into future management decisions to accelerate genetic progress of the herd. The sustainability of the US dairy industry will become more dependent upon the effective implementation and strategic use of genomic selection of desirable complex phenotypic traits.

**Key Words:** genetics, dairy, selection

**M13 Effects of rubber coverings for slatted floor facilities on cattle performance and cleanliness.** Derrick S. Smith\*, Bryant R. Chapman, Colleen N. Curtiss, Monica J. Atkin, Steven R. Rust, and Dan L. Grooms, *Michigan State University, East Lansing, MI.*

Approximately 1 to 3% of steers housed on slatted floors must be removed for lack of mobility and foot problems. The objective of this study was to compare animal performance and hide soiling of cattle housed on slatted floors with (RC) and without (NC) rubber coverings. Angus-based steers (250–300 kg) were randomly assigned to pens with NC (n = 4) or RC (n = 4). Each pen contained 7 steers at a stocking density of 6.9 m<sup>2</sup>/steer. Locomotion scores, tail lesions, left carpal joint circumference, and hoof dimensions were recorded and reported in a companion abstract. Average daily gain, feed conversion efficiency, cleanliness, and carcass traits were collected and reported. Hide cleanliness was graded on a scale of 0 to 9, with 0 being less than 5% soiled and 9 being completely soiled. Cattle housed on the rubber-covered slats had more soiling of the hide ( $P < 0.05$ ) and slightly lower dressing percentages ( $P < 0.13$ ) than steers on concrete slats. Average daily gain, dry matter intake and feed conversion efficiency were similar among treatments. Cattle on NC tended to have more backfat thickness ( $P < 0.06$ ) and high yield grade scores ( $P < 0.05$ ) compared with RC. There were negative correlations between toe angles of the front ( $r = -0.86$ ;  $P = 0.006$ ) and rear ( $r = -0.72$ ;  $P = 0.04$ ) hooves to carcass adjusted feed conversion efficiency. In summary, provision of rubber covers for concrete slats had minor effects on overall cattle performance. Cattle fed on rubber-covered slats may have more soiling of the hide and lower dressing percentages.

**Key Words:** feed conversion efficiency, slatted floor, dressing percent

**M14 Expression of aryl hydrocarbon receptor in the endometrium of dairy heifers during early pregnancy and the estrous cycle.** Michelle C. Hartzell\*, Troy L. Ott, Manasi M. Kamat, and Sreelakshmi Vasudevan, *Pennsylvania State University, State College, PA.*

Early embryo loss in dairy cattle is a major economic cost to dairy producers. A dairy operation with a low herd conception rate must use more feed and resources to be as productive as a herd with a higher conception rate and shorter calving interval. Our overall hypothesis is that a portion of these early embryo losses are mediated by aberrant response of the uterine immune system to the embryo. This research seeks to improve our knowledge of a potential cellular mechanism that may be involved in maintaining pregnancy, the aryl hydrocarbon receptor (AhR). The AhR is a ligand activated transcription factor involved in differentiation of T regulatory cells (T reg). We hypothesize that activation of T reg by AhR causes suppression of the maternal immune response to the allogeneic embryo. Holstein dairy heifers (n = 22) were estrous synchronized and bred by artificial insemination (Day = 0) or remained cyclic. Uterine endometrium was collected from heifers on Day 17 of the estrous cycle (n = 9), and Days 17 (n = 9) and 20 (n = 4) of pregnancy. Total cellular RNA was isolated from endometrial tissue using Trizol. The RNA was converted to cDNA and subjected to PCR using primers designed to recognize the bovine AhR. A single amplicon was visualized by gel electrophoresis and the DNA was excised, purified, and sequenced to confirm its identity. Purified cDNA from the amplicon was used to create a standard curve and a qPCR assay was developed and validated with a slope of -3.5 and efficiency of 93%. Analysis of steady-state mRNA abundance for AhR was conducted using RPL19 as the reference gene. Critical threshold (Ct) data were adjusted for RPL19 and  $2^{-\Delta Ct}$  values were analyzed using PROC Mixed and orthogonal contrasts. Although AhR was abundantly expressed in the endometrium, there was no difference in AhR mRNA abundance between pregnant and nonpregnant heifers nor between d 17 and d 20 pregnant heifers ( $P > 0.1$ ). Studies are ongoing to localize the AhR in endometrial tissue using immunofluorescence analysis. This study confirms the presence of the AhR gene in the bovine endometrium during the estrous cycle and early pregnancy.

**Key Words:** aryl hydrocarbon receptor (AhR), embryo

**M15 Absence of milk residues after using a hydrogen peroxide pre-dip.** Jamie E. Leistner\*, Joey D. Clark, and Jeffrey M. Bewley, *University of Kentucky, Lexington, KY.*

Some pre-dips leave residues in bulk tank after milking. Hydrogen peroxide is digested by lactoperoxidase, an enzyme released from the mammary gland during milking, which would mean this teat dip would not leave residues in the milk. The objective of this study was to determine if hydrogen peroxide-based teat dip a (OxyCide, GEA, Naperville, IL) leaves a residue in the bulk tank milk after use as a pre-dip. The study was conducted using 97 Holstein cattle at the University of Kentucky Coldstream Dairy. Three samples were collected from the bulk tank 20 different times, 10 from the 3:30 a.m. and 10 from the 4:00 p.m. milking sessions; once at the beginning of milking, once during the middle, and once at the end of milking. The bulk tank is collected every other day

so the dates of sampling were determined to have 10 a.m. and 10 p.m. samples before and after the bulk tank was emptied. The pre-dip was applied to the teats using teat dip cups. Then, 236.8 mL of milk from the bulk tank were collected into the same liquid measuring cup each sample. Peroxide test strips (Em Quant, Santa Ana, CA) were used to determine if hydrogen peroxide residues were present in the sample. In all samples collected throughout the study, the test strips tested negative for hydrogen peroxide residue. This study demonstrated that the enzymes in the milk successfully digested all detectable hydrogen peroxide from the pre-dip. This study suggests hydrogen peroxide can be used as a pre-dip to prevent any risk of residue left in the milk after milking.

**Key Words:** milk residue, hydrogen peroxide, pre-dip

**M16 Effects of adding a pasteurized milk balancer on dairy calf growth and performance.** Lydia H. Hoene\*<sup>1</sup>, B. Houin<sup>2</sup>, and Tamilee Nennich<sup>1,3</sup>, <sup>1</sup>*Purdue University, West Lafayette, IN*, <sup>2</sup>*Homestead Dairy, Plymouth, IN*, <sup>3</sup>*Famo Feeds, Freeport, MN.*

Milk is one of the largest costs of raising dairy calves. Therefore, increasing growth performance and efficiency of dairy calves is very important. The objective of this study was to determine the effect of adding a protein based pasteurized milk balancer (PMB) on the growth performance of dairy calves over the summer months. A total of 90 Holstein heifers were randomly assigned at 1 d of age to 1 of 3 treatments. The treatments were (1) 7.6 L/d of pasteurized milk (CONT), (2) 7.6 L/d of pasteurized milk supplemented with a PMB (PMB), and (3) pasteurized milk supplemented with a PMB and fed at a reduced volume amount of 5.4 L/d (LESS). The calves were fed milk 3 times a day and allowed ad libitum access to 20% CP starter. Calves were weighed at birth and measured every 2 wk until 8 wk of age for body weight (BW), hip height, heart girth circumference (HGC), and hip width. Starter intakes were measured weekly. Milk intake of calves was reduced to one-third of their milk allowance at 7 wk and they were weaned at 8 wks. Data were analyzed using Proc Mean of SAS with calf as the experimental unit. At 8 wk, BW were similar ( $P = 0.18$ ) for all calves on the CONT, PMB, and LESS treatments (81.2, 84.9, 84.1 kg of BW, respectively). Average daily gain (ADG) was similar regardless of treatment and averaged 0.78 kg/d over the 8 wk. Total starter DM intake was similar for all treatments ( $P = 0.71$ ) over the 8 wk, resulting in similar overall feed efficiency among treatments ( $P = 0.53$ ) at 0.44, 0.44, and 0.41 kg of starter per kg of gain for CONT, PMB, and LESS, respectively. Hip heights at 8 wk tended ( $P = 0.10$ ) to be greater for LESS compared with CONT (97.6 and 96.0 cm, respectively). There also tended ( $P = 0.09$ ) to be a difference in HGC with PMB calves having a HGC of 104.4 cm compared with 102.3 cm for CONT, with LESS calves being intermediate at 103.4 cm. Hip widths were similar between treatments ( $P = 0.26$ ). At 8 wk of age, supplementing calves with PMB tended to increase the HGC of calves, and feeding calves a reduced amount of milk with PMB added tended to result in taller calves compared with when calves were fed only pasteurized waste milk.

**Key Words:** dairy calves, pasteurized milk balancer, waste milk

# Animal Behavior and Well-Being I

**M17 Associations between feed push-up frequency, lying and feeding behavior, and milk composition of dairy cows.** Emily K. Miller-Cushon\*<sup>1</sup> and Trevor J. DeVries<sup>2</sup>, <sup>1</sup>*Department of Animal Sciences, University of Florida, Gainesville, FL*, <sup>2</sup>*Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada*.

The objectives of this study were to assess the effects of feed push-up frequency on lying behavior and feed sorting of dairy cows, and to determine associations between behavior and milk composition. Lactating Holstein dairy cows ( $n = 30$ ; parity =  $1.9 \pm 1.1$ ; mean  $\pm$  SD) were individually housed in tie stalls, milked 2 $\times$ /d, and offered ad libitum access to water and TMR (containing on DM basis: 25% corn silage, 25% grass/alfalfa haylage, 30% high-moisture corn, and 20% protein/mineral supplement), provided 1 $\times$ /d. Cows were divided into 2 groups of 15 (balanced by DIM, milk production, and parity) and individually exposed to 2 treatments in a crossover design with 21-d periods: (1) infrequent feed push up (3 $\times$ /d), and (2) frequent feed push up (5 $\times$ /d). During the last 7 d of each period, DMI and milk production were recorded and lying behavior was monitored using electronic data loggers. During the last 2 d of each period, milk samples were collected for analysis of protein and fat content and feed samples were collected for particle size analysis. The particle size separator had 3 screens (19, 8, and 1.18 mm) and a bottom pan, resulting in 4 fractions (long, medium, short, fine). Sorting was calculated as the actual intake of each particle size fraction expressed as a percentage of the predicted intake of that fraction. Data were analyzed in multivariable mixed-effect regression models. Feed push up frequency had no effect ( $P > 0.3$ ) on lying time (11.4 h/d; SE = 0.37) or feed sorting; cows sorted ( $P < 0.001$ ) against long particles ( $78.0 \pm 2.2\%$ ; mean  $\pm$  SE) and for short ( $102.6 \pm 0.6\%$ ) and fine ( $108.4 \pm 0.9\%$ ) particles. Milk fat content increased ( $P < 0.001$ ) by 0.1% for every 10% increase in sorting for long particles and was not associated with lying behavior or other cow-level factors. Milk protein content increased ( $P < 0.003$ ) by 0.07% for every 1 h/d increase in lying time and by 0.05% for every 10% increase in sorting in favor of long particles. These results suggest that reduced lying time and sorting against long ration particles negatively affected milk composition. Further, this study did not find that altering feed push-up frequency affected feed sorting.

**Key Words:** feed sorting, lying behavior, milk

**M18 Effect of timing of feed delivery on feeding behavior and productivity of dairy cows.** Meagan T. M. King\*, Robin E. Crossley, and Trevor J. DeVries, *Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada*.

The objective of this work was to assess the effect of timing of feed delivery on behavior and productivity of cows milked 3 $\times$ /d. Twenty-four lactating Holstein dairy cows (7 primiparous and 17 multiparous), milked 3 $\times$ /d (at 1400, 2100, and 0700 h), were individually exposed to each of 2 treatments (over 21-d periods) in a replicated crossover design. Treatments were the manipulation of timing of TMR delivery, 2 $\times$ /d, in relation to milking time: (1) feeding at milking time (at 1400 and 0700 h) and (2) feeding halfway between milking times (at 1730 and 1030 h). Milk production, feeding, and rumination behavior were electronically monitored for each animal for the last 7 d of each treatment period. Milk samples were collected for 2 of the last 4 d of each period for milk component analysis. Data were then summarized across treatment period by cow and analyzed in a general linear mixed model. With a feed delay, DMI was lower (26.3 vs. 27.6 kg/d; SE = 0.6;  $P =$

0.04). Although there was no difference in feeding time (230.2 min/d), cows fed with a delay consumed their feed slower (0.12 vs. 0.13 kg DM/min; SE = 0.003;  $P < 0.001$ ) and had more meals (10.1 vs. 9.2 meals/d; SE = 0.3;  $P = 0.02$ ), which were smaller in size (2.8 vs. 3.1 kg/meal; SE = 0.1;  $P = 0.008$ ) and shorter (27.9 vs. 30.8 min/meal; SE = 1.3;  $P = 0.03$ ). Rumination time (8.7 h/d) and lying time (9.9 h/d) were similar between treatments. Cows without fresh feed at their 1400 h milking stood for less time following that milking (72.2 vs. 86.4 min; SE = 6.9;  $P = 0.045$ ), however, no differences in this latency to lie down were seen for either of the other 2 milking times. Milk yield (46.3 kg/d), milk fat content (3.65%), milk protein content (2.90%) were similar between treatments. Given the difference in DMI and no change in yield, with a feed delay, efficiency of production was improved (1.80 vs. 1.69 kg milk/kg DMI; SE = 0.04;  $P = 0.01$ ). These data suggest that moving the timing of feed delivery away from milking resulted in cows consuming their feed more slowly in smaller, more frequent meals, contributing to an improvement in efficiency of production.

**Key Words:** feed delivery, meal pattern, behavior

**M19 Relationships between dry matter intake, rumination time, lying behavior, and milk yield in lactating cows.** Dafu Shao\*, Fengxia Wang, YaJing Wang, Shengli Li, and Zhijun Cao, *State Key Laboratory of Animal Nutrition, College of Animal Science and Technology, China Agricultural University, Beijing, China*.

The objective of this study was to investigate the relationship between dry matter intake (DMI) and rumination time (RT), lying behavior (lying time and frequency), and milk yield in dairy cows. Data were collected from 32 mature Holstein dairy cows (16 primiparous, PR; 16 multiparous, MP, parity =  $1.7 \pm 0.8$ ; milk production =  $10,019 \pm 166$  kg/year; mean  $\pm$  SD). DMI was monitored using Insentec feed bins (Insentec BV, Marknesse, the Netherlands). RT was monitored electronically using HR-Tags (SCR Engineers Ltd., Netanya, Israel). Lying behavior was monitored using an activity logger (HOBO Pendant G; Onset Computer Corp., Cape Cod, MA). Data were analyzed by the R programming language (Mathsoft). According to the level of RT, cows were assigned to the low-RT group (L: RT =  $277.65 \pm 39.61$  min/d; 8 PR and 8 MP) and the high-RT group (H: RT =  $413.21 \pm 46.89$  min/d; 8 PR and 8 MP). One-way ANOVA was used to analyze the difference between H and L groups. There was no significant difference between H and L group in DMI (H:  $20.59 \pm 2.63$  kg/d; L:  $20.31 \pm 3.34$  kg/d;  $P = 0.79$ ), lying behavior (lying time, H:  $690.44 \pm 155.46$ ; L:  $773.00 \pm 163.41$ ,  $P = 0.20$ ; Lying frequency, H:  $11.25 \pm 3.14$ , L:  $10.79 \pm 3.39$ ,  $P = 0.43$ ), and milk yield (H:  $32.04 \pm 6.00$  kg/d; L:  $32.15 \pm 4.31$  kg/d;  $P = 0.95$ ). Pearson correlation was used to describe associations among DMI, RT, lying behavior (lying time and frequency) and milk yield. Multiple Linear Regression analysis was performed with lm package of the R programming language. A significant positive relationship existed between DMI and milk yield ( $P = 0.01$ ,  $r^2 = 0.93$ ). Rumination time ( $P = 0.34$ ) and lying behavior (Lying time,  $P = 0.36$ ; and Lying frequency,  $P = 0.14$ ) were not affected by DMI. Only the milk yield ( $P < 0.01$ ) was significantly affected by DMI, and the equation was  $DMI = 8.97934 + 0.35762 \times MY$ ,  $R^2 = 0.3865$ , and which MY represents milk yield. These results indicate that milk yield can be used to estimate DMI; however, RT and lying behavior are independent variables and not the suitable indicators for DMI.

**Key Words:** rumination time, lying behavior, DMI



**M20 Association between lying behavior and subclinical ketosis in transition dairy cows.** Emily I. Kaufman<sup>\*1</sup>, Stephen J. LeBlanc<sup>2</sup>, Brian W. McBride<sup>1</sup>, Todd F. Duffield<sup>2</sup>, and Trevor J. DeVries<sup>1</sup>, <sup>1</sup>*Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada*, <sup>2</sup>*Department of Population Medicine, University of Guelph, Guelph, ON, Canada*.

The objective of this study was to characterize the relationship between lying behavior and subclinical ketosis (SCK) in transition dairy cows. A total of 339 dairy cows (107 primiparous and 232 multiparous) on 4 commercial dairy farms, in Eastern Ontario, Canada, were monitored for lying behavior and SCK from 14 d before calving until 28 d after calving. Daily lying time was measured using automated data-loggers 24 h/d. A blood sample was taken from the coccygeal vein of each cow for measurement of  $\beta$ -hydroxybutyrate (BHBA) 1x/wk. Cows with BHBA  $\geq 1.2$  mmol/L postpartum were considered SCK. Cases of retained placenta, metritis, milk fever, or mastitis during the study period were also recorded. Cows were categorized into 1 of 3 groups: healthy (H) cows had no SCK or any other health issue ( $n = 139$ ); SCK (K) cows with no other health problems during transition ( $n = 97$ ); or ketotic plus (K+) cows that had SCK and one or more other health problems ( $n = 53$ ). Data were summarized by wk and analyzed in a repeated measures general linear mixed model. A 3-way interaction was found between parity, disease status, and wk ( $P < 0.001$ ); thus, first lactation and multiparous (MP) cows were analyzed separately. There was no difference among H, K and K+ ( $P = 0.5$ ) in daily lying time from 2 wk before (-2) to 4 wk after calving (+4) for first-lactation cows. In MP cows, an interaction of disease status and week was detected ( $P < 0.001$ ). Prior to calving, lying time was similar for MP cows ( $746 \pm 32.8$  min/d), decreasing ( $P < 0.001$ ) after calving to an average of  $598 \pm 10.8$  min/d. Differences in lying time for MP cows were seen in wk +1 when K+ cows spent  $92 \pm 24.0$  min/d more time ( $P < 0.001$ ) lying down than H cows, and during wk +2 where K+ cows tended ( $P = 0.07$ ) to spend  $38 \pm 21.3$  min/d more time lying down than H cows. Differences in lying time were also seen in MP cows in wk +3 and +4 where K cows spent  $44 \pm 16.7$  min/d and  $41 \pm 18.9$  min/d, respectively, more time ( $P \leq 0.03$ ) lying down than H cows. Overall, these results suggest that monitoring lying time might contribute to identifying MP cows experiencing multiple health issues, including SCK, after calving.

**Key Words:** transition cow, lying behavior, subclinical ketosis

**M21 Metritic heifers search for a safe place.** Julia C. Lomb<sup>1</sup>, Julie M. Huzzey<sup>2</sup>, Heather W. Neave<sup>\*1</sup>, Daniel M. Weary<sup>1</sup>, Bianca Costa<sup>1</sup>, and Marina A. G. von Keyserlingk<sup>1</sup>, <sup>1</sup>*University of British Columbia, Vancouver, BC, Canada*, <sup>2</sup>*California Polytechnic State University, San Luis Obispo, CA*.

Isolation from the group is a common behavioral response to illness in social animals. Modern freestall barns do not provide a refuge area for sick cows, but animals feeling ill may be able to use the lying stall itself to get away from herd mates. First-lactation cows are smaller, and thus better able to stand with all 4 feet in a stall and benefit from its protection. The objective of this study was to determine if these cows increase standing in the stall when they develop metritis, a common uterine infection. Metritis diagnosis was based on vaginal discharge, scored on every third day between calving and 21 d in milk (DIM). First-lactation cows identified as metritic (M,  $n = 8$ ) were paired with healthy individuals (H,  $n = 8$ ), based on body weight and days in milk. Cows were housed in a dynamic, mixed-parity group of 20 and had access to 24 lying stalls. Cameras installed above the experimental pen allowed for 24 h of continuous observation. Time spent standing with either the 2 front feet or all 4 feet in the stall was measured using 5-min

scan sampling of the video recordings. The observation period included the 3 d before diagnosis in the metritic animals and the corresponding DIM for healthy pairs. A Mixed procedure in SAS (SAS Institute Inc., Cary, NC) was used for statistical analysis. Time standing with 4 feet in the stall was normalized using a log-transformation; values reported are back-transformed geometric means and 95% CI. Metritic heifers spent more time standing with all 4 feet in the stall ( $102.5$ , 49–214 min/d; mean, 95% CI) than the paired healthy heifers ( $10.5$ , 5–22 min/d; mean, 95% CI;  $P < 0.001$ ). Differences in time spent perching with the 2 front feet in the stall were not significant (M:  $209.6 \pm 31$ ; H:  $284 \pm 31$ ; LS means  $\pm$  SE;  $P > 0.1$ ). There was no significant difference in overall time spent standing in the stall. We suggest that cows experiencing malaise with the onset of metritis spend more time standing with 4 feet in the stall because they perceive this to be a protective environment.

**Key Words:** sickness behavior, disease, standing behavior

**M22 Feeding and social behaviors change prior to metritis diagnosis in transition dairy cows.** Heather W. Neave<sup>\*</sup>, Julia C. Lomb, Julie M. Huzzey, Daniel M. Weary, and Marina A. G. von Keyserlingk, *University of British Columbia, Vancouver, BC, Canada*.

Metritis is common in the days after calving and can reduce milk production and reproductive performance. Early identification of metritic animals may improve the welfare of affected dairy cows and the economic viability of the farm. The aim of this study was to identify feeding and social behaviors that could be used for the early detection of metritis. Healthy Holstein cows were enrolled in the study 3 wk before calving and all behaviors were recorded using an electronic feeding system. Metritis was diagnosed based on condition of vaginal discharge assessed on d 6 after calving. Twenty-one primiparous and 12 multiparous cows were diagnosed with metritis (more than 50% pus, or watery and red-brown in color with putrid smell) with no other health conditions; these were compared with 49 healthy primiparous and 96 healthy multiparous cows. In the 5 d leading up to clinical diagnosis, metritic primiparous cows ate less (metritic:  $13.0 \pm 0.42$  kg DM/d; healthy:  $14.6 \pm 0.27$  kg DM/d), spent less time eating (metritic:  $137.7 \pm 5.9$  min/d; healthy:  $152.1 \pm 3.8$  min/d) and had fewer visits to the feed bins per day (metritic:  $53.8 \pm 3.1$ ; healthy:  $65.5 \pm 2.0$ ) compared with healthy primiparous cows; healthy and metritic multiparous cows did not differ in these measures. No differences were found between healthy and metritic primiparous or multiparous cows in feeding rate and number of meals per day. Metritic multiparous cows were more likely than healthy multiparous cows to be competitively replaced at a feed bin by another cow (proportion of total feed bin visits where cow was replaced; metritic:  $0.25 \pm 0.02$  vs. healthy:  $0.20 \pm 0.01$ ). We conclude that cows at risk for metritis can be identified by changes in feeding and social behavior in the days before diagnosis.

**Key Words:** sickness behavior, disease, intake

**M23 LPS injection in pregnant ewes and the number of lambs born affect maternal behavior and the time to first suckling.** Cristiane G. Titto<sup>\*1</sup>, Fábio L. Henrique<sup>1</sup>, Evaldo A. L. Titto<sup>1</sup>, Adroaldo J. Zanella<sup>2</sup>, Henrique B. Hooper<sup>1</sup>, Lina F. P. Rodriguez<sup>1</sup>, Ana Luisa S. Longo<sup>1</sup>, Thays M. C. Leme-dos Santos<sup>1</sup>, Raquel F. Calviello<sup>1</sup>, Jessica C. Veronezi<sup>1</sup>, and Alfredo M. F. Pereira<sup>3</sup>, <sup>1</sup>*Faculdade de Zootecnia e Engenharia de Alimentos, Universidade de São Paulo, Pirassununga, São Paulo, Brazil*, <sup>2</sup>*Faculdade de Medicina Veterinária e Zootecnia, Universidade de São Paulo, Pirassununga, São Paulo*,

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Stress during gestation and multiple births can alter postpartum maternal care in sheep. The objective of this study was to determine the effect of LPS injection during the second and last third of gestation and the number of lambs born per ewe on maternal behavior and time to first suckling. Pregnant multiparous crossbred Santa Ines ewes were challenged with an intravenous administration of 0.8 µg·kg<sup>-1</sup> of lipopolysaccharide *E. coli*, or saline. Forty-two ewes were placed in individual pens with full open view of other animals, inside of a barn, during an adaptation period of 5 d. Ewes had free access to water, shade and food. Ewes were divided into 3 groups: CG - control group, (n = 14) that were not subjected to LPS injection; SG – stress at second third of gestation (70 d; n = 14); LG – LPS injection during the last third of their gestation period (120 d; n = 14). Behavior was recorded as positive (facilitated and encouraged suckling) and negative maternal behavior (aggressive, lack of co-operation with the lamb's sucking attempts) using 1-min scan sampling interval. Data were recorded from the time of birth until the first effective suckling event. If there were multiple births (2 lambs), each lamb was assessed separately. Data were analyzed as a completely randomized design using MIXED/SAS, and the ANOVA model had gestation LPS injection and the number of lambs born as the fixed effects as their interaction. Differences between LPS injection periods x birth were significant at a  $P < 0.05$ . Mother-offspring positive interactions were observed more than negative interactions ( $P < 0.05$ ), except for the LG with multiple births, where the opposite occurred. Negative interactions were only observed in ewes with multiple births ( $P < 0.05$ ). Lambs born from ewes in SG and LG groups, in twin births, spent more time until the first suckling ( $P < 0.05$ ). It is likely that the delay in colostrum intake decreases the immunological capacity of the lamb, and the success of the ewe and lamb bond. LPS injection, which mimics a disease process, in late pregnancy is more harmful to the newborn lamb increasing negative maternal behavior and the time to first suckling.

**Key Words:** LPS, sheep, welfare

**M24 Effect of increasing levels of babassu flour starch on feeding behavior of feedlot lambs.** Michelle de Oliveira Maia Parente<sup>1</sup>, Osman José de Aguiar Gerude Neto<sup>1</sup>, Paull Andrews Carvalho Santos<sup>1</sup>, Henrique Nunes Parente<sup>1</sup>, Miguel Arcaño Moreira Filho<sup>1</sup>, Ruan Mourão da Silva Gomes\*<sup>1</sup>, Itamara Gomes França<sup>1</sup>, Arnaud Azevedo Alves<sup>2</sup>, and Valdi Lima Júnior<sup>3</sup>, <sup>1</sup>Universidade Federal do Maranhão, Chapadinha, Maranhão, Brazil, <sup>2</sup>Universidade Federal do Piauí, Teresina, Piauí, Brazil, <sup>3</sup>Universidade Federal do Rio Grande do Norte, Natal, Rio Grande do Norte, Brazil.

Babassu (*Orbignya phalerata* Mart.) is a palm tree native to Brazil's northeast and north states that appears between Cerrado and the Amazon rain forest. Usually, each tree produces 15 to 25 bunches of fruit, each fruit weighting 98 to 280 g. The average weights of each component of the babassu coconut are 11% exocarp, 23% mesocarp, 59% endocarp and 7% kernels. From mesocarp can be extracted a flour, whose composition is 60% starch. The use of babassu flour starch can be used as an energy source for ruminants, however because of composition, it is important to study the feeding behavior. Twenty crossbred lambs (initial BW of 21.6 ± 3.5 kg and 120 d old) were used in a randomized complete block design according to initial BW and age. Lambs were penned individually during 50 d and fed an isonitrogenous diet (16.5 ± 0.2 CP, DM basis) containing 70% concentrate and 30% coastcross hay. Increasing levels of BSF were 0, 10, 20 and 30%, DM basis, corresponding to the experimental diets BFS0, BFS10, BFS20 and BFS30, respectively. Animals were monitored

every 5 min during 24 h, on the 25th day of the experiment, according to the activities: eating, rumination, idle and other activities. The feed and rumination efficiencies, expressed as g DM/hour were obtained by dividing the average daily intake of DM by the total time spent eating and ruminating in 24 h, respectively. Orthogonal polynomials for diet responses were determined by linear and quadratic effects. Effects were declared significant at  $P \leq 0.05$ . There was a quadratic response ( $P \leq 0.05$ ) for time spent in eating (164.0, 231.0, 270.0 and 250.0 min/d for BFS0, BFS10, BFS20 and BFS30), time spent in other activities (289.0, 206.0, 179.0 and 168.0 min/d for BFS0, BFS10, BFS20 and BFS30) and eating efficiency on DM (402.9, 331.6, 234.5 and 180.7 g DM/hour for BFS0, BFS10, BFS20 and BFS30). The increasing levels of BFS did not affect ( $P \geq 0.05$ ) the time spent in rumination, rumination efficiency and time spent in idle. It is concluded that increasing BFS levels changed the feeding behavior of lambs, especially for time spent for eating and feed efficiency.

**Key Words:** feed efficiency, mesocarp, rumination

**M25 Residual feed intake selection: Effect on gilt behavior in response to a lipopolysaccharide challenge.** Samaneh Azarparajouh\*<sup>1</sup>, Jessica Colpoys<sup>1</sup>, Anoosh Rakhshandeh<sup>1,2</sup>, Jack Dekkers<sup>1</sup>, Caitlyn Abell<sup>3</sup>, Nicholas Gabler<sup>1</sup>, and Anna Johnson<sup>1</sup>, <sup>1</sup>Department of Animal Science, Iowa State University, Ames, IA, <sup>2</sup>Department of Animal and Food Sciences, Texas Tech University, Lubbock, TX, <sup>3</sup>DNA Genetics, Columbus, NE.

Increasing feed efficiency in swine is important for increasing sustainable food production and profitability for producers. However, it is unknown if selection for improved feed efficiency impacts the expression of sickness behavior. The objective of this study was to characterize gilt behaviors and postures when challenged with lipopolysaccharide (LPS). This work was conducted with 7 low residual feed intake (LRFI; more feed efficient) and 8 high RFI (HRFI; less feed efficient) gilts (63 ± 4 kg BW) from the 8th generation of the ISU Yorkshire RFI selection lines. All gilts were individually housed in metabolism crates. Gilts were challenged I.M. with 30 µg/kg BW *Escherichia coli* O5:B55 LPS at 10:00 ± 1 h. Gilts were video recorded one day before LPS challenge (baseline) and on the treatment day (LPS challenge). Video was analyzed using a 1-min scan sample interval at 2 time points; 1) for 2 h starting at the time of treatment injection and 2) for one hour starting at the evening feeding time (~17:00h). Standing, sitting, lying, eating, and drinking were recorded. Data were analyzed using the GLIMMIX procedure of SAS. The model included line, treatment, time, and the interaction, with a random effect of pig nested within replicate. There was no line by treatment interaction for behaviors and postures ( $P \geq 0.32$ ). There were no selection line behavioral and postural differences in response to the LPS challenge ( $P \geq 0.45$ ). Regardless of selection line, after the LPS challenge gilts laid more ( $P < 0.0001$ ) and stood less ( $P < 0.0001$ ). For the other behaviors and postures there were no treatment differences ( $P \geq 0.16$ ). In conclusion regardless of divergent selection for RFI, the LPS challenge affected lying and standing behavior in gilts in the same way. This project was supported by USDA-AFRI Grant no. 2011-68004-30336.

**Key Words:** gilt, lipopolysaccharide challenge, residual feed intake



**M26 Effects of light programs and tryptophan supplementation on stress indicator parameters and growth of weaned piglets.**

Lívea Maria Gomes<sup>\*1</sup>, Gabriela de Mello Miassi<sup>1</sup>, Luan Sousa Santos<sup>2</sup>, Marcos Livio Panhoza Tse<sup>1</sup>, and Dirlei Antonio Berto<sup>1</sup>, <sup>1</sup>Faculdade de Medicina Veterinária e Zootecnia – FMVZ/UNESP, Botucatu, SP, Brazil, <sup>2</sup>Faculdade de Ciências Agrárias e Veterinárias – FCAV/UNESP, Jaboticabal, SP, Brazil.

Stressors such as weaning, social structure and environmental changes reduce piglet performance. Tryptophan (Trp) as a precursor of cerebral 5-hydroxytryptamine (5HT, serotonin) and melatonin may mitigate stress of animals. The photoperiod controls the transformation of Trp in its metabolites which in turn influence the consumption and welfare of piglets. This study aimed to evaluate the effects of light program and Trp levels on growth performance, blood glucose and plasma cortisol profile of piglets. Seventy-two 21-d-old piglets (initial BW = 6.6 ± 2.33 kg) were allotted to 4 treatments stratified by sex and initial BW with 6 pens/treatment and 3 piglets/pen. Data were analyzed as randomized complete block design in a 2 × 2 factorial arrangement (light programs: 12 or 23 h of light/d, and 2 digestible Trp levels: 2.6 or 5.2 g of L-Trp/kg of diet in pre-starter 1 from d 1 to d 14, and 2.4 or 4.8 g of L-Trp/kg of diet in pre-starter 2 from d 15 to d 24). The trial was carried out in a room with controlled temperature (26 and 23°C during the first 14 and during the last 10 d, respectively). Blood glucose was determined on d 0 and 24, and plasma cortisol on d 0 and 8. The baseline glucose and cortisol concentrations were used as a covariate adjustment in all analyses. No effects of light program and Trp levels interaction were observed on performance and blood glucose. From d 0 to 14 and 0 to 24, treatments did not influence ADG, ADFI and glucose, while G:F decreased ( $P < 0.05$ ) from d 0 to 14 for pigs receiving 12 h of light/d compared with the ones receiving 23 h of light/d, but improvement was offset in the total period. Light program and Trp levels interaction was observed for plasma cortisol. Pigs receiving 23 h of light/d presented higher plasma cortisol ( $P < 0.05$ ) than piglets receiving 12 h of light/d when fed diet with 2.6 g L-Trp/kg, however there was no difference on plasma cortisol ( $P > 0.05$ ) in both light programs for pigs fed 5.2 g L-Trp/kg. In conclusion, higher levels of dietary Trp are recommended for piglets receiving light program of 23 h.

**Key Words:** cortisol, nursery pigs, photoperiod

**M27 The effects of ethyl alcohol as a tool for pain management in neonatal pigs during castration.**

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Castration without anesthesia is known to be a stressful procedure in neonatal pigs. The objective of this study was to examine the analgesic effects of ethyl alcohol (ETOH) during castration. Fifty boars across 14 litters (n = 14) were randomly assigned in a completely randomized design to 1 of 4 injection treatments: ETOH, Lidocaine, saline, or sham. One gilt from each litter was also selected as a handling control. At 3 days of age (doa), 1 ml of the respective treatment was injected into each testicle, and boars were castrated at 14 doa. Behaviors related to social cohesion, pain, and non-specific actions were continuously recorded in pens and during treatment injection and castration. Social cohesion behaviors identified how individuals acted in relation to the litter, pain-related behaviors quantified behaviors associated with pain, and non-specific behaviors signified general behaviors not associated

with pain. Body weight (BW) and testicular wound scores were recorded daily post-injection and post-castration. All data were analyzed with the use of a mixed effects model with treatment as a fixed effect and pen as a random effect. The frequency of pain-related behaviors post-injection was significantly increased in the ETOH and Lidocaine groups ( $P < 0.05$ ), but these groups had lower behavioral frequencies than the saline and sham treatments post-castration ( $P < 0.05$ ). There were no significant differences in the frequency of non-specific behaviors after injection ( $P = 0.252$ ) or castration ( $P = 0.456$ ). The frequency of social cohesion behaviors did not differ across groups post-injection, but all groups had significantly greater frequencies than gilts post-castration ( $P < 0.05$ ). Pigs treated with ETOH had increased wound scores post-injection versus all groups ( $P < 0.05$ ). Wound healing scores did not differ across treatments post-castration ( $P = 0.37$ ) and BW did not differ across treatments overall ( $P = 0.40$ ). Pigs treated with ETOH behaved similarly to Lidocaine-treated pigs, but further analyses are needed to determine the effectiveness of ETOH as a practical solution to address the pain and welfare concerns of castration in pigs.

**Key Words:** pig, castration, pain

**M28 Effect of surgical and band castration on indicators of chronic pain in 0-, 2-, and 4-month-old beef calves.**

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Three experiments were conducted to evaluate the effects of band and surgical castration on behavioral and physiological indicators of pain in beef calves at 3 different ages (36 calves/age group): newborn (Exp. 1, 4 ± 1.15 d of age, 43 ± 1.13 kg BW); 2 mo of age (Exp. 2, 63 ± 2.35 d of age, 92 ± 1.75 kg BW); or 4 mo of age (Exp. 3, 125 ± 4.65 d of age, 160 ± 3.36 kg BW). In each experiment calves were randomly assigned to sham (CT), band (BA) or surgical (SU) castration. Exp. 1, 2 and 3 ended when the testicles of banded calves had sloughed off (68, 49, and 42 d, respectively). Animal BW and rectal temperature were recorded weekly over the experimental period. Salivary cortisol, scrotal area temperature using infrared thermography, visual evaluation of swelling (5-point scale), and gait stride length was collected on d 1 and immediately before castration, and weekly thereafter until the end of the study. Hair samples were collected 1 d before and 28 d after castration, and at the end of the study for cortisol concentration. Standing and lying behavior was recorded over a 28-d period immediately after castration. At 4 mo of age, salivary cortisol concentration, scrotal area temperature, and stride length were greater ( $P \leq 0.05$ ) in BA compared with CT calves, and lying time was lower ( $P \leq 0.001$ ) in SU than CT calves. At 2 and 4 mo, hair cortisol tended to be greater ( $P \leq 0.07$ ) in BA and SU compared with CT calves. The maximum degree of swelling ( $P < 0.05$ ) caused by BA and SU was 1.5 at age 0; 3 and 3.5, respectively, at 2 mo; and 4.5 and 4, respectively, at 4 mo. At 2 and 4 mo of age swelling was observed between d 7 and 14 in SU, and between d 21 and 28 in BA. Newborn calves had maximum swelling between d 35 and d 42 in BA and between d 14 and d 21 for SU. Overall, band castration resulted in more chronic indicators of pain than surgical and sham methods. However, as calves got older, indicators of pain increased sequentially in band castrated calves.

**Key Words:** chronic pain, castration, beef calves

**M29 Characteristics of agonistic behavior of commercially housed pigs after mixing.** Shin-Jae Rhim\*<sup>1</sup>, Hyun-Su Hwang<sup>1</sup>, Seung-Hun Son<sup>1</sup>, Hojeong Kang<sup>1</sup>, and Joon-Ki Hong<sup>2</sup>, <sup>1</sup>*Chung-Ang University, Ansung, Gyeonggi, South Korea*, <sup>2</sup>*National Institute of Animal Science, Cheonan, Chungnam, South Korea*.

This study was conducted to compare the aggressive behaviors of commercially housed pigs when mixed at different times after weaning. The behavioral patterns of 36 groups of pigs (a total of 360 animals) were observed by continuous monitoring over 3 consecutive days directly after weaning ( $25 \pm 1.2$  d of age), and 50 and 75 d with the aid of video technology. The pigs were not mixed previously and were same ages. The data were analyzed by ANOVA, Tukey's post hoc tests, and Wilcoxon rank-sum test using the SAS software. Fight latency and total duration and frequency of fighting were significantly different among the age groups. The agonistic behaviors decreased in 75-d old pigs if compared with 25- and 50-d-old animals. Moreover, dominance index (DI, DI was defined as the sum of wins minus defeats divided by the sum of wins, defeats, and stand-off outcomes) was higher in 25-d-old and lower in 75-d-old pigs. A comparison of dominant (DI >0) and submissive (DI <0) pigs showed significant differences ( $P < 0.05$ ) for major aggressive behaviors in all age groups. Higher frequency and longer duration of aggressive interactions mean that dominant animals were more active than the submissive ones. Moreover, pigs with a positive dominance index initiated more fights ( $P < 0.05$ ). Dominant pigs were involved in more aggressive interactions, had longer fights, and initiated more fights than submissive pigs. Moreover, older pigs have shorter fights after mixing and that they also sustain fewer injuries from these fights. Post-mixing aggressive behavior changed over time. Early experience of mixing in the rearing conditions might contribute to reduced aggressive behavior of growing pigs. It may have been occurred by more energetically efficiency strategy for stable social structure. This finding has potentially important consequences for animal welfare and economic production of commercial swine production.

**Key Words:** agonistic behavior, dominance index, interaction

**M30 Determining feeder space requirement for growing-finishing pigs.** Yuzhi Li\*<sup>1</sup>, Kimberly McDonalds<sup>2</sup>, and Harold Gonyou<sup>2</sup>, <sup>1</sup>*West Central Research and Outreach Center, University of Minnesota, Morris, MN*, <sup>2</sup>*Prairie Swine Center Inc., Saskatoon, SK, Canada*.

A study was conducted to determine feeder space requirement for growing and finishing pigs based on the amount of time needed to maintain their feed intake and growth performance. The first trial used 16 pens of 12 pigs. Four pens were randomly assigned to each of 4 treatment combinations: mash diets fed from a dry feeder (DM), mash diets fed from a wet/dry feeder (WM), pelleted diets fed from a dry feeder (DP) and pelleted diets fed from a wet/dry feeder (WP). Both the dry and wet/dry feeder provided single feeding space and were similar in design. Eating behavior was video-recorded for 24 h in all pens when pigs were 35 to 45 kg and 90 to 100 kg, respectively. The videos were analyzed using scan-sampling at 5-min intervals to determine time spent eating for each size of pigs. In the second trial, 564 pigs in 32 pens were used to evaluate 4 levels (80%, 95%, 110%, and 125%) of feeder capacity for growing and finishing pigs, with 2 pens assigned to each level of feeder capacity under the same treatments. Feeder capacity was defined 100% when the feeder was expected to be used 100% of the time by pigs according to the eating behavioral data from trial one. The different levels of feeder capacity were generated by varying the number of

pigs in a pen and modifying the pen size accordingly to maintain floor space allowance consistent cross treatments. Pigs fed WM spent less time eating (72 vs. 107 min/pig/d for growers, SE = 2.68;  $P < 0.001$ ) and 67 vs. 106 min/pig/d for finishers, SE = 3.39;  $P < 0.001$ ) compared with pigs fed DM. Likewise, pigs fed DP spent less time eating than pigs fed DM ( $P < 0.001$ ) during both growing and finishing phases. There was no difference in total duration of eating between pigs fed DP and WP. Cross treatments, ADG decreased from 793 to 693 g during the grower phase (SE = 9;  $P < 0.001$ ) and from 941 to 667 g (SE = 27;  $P < 0.001$ ) during the finishing phase when feeder capacity increased from 80 to 125%, respectively. To maintain 80% feeder capacity, the estimated feeder requirement is 11 pigs per feeder space for pigs fed DM during both growing and finishing phases. For pigs fed WM, DP or WP diets, the estimated feeder requirement is between 14 and 16 pigs for the growing phase, and between 17 and 18 pigs for the finishing phase.

**Key Words:** eating behavior, feeder space, pig

**M31 Effects of different number of animals relative to a single feeding space on performance and behavior in Holstein bulls fed high-concentrate diets.** Maria Devant\*<sup>1</sup>, Alex Bach<sup>2,1</sup>, and Marçal Verdú<sup>1</sup>, <sup>1</sup>*IRTA-Ruminant Production, Animal Nutrition, Management, and Welfare Research Group, Caldes Montbui, Spain*, <sup>2</sup>*ICREA, Barcelona, Spain*.

One hundred and eight Holstein bulls ( $155 \pm 2.0$  kg BW and  $121 \pm 0.8$  d age) were randomly allocated to one of 6 pens and assigned either in groups of 16 (Ratio16) or in groups of 20 (Ratio20) to a single space feeder with lateral protections. Each pen (6 m × 12 m) had also 1 straw feeder, and 1 drinker. Concentrate intake was recorded daily, straw consumption weekly, and BW fortnightly (total 15 periods). Animal behavior was registered every 28 d by scan sampling. Animals were slaughtered after 219 d and HCW and carcass quality recorded. Data were analyzed using a mixed-effects model with repeated measures. Mean concentrate intake ( $6.77 \pm 0.168$  kg/d), concentrate efficiency ( $0.22 \pm 0.016$  kg/kg), and carcass weight ( $271 \pm 2.2$  kg) were not affected by treatments. An interaction between treatment and time was observed in most of eating behavior parameters. During the growing (period 1 to 11) in Ratio16 bulls the number of visits to the feeder tended ( $P = 0.08$ ) to increase, total daily time devoted to eat was greater ( $P = 0.01$ ), whereas meal size ( $P = 0.05$ ), meal duration ( $P = 0.001$ ), and eating rate (period 5 to 11;  $P = 0.001$ ) were lesser compared with Ratio20 bulls. However, at the end of the fattening period (periods 12 to 15) no differences between treatments were observed. In Ratio16, the percentage of animals drinking was greater ( $P = 0.05$ ) compared with Ratio20. Moreover, an interaction between treatment and time was observed in the percentage of animals performing self-grooming ( $P < 0.05$ ) and attempting to mount ( $P = 0.10$ ). In conclusion, when the ratio of number of animals to a single feeder was reduced by 20%, the number of visits tended to increase, animals had shorter meals and meal sizes, and eating rate decreased; however, these differences were only observed during the growing period. These data may suggest that feeding space to animal ratio has an effect on eating behavior during the growing period, where social facilitation may be important. However, at the finishing as eating behavior becomes more individualized and/or animals learn to take turns, the feeding space to animal ratio is less critical.

**Key Words:** bulls, feeding space to animal ratio, eating behavior

**M32 Effects of stocking density and source of forage fiber on short-term behavioral and lactational responses of Holstein dairy cows.** Mackenzie A. Campbell<sup>1,2</sup>, Kurt W. Cotanch<sup>1</sup>, Catherine S. Ballard<sup>1</sup>, Heather M. Dann<sup>1</sup>, Dave M. Barbano<sup>3</sup>, Alyssa M. Couse<sup>1</sup>, and Richard J. Grant<sup>1</sup>, <sup>1</sup>William H. Miner Agricultural Research Institute, Chazy, NY, <sup>2</sup>The University of Vermont, Department of Animal Science, Burlington, VT, <sup>3</sup>Cornell University, Ithaca, NY.

Understanding the interaction of stocking density and diet is vital for the improvement of dairy cow well-being and productivity. Multiparous (n = 48) and primiparous (n = 20) Holstein cows were assigned to 1 of 4 pens (n = 17 cows/pen, based on previous response variability) to determine the short-term effects of stocking density and source of forage fiber on behavior and lactational performance. Pens were assigned to treatments in a 4 × 4 Latin square with 14-d periods using a 2 × 2 factorial arrangement. Two stocking densities (STKD; 100 or 142% of stalls and headlocks) and 2 diets (straw; S and no straw; NS) resulted in 4 treatments: 1) 100NS, 2) 100S, 3) 142NS, and 4) 142S. Dietary forage content consisted of 39.7% corn silage and 6.9% haycrop silage versus 39.7% corn silage, 2.3% haycrop silage, and 3.5% chopped straw (dry matter; DM basis) for NS and S, respectively. Both diets were formulated for 16% crude protein, 28% neutral detergent fiber (NDF), and 28% starch (DM basis). Alterations in forage fiber source resulted in physically effective NDF values of 18.8% and 20.1% for NS and S, respectively. Pen intake and milk yield were measured on d 8–14 of each period. Time spent feeding, ruminating, and lying were measured using 72-h direct observation on d 8–10 of each period. Milk component yields were quantified for 6 milkings on d 13 and 14 of each period. Data were analyzed using the MIXED procedure in SAS with pen as the experiment unit. Eating time (238 min/d, SEM = 4), rumination time (493 min/d, SEM = 9), and pen intake (25.3 kg/cow/d, SEM = 0.4) did not differ ( $P > 0.10$ ) among treatments. Milk, true protein and lactose yields tended to decrease with S. Although higher STKD decreased lying time, diet had no effect. The dietary addition of straw had minimal effects on short-term production and behavioral responses at varying STKD.

**Table 1 (Abstr. M32).** Production of Holstein cows assigned to 2 stocking densities (STKD; 100 or 142% of stalls and headlocks) and 2 diets (straw; S and no straw; NS)

Item	100% STKD		142% STKD		SEM	P-value		
	NS	S	NS	S		STKD	Diet	STKD × Diet
Milk, kg/d	41.2	40.4	40.7	40.0	0.7	0.21	0.06	0.79
Fat, kg/d	1.29	1.30	1.30	1.25	0.02	0.48	0.12	0.08
True protein, kg/d	1.02	1.01	1.02	0.99	0.01	0.25	0.07	0.42
Lactose, kg/d	1.41	1.39	1.40	1.37	0.04	0.25	0.08	0.61
Lying, min/d	832	827	779	796	11	<0.01	0.56	0.31

**Key Words:** physically effective fiber, overcrowding

**M33 Effect of temperament on feedlot performance and carcass traits in purebred and crossbred Nellore cattle.** Aline C. Sant'Anna<sup>1</sup>, Fernanda M. Benez<sup>2</sup>, Janaina S. Braga<sup>2</sup>, Arquimedes J. R. Pellechia<sup>2</sup>, and Mateus J. R. Paranhos da Costa<sup>1</sup>, <sup>1</sup>São Paulo State University, Department of Animal Science, Faculty of Agricultural

and Veterinarian Sciences, Jaboticabal, São Paulo, Brazil, <sup>2</sup>São Paulo State University, Post Graduate Program in Animal Science, Faculty of Agricultural and Veterinarian Sciences, Jaboticabal, São Paulo, Brazil.

The aim of this study was to assess the effect of temperament on feedlot performance and carcass traits in Nellore and crossbred cattle (F<sub>1</sub> Angus × Nellore and Caracu × Nellore). A total of 450 bulls raised on pasture and finished in feedlot were kept in outdoor feedlot pens with 12 m<sup>2</sup>, during 87 d of confinement. Cattle temperament was assessed using the crush test (REA, recording the movements and tension inside the crush in a 7-point scale); and the flight speed test (FS, m/s), measuring the speed at which the animals exited the crush, measured in the beginning (d 0, initial) and in the end of the feedlot period (final). Performance and carcass traits used were: final BW, ADG, hot carcass weight (HCW), carcass yielding (CY), subcutaneous fat score (SFS), number of bruises in the carcass (BRU) and ultimate meat pH (pH<sub>24h</sub>). Linear mixed models were fitted to assess the effects FS and REA on dependent traits. Final FS affected ( $P < 0.05$ ) BW and ADG, with an estimated reduction of 7.92 kg in BW and 0.13 kg/d in ADG for an additional unity in FS (Table 1). Final FS had significant effects on most of the carcass traits assessed, resulting in reduction of CW ( $P < 0.05$ ), SFS ( $P < 0.01$ ), and increased BRU ( $P < 0.05$ ) and meat pH<sub>24h</sub> ( $P > 0.05$ ). Final REA affected only CY ( $P < 0.05$ ). Initial FS and REA had less pronounced effects on performance traits, with initial FS effecting only BRU ( $P < 0.01$ ), and initial REA affecting ADG ( $P < 0.05$ ). We conclude that more excitable temperament (faster FS) may have negative effects on cattle feedlot performance, carcass traits and meat pH<sub>24h</sub>. The FS assessed at the end of feedlot period was the best predictor of the detrimental effects of temperament on performance of Nellore and crossed cattle. Financial support: FAPESP (2013/20036–0).

**Table 1 (Abstr. M33).** Effects of initial and final flight speed (FS) and reactivity score (REA) on feedlot performance and carcass traits, expressed as regression slopes (β)

Trait	β FS initial	β REA initial	β FS final	β REA final
Final BW, kg	-4.58	-1.12	-7.92*	-0.91
ADG, kg/d	0.00	-0.05*	-0.13*	0.00
Carcass weight, kg	-1.70	-0.31	-6.23*	-1.52
Carcass yielding, %	0.23	-0.12	-0.10	-0.28*
Subcutaneous fat score	-0.29	-0.02	-0.66†	0.07
Bruises	0.25†	-0.01	0.22*	0.02
pH <sub>24h</sub>	0.00	0.00	0.02*	0.00

\* $P < 0.05$ ; † $P < 0.01$ .

**Key Words:** flight speed, reactivity, beef cattle

**M34 Agitated temperament related to worse carcass quality in feedlot cattle.** Désirée Ribeiro Soares<sup>1</sup>, Karen S. Schwartzkopf-Genswein<sup>2</sup>, Joslaine N. Dos Santos Gonçalves Cyrillo<sup>3</sup>, and Mateus J. Rodrigues Paranhos da Costa<sup>1</sup>, <sup>1</sup>Faculdade de Ciências Agrárias e Veterinárias, UNESP, Jaboticabal, São Paulo, Brasil, <sup>2</sup>Agriculture and Agri-Food Canada, Lethbridge Research Centre, Lethbridge, Alberta, Canada, <sup>3</sup>Centro APTA Bovinos de Corte, Instituto de Zootecnia, IZ, Sertãozinho, São Paulo, Brasil.

The aim of this study was to determine the influence of flight speed (a proxy for cattle temperament) on dry matter intake (DMI), residual feed intake (RFI), performance and carcass composition. A total of 284 bull Nellore calves (256.9 ± 26.5 d of age) were evaluated during 2 feed



efficiency tests in 2012 and 2013. Calves were housed over a period of 93 and 87 d, respectively, in 2 feedlot pens (41 m<sup>2</sup>·animal<sup>-1</sup>·pen<sup>-1</sup>), containing 8 feed troughs per pen equipped with a radio frequency system (GrowSafe System Ltd., Airdrie, AB, Canada) used to measure DMI (kg·d<sup>-1</sup>), RFI (kg·d<sup>-1</sup>) and feed conversion ratio (kg gain·kg DMI<sup>-1</sup>). The diet was offered ad libitum, 3 times daily and consisted of corn silage, ground corn, soybean meal and mineral supplement. Calves were weighed every 14 d to calculate average daily gain (ADG; kg·d<sup>-1</sup>) for each individual. Carcass composition was recorded using ultrasound between the 12th and 13th ribs, on the last day of the feeding period to obtain ribeye area (REA; cm<sup>2</sup>). Flight speed (FS; m·s<sup>-1</sup>) was assessed every 28 d at the time of weighing. Calves were classified for temperament type according to their flight speed as follows: Calm = calves within the slowest tertile; Agitated = calves within the fastest tertile; and Intermediary = calves between the slowest and fastest tertile. Comparison of temperament type was analyzed with the PROC MIXED procedure (SAS Inst. Inc., Cary, NC). The model statement contained the effects of temperament class, feed efficiency test, and age on d 1 of the feeding period, as a covariate. No FS effect ( $P > 0.05$ ) was observed on RFI and ADG; however, calves categorized as Agitated temperament had lower ( $P < 0.05$ ) DMI ( $7.20 \pm 0.09$ ,  $7.40 \pm 0.09$ , and  $7.16 \pm 0.09$  kg·d<sup>-1</sup>) and REA ( $62.90 \pm 0.71$ ,  $62.05 \pm 0.72$ , and  $60.69 \pm 0.72$  cm<sup>2</sup>) than Calm and Intermediary groups, respectively. Based on these results we conclude that Nellore cattle characterized as having agitated temperaments during handling, consumed less feed, and had worse carcass composition.

**Key Words:** behavior, confinement, reactivity

**M35 Effects of handling before and during processing on behavior and ADG of feedlot steers.** Ruth H. Woiwode\*<sup>1</sup>, Temple Grandin<sup>1</sup>, Brett Kirch<sup>1</sup>, and John Paterson<sup>2</sup>, <sup>1</sup>Colorado State University, Fort Collins, CO, <sup>2</sup>National Cattlemen's Beef Association, Centennial, CO.

The objective of this study was to investigate if a relationship exists between handling, and behavior and ADG of feedlot cattle. Upon arrival at a commercial feedlot in Kansas, Hereford steers ( $n = 496$ ; initial BW =  $304 \pm 35.6$  kg) of similar genetic background were sorted into 4 pens to determine the effects of handling on behavior and ADG. Two handling conditions before processing and 2 conditions of release from the squeeze chute were imposed. Prior to processing, handlers were required to quietly walk all steers from their home pen to the processing area (SLOW); or handlers were permitted to bring steers to the processing area in the normal fashion (FAST). Individual steers were randomly assigned to one of 2 conditions of release from the squeeze chute. The first was a delay no longer than 30 s following the completion of procedures to allow cattle to stop struggling (DELAY); the second was release immediately following the completion of procedures (NORM). Vocalization, chute temperament, exit speed and exit behavior scores were assigned to all steers during intake processing. Vocalization was scored on a yes/no basis, and was recorded before procedures. Temperament scores were assigned after head gate capture, on a 5-point scale (1 = calm, 2 = shifting; restless, 3 = squirming; shaking squeeze chute, 4 = continuous, vigorous movement, 5 = rearing, struggling violently). Exit

speed was scored on a 3-point scale (walk; trot; run), and exit behavior was scored on a 2 point scale with cattle classified as high or low on a reactivity scale (L = No behaviors other than exit speed, H = Stumble, rear, jump). Paired *t*-tests determined that cattle exiting the chute at a walk or trot vs. a run tended ( $P = 0.08$ ) to have higher ADG. Cattle vocalizing during restraint had lower ( $P = 0.04$ ) ADG than those that did not vocalize. The FAST group showed a tendency to vocalize more frequently than the SLOW group. Pearson correlation analysis showed a significant, positive correlation between exit speed and vocalization ( $P = 0.0021$ ,  $r = 0.14256$ ), and a significant, negative correlation between exit speed and ADG ( $P = 0.0036$ ,  $r = -0.13542$ ). Using this approach, handling was correlated with behavior and ADG.

**Key Words:** behavior, vocalization, ADG

**M36 Temperament of beef cattle receiving supplementation on grazing system: daily gain, ultrasound measures, and intake behavior.** C. L. Francisco\*, A. M. Castilhos, D. C. M. Silva, F. M. Silva, and A. M. Jorge, Universidade Estadual Paulista-FMVZ, Botucatu, SP, Brazil.

A study was conducted to evaluate the temperament of beef cattle on grazing system regarding daily gain, ultrasound measures, and intake behavior. Twenty growing animals [Nellore; non-castrated males;  $220 \pm 3.73$  kg initial body weight (BW);  $10 \pm 1$  mo of age] on rotational stocking system (*Brachiaria brizantha* 'Xaraés') were used. Animals were evaluated for BW and temperament at the beginning of the trial period. Individual temperament scores were calculated by averaging animal chute score and exit score [adequate (ADQ)  $\leq 3$ ; or excitable (EXC)  $> 3$ ]. Animals were ranked by BW and assigned to receive ( $n = 10$ ) or not (control = CON;  $n = 10$ ) a supplementation treatment (SUP; 0.6% of BW; corn, urea and mineral salt). BW were measured on d 0 and d 84 to determine the total gain (TG) and average daily gain (ADG). Hip height (HH) and ultrasound evaluations (ribeye area; backfat thickness) were carried out on d 0 and 84. Intake behavior evaluations were obtained via direct observation (8 d; 24-h per day) with an interval of 15 min between observations (time of grazing, rumination, resting, and drinking water). Data were analyzed with PROC MIXED in SAS with fixed effects of temperament, treatment and the resultant interaction. No interaction was significant. ADQ animals had greater final BW ( $P = 0.01$ ) and HH ( $P = 0.01$ ) than EXC animals (310.78 vs. 273.06 kg for BW; 1.36 vs. 1.32 m for HH, for ADQ and EXC animals, respectively). SUP treatment had greater final BW ( $P = 0.04$ ), TG ( $P < 0.01$ ), ADG ( $P < 0.01$ ), final ribeye area ( $P < 0.01$ ), and HH ( $P = 0.03$ ) than CON treatment (305.80 vs. 278.04 kg for BW; 79.60 vs. 57.43 kg for TG; 0.95 vs. 0.69 kg for ADG; 37.25 vs. 30.84 cm<sup>2</sup> for ribeye area; 1.35 vs. 1.32 m for HH, for SUP and CON treatments, respectively). For intake behavior outcomes, ADQ animals had greater time of rumination ( $P = 0.01$ ) than EXC animals (456.30 vs. 440.31 min, for ADQ and EXC animals, respectively). In conclusion, some characteristics of performance and intake behavior are associated with temperament and it is independent of differences in supplementation treatment in Nellore cattle. Supported by FAPESP#2014/07406-5.

**Key Words:** Nellore cattle, pasture, temperament

## Animal Health: Beef cattle

### **M37 Functional capacities of blood neutrophils are influenced by both acute and chronic dexamethasone stress models in beef steers.**

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<sup>1</sup>Department of Animal and Food Sciences, Texas Tech University, Lubbock, TX, <sup>2</sup>USDA-ARS, Lubbock, TX, <sup>3</sup>Department of Agricultural Sciences, West Texas A&M University, Canyon, TX.

This study investigated the effects of acute and chronic stress models on the functional capacity of blood neutrophils in beef steers. Steers ( $n = 32$ ;  $209 \pm 8$  kg) were blocked by BW and assigned to 1 of 3 treatments: (1) Control (CON), no dexamethasone (DEX); (2) Chronic stress (CHR), 0.5 mg/kg BW DEX administered i.v. at 1000 h on d 3 to 6; or (3) Acute stress (ACU), 0.5 mg/kg BW DEX administered i.v. at 1000 h on d 6 only. Multiple blood samples were collected from jugular catheters to profile hematology. A blood sample collected at 2 h after the 4th DEX injection in the CHR treatment and 2 h after the only DEX injection in the ACU treatment was analyzed for functional capacities of neutrophils, which included surface expression of L-selectin (CD62-L) and the phagocytic and oxidative burst capabilities to an environmental *Escherichia coli*. There was a treatment  $\times$  time interaction ( $P \leq 0.001$ ) for neutrophil concentrations in peripheral circulation. The concentration of neutrophils increased 24 h after the 1st DEX injection among the CHR steers when compared with CON ( $10.6$  vs.  $2.8 \pm 0.62 \times 10^6/\text{mL}$ ;  $P \leq 0.001$ ) and remained greater until 72 h after the 4th DEX injection. Neutrophil concentrations also increased rapidly, within 2 h of the DEX, in the ACU steers. Treatment influenced ( $P \leq 0.001$ ) the expression of L-selectin on the surface of neutrophils ( $119^a$ ,  $138^b$ , and  $61^c \pm 5.2$  MFI) for CON, ACU, and CHR steers, respectively. The percentages of neutrophils phagocytizing and producing an oxidative burst were suppressed ( $P \leq 0.001$ ) among the CHR steers only ( $72^a$ ,  $71^a$ , and  $55^b \pm 4.2\%$ ), whereas the intensity of the oxidative burst was suppressed ( $P \leq 0.001$ ) for both ACU and CHR steers ( $170^a$ ,  $131^b$ ,  $63^c \pm 11.7$  MFI) for CON, ACU, and CHR steers, respectively. In contrast, the intensity of neutrophil phagocytosis was not influenced by treatment ( $P = 0.439$ ). These data indicate that chronic DEX suppresses neutrophil L-selectin as well as neutrophil phagocytosis and oxidative burst, whereas the acute DEX may initially prime neutrophil L-selectin expression although the oxidative burst intensity was already partially suppressed.

**Key Words:** immunity, neutrophil, stress

### **M38 Transcriptome profiling of the endometrium of healthy beef cows postpartum.** Robmay Garcia\*, Dianelys Gonzalez-Pena, and Sandra L. Rodriguez-Zas, *University of Illinois at Urbana-Champaign, Urbana, IL.*

The uterine cervix remains open for 7 to 10 d postpartum, which facilitates bacterial infection resulting in \$650M in losses to the US dairy industry. Understanding the molecular profiles of healthy endometrial involution and regeneration can help understand bacterial clearance and inflammation resolution mechanisms. The objective of this study is to characterize the endometrium transcriptome of beef cows at 15d and 30d postpartum. Individual paired-end reads libraries were mapped to the *Bos taurus* reference genome (Btau\_4.6.1) using Tophat v2.0.12. In total 8,282 isoform transcripts pertaining to 8,124 genes were identified and 1002 isoform transcripts pertaining to 995 genes were found to be differentially expressed (false discovery rate-adjusted  $P$ -value  $< 0.05$ )

between 15d and 30d using Cufflinks v2.2.1. Among the top 50 differentially abundant transcripts T-Box 21 (TBX21), T cell receptor associated transmembrane adaptor 1 (TRAT1) and indoleamine 2,3-dioxygenase (IDO) were overexpressed at 15d relative to 30d. TBX21 controls the expression of the hallmark Th1 cytokine interferon-gamma IFNG. TRAT1 Stabilizes the TCR T-cell antigen receptor / CD3 complex at the surface of T-cells and IDO play a role in processes such as antimicrobial defense and immunoregulation. Functional analysis of the differentially expressed genes using DAVID identified 10 enriched (enrichment score  $> 2$ ) functional category clusters including the Gene Ontology molecular functions for regulation of cell death and apoptosis, cytokine and chemokine activity, inflammatory response; Gene Ontology biological processes of immune system development, leukocyte activation, proliferation and differentiation, among associated immunological. These categories confirm that endometrial involution elicits changes associated with the inflammatory response and immunological activation. Our results offer insights on the transcriptome changes during normal endometrium involution.

**Key Words:** endometrium, involution, transcriptome

### **M39 Cardiac damage assessment in beef cattle receiving different dosages of monensin in finishing diets as measured by Creatine-Kinase Myocardic kit.**

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Monensin (MN) has affinity for monovalent cations such as Na and disruption of Na dynamics in tissues could affect the permeability of cardiac muscle cells. High or continued doses of MN have been suggested to result in cardiac myopathy, blood stasis and interstitial pulmonary edema. The objective of this study was to evaluate cardiac damage, not clinically detected, using a Creatine-Kinase Myocardic (CK-MB) commercial kit. Thirty Angus  $\times$  Hereford steers, (BW =  $205 \pm 12$  kg) were used to evaluate the effect of 2 doses of dietary MN during finishing in feedlot. Steers were randomly allotted into 3 groups of 10 each for a feedlot period of 168 d. Group 1: un-supplemented MN (G1: Control, CO), group 2: supplemented with 1 mg of MN/kg of body weight (BW) (G2: 1 mg/kg) and group 3, supplemented with MN 1.5 mg of MN/kg of BW (G3: 1.5 mg/kg). Steers were weighed on d 0, 63, 98, 126 and 168, with blood sampling at each time. Steers that received MN were 25 and 30 kg heavier ( $P < 0.05$ ) than the controls (G2 and G3, respectively). Concentration of CK-MB increased linearly ( $P < 0.05$ ) for the 3 groups (Table 1). Treatment effects were not that evident. Concentration of CK-MB was greater for G3 than G1 and G2 on sampling at d 63 and 168 ( $P < 0.05$ ). No macroscopic lesions either on lungs or cardiac muscle were observed at harvest.

*Contd.*

**Table 1 (Abstr. M39).** Concentration means and standard deviations for CK-MB throughout the trial

Group	Day 1	Day 63	Day 98	Day 126	Day 168
G1: Control	25.1 ± 10.0 <sup>A</sup>	40.4 ± 22.4 <sup>A</sup>	68.2 ± 9.0 <sup>B</sup>	89.6 ± 18.3 <sup>B</sup>	72.9 ± 20.3 <sup>A</sup>
G2: 1 mg/kg BW	32.1 ± 6.3 <sup>A</sup>	40.4 ± 14.7 <sup>A</sup>	49.0 ± 13.7 <sup>A</sup>	62.1 ± 22.5 <sup>A</sup>	61.2 ± 23.9 <sup>A</sup>
G3: 1.5 mg/kg BW	26.8 ± 13.1 <sup>A</sup>	61.7 ± 21.4 <sup>B</sup>	57.0 ± 11.3 <sup>A</sup>	76.4 ± 25.0 <sup>A</sup>	92.6 ± 18.4 <sup>B</sup>

<sup>A,B</sup> Treatment means followed by different letters denote statistical differences at  $P \leq 0.05$ .

**Key Words:** beef, monensin toxicity, respiratory symptomatology.

**M40 Estimating glucose requirements of an activated immune system in Holstein steers.** Sara K. Stoakes\*, Erin A. Nolan, David J. Valko, Mohannad Abuajamieh, Maria V. Sanz Fernandez, and Lance H. Baumgard, *Iowa State University, Ames, IA.*

Activated immune cells are obligate glucose utilizers and a large lipopolysaccharide (LPS) IV dose causes severe hypoglycemia. Therefore, study objectives were to use the quantity of glucose needed to maintain euglycemia during an IV bolus endotoxin challenge as a proxy for the immune system's glucose requirement. Fasting growing Holstein steers (148 ± 9 kg) were jugular catheterized bilaterally and assigned to 1 of 3 treatments: control (CON; 3 mL sterile saline; n = 5), LPS-infused (LPS; *E. coli* 055:B5; 1.5 µg/kg BW; n = 5), and LPS + euglycemic clamp (LPS-Eu; 1.5 µg/kg BW; 50% dextrose infusion to maintain euglycemia; n = 5). Following infusion, blood glucose was determined every 10 min and dextrose infusion rates were adjusted in LPS-Eu calves to maintain euglycemia for 12 h. Plasma samples were obtained 3, 6, 9, and 12 h relative to bolus infusion for further analysis. All calves survived the LPS challenge. Rectal temperature was increased in LPS (0.6°C,  $P = 0.03$ ) and tended to be increased in LPS-Eu (0.5°C,  $P = 0.06$ ) relative to CON calves. LPS and LPS-Eu calves were hyperglycemic for 3 h post-bolus, likely due to hepatic glycogenolysis. Thereafter, blood glucose was markedly decreased in LPS relative to both CON and LPS-Eu calves (30%,  $P < 0.01$ ). Blood lymphocytes, and platelets were increased in LPS and LPS-Eu calves relative to CON (53 and 56%, respectively;  $P < 0.01$ ). Ionized calcium was decreased in both LPS and LPS-Eu calves relative to CON (18%,  $P < 0.01$ ). White blood cells decreased or tended to decrease in LPS-Eu and LPS (59%,  $P = 0.03$ ; 44%,  $P = 0.06$ ; respectively) relative to CON. During the 12 h, 516 ± 65 g of infused glucose was required to maintain euglycemia. If the amount of glucose required to maintain euglycemia can be used as a proxy, then the glucose requirements of an activated immune system are approximately 43 g/h in growing ruminants.

**Key Words:** lipopolysaccharide, immune challenge, glucose homeostasis

**M41 A comparison of rumen bacterial communities in bloated and non-bloated cattle grazing alfalfa.** Elnaz Azad\*<sup>1</sup>, Robert Forster<sup>2</sup>, Surya Acharya<sup>2</sup>, Tim McAllister<sup>2</sup>, and Ehsan Khafipour<sup>1</sup>, <sup>1</sup>*Department of Animal Science, University of Manitoba, Winnipeg, Manitoba,* <sup>2</sup>*Agriculture and Agri-Food Canada, Lethbridge Research Centre, Lethbridge, Alberta, Canada.*

To study bacterial shifts during the bloat process, 2 dietary interventions for alleviating the severity of bloat on rumen bacterial composition were studied in 12 rumen-cannulated cattle in a 3-phase crossover study. Cattle were subjected to 1 of 3 treatments: (1) pure alfalfa pasture (PA), (2) pure

alfalfa pasture supplemented with Alfasure (AA), and (3) alfalfa/sainfoin mixed pasture (AS). A 7-d washout interval was provided between phases during which cattle received a baseline diet. Rumen liquid and solid fractions were collected from non-bloated (NB; bloat score = 0) and bloated (B; bloat scores = 1–3) cattle within each treatment group and subjected to genomic DNA extraction and Illumina sequencing of the V3-V4 regions of bacterial 16S rRNA gene. On average, 47,211 high-quality sequences were generated per sample. Taxonomic classification revealed the presence of 20 different bacterial phyla among which Bacteroidetes followed by Firmicutes, Fibrobacteres, Cyanobacteria, and Spirochaetes were identified as predominant (>1% of population) members of rumen microbiota. The PERMANOVA analysis of UniFrac distances revealed distinct ( $P < 0.05$ ) clustering patterns when comparing PA(B), AA(NB) and AS(NB)-associated microbiota. Statistical analysis (LEfSe) of phylogenetic data for rumen liquid and solid fractions also revealed significant associations between several members of microbiota and treatment groups ( $P < 0.05$ ; Table 1). Further investigation can focus on manipulation of rumen microbiota in favor of increased bloat-resistant functional properties.

**Table 1 (Abstr. M41).** Association of bacterial genera with treatment groups<sup>1</sup>

Genus	Phylum	Treatment group <sup>2</sup>
Rumen liquid fraction		
<i>Succinivibrio</i>	Proteobacteria	PA(B)
<i>Succiniclasticum</i>	Firmicutes	PA(B)
<i>Butyrivibrio</i>	Firmicutes	AS(NB)
<i>Coprococcus</i>	Firmicutes	AS(NB)
<i>Fibrobacter</i>	Fibrobacteres	AA(NB)
<i>Ruminococcus</i>	Firmicutes	AA(NB)
Rumen solid fraction		
<i>Butyrivibrio</i>	Firmicutes	PA(B)
<i>Streptococcus</i>	Firmicutes	PA(B)
<i>Prevotella</i>	Bacteroidetes	AS(NB)
<i>Fibrobacter</i>	Fibrobacteres	AA(NB)

<sup>1</sup>Linear discriminant analysis effect size (LEfSe) was used to test for significant associations ( $P < 0.05$ , and >3 log fold increase in the relative abundance).

<sup>2</sup>PA(B) = pure alfalfa (bloated); AS(NB) = alfalfa-sainfoin (non-bloated); AA(NB) = alfalfa-Alfasure (non-bloated).

**Key Words:** frothy bloat, alfalfa, rumen microbiota

**M42 Efficacy of dosing toltrazuril 5% as a coccidiostat for cattle.** Rafahel C. Souza<sup>1</sup>, Rogério C. Souza<sup>1</sup>, Renato O. Santos<sup>1</sup>, Sérgio V. G. Ribeiro<sup>1</sup>, Andre B. D. Pereira\*<sup>3</sup>, Thiago M. Soares<sup>2</sup>, and Maria I. V. Melo<sup>1</sup>, <sup>1</sup>*Pontifícia Universidade Católica de Minas Gerais, Betim, MG, Brazil,* <sup>2</sup>*Ourofino Agronegocios, Cravinhos, SP, Brazil,* <sup>3</sup>*University of New Hampshire, Durham, NH.*

Coccidiosis is a protozoan disease that negatively affects animal production and can result in economic losses. *Eimeria* spp. is the most common parasite. Infection occurs mainly in young cattle and can lead to gastrointestinal problems and clinical signs as bloody diarrhea and hemorrhagic enteritis. The objective of this study was to evaluate fecal infestation and average daily gain of 100 Nelore heifers (Average weight of 191.22 kg) randomly distributed, as a completely randomized design, in 1 of 2 treatments: (1) Control (C, no coccidiostat), and (2) Treatment (T, dosage of 15 mg/kg of toltrazuril 5%, dosed once at weaning, 210 d after birth). The experiment was conducted in the Arrojo Farm (Teófilo Otoni, MG, Brazil). Animals were weighed in the first day of the experiment and then 30 and 60 d after. Fecal samples were collected



for laboratory analysis in the first and last days of the experiment, for counting oocysts per gram of feces (OOPG). Data were analyzed using the MIXED procedure of SAS and Tukey as post hoc tests for separation of means. Friedman nonparametric test was done for repeated analysis and count of OOPG. There was no difference between treatments for average daily gain (596 g/d for C and 656 g/d for T,  $P > 0.05$ ). However, presence of OOPG was lower for cattle in the treatment group after 60 d when compared with samples from the beginning of the experiment ( $P = 0.01$ ), which did not happen with the control group ( $P = 0.86$ ). Results of this experiment suggest that toltrazuril 5% in the dose of 15 mg/kg can reduce parasite infestation in the gastrointestinal tract. Infestation in the beginning of the experiment was not deleterious enough to cause changes in average daily gain.

**Table 1 (Abstr. 42).** Effects of dosing 15 mg/kg of toltrazuril 5% to cattle

Item	Group		SEM	P-value
	Control	Treatment		
ADG, g/d <sup>1</sup>	596 <sup>a</sup>	656 <sup>a</sup>	247.67	NS
Oocysts per gram of feces, d 1	982.76 <sup>a,A</sup>	988.89 <sup>a,B</sup>	108.94	0.91
Oocysts per gram of feces, d 60	786.21 <sup>a,A</sup>	730.56 <sup>a,A</sup>	113.34	0.30

<sup>a,b,A,B</sup>Values with lowercase letters indicate differences between treatments; those with uppercase letters indicate difference between d 1 and 60 (Friedman nonparametric test).

<sup>1</sup>Tukey post hoc test.

**Key Words:** toltrazuril, coccidiosis, oocysts per gram of feces

**M43 In vitro evaluation of the antimicrobial activity of plant extracts from *Ruta graveolens* and *Annona muricata*.** Yadileiny Portilla<sup>1</sup>, María Dolores Carro<sup>2</sup>, Grethel Milián<sup>1</sup>, Conrado Camacho<sup>1</sup>, Aymara Valdivia<sup>1</sup>, Alexey Díaz<sup>3,4</sup>, Cristina Saro<sup>3</sup>, Iván Mateos<sup>3</sup>, and María José Ranilla<sup>\*3,4</sup>. <sup>1</sup>Center for Biotechnological Studies, University of Matanzas, Matanzas, Cuba, <sup>2</sup>Agriculture Production Department, Technical University of Madrid, Madrid, Spain, <sup>3</sup>Animal Production Department, University of León, León, Spain, <sup>4</sup>IGM (CSIC-ULE), Finca Marzanas s/n, Grulleros, León, Spa.

Resistance of microorganisms to commercial drugs is increasing worldwide, and therefore the search for new antimicrobial agents is a key issue. The aim of this study was to identify the potential of plant extracts from *Ruta graveolens* and *Annona muricata* as candidates for the development of new antimicrobials. Plant extracts were obtained by the Soxhlet method and their biological evaluation was carried out by the agar diffusion method, with 4 doses assayed (6.25, 25, 50 and 100 mg/mL) and 4 replicates per dose. Eight bacterial strains from American Type Culture Collection (ATCC) were tested: *Escherichia coli* O157 (ATCC 43894), *Streptococcus agalactiae* (ATCC 13813), *Salmonella enteritidis* (ATCC BBA664), *Enterobacter aerogenes* (ATCC 13048), *Staphylococcus aureus* (ATCC 13565), *Klebsiella pneumoniae* (ATCC 4352), *Proteus mirabilis* (ATCC 14153) and *Proteus vulgaris* (ATCC 9484). Extracts from both plants showed antibacterial activity against all bacteria tested, with the exception of *A. muricata* extract against *S. enteritidis*. Minimum inhibitory concentration for both extracts was 6.25 mg/mL for *E. aerogenes*, *S. agalactiae*, *S. aureus*, and *K. pneumoniae*, 25 mg/mL for *E. coli*, *P. mirabilis*, and *P. vulgaris*, and 50 mg/mL of *R. graveolens* for *Salmonella enteritidis*. There were no differences between extracts in their antibacterial activity against *P. vulgaris* ( $P = 0.91$ ) and *K. pneumoniae* ( $P = 0.37$ ), but *R. graveolens* extract showed greater ( $P < 0.001$ ) antibacterial activity against *E. coli* and *S. agalactiae* than *A. muricata* extract, and a trend was also observed for *E. aerogenes* ( $P = 0.064$ ). In contrast, *A. muricata* extract tended to have greater ( $P$

$= 0.094$ ) antibacterial activity against *P. mirabilis* compared with *R. graveolens* extract. The results suggest that these extracts have active ingredients that could help to develop new antimicrobial products for the improvement of animal production and health.

**Key Words:** *Ruta graveolens*, *Annona muricata*, gram-positive

**M44 OmniGen-AF affects expression of immune-related genes in whole blood of healthy Angus heifers.** S. A. Armstrong<sup>\*1,2</sup>, D. J. McLean<sup>1</sup>, T. H. Schell<sup>1,2</sup>, G. Bobe<sup>2</sup>, and M. Bionaz<sup>2</sup>, <sup>1</sup>Phibro Animal Health, Corvallis, OR, <sup>2</sup>Department of Animal and Rangeland Sciences, Oregon State University, Corvallis, OR.

Purebred Angus heifers were used to determine the effect of OmniGen-AF (OG) supplementation on expression of cytokines, chemokines, and associated receptors involved in the inflammatory response in whole blood cells of healthy Angus heifers within the first 28 d of supplementation. Heifers were randomly assigned to control or supplemented daily with 56 g OG group ( $n = 4/\text{group}$ ), and fed a diet including grass hay, alfalfa and ground corn. Heifers were housed in a freestall barn and fed via Calan Broadbent system. Blood was collected via jugular before the study started (0) and on d 3, 5, 10, 14, 21, and 28 of supplementation. The qRT-PCR was performed using the Cow Inflammatory Cytokines and Receptor qPCR array (Qiagen). Data were analyzed using LinReg software to account for efficiency of amplification and normalized by 3 internal control genes (*HPRT1*, *TBP*, and *YWHAZ*). qRT-PCR data were log-transformed and the samples with Studentized residuals  $t > 2$  removed. The final data set (82 genes) was subjected to ANOVA analysis with treatment, time, and treatment  $\times$  time as main effect and animal as random using JMP Genomics of SAS. Significance was deemed with a false discovery rate-adjusted  $P$ -values  $< 0.10$ . Genes coding for chemokine receptors (*CX3CR1*, *CXCR1*), stress response (*NAMPT*), osteoclastogenesis (*TNFRSF11B*), and angiogenesis (*VEGFA*) were affected by treatment  $\times$  time. Thirteen genes coding for interleukins and interleukin receptors (*IL1B*, *IL9*, *IL1RN*, *IL1R1*, *IL10RB*, *IL10RA*), chemokine ligand and receptors (*CCR2*, *CXCL2*, *CXCR1*, *CCL26*, *CCR1*), macrophage function (*CSF1*), and secondary immune response (*BMP2*) were downregulated and *CCL1* was upregulated by OG supplementation. Of the 23 receptors evaluated, 9 (39%) were influenced by OmniGen supplementation. Overall, the data suggest a transcriptional inhibition of genes related to inflammatory response by OG during the first 28 d of supplementation of healthy Angus heifers.

**Key Words:** OmniGen-AF, immune, cytokine

**M45 Influence of hydrolysable tannin extract on nematode egg count in feces of receiving beef cattle.** Melissa B. Corona<sup>1</sup>, Eva X. Murillo<sup>1</sup>, Billy J. Cervantes<sup>2</sup>, Nohemi Castro<sup>1</sup>, Javier A. Romo<sup>1</sup>, Soila M. Gaxiola<sup>1</sup>, and Rubén Barajas<sup>\*1</sup>, <sup>1</sup>FMVZ-Universidad Autónoma de Sinaloa, Culiacán, Sinaloa, México, <sup>2</sup>Ganadera Los Migueles, S.A. de C.V., Culiacán, Sinaloa, México.

The nematode parasites decline productivity of beef cattle. The nematode egg count is decreased in feces of cattle grazing plants with high hydrolysable tannin content. There is little information of effect of added tannins to the diet on nematodes presence in beef cattle. In this experiment 40 receiving bull-calves were involved to determine the influence of hydrolysable tannin extract on nematode egg count in feces of receiving beef cattle. Bull-calves were placed in 8 dirt-floor pens, and during 3 continuous days, fecal samples were taken from each. They were randomly assigned to treatments: (1) 70% roughage (16.1% CP; 1.27 Mcal NE<sub>m</sub>/kg DM) corn silage-based diet (Control); (2) Control

plus 1.5% of hydrolysable tannin extract DM basis (HT). After 28 d on treatment diets, fecal samples were taken during 3 continuous days again, and nematode eggs per gram of feces (EGF) were counted. Before statistical analyses, data were normalized by transforming to  $\log_{10} x + 17$  EGF. Results were analyzed by ANOVA for a completely randomized design. Additionally, both in Control and HT, EGF before and after treatments were compared using paired *t*-test. *Haemonchus* spp. and *Cooperia* spp. were most frequently genus found (82.5 and 75%, respectively). At start of experiment *Haemonchus* spp. EGF was similar ( $223 \pm 77$  EGF) between treatments ( $P > 0.87$ ). After 28 d of receiving treatments, HT decreased 67.9% ( $P = 0.02$ ) *Haemonchus* spp. EGF comparatively with Control (62 vs. 196 EGF). The paired *t*-test results indicated that *Haemonchus* spp. EGF were similar ( $P = 0.47$ ) before and after in Control, but in HT were 70% lower ( $P < 0.01$ ) after treatments (210 vs. 62 EGF). At arriving *Cooperia* spp. EGF was similar between treatments ( $P = 0.94$ ), but after 28 d, EGF count was reduced 67.8% ( $P = 0.04$ ) by HT relative to Control (39 vs. 122 EGF). The paired *t*-test indicated that in Control, EGF count was similar before and after 28 d ( $P = 0.29$ ), but in HT EGF decreased ( $P < 0.01$ ) 53% (64 vs. 136 EGF). Results suggest that HT addition in to the diet contributes to decrease fecal shedding of nematode eggs in receiving feedlot cattle.

**Key Words:** bovine, nematode, tannin

**M46 Effects of bambermycin or monensin on health and performance of receiving cattle.** William Galyen<sup>\*1</sup>, Tom Hess<sup>2</sup>, Don Hubbell<sup>2</sup>, Shane Gadberry<sup>3</sup>, Elizabeth Kegley<sup>1</sup>, Matt Cravey<sup>4</sup>, Jeremy Powell<sup>1</sup>, Elizabeth Backes<sup>1</sup>, Laura Meyers<sup>1</sup>, and Paul Beck<sup>5</sup>, <sup>1</sup>University of Arkansas Department of Animal Science, Fayetteville, AR, <sup>2</sup>University of Arkansas LFRS, Batesville, AR, <sup>3</sup>University of Arkansas Cooperative Extension Service, Little Rock, AR, <sup>4</sup>Huvepharma, Inc, Amarillo, TX, <sup>5</sup>University of Arkansas SWREC, Hope, AR.

Growing steers and bulls, were received in 3 blocks (Block 1,  $n = 150$ ,  $BW = 208 \pm 12.4$  kg; Block 2,  $n = 99$ ,  $BW = 213 \pm 16.7$  kg; Block 3,  $n = 149$ ,  $BW = 219 \pm 14.9$  kg) to evaluate the effects of supplying 20 mg of bambermycin (Gainpro; Huvepharma, Inc., Sofia Bulgaria) or 0.77 mg/kg BW monensin (Rumensin; Elanco Animal Health, Indianapolis IN) in receiving supplements (20% CP and 78% TDN) compared with non-medicated supplements (Control) on cattle morbidity, performance, and coccidia infection. Upon receiving, bulls were castrated, and calves were weighed on 2 consecutive days. Calves were then stratified by BW and arrival castrate status and randomly allocated to receiving pens ( $n = 12-0.4$  ha pens in Block 1 and  $n = 6$  pens in Blocks 2 and 3). Calves received 0.9 kg of supplement daily and ad libitum access to moderate quality hay. Fecal samples were collected from 6 steers/pen on d 0, 14, and 28 to evaluate coccidia infection. Water in the Gainpro pens was treated from d 14 to d 19 with 10 mg/kg BW amprolium (Corid; Merial, Duluth GA). Steers remained on treatment for 56 to 84-d for Block 1, 49-d for Block 2 and 42-d for Block 3. Data were analyzed as a randomized complete block design using the mixed procedure of SAS (SAS Inst. Inc., Cary, NC). Cocci counts were log-transformed before analysis as a repeated measure in time. There were no differences ( $P \leq 0.36$ ) in morbidity, mortality, or animals identified as chronically morbid. There was no treatment by day interaction ( $P = 0.12$ ) for cocci oocysts counts, and monensin decreased coccidia ( $P \leq 0.03$ ) compared with Control and bambermycin, which did not differ ( $P = 0.85$ ). No cattle were observed with or treated for symptoms of coccidiosis (bloody scours and diarrhea). At the end of receiving, BW and ADG for Control ( $237 \pm 5.1$  kg/steer and  $0.49 \pm 0.272$  kg/d, respectively) was less than ( $P \leq 0.04$ ) bambermycin ( $243 \pm 5.1$  kg/steer and  $0.60 \pm 0.272$  kg/d) and monensin ( $247 \pm 5.1$  kg/steer and  $0.68 \pm 0.272$  kg/d), and monensin tended ( $P \leq 0.10$ )

tended to be greater than bambermycin. The results of this experiment indicate both bambermycin and monensin increased receiving cattle gain performance compared with Control, and monensin also provided greater benefits in reduction of coccidia counts.

**Key Words:** bambermycin, monensin, receiving steers

**M47 Influence of Papaveraceae plant preparation on nematode egg count in feces of receiving bull-calves.** Rubén Barajas<sup>\*1</sup>, Melissa B. Corona<sup>1</sup>, Eva X. Murillo<sup>1</sup>, Billy J. Cervantes<sup>2</sup>, Ingo Rogge<sup>3</sup>, Nohemi Castro<sup>1</sup>, and Luis E. Soto<sup>4</sup>, <sup>1</sup>FMVZ-Universidad Autónoma de Sinaloa, Culiacán, Sinaloa, México, <sup>2</sup>Ganadera Los Migueles, S.A. de C.V., Culiacán, Sinaloa, México, <sup>3</sup>Phytobiotics Futtermittelwerke GmbH, Eltville, Germany, <sup>4</sup>FA-Universidad Autónoma de Sinaloa, Culiacán, Sinaloa, México.

Quaternary benzophenanthridine alkaloids and protopine alkaloids (QBA + PA) have systemic anti-inflammatory effects. However, its activity against nematode parasites of the gastro intestinal tract is not well documented. In the actual experiment, 20 receiving bull-calves  $236 \pm 4.12$  kg were utilized to evaluate the influence of a standardized QBA + PA *Papaveraceae* family plant preparation named Sangrovit RS (SANG; Phytobiotics, Eltville, Germany) on fecal shedding of parasitic nematode eggs. In groups of 5, bull-calves were allotted in dirt-floor pens, fed with 70% roughage corn silage-diet. During 3 consecutive days fecal samples were taken from each animal and nematode eggs per gram of feces (EGF) were counted by McMaster method. Pens were randomly sorted to receive 10 g/d of Sangrovit RS (SANG) or not (Control) supplied on the feed bunk as top dressing. Once completed the 28 d period treatments, fecal samples were taken again during 3 consecutive days, and EGF count was performed. Results were analyzed by ANOVA for a completely randomized design. In fecal samples taken before applied treatments, the nematode EGF count was similar ( $P > 0.30$ ) between treatments. After 28 d total nematode EGF was lower ( $P = 0.04$ ) in the SANG group compared with the Control group (33 vs. 113 EGF). The amount of *Haemonchus* sp. EGF (an abomasum parasite) was lower ( $P = 0.05$ ) in bull-calves fed SANG compared with Control group (12 vs. 48 EGF). The EGF count of *Cooperia* spp., a parasite of the small intestine, was inferior ( $P = 0.03$ ) in SANG supplemented bull-calves in relationship to unsupplemented bull-calves (13 vs. 52 EGF). Results suggest that fed QBA + PA plant preparation contributes to decrease parasites nematode eggs shedding in receiving bull-calves.

**Key Words:** bovine, nematode, alkaloid

**M48 Effect of Safeguard on fecal egg count and performance in received beef calves.** Antonio Jose Neto<sup>\*1</sup>, Curt J. Bittner<sup>1</sup>, Galen E. Erickson<sup>1</sup>, and Brandon L. Nuttelman<sup>2</sup>, <sup>1</sup>Department of Animal Science; University of Nebraska, Lincoln, NE, <sup>2</sup>Merck Animal Health, De Soto, KS.

Parasite infestations can reduce or limit feed intake and subsequently depress the performance due to decreased absorption of nutrients. Fenbendazole (Safeguard, Merck Animal Health) is indicated for use in cattle for removal and control of lungworms, stomach worms and intestinal worms. The objective of this study was to measure the effects of Safeguard on fecal egg count (FEC) and performance of newly received calves in the feedlot over the first 25 d. Treatments were applied to steers at arrival and were: Safeguard and Doramectin (Dectomax, Zoetis Animal Health) injectable (SG+DTX) or only Dectomax injectable (DTX). Three hundred sixty-eight ( $265 \pm 20$  kg) steers were used in a completely randomized design with 16 pens (8 replications per

treatment and 23 steers per pen). The basal diet consisted 30% dry-rolled corn, 36% sweet bran, 30% alfalfa hay, and 4% supplement. Steers were assigned to pen based on processing order, with every other steer assigned to either SG+DTX or DTX. Once a pen replicate was filled, new pen replicates were started until all steers were assigned. On d 1, steers were weighed, and individual fecal sample collected. On d 19, fecal samples were collected per pen (10 samples/pen). Fecal samples were analyzed for FEC at a commercial laboratory. At the end of the receiving period, steers were limit-fed a diet consisting of 50% sweet bran and 50% alfalfa hay at 2% of BW for 5 d before being weighed. Data were analyzed using the Proc MIXED of SAS, with pen as the

experimental unit. There were no differences in initial BW ( $P = 0.13$ ), ending BW ( $P = 0.33$ ), DMI ( $P = 0.41$ ), ADG ( $P = 0.94$ ), and G:F ( $P = 0.43$ ) between SG+DTX or DTX. No difference for initial FEC ( $P = 0.45$ ) was observed between treatments and averaged 16.9 eggs per 3 g of feces. However, FEC on d 19 was lower ( $P = 0.03$ ) for animals receiving SG+DTX (FEC = 0.06 eggs per 3 g feces) compared with DTX (FEC = 0.50 eggs per 3 g feces). A combination of Safeguard and Dectomax reduced FEC of newly received calves in feedlot.

**Key Words:** dewormer, fecal egg count, performance

## Animal Health: Immunology

**M49 Comparison of antibody response, bacteriological culture and PCR based diagnostic methods in *Brucella ovis* inoculated rams.** Ariel O. Miranda\*<sup>1</sup>, Hernán Romero Harry<sup>1</sup>, Valeria N. Baldone<sup>1</sup>, Marcy Owens<sup>2</sup>, and Scott Pratt<sup>2</sup>, <sup>1</sup>INTA, Anguil, La Pampa, Argentina, <sup>2</sup>Clemson University, Clemson, SC.

Twenty-seven *Brucella ovis* (*B. ovis*)-negative adult Pampinta rams were used to evaluate 3 diagnostic methods for *B. ovis* detection post-inoculation. Twenty-four rams were inoculated by the conjunctival route and intraprepuccially with a total of  $3 \times 10^9$  cfu/ram of *B. ovis* (strain, INTA Bariloche, Argentina). Three rams were inoculated with a saline placebo. Blood and semen samples were taken for serological antibody detection, bacteriology culture and PCR at 0, 12, 24, 62 and 97 d post inoculation. Modified Thayer Martin medium was used to detect presence of *B. ovis* in semen. Serum samples were tested for antibodies to *B. ovis* using a commercial ELISA kit. *B. ovis* DNA from semen samples was extracted using a commercial kit (Qiagen) and PCR performed using primers pairs specific to *B. ovis*. The proportion of agreement (Kappa value,  $\kappa$ ) was calculated. *B. ovis* was not detected 12 d post-inoculation by any detection method. Nine rams were positive for *B. ovis* 24 d post-inoculation detected by ELISA; however, the rams were negative for *B. ovis* using the other 2 detection methods. Further, we observed a continued increase toward the end of the study. The intermittent elimination of *B. ovis* by semen agrees with the lower result response in culture and PCR. The low percentage of positive animals in culture on the 97 d could have been given to false negatives or methodology. Control group was negative to the 3 methods over the study. Seroconversion confirmed by ELISA is the most sensitive diagnostic measure of *B. ovis* exposure.

**Table 1 (Abstr. M49).** Number and percentage of positive rams by different methods and  $\kappa$  value between them throughout the study

Test diagnostic	Sampling day				
	0	12	24	62	97
ELISA positive	0	0	9 (38%)	21 (88%)	23 (96%)
Culture positive	0	0	0	14 (58%)	2 (8%)
PCR positive	0	0	0	18 (75%)	17 (71%)
	Agreement (%), $\kappa$ -value (SE)				
ELISA vs. culture	100, 1.0 (-)	100, 1.0 (-)	62, 0.11 (0.10)	70, 0.33 (0.16)	12, 0.00 (0.01)
ELISA vs. PCR	100, 1.0 (-)	100, 1.0 (-)	62, 0.11 (0.10)	70, 0.07 (0.20)	75, 0.19 (0.17)
Culture vs. PCR	100, 1.0 (-)	100, 1.0 (-)	100, 1.0 (-)	83, 0.63 (0.15)	37, 0.07 (0.05)

**Key Words:** *Brucella ovis*, diagnostic tools, PCR diagnostic

**M50 Prevalence of brucellosis in Iraq and control through a vaccination campaign.** Alaa Khalil Ismaiel\*, Ministry of Agriculture, Veterinary Directorate, Central Veterinary Laboratory, Baghdad, Iraq.

*Brucella melitensis* is primarily a cause of abortions in sheep and goats. It can be isolated from cattle, water buffalo, and camels, and transmits to humans through unpasteurized milk and cheeses. Little was known about the distribution of this organism around Iraq, which was needed for developing an effective control campaign. The goals of this

study, initiated in 2005 in collaboration with the Food and Agriculture Organization (FAO), were to determine the prevalence of *Brucella* in sheep, goats, cattle, water buffalo, and camels across Iraq, and to test the effectiveness of a whole cell vaccine. Villages for sampling were selected from 15 governorates that had more than 2000 animals. In each governorate, goats and sheep were sampled in 17 villages, cattle and water buffalo in 11 villages, and camels in 9 villages; 60 animals were sampled in each category per village. Serum samples initially screened by the Rose Bengal test were further confirmed by ELISA. These results indicated that 6.5% (6.1–6.9, 95% CI), 1.0% (0.78–1.2, 95% CI) and 1.5% (1.2–1.8, 95% CI) of sheep and goats, cattle, and water buffalo were positive for *B. melitensis*, respectively. Prevalence for sheep and goats ranged from 0.2% in Murhana to 16.4% in Kirkuk, for cattle from 0.02% in Thyqar to 11.1% in Muthana, and for water buffalo from 0% in Babylon to 5.4% in Basrah. Approximately  $1$  to  $2 \times 10^9$  cfu of the live attenuated *B. melitensis* vaccine, Rev1, was used to vaccinate 1.8 to 2.5 million lambs and kids each year, across all 15 governorates, from 2009 to 2014. During this time, cases of abortion in sheep and goats fell from 5090 in 2011, to 322 in 2013. This study concluded that the incidence of *Brucella* varies across different regions of Iraq, which can be related to animal handling practices. An aggressive strategy of vaccination was effective in reducing abortions in sheep and goats by over 90%. During the course of the vaccine intervention in this study, the Iraqi Ministry of Health reported that the incidence of brucellosis in humans decreased from 24 to 7 incidences per 100,000 people, further indicating an effective vaccination program.

**Key Words:** *Brucella*, vaccination, pathogen control

**M51 Maternal undernutrition increases acylated ghrelin concentrations in the umbilical artery and vein of the twin ovine fetus.** Sahng-Wook Hahm\*<sup>1</sup>, Meghan Field<sup>1</sup>, Russell V. Anthony<sup>2</sup>, and Hyungchul Han<sup>1</sup>, <sup>1</sup>Department of Animal Sciences, Colorado State University, Fort Collins, CO, <sup>2</sup>Department of Biomedical Sciences, Colorado State University, Fort Collins, CO.

Maternal undernutrition can induce intrauterine growth restriction and contribute to the development of adult metabolic diseases. Ghrelin, a peptide hormone purified from the gastric mucosa, activates food intake and energy homeostasis. Ghrelin exists in 2 forms, the non-acylated ghrelin and acylated ghrelin (active form). The acylation of ghrelin is mediated by membrane bound O-acyltransferase 4 (Mboat4). We hypothesized that maternal undernutrition would increase the acylated ghrelin in the blood of the fetus during gestation. Twin bearing western whiteface ewes were either fed 100% (C,  $n = 12$ ), or 50% of their global nutrient requirements from 28 to 78 d of gestational age (dGA) and readjusted to 100% beginning at 79 dGA (LC,  $n = 12$ ), or continuously restricted until 135 dGA (LL,  $n = 12$ ). At 135 dGA, umbilical artery and vein plasma, and fetal abomasum samples were collected. Umbilical arterial and venous ghrelin concentrations were measured by radioimmunoassay. Mboat4 gene expression in fetal abomasum was measured using quantitative real-time PCR. Mboat4 protein concentration in the fetal abomasum was analyzed by Western blot analysis. Each ewe was treated as one experimental unit and twin fetuses were nested within the ewe. All data are presented as least squares means  $\pm$  SEM using the PROC MIXED model of SAS. The umbilical arterial active ghrelin concentration tended to be greater in LL fetuses ( $41.63 \pm 5.49$  pg/mL) compared with control ( $27.17 \pm 5.76$  pg/mL;  $P = 0.0523$ ) and LC fetuses ( $22.81 \pm 5.28$  pg/mL;  $P = 0.0197$ ). The active to total ghrelin ratio was



higher in LL than LC fetuses ( $P < 0.05$ ). Umbilical vein active ghrelin concentration tended to be greater in LL, when compared with LC fetuses ( $P = 0.0812$ ). No significant difference was observed in Mboat4 mRNA expression in fetal abomasum. Mboat4 protein concentration tended to be greater in fetal abomasum of LC and LL fetuses compared with control fetuses ( $P = 0.0584$ ). These results are interpreted to mean that elevated active ghrelin but not total ghrelin in fetal circulation may be an adaptation of the fetus to prolonged undernutrition during gestation. USDA-AFRI Grant no. 2009–65203–05670.

**Key Words:** fetus, ghrelin, membrane bound O-acyltransferase 4 (Mboat4)

**M52 Evaluating udder health in dairy goats: An old but still unsolved issue.** Andrea Bezerra<sup>1</sup>, Candice De Leon<sup>1</sup>, Magda Fernandes<sup>1</sup>, Bryan White<sup>2</sup>, Juan Loo<sup>2</sup>, and Celso Oliveira<sup>\*1,2</sup>, <sup>1</sup>Federal University of Paraiba (UFPB), Brazil, Areia, PB, Brazil, <sup>2</sup>The University of Illinois at Urbana-Champaign, Urbana, IL.

Despite the high economical importance of goat milk production in certain regions, the real burden of subclinical mastitis caused by intramammary infections (IMI) is still unknown. This is mainly caused by the lack of reliable diagnostic tests, since the accuracy of somatic cell count (SCC) as an indirect indicator of udder health is questionable in this species. This study aimed to investigate the correlation among and somatic cell count (SCC), total bacterial count (TBC), California Mastitis test (CMT) and microbiological culture (MC) as indicators of udder health in dairy goats in Northeastern Brazil, the leading goat milk producing region in South America. From 6 farms, a total of 396 milk samples were individually collected from each teat of 66 goats at different lactation periods (beginning, mid, end). Out of 146 (37%) positive samples, coagulase negative (CoNS, 73%) and positive *Staphylococcus* (21%), gram-positive bacilli (6%), streptococci (3.4%) and Enterobacteriaceae (1.4%) were identified. SCC and TBC were correlated ( $r = 0.47$ ;  $P < 0.01$ ) but a weak correlation (0.20;  $P < 0.01$ ) was observed between SCC and infection by CoNS infection. Although a positive association ( $P < 0.05$ ) was seen between SCC and MC using percentile 75 (6.05 log SCC/mL) as threshold, SCC was weakly (0.29) correlated with MC. Interestingly, the correlation was strong at the beginning of lactation (0.49,  $P < 0.001$ ) but negligible at mid (0.3;  $P = 0.79$ ) and end lactation (0.12;  $P = 0.14$ ). Using MC as the gold standard, sensitivity values for CMT and SCC were 39.2 and 40.2%, whereas specificity reached 80 and 89.5%. SCC in non-infected goats increased ( $P < 0.01$ ) at the end lactation. The low agreement among the diagnostic methods and the large physiological variations in SCC during lactation reinforce the limitations of the current methods to accurately predict udder health in goats, especially at the animal level. A better knowledge about the glandular tissue responses against IM agents is strongly needed. We have currently been using metagenomic approaches to investigate in-depth changes in the microbiome of naturally infected animals to bring new insights about mastitis in goats.

**Key Words:** goat mastitis, somatic cell, goat milk

**M53 Effect of FMD vaccine on seminal traits of HF bulls.** Mohua Das Gupta<sup>\*1</sup>, Shivaji Hanmantrao Sontakke<sup>1</sup>, Gunjan Rathi<sup>1</sup>, Vinod Haribhau Shende<sup>1</sup>, Mohammed Mushtaque<sup>1</sup>, Samir Kumar Dash<sup>1</sup>, Suresh B. Gokhale<sup>1</sup>, Arun P. Phatak<sup>2</sup>, Hemant Dasharath Kadam<sup>1</sup>, Narayan Laxman Phadke<sup>1</sup>, and Jayant Ramachandra Khadse<sup>1</sup>, <sup>1</sup>BAIF Development Research Foundation, Central Research Station, Uruli Kanchan, Pune, Maharashtra, India, <sup>2</sup>601 Curran Drive, Waterford, CA.

An attempt was made to study the effect of Foot and Mouth Disease vaccination on seminal traits of 30 Holstein Friesian bulls ranging between 3 and 5 years age maintained at BAIF Central Research Station Uruli Kanchan, Pune, India. Study period was from August to September 2013 and observations on semen were of bulls maintained under identical feeding and management regimens. Semen collections 15 d before vaccination, 15, 30 and 45 d after vaccination were evaluated for seminal traits such as fresh and post-thaw sperm motility, sperm concentration, semen volume, live and dead count, plasma integrity. Data generated were analyzed using ANOVA. It revealed that vaccination had significant ( $P < 0.05$ ) effect on post-thaw motility and highly significant ( $P < 0.01$ ) deleterious effect on host test, head and mid piece abnormalities as it causes derangement in spermatogenesis and epididymal function due to rise in testicular and body temperature; however, no effect was noted on seminal traits such as volume, concentration, initial motility viability and tail abnormalities. There was general decline observed in performance of seminal traits of bulls, confirming the reports of previous studies, which state that the secondary activities following vaccination remain unaltered. Therefore it can be concluded that FMD vaccination causes alteration in spermiogram but functions of accessory sex glands remain unaltered. Hence for better semen picture and to counter the effect, preventive immune enhancement before vaccination and sexual rest for highly affected bulls can be adopted.

**Table 1 (Abstr. M53).** Sperm characteristics before and after vaccination with FMD vaccine

Parameter	Pre-vaccination	Post-vaccination	P-value
Volume (mL)	6.63 ± 0.31	6.42 ± 0.17	0.59
Concentration (millions/mL)	1705.69 ± 86.09	1688.40 ± 43.29	0.88
Initial motility (%)	76.53 ± 0.38	76.3 ± 0.21	0.41
Live and dead (%)	82.89 ± 1.63	80.64 ± 1.03	0.93
Post-thaw motility (%)	58.19 ± 0.41	57.5 ± 0.24	0.03*
Host test (%)	61.53 ± 0.43	60.59 ± 0.21	0.0003**
Morphology			
Head (%)	2.78 ± 0.17	3.3 ± 0.12	0.01**
Mid (%)	1.78 ± 0.16	2.41 ± 0.11	0.0001**
Tail (%)	2.08 ± 0.18	2.19 ± 0.09	0.61

**Key Words:** FMD vaccine, seminal trait

**M54 Development of an effective oral animal vaccine using M cell targeting strategy.** Sangkee Kang<sup>\*1,2</sup>, Yoonseok Lee<sup>2</sup>, Jinduck Bok<sup>2</sup>, Chongsu Cho<sup>3</sup>, and Yunjae Choi<sup>2,3</sup>, <sup>1</sup>Graduate School of International Agricultural Technology, Seoul National University, Pyeongchang, Republic of Korea, <sup>2</sup>Institute of Green-Bio Science & Technology, Seoul National University, Pyeongchan, Republic of Korea, <sup>3</sup>Department of Agricultural Biotechnology, Seoul National University, Seoul, Republic of Korea.

Development of oral vaccine is necessary in animal husbandry field because of not only its convenience in treatment but also its capability inducing mucosal immune response. M cells are well-known as antigen collecting portals for GALT (gut associated lymphoid tissue) in intestinal tract, thus targeted delivery of vaccine molecules to the M cells could be a promising strategy to improve efficiency of oral vaccine. Previously, our research group have identified an M cell targeting peptide moiety, CKS9, by phage display technique using in vitro M cell model consisting of coculture system with Caco-2 (human colon carcinoma cells) and human Raji B cells. In this study, we constructed a recombinant lactic acid bacteria, *Lactobacillus plantarum* producing a model

antigen, M-BmpB, the BmpB (surface membrane protein originated from *Brachyspira hyodysenteriae*) conjugated with CKS9, to validate its potency as an efficient animal oral vaccine. We ascertained that the fusion protein, M-BmpB, was expressed as soluble form in the cytoplasm of *L. plantarum* by SDS PAGE and Western blot and confirmed its M cell targeting property in contrast to original BmpB (without CKS9) and P-BmpB (with unrelated peptide ligand) by in vivo closed ileal loop assay. In in vivo immunization assay (Balb/C, n = 5 in each group), Oral administration of *L. plantarum* producing M-BmpB (LP25-M-BmpB) to mice revealed significant improvement in induction of both serum IgG ( $P < 0.05$ ) and fecal IgA ( $P < 0.01$ ) against BmpB compared with control groups. Our results suggest that the recombinant lactic acid bacteria, such as *L. plantarum*, producing certain pathogenic antigen with M cell targeting strategy could have a great potential to develop an effective and convenient oral animal vaccine system.

**Key Words:** oral vaccine, M cell targeting, mucosal immunity

**M55 Amino acid supplementation and lipopolysaccharide challenge alters bovine blood polymorphonuclear leukocytes response in vitro.** M. Garcia<sup>\*1</sup>, T. H. Elsasser<sup>2</sup>, Y. Qu<sup>1</sup>, L. Juengst<sup>1</sup>, B. J. Bequette<sup>1</sup>, and K. M. Moyes<sup>1</sup>, <sup>1</sup>Department of Animal and Avian Sciences, University of Maryland, College Park, MD, <sup>2</sup>Agricultural Research Service, Animal Biosciences and Biotechnology Laboratory, U.S. Department of Agriculture, Beltsville, MD.

Glutamine is the preferred amino acid (AA) utilized by polymorphonuclear leukocytes (PMNL) during the inflammatory response. However, the effect of other AA on bovine PMNL response during inflammation and how this is altered by stage of lactation are currently unknown. The objective of this study was to determine the effect of additional AA supplementation (pool of AA excluding glutamine) on AA profile, transcriptomic, and inflammatory function of PMNL from dairy cows in early and mid-lactation in vitro. Twenty Holstein dairy cows in early (n = 10; DIM = 17 ± 3.1) and mid-lactation (n = 10; DIM = 168 ± 14.8) were used for this study. PMNL were isolated and diluted using RPMI, containing basal concentrations of glucose (7.2 mM) and amino acids (3.1 mM). Working solutions of AA (0 mM or 4 mM of AA) and LPS (0 or 50 µg/mL) were added and tubes were incubated for 2 h at 37°C and 5% CO<sub>2</sub>. Data were analyzed as a randomized block design. Stage of lactation did not alter PMNL responses in vitro. AA in combination with LPS increased ( $P \leq 0.02$ ) the concentration of alanine and methionine and tended ( $P < 0.10$ ) to increase that of leucine, isoleucine, threonine, and phenylalanine. Regardless of LPS challenge, AA supplementation downregulated ( $P < 0.05$ ) the expression of genes associated with inflammation such as *NFKB1*, *IL10*, *IL1B*, *IL6*, *TNFA*, *LYZ*, *SOD2*, and *SLC2A3* but tended ( $P < 0.10$ ) to increase the expression of *TLR6*, *G6PD*, *LDHA*, and *PDHA1*. Supplementation of AA reduced the concentration of TNF- $\alpha$  (104.0 vs. 34.9 ng/mL,  $P = 0.01$ ) in medium but did not affect chemotaxis and phagocytic functions of PMNL. Metabolic profiles for cows in early lactation did not parallel those for cows during the early postpartum period and may partly explain the lack of stage of lactation effects. This study identified AA other than glutamine that may alter PMNL response during inflammation in vitro that may lead to new avenues to improve immune response during inflammation in vivo.

**Key Words:** amino acid, cow, polymorphonuclear leukocyte

**M56 In vitro activity of *Pithecellobium dulce* and *Lysiloma acapulcensis* on exogenous development stages of sheep gastrointestinal strongyles.** A. Olmedo-Juárez<sup>1</sup>, R. Rojo-Rubio<sup>\*1</sup>, J. Arece-García<sup>2</sup>, C. Marie-Magdeleine<sup>3</sup>, and J. F. Vázquez-Armijo<sup>1</sup>,

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Parasitic nematodes of the digestive tract remain one of the main constraints in small ruminants in subtropical countries. In these regions there are a lot of browsed plant species with antihelminthic activity; in this way an experiment was conducted to evaluate the effects of 2 lyophilised aqueous extracts of *Lysiloma acapulcensis* (LAE) and *Pithecellobium dulce* (PDE) tree leaves on in vitro assessment of hatching of eggs, larval development and migration of gastrointestinal nematodes of sheep using a general linear model. Treatments contained extracts from both species at concentrations of 0, 125, 250 and 500 µg/mL. Both albendazole and levamisole were used at a level of 1% as positive control. The extract of LAE, compared with PDE, showed better inhibition ( $P < 0.05$ ) of egg hatching. Different doses of both the LAE and PDE extracts showed a larvicidal effect ( $P < 0.05$ ) on all larvae exposed to different doses of the extracts. In the larval migration assay, a similar effect with levamisole occurred with the LAE extract at doses of 250 and 500 µg/mL. The extract of *P. dulce* had a lower larvicidal effect ( $P < 0.05$ ) than levamisole and *L. acapulcensis* extracts. The use of aqueous extract of *L. acapulcensis* could be a promising alternative to synthetic anthelmintics as treatments of gastrointestinal nematodes of sheep in organic and conventional production systems under subtropical conditions.

**Key Words:** anthelmintic, extract, sheep

**M57 Changes in transcriptome of bovine monocytes-derived macrophage challenged with *Mycobacterium bovis*.** Dianelys Gonzalez-Pena<sup>\*</sup>, Robmay Garcia, and Andrew J. Steelman, and Sandra L. Rodriguez-Zas, University of Illinois at Urbana-Champaign, Urbana, IL.

Macrophages pertain to the mononuclear phagocytes system as part of the innate and acquired immunity responses. The interaction of macrophages with *Mycobacterium bovis* concludes with the formation of granulomas. However, the host-immune response to *M. bovis* could be compromised by the suppression of immuno-regulatory pathways leading to active tuberculosis. Therefore, transcriptome analysis of the macrophage response to *M. bovis* can offer insight into the host-pathogen interaction. Bovine monocyte derived macrophage (MDM) of peripheral blood was obtained from 7 non-infected Holstein cows, cultivated and infected with *M. bovis*. The RNA was isolated 24 h after infection and single-end reads were mapped to the *Bos taurus* (UCSC\_bosTau7) reference genome using Tophat v2.0.12. Using Cufflink v2.2.1, 7,664 transcripts from 7,505 genes were tested and 1,192 transcripts from 1,187 genes were differentially expressed between the infected and non-infected MDM (false discovery rate adjusted  $P$ -value  $< 0.05$ ). Among the transcripts, 61% were upregulated in the infected relative to non-infected MDM. Interferon gamma (IFNG), indoleamine 2,3-dioxygenase 1 (IDO1), chemokine (C-X-C motif) ligand 10 (CXCL10), and chemokine (C-X-C motif) ligand 9 (CXCL9) were overexpressed in infected relative to non-infected MDM. The IFNG is a potent activator of macrophages, while IDO1 encoded an important rate-limiting enzyme in the kynurenine pathway, activated during the inflammation process that favors immune suppression and tolerance. The CXCL9 and CXCL10 are critical during the early stages of the immune response to *M. bovis*. The top 5 ranking category clusters detected by the functional analysis of the differentially expressed genes using DAVID (enrichment score  $> 5$ ) included immune defense, and the activation and proliferation of leukocyte, lymphocyte, mononuclear cells, and T and B cells as well

as cytokines production and apoptosis regulation. This study unveils details of the host response of MDM to *M. bovis*.

**Key Words:** transcriptome, macrophage, *Mycobacterium*

**M58 Phytochemicals in corn distillers grains.** Adebola Dar-amola and Byungrok Min\*, *University of Maryland Eastern Shore, Princess Anne, MD.*

Corn is a major feedstock for the production of fuel ethanol as well as corn distillers grains that are widely used for animal feed. Corn is one of the most abundant sources of health-beneficial phytochemicals among crops. Little information is, however, available for phytochemicals in corn distillers grains. The objective was to determine amounts, availability, and antioxidant capacities of phytochemicals in corn distillers grains: dried distillers grains with solubles (DDGS), wet or modified wet distillers grains (WDG/MWDG), and condensed distillers solubles (CDS), compared with corn. Freeze-dried samples were extracted with 80% ethanol to obtain soluble phenolics. Residues were alkali-treated and extracted with ethyl acetate for cell-wall-bound phenolics. Amounts (total phenolic and flavonoid contents) and antioxidant capacities (DPPH radical scavenging, oxygen radical absorbance, hydroxyl radical averting, and iron chelating capacities) in both phenolics were evaluated. Phenolic acids and carotenoids were also identified and quantified using HPLC. Data were analyzed by ANOVA and sample means were compared using SNK multiple range test. Amounts and antioxidant capacities of soluble phenolics were the highest in DDGS, followed by CDS, WDG/MWDG, and corn ( $P < 0.05$ ). Those of cell-wall-bound ones were the highest in DDGS, followed by WDG/MWDG, corn, and CDS ( $P < 0.05$ ). Ferulic acid was the predominant phenolic acid (~90%) and its amount was the highest in DDGS ( $P < 0.05$ ). Carotenoid content was the highest in DDGS, followed by WDG/MWDG, CDS, and corn ( $P < 0.05$ ). Total phenolic, phenolic acid, and carotenoid contents in DDGS were over 3 times higher than in corn. It is known that, due to conversion of starch to ethanol and CO<sub>2</sub>, macronutrients in corn are generally concentrated into DDGS over 3 times. The results indicate that phytochemicals in corn are well concentrated during the processing, thus DDGS have great potential as a source of phytochemicals to improve farm animal health and performance. However, cell-wall-bound phenolic contents in DDGS were 2–5 times higher than those of soluble ones, indicating that the majority of phenolics in DDGS are not readily absorbable. Hence, technologies to liberate them are needed to maximize health-beneficial potentials of DDGS.

**Key Words:** phytochemical, DDGS, corn

**M59 Characterization of the binding potential of pathogenic bacteria to yeast probiotics and paraprobiotics.** Janet R. Donaldson\*<sup>1</sup>, Gabe Posadas<sup>1</sup>, Jeffery A. Carroll<sup>2</sup>, Paul R. Broadway<sup>2</sup>, Amanda Lawrence<sup>1</sup>, and Jimmie Corley<sup>3</sup>, <sup>1</sup>Mississippi State University, Mississippi State, MS, <sup>2</sup>USDA-ARS, Lubbock, TX, <sup>3</sup>Phileo, Lesaffre Animal Care, Milwaukee, WI.

Probiotics and their associated derivatives (paraprobiotics) are frequently utilized to improve animal health and productivity. However, their mechanisms of action are not fully characterized, especially in regards to the interactions with pathogenic bacteria in the gastrointestinal tract. This study tested the hypothesis that yeast probiotics and paraprobiotics directly interact with pathogenic bacteria differently. To test this hypothesis, the binding capability of 5 different yeast probiotics or paraprobiotics to gram-negative bacteria (*Salmonella* sp. and *Escherichia coli* O157:H7) and gram-positive bacteria (*Listeria monocytogenes* and

*Clostridium* sp.) were analyzed. Yeast and bacteria were co-incubated on coverslips, washed extensively, and examined by scanning electron microscope to determine the extent of binding between products and pathogens. Membrane filtration was also used to quantitate the amount of bacteria capable of binding to the yeast product; yeast products and bacteria were co-incubated, filtered using 3µM membrane filters, and the resulting filtrates were assessed for viable bacteria by plate counts. All bacteria tested bound with nearly equivalent efficiencies ( $P > 0.05$ ) against the live yeast probiotics tested (~26%). However, much variation was observed in the binding efficiencies with the paraprobiotics. The gram-positive bacteria had, as a group, a preference for binding to one paraprobiotic in comparison to the other 2 products analyzed (25% adhered vs. 47% adhered;  $P < 0.05$ ), whereas the gram-negative bacteria had greater efficiency to bind to 2 paraprobiotics (40% adhered;  $P < 0.001$ ). These data suggest that the use of probiotics and paraprobiotics as therapies needs to be specific to the pathogen of interest; thus indicating a need for “designer” probiotic/paraprobiotic feeding strategies. Further research is needed to analyze specific binding efficiencies of probiotics and paraprobiotics against infectious agents in vivo.

**Key Words:** probiotic, bacteria, paraprobiotic

**M60 Effects of polybrene and puromycin on equine infectious anemia virus replication.** Dustin A. Therrien, Rebecca D. Parr, and Sarah C. Canterberry\*, *Stephen F. Austin State University, Nacogdoches, TX.*

Equine infectious anemia virus (EIAV) is a lentivirus that infects members of the family Equidae. Despite extensive study, there is no method for prevention or treatment of this disease. Novel mechanisms, such as RNA interference (RNAi), have been used to decrease replication of many viruses. Results of previous in vitro studies have indicated that in the process of generating transgenic cells, EIAV replication was inhibited. Thus, these data were inconclusive as to the efficacy of RNAi against EIAV. To further investigate the effectiveness of RNAi, it is essential to discern which aspects of generating transgenic cell lines affects viral replication. Here, we have investigated 2 common reagents: polybrene (Pol), a polycationic polymer used to increase initial cell transduction, and puromycin (Pur), an antibiotic selecting agent. Non-transgenic feline adenocarcinoma cells, persistently infected with EIAV<sub>19</sub> (a laboratory adapted strain of the virus), were exposed to high and low concentrations of Pol (8 and 16 µg/mL), and Pur, (1 and 2 µg/mL). Control cell lines were exposed to neither reagent. Supernatants were collected for 17 d and viral replication was quantified using a reverse transcriptase (RT) Assay. The RT values were analyzed using R (R, v. 2.15.3, R Development Core Team, 2012) to generate 9999 permutations of the data. Mean RT values (Table 1) for experimental groups were not significantly different from the mean RT values for the control groups. These data indicate that Pol and Pur alone have no effect on viral replication in these cells. Additional studies are needed to determine which reagents used in establishing transgenic cells is responsible for the observed decreases in viral replication.

*Contd.*



**Table 1 (Abstr. M60).** Mean RT values per 100,000 cells per day of viral accumulation

Day	Control	Pol High	Pol Low	Pur High	Pur Low
2	3.2E6 ± 0.5E6	3.8E6 ± 0.4E6	4.9E6 ± 0.9E6	13.6E6 ± 0.4E6	13.4E6 ± 4.9E6
4	12.7E6 ± 2.7E6	13.7E6 ± 1.5E6	15.2E6 ± 1.3E6	720.6E6 ± 341.6E6	59.1E6 ± 21.8E6
7	8.6E6 ± 1.7E6	13.8E6 ± 2.5E6	17.4E6 ± 3.3E6	527.1E6 ± 210.1E6	21.6E6 ± 5.9E6
9	27.9E6 ± 1.8E6	27.7E6 ± 1.5E6	28.4E6 ± 6.5E6	529.4E6 ± 104.4E6	27.4E6 ± 1.5E6
13	9.7E6 ± 0.8E6	12.0E6 ± 3.1E6	9.9E6 ± 0.9E6	215.0E6 ± 13.5E6	6.5E6 ± 0.8E6
17	5.2E6 ± 1.0E6	7.0E6 ± 1.7E6	6.4E6 ± 0.8E6	198.6E6 ± 11.7E6	4.2E6 ± 0.3E6

**Key Words:** equine infectious anemia virus (EIAV), RNA interference (RNAi)

**M61 Co-aggregation ability of cell wall components of *Saccharomyces cerevisiae* to pathogenic bacteria.** Marlén Rodríguez<sup>1</sup>, Ana Julia Rondón<sup>1</sup>, Yadileiny Portilla<sup>1</sup>, Ramón Bocourt<sup>2</sup>, María José Ranilla<sup>3,5</sup>, María Dolores Carro<sup>4</sup>, Alexey Diaz<sup>3,5</sup>, and Grethel Milián<sup>1</sup>, <sup>1</sup>Center for Biotechnological Studies, University of Matanzas, Matanzas, Cuba, <sup>2</sup>Institute of Animal Science, Mayabeque, San José de las Lajas, Cuba, <sup>3</sup>Animal Production Department, University of León, León, Spain, <sup>4</sup>Agriculture Production Department, Technical University of Madrid, Madrid, Spain, <sup>5</sup>5IGM (CSIC-ULE), Finca Marzanas s/n, Grulleros, León, Spain.

Autoaggregation in bacteria is the phenomenon of aggregation between cells of the same strain, whereas coaggregation is due to aggregation occurring among different species. Aggregation ability of prebiotic bacteria is related to adhesion ability, which is a prerequisite for the colonization and protection of the gastrointestinal tract in all animal species; however, coaggregation ability of prebiotic bacteria offers a possibility of close interaction with pathogenic bacteria. Coaggregation ability of cell wall components of *Saccharomyces cerevisiae* is known, because of their mannan content, but literature offers little information on this topic. The aim of this experiment was to assess the ability of coaggregation of 2 preparations of *S. cerevisiae* cell walls to 3 pathogenic bacteria (*Staphylococcus aureus* hemolytic enterotoxin A, *Salmonella enteritidis* and *Escherichia coli* serotype O157:H7). Cell wall preparations consisted on either the distillery cream (DT), a byproduct of sugar cane, or a hydrolyzate (HT) obtained by enzymatic methods. Pathogens were grown in nutritive broth medium for 18 h at 37°C. After that, cultures were diluted (1:1) with DT and HT, and absorbance (560λ) was measured at 0 and 5 h. Both DT and HT showed the ability of coaggregate to the 3 pathogenic strains, and no bacterial strain × cell wall preparation interaction ( $P = 0.379$ ) was detected. Coaggregation was higher ( $P < 0.001$ ; SEM = 0.36) with HT (mean values of 85.3,

78.6 and 77.8% for *S. aureus*, *S. enteritidis*, and *E. coli*, respectively) compared with DT (mean values of 16.5, 5.8 and 6.0% for *S. aureus*, *S. enteritidis*, and *E. coli*, respectively). If confirmed with other pathogen species, these results support further research on the use of the HT from *S. cerevisiae* as a possible prebiotic additive for animal feed.

**Key Words:** coaggregation, pathogenic bacteria, *Saccharomyces cerevisiae*

**M62 In vitro efficacy of chitosan against *Cryptosporidium parvum* and validation on infected goat kids.** Karim Adjou<sup>1</sup>, Jean-Philippe Marden<sup>2</sup>, Eric Auclair<sup>2</sup>, Christian Mage<sup>3</sup>, and Isabelle Vallée<sup>1</sup>, <sup>1</sup>UMR BIPAR Anses-ENVA, Maisons-Alfort, France, <sup>2</sup>Phileo Lesaffre Animal Care, Marcq en Baroeul, France, <sup>3</sup>Mage Consultant, Estivaux, France.

The aims of this study were to investigate (1) the efficacy of chitosan in 2 forms, the monomer *N*-acetyl glucosamine (NAG) and a chloride salt of chitosan (MIX) in culture systems HCT-8 and Caco-2 cell lines in vitro for *Cryptosporidium parvum* compared with a positive control, paromomycin (PARO) a classical drug used in veterinary medicine; (2) the action of a chitosan-yeast-bacteria based product on neonatal diarrhea and mortality in goat kids. Cryptosporidiosis is considered as an economically important disease with clinical signs and death in young ruminants. The usual clinical symptom is acute diarrhea affecting animals from 1 to 3 weeks old. As no drugs are fully effective in the treatment of cryptosporidiosis in man and animals, the research for new therapeutic agents is crucial. Chitosan is a sugar that is obtained from the hard outer skeleton of shellfish, including crab and shrimp and it is used in medicine. It has been found to be active against a variety of diseases including antimicrobial and anti-tumoral effects. Immunofluorescence technique was used for the identification and enumeration of the parasites. The results showed a significant reduction of viability of *Cryptosporidium* oocysts (>95%) after pre-incubation of 24h at 37°C with PARO ( $P < 0.001$ ), MIX and NAG ( $P < 0.001$ ). Additionally, PARO, MIX and NAG inhibited significantly the development of *C. parvum* in HCT-8 and Caco-2 cell lines ( $P < 0.005$ ). These effects were dose-dependent. Synergistic effects were obtained when NAG treatment was associated with Paromomycin. The efficacy of MIX in combination with yeast and bacteria (Optisaf FIRST, Phileo, France) was evaluated experimentally in goat neonates inoculated with *C. parvum* oocysts (10<sup>6</sup> oocysts/mL) per oral route. Preliminary results showed a significant reduction in oocyst shedding and diarrhea score in goat kids and mortality was significantly reduced (36%) in treated animals ( $P < 0.05$ ) compared with the control group (90%). In conclusion, these findings provide evidence of in vitro inhibitory activities of chitosan against *C. parvum* and its combination with yeast-based products revealed promising in lessening the incidence of neonatal diarrhea in young ruminants.

**Key Words:** chitosan, yeast, goat



## Bioethics

**M63 US consumer perceptions of and willingness to pay for “local” pork chops and chicken breasts.** Elizabeth Byrd\*, Nicole Widmar, and Michael Wilcox, *Purdue University, W. Lafayette, IN.*

As the debate continues about what animal agriculture “should” look like, it is important to understand what consumers perceive attributes and buzzwords such as “local” to mean. In our study, when asked how they defined “local” food, 37% of US consumers described local food as having been produced within 10 miles of their home, 21% described local to mean within 20 miles of their home, and 17% described local food as having been produced within the state. Several demographics were found to be correlated with perceptions of “local”; being male is correlated with defining local food as having been produced within 20 miles of home, whereas living in the Northeast is positively correlated with defining local food as produced within 10 miles of home. Households earning less than \$40,000 more often defined local food as having been produced within 10 miles of home. Having visited a beef or dairy farm in the last 5 years is negatively correlated with defining local food as having been produced within 10 miles of home. Expressing concern for the welfare of beef or dairy cattle is positively correlated with defining local food as having been produced within 100 miles of home. Are consumers willing to pay (WTP) for verified locally grown pork chops and chicken breasts? Preliminary results of a simulated purchasing scenario indicate that consumers are WTP up to \$2.02/lb. for USDA-verified locally produced chicken breasts, but are not WTP for verified locally produced pork chops. Is it possible consumers feel differently about pigs and chickens? But, when asked, 37% and 38% of consumers express concern for the welfare of farmed pigs and chickens, respectively. Do consumers exhibit more trust in different verification agencies? While consumers were WTP \$2.02/lb. for the USDA to verify local production in chicken breasts, those same consumers were only WTP \$0.37 for the poultry industry to verify that chicken breasts were produced locally. Knowledge about consumer perceptions of local production can help the animal industries make decisions about marketing livestock products.

**Key Words:** consumer demand, local food, preferences

**M64 US consumer awareness of animal diseases and animal health requirements: Are we meeting the demands of the US public?** Elizabeth Byrd\*, Nicole Widmar, and John Lee, *Purdue University, W. Lafayette, IN.*

Have various segments of a representative sample of US consumers heard of prominent animal diseases such as bovine spongiform encephalopathy (BSE), porcine epidemic virus diarrhea (PEDV), tuberculosis (TB), chronic wasting disease (CWD), leptospirosis, rabies, and food and mouth disease (FMD)? We have found that 30% of respondents reported having heard of BSE, 17% heard of PEDV, and 27% heard of CWD. Various demographic factors have been found, through cross-tabulations, to be associated with the awareness of animal diseases. Being a college graduate, someone who regularly hunts, a pet owner, or a member of HSUS or PETA were all positively correlated with having heard of BSE, PED-v, and Leptospirosis. Reporting concern for the welfare of beef cattle, dairy cattle, or pigs was positively correlated with being aware of all diseases analyzed. Do people who have recently visited farms raising cattle, deer, or pigs show an increased awareness for their associated diseases? Having visited a beef cattle farm in the

last 5 years was positively correlated with animal disease awareness. Visiting a dairy or pig farm in the past 5 years was positively associated with awareness of BSE, PED-v, CWD, and Leptospirosis. Consumers who raise their own food, either through gardening, raising chickens for eggs or meat, or raising animals for meat, are more likely to be aware of BSE and PEDV. Related to disease awareness is inspection; respondents were asked about veterinary inspection for transport. Sixty-eight percent of respondents agreed that livestock should be required to undergo a veterinary inspection before being transported within or outside the state. Likewise, 75% of US consumers who were aware of BSE agree that beef carcasses should be tested for BSE before being used for human food consumption. Understanding the demographics the predispose consumers to be more aware and sensitive to animal disease/health issues is important for animal industry communication to consumers to ensure consumer confidence in the food they choose to purchase.

**Key Words:** consumer demand, preference

**M65 Consumers know food, but they don’t know ag (how does that work?).** Ann Cummins\*, Nicole Widmar, Candace Croney, and Joan Fulton, *Purdue University, W. Lafayette, IN.*

This research examined a gap in some consumers’ minds between being educated food consumers and being educated about agriculture. This research uses data from a survey that is representative of national household consumers in terms of age, sex, income, and region of residence, with a sample size of 1004. This research identified that to some consumers, being educated about food consumption did not necessarily mean being educated about agriculture. Results include that 37% of people thought they were educated about agriculture but 76% thought they were educated about food. Food consumption and agriculture are connected to one another, but how are we as food providers communicating if in many consumer minds being educated about food and educated about agriculture is not the same. This research examines individuals self-reported level of education about food consumption and education level about agriculture. We examine the differences in these self reported values and demographics, knowledge and perception about as livestock production, and personal consumption behaviors. This research examines the demographic, educational, and perceptions of livestock operation differences that are associated with the different groups, including, those who self-identified as more educated about agriculture than food, those who self-identified as very educated about both food and agriculture, and those who self identified as being more educated about agriculture than food. The methods used for this analysis were both Spearman and Pearson correlations, cross tabs and z-scores (all completed in SPSS). The majority of respondents, 51%, self-reported being more educated about food consumption than agriculture, 2.7% of respondents indicated that they were more educated about agriculture than about food consumption, and 35% of respondents indicated that they consider themselves to be highly educated about both food consumption and about agriculture. Of those who reported to be more educated about food consumption than agriculture, we see that they tend to be female, older in age, not a pet owner, and not visiting tourist attractions.

**Key Words:** consumers, education, perceptions of agriculture

**M66 Ergonomic assessment of the milking routine in large dairy operations of the US Southwest.** Jose A. Garcia Buitrago\*<sup>1</sup>, Gerrit R. Hagevoort<sup>1</sup>, David Gimeno<sup>2</sup>, and David I. Douphrate<sup>2</sup>, <sup>1</sup>*New Mexico State University, Clovis, NM*, <sup>2</sup>*University of Texas, San Antonio, TX*.

The US dairy industry has experienced a rapid transformation from small dairy farms to large operations. In southwest regions the majority of commercial dairy herds are large operations. Current trends are larger herds where a great number of cows are attended for few employees. Reasons for these trends are beyond the scope of this abstract. This transformation has led to significant changes in work tasks and in ergonomic challenges due to the repetitive work nature of the milking process. Despite the improvement in modern equipment and design of milking parlors, it seems that the musculoskeletal workload of dairy workers remains high. Field-based direct measures of physical exposures have been limited in these challenging work environments. To assess quantitatively the posture, muscle activity and muscular fatigue a throughout milking shift, we have recruited 30 milking parlor workers representing large-herd (1000+ head) dairy operations in 3 southwest US states. All

participants were Latino males between 21 and 45 years of age, who were free of pain or upper extremities pathologies. Shoulder elevation and the trunk inclination angles, were estimated using wireless triaxial accelerometers. Surface electromyography was continually recorded of the upper trapezius, finger flexors, finger extensors and anterior deltoid muscles, while workers performed milking tasks. These muscles were chosen for their relevance when performing milking tasks. Preliminary findings suggest milking workers may be exposed to awkward postures, high velocity and repetitive movements, high repetition, and high muscle forces. These physical exposures have been often associated with the development of upper limb pathology. These results warrant the need for continued research in these working environments, for development of ergonomic strategies that permit improving work conditions and to reduce the negative effect of postures during the milking routine on the musculoskeletal system, as well as to prevent fatigue that would impair dairy workers performance.

**Key Words:** milking routine, ergonomic, electromyography

# Breeding and Genetics: Application and methods in animal breeding— Swine, poultry, and other species

**M67 Accuracy of estimated breeding values for males and females with genomic information on males, females, or both: A broiler chicken example.** Daniela A. L. Lourenco\*<sup>1</sup>, Breno O. Fragomeni<sup>1</sup>, Shogo Tsuruta<sup>1</sup>, Ignacio Aguilar<sup>2</sup>, Birgit Zumbach<sup>3</sup>, Rachel J. Hawken<sup>3</sup>, Andres Legarra<sup>4</sup>, and Ignacy Misztal<sup>1</sup>, <sup>1</sup>University of Georgia, Athens, GA, <sup>2</sup>INIA, Las Brujas, Uruguay, <sup>3</sup>Cobb-Vantress Inc., Siloam Springs, AR, <sup>4</sup>INRA, Castanet-Tolosan, France.

Phenotypes were available on 4 production traits recorded for up to 196,613 broiler chickens. Heritabilities ranged from 0.22 to 0.49. Among all phenotyped birds, 15,723 were genotyped for 39,102 segregating SNP. Traditional and genomic evaluations were run in a multiple-trait model. Single-step genomic BLUP (ssGBLUP) was used for genomic evaluations with 3 different reference sets including only males (4648), only females (8100), and both sexes (12,748). Realized accuracy of genomic EBV (GEBV) was used to evaluate the inclusion of genotypes for different reference sets on predictive ability of young genotyped males (1501), females (1474), and both sexes (2975). Using male genotypes as reference, the average increase in accuracy of GEBV over EBV for males and females was 12 and 1 percentage point, respectively. When the reference population included only female genotypes, the increase for males and females was 1 and 18 percentage points, respectively. Using genotypes on both sexes as reference increased accuracies by 19 points for males and 20 points for females. Adding genotypes for females without phenotypes did not improve predictions. For one trait, EBV and GEBV accuracies for females were much lower than for males. For another trait with similar heritability, both accuracies were higher, and females had higher accuracy than males. For validation animals  $GEBV \approx w_1PA + w_2DGV$ , where PA is parent average, DGV is genomic prediction, and the  $w$  terms are the weights for each component. When the number of genotyped animals is high, the highest weight is for  $w_2$ . When an animal is genotyped, the increase in accuracy comes mainly from the DGV portion of GEBV and marginally from improved PA. For non-genotyped animals there is no improvement in accuracy due to DGV. Accuracies for animals of one sex increase with genotypes of the other sex when that sex has independent phenotypic information and when the evaluation methodology avoids double counting. Realized accuracies are biased down by selection, and analysis of realized accuracies can reveal different selection pressure for traits and sex.

**Key Words:** genomic prediction, genotyping strategy

**M68 Genetic parameters for length of productive life and lifetime production traits of purebred Landrace and Yorkshire sows in northern Thailand.** Udomsak Noppibool\*<sup>1,2</sup>, Skorn Koonawootrittriron<sup>1</sup>, Mauricio A. Elzo<sup>2</sup>, and Thanathip Suwanasopee<sup>1</sup>, <sup>1</sup>Kasetsart University, Chatuchak, Bangkok, Thailand, <sup>2</sup>University of Florida, Gainesville, FL.

The objective of this research was to estimate genetic parameters and trends for length of productive life (LPL), lifetime piglets born alive (LBA), lifetime piglets weaned (LPW), lifetime piglets' birth weight (LBW), lifetime piglets' weaning weight (LWW) in a commercial swine farm in Northern Thailand. Phenotypic records came from 1,983 Landrace (L) and 745 Yorkshire sows (Y) collected from July 1989 to August 2013. Variance and covariance components, heritabilities and correlations were estimated using a multiple-trait AIREML procedure. The

5-trait mixed animal model contained the fixed effects of first farrowing year-season, breed group (L and Y), and age at first farrowing. Random effects were sow and residual. Medium heritabilities were estimated for all 5 traits (LPL =  $0.16 \pm 0.04$ ; LBA =  $0.18 \pm 0.04$ ; LPW =  $0.22 \pm 0.04$ , LBW  $0.18 \pm 0.04$  and LWW =  $0.22 \pm 0.04$ ). Genetic correlations among these traits were positive and favorable (greater than 0.91;  $P < 0.05$ ). Genetic correlation estimates were  $0.94 \pm 0.02$  for LPL-LBA,  $0.98 \pm 0.03$  for LPL-LPW,  $0.92 \pm 0.03$  for LPL-LBW,  $0.93 \pm 0.02$  for LPL-LWW,  $0.96 \pm 0.01$  for LBA-LPW,  $0.96 \pm 0.01$  for LBA-LBW,  $0.93 \pm 0.02$  for LBA-LWW,  $0.93 \pm 0.02$  for LPW-LBW,  $0.97 \pm 0.01$  for LPW-LWW and  $0.94 \pm 0.02$  for LBW-LWW. Dam genetic trends were positive, small and significant only for LBA ( $0.18 \pm 0.05$  piglets/yr;  $P = 0.0024$ ), LPW ( $0.12 \pm 0.05$  piglets/yr;  $P = 0.0153$ ), LBW ( $0.35 \pm 0.09$  kg/yr;  $P = 0.0009$ ), and LWW ( $1.36 \pm 0.40$  kg/yr;  $P = 0.0024$ ). Genetic trends for sows and sires were mostly small, negative and not significant for any trait. Thus, the selection program in this commercial herd was ineffective to improve LPL in sows, sires, and dams, and lifetime production traits in sows and sires. This program was only effective to improve lifetime productive traits in dams.

**Key Words:** genetic parameter, lifetime production trait, swine

**M69 A study on *PIT1* gene polymorphism and its association with growth traits in pigs.** S. Mohana Devi\*<sup>1</sup>, V. Balachandar<sup>2</sup>, and I. H. Kim<sup>1</sup>, <sup>1</sup>Department of Animal Resource & Science, Dankook University, Cheonan, Chungnam, South Korea, <sup>2</sup>Department of Human Genetics and Molecular Biology, Bharathiar University, Coimbatore, Tamil Nadu, India.

The pituitary transcription factor (PIT1) (or POU1F1) protein belongs to pituitary-specific transcription factors localized on chromosome 13 in porcine. The PIT1 gene was used as a candidate gene for selecting animals for growth and carcass traits. Thus, the aim of this study was to identify and compare the Rsa I polymorphism of the PIT1 gene in finishing pigs to evaluate mutations in the meat quality of porcine by screening to novel gene markers PIT-1 and to investigate the genotypic alteration of the pigs, which may help to locate the chromosomal regions that may be linked with candidate genes that are associated with pig muscle growth and pork quality. The genomic DNA from 460 pigs was extracted and the genotypes for polymorphism analysis were determined using PCR-RFLP. The PIT1 genotype frequencies analyzed were 42.39% for AA, 39.13% for AB and 18.48% for BB. Several significant associations of PIT1 gene polymorphisms with some of the growth traits were observed. The pigs with BB genotype showed the highest average daily gain. Several significant differences were observed in meat color (A, B and L\*;  $P < 0.05$ ). Meat carcass traits LMP and LMC seem to be lower in pigs with BB genotype compared with AA genotype animals. The AA finishers presented a lower level of back fat thickness compared with AB genotypes. The PIT1 are potential candidate genes, influencing quality traits as they encode proteins, which causes essential effects in its functions and the present study tries to confirm the association of its polymorphism with its economically important traits in pigs. Therefore the assumption that genetic unpredictability of the PIT1 gene could be associated with swine population in commercial traits.

**Key Words:** *PIT1* gene, growth trait, pig



**M70 Recombination rates in layer chickens.** Zi-Qing Weng<sup>\*1</sup>, Anna Wolc<sup>1,2</sup>, Rohan L. Fernando<sup>1</sup>, Jack C. M. Dekkers<sup>1</sup>, Jesus Arango<sup>2</sup>, Petek Settar<sup>2</sup>, Janet E. Fulton<sup>2</sup>, Neil P. O'Sullivan<sup>2</sup>, and Dorian J. Garrick<sup>1</sup>, <sup>1</sup>*Department of Animal Science, Iowa State University, Ames, IA*, <sup>2</sup>*Hy-Line International, Dallas Center, IA*.

Recombination events, which occur during meiosis, vary in frequency across chromosomes, and among individuals. Recombinations are more common in certain genomic locations known as hotspots and these are controlled by genes. The objective of this study was to assess recombination events across the genome, and identify quantitative trait loci (QTL) that influence recombination frequency in white and brown layer chickens. This study included 1,200 white layers hatched between 2006 and 2012, genotyped with a 600K single nucleotide polymorphisms (SNP) panel, and 5,108 brown layers hatched between 2003 and 2011, genotyped with a 40K SNP panel. FImpute was used to impute missing genotypes. After quality control, 173,224 and 23,098 segregating SNPs remained. There were 492 half-sib families in white layers averaging  $3.0 \pm 3.0$  birds, and 1717 half-sib families averaging  $5.4 \pm 4.9$  brown layer birds. Recombinations were identified within half-sib families using LINKPHASE. Total recombination rates within each 1-Mb window was calculated across 28 chromosomes (Chr). Windows with recombination rates greater than 0.03 ( $\geq 1.5$  SD from the mean) were considered to be recombination hotspots, while those with no recombinations were cold-spots. Genome-wide recombination numbers of parents were analyzed in a weighted BayesB model. Windows that explained  $>1\%$  genetic variance were considered to harbor QTL. There were 14,746, and 230,701 recombination events detected in white layers and brown layers, respectively. There were 163 and 281 windows with hotspots detected in white and brown layers, respectively, of which 66 were in common. There were 48 common cold-spots in these 2 breeds. Genome-wide recombination number (GRN) differed by breed and sex. White layers ( $10.9 \pm 4.1$ ) had smaller GRN than brown layers ( $24.1 \pm 3.9$ ). In white layers, females ( $13.6 \pm 3.7$ ) had higher GRN than males ( $9.3 \pm 3.5$ ) but in brown layers GRN were similar in females ( $24.2 \pm 4.2$ ) and males ( $23.9 \pm 3.4$ ). A total of 14 and 6 significant windows, which harbor candidate genes influencing genome-wide recombination, were detected in the 2 breeds. No common QTL windows were found in the 2 breeds. Sample size, marker density, inbreeding level and population structure lead to differences in detection of recombination events and QTL in the 2 breeds.

**Key Words:** recombination, layer chicken

**M71 Genetic selection tool for number born alive and stillbirth piglets in commercial Thai populations.** Thanathip Suwanasopee<sup>\*1</sup>, Skorn Koonawootrittriron<sup>1</sup>, and Mauricio A. Elzo<sup>2</sup>, <sup>1</sup>*Department of Animal Science, Faculty of Agriculture, Kasetsart University, Bangkok, Thailand*, <sup>2</sup>*Department of Animal Sciences, University of Florida, Gainesville, FL*.

Genetic markers are widely used for selection and prediction in commercial pig populations. Number of piglets born alive (NBA) and stillborn piglets (STB) within litters have a major effect on overall economic returns. The aim of this study was to determine the association between estrogen receptor gene (*ESR*) and paternally expressed gene 1 (*PEG1*) and NBA and STB. The data set included information from 2 commercial pig populations, one in Northern Thailand (210 sows; 123 Landrace and 87 Yorkshire) and another one in Northeastern Thailand (130 sows; 70 Landrace and 60 Large White). Each population was analyzed separately using 2 models. Model 1 included farrowing year-season, parity, age at first farrowing and breed group as fixed effects, and animal and

residual as random effects. Model 2 included all effects from model 1 plus genotypes for *ESR* (AA, AB, and BB) and *PEG1* (AA, GA, and GG) as fixed effects. Association between predicted EBV from models 1 and 2 were analyzed using Spearman rank correlations. Genotype frequencies were 0.67, 0.29 and 0.04 for *ESR* and 0.01, 0.40, and 0.59 for *PEG1* in the Northern population, and 0.42, 0.34, and 0.24 for *ESR* and 0.05, 0.25, and 0.70 for *PEG1* in the Northeastern population. The *ESR* genotype influenced the EBV of Yorkshire sows for NBA ( $P < 0.05$ ) and STB ( $P < 0.01$ ). Yorkshire *ESR* AA sows had the highest average EBV for NBA ( $0.16 \pm 0.03$  piglets), whereas Yorkshire *ESR* AB sows had the lowest average EBV for STB ( $-0.01 \pm 0.01$  piglets). The *PEG1* genotype affected the EBV of Landrace, Yorkshire and Large White sows for NBA and STB ( $P < 0.05$ ). Large White *PEG1* GG sows had the highest average EBV for NBA ( $0.43 \pm 0.06$  piglets), and Landrace *PEG1* AA sows had the lowest average EBV for STB ( $-0.09 \pm 0.09$  piglets). Positive rank correlations between EBV from models 1 and 2 existed in the 2 populations for all breeds (0.41 to 0.63;  $P < 0.01$ ), except for Landrace sows in the Northern population ( $-0.08$ ;  $P > 0.05$ ). These rank correlations indicated that selecting sows with higher NBA and lower STB by using EBV from models with and without *ESR* and *PEG1* marker effects would likely yield similar outcomes.

**Key Words:** pig, genetic marker, selection

**M72 Genomic correlation between piglet preweaning mortality and individual birth weight using a bivariate threshold-linear maternal effect model.** Shogo Tsuruta<sup>\*1</sup>, Ching-Yi Chen<sup>2</sup>, William O. Herring<sup>2</sup>, and Ignacy Misztal<sup>1</sup>, <sup>1</sup>*University of Georgia, Athens, GA*, <sup>2</sup>*PIC North America, Hendersonville, TN*.

The objective of this study was to predict genomic breeding values for preweaning mortality and birth weight using a bivariate threshold-linear maternal effect model.

The data for preweaning mortality and birth weight, and genotypes were obtained from PIC North America and contained 123,163 phenotypic records and 135,530 animals in pedigree. The genotype file contained 42,787 single nucleotide polymorphism (SNP) markers for 13,566 pigs. Birth weight and preweaning mortality were recorded at piglet level. The model included fixed contemporary group, sex, parity, and number of total born per litter effects, random direct and maternal genetic effects, random maternal permanent environmental effects (MPE), and random residual effects (R). First, direct and maternal heritabilities for preweaning mortality and birth weight and genetic correlations were estimated with the Gibbs sampling THRG-IBBS1F90 program. Second, genomic breeding values (GEBV) were predicted with the CBLUP90IOD program, which uses preconditioning conjugate gradients and iteration-on-data for a large number of categorical records, using a single-step genomic BLUP. Direct heritability estimates for preweaning mortality and birth weight were 5.4 and 14.0%, respectively. Maternal heritability estimates were 2.1 and 17.2%. Direct and maternal genetic correlations between the two traits were  $-0.18$  and  $-0.63$ , respectively. Environmental correlations were also negative ( $-0.34$  and  $-0.41$  for MPE and R). Correlations of GEBV for 13,566 genotyped animals between the two traits were  $-0.17$  and  $-0.84$  for direct and maternal genomic effects, respectively. Preweaning mortality is negatively affected by birth weight especially through maternal genetic components.

**Key Words:** preweaning mortality, birth weight, genotype



**M73 Genetic parameters of lifetime characteristics of pre-weaning production traits of Landrace sows raised under tropical conditions.** Teerapong Jaichansukkit<sup>1</sup>, Thanathip Suwanasopee\*<sup>1</sup>, Skorn Koonawootrittriron<sup>1</sup>, and Mauricio A. Elzo<sup>2</sup>, <sup>1</sup>*Kasetsart University, Bangkok, Thailand*, <sup>2</sup>*University of Florida, Gainesville, FL*.

Lifetime preweaning production traits are important for increasing profitability in commercial swine operations. Sows with larger preweaning production from the first to the last parity in her lifetime are preferred. Thus, it is important to determine how heritable lifetime preweaning production traits are in swine populations under tropical conditions in Thailand. Consequently, the objective of this study was to estimate genetic parameters for characteristics of lifetime preweaning production traits of Landrace (L) sows raised in an open-house system in Thailand. The preweaning production traits were number of piglet born alive (NBA), number of piglets weaned (NPW), average weight of piglets at birth (ABW), and average weight of piglets at weaning (AWW). The characteristics considered for these traits were first-parity value (FPV), peak-parity value (PPV), number of parities from first-parity to peak-parity (P1P), and persistency from the third to the last parity (regression coefficient; P3L). The data set contained 6,075 performance records from 941 sows that farrowed from 2004 to 2012. Variance components for each characteristic were estimated separately for the 4 traits using an AI-REML procedure. The 4-trait animal model for each characteristic included first-farrowing year-season and age at first farrowing as fixed effects, and sow and residual as random effects. Heritability estimates ranged from 0.04 (NPW) to 0.10 (ABW) for FPV, 0.06 (NPW) to 0.20 (NBA) for PPV, 0.01 (NBA) to 0.04 (ABW) for P1P, and 0.03 (NPW) to 0.17 (ABW) for P3L. Genetic correlation estimates ranged from -0.47 (NPW-AWW) to 0.87 (ABW-AWW) for FPV, -0.10 (NPW-AWW) to 0.85 (ABW-AWW) for PPV, -0.73 (NBA-NPW) to 0.58 (NBA-AWW) for P1P, and -0.70 (NPW-AWW) to 0.13 (NPW-ABW). The low estimates of heritability for all characteristics in these 4 lifetime preweaning production traits indicated that genetic improvement for these characteristics in all these traits would either be slow or nonexistent in this Landrace population.

**Key Words:** pig, lifetime production, tropics

**M74 General and specific combining abilities for reproductive and growth performance of three color variants of Nigerian indigenous turkeys.** Matthew A. Adeleke\*<sup>1</sup>, Rasheed O. Ojo<sup>1</sup>, Sunday O. Peters<sup>2</sup>, and Michael O. Ozoje<sup>1</sup>, <sup>1</sup>*Department of Animal Breeding and Genetics, Federal University of Agriculture, Abeokuta, Ogun State, Nigeria*, <sup>2</sup>*Department of Animal Science, Berry College, Mount Berry, GA*.

Assessment of combining abilities is important in evaluation of hybrid combinations for genetic improvement. Our objective in this study was to determine combining abilities of 3 color variants of Nigerian indigenous turkeys. Two hundred fifty poults consisting of 41 White × White (WW), 40 Black × Black (BB), 32 Lavender × Lavender (LL) purebreds; 26 White × Black (W×B), 24 Black × White (B×W), 22 White × Lavender (W×L), 20 Lavender × White (L×W), 23 Black × Lavender (B×L) and 22 Lavender × Black (L×B) crossbreds were generated from matings among White, Black and Lavender color types. The poults were raised from day-old to 20 weeks. A 3 × 3 diallel design was set up and data analyzed using SAS (2005) to estimate general combining ability (GCA) and specific combining ability (SCA) for fertility, hatchability, weak-in-shell, dead-in-shell, dead-in-germ, body weight, breast girth, body length and thigh length. The highest GCA was recorded for fertility (4.44), hatchability (5.39) and dead-in-shell (2.60) in BB, LL and WW respectively. SCA for fertility was the highest in W×B (0.07). W×L had

highest SCA (3.28) for hatchability. Least SCA for weak-in-shell (-0.50), dead-in-germ (-2.87) and dead-in-shell (-47.17) were observed in W×L crossbred. The highest GCA for body weight at wk 20 was recorded for WW (90.83). The highest SCA for body weight at wk 20 was recorded in W×L (0.62). BB had highest GCA values for breast girth, body length and thigh length at wk 20 (1.46, 0.46 and 0.41 respectively). B×W recorded the highest SCA (0.94) for breast girth; W×L gave the highest SCA (2.34) for body length while the highest SCA (0.89) for thigh length was observed in B×L turkey genotype. Based on genetic parameter estimates, additive variance was more important for growth parameters, fertility and dead-in-germ while dominance variance was higher and more important in controlling hatchability, weak-in-shell and dead-in-shell than additive variance. Dams from B×W crossbred local turkey can be used to improve growth performance while W×L can be used to achieve best combiners for improvement of reproductive traits in Nigerian indigenous turkeys.

**Key Words:** combining ability, turkey, plumage color

**M75 Association with disease resistance markers and economic traits in Korean native chickens.** Boyeong Park\*, Anh Duc Truong, Jihye Ban, and Yeong Ho Hong, *Chung-Ang University, Anseong, Gyeonggi, Korea*.

Avian coccidiosis is a mucosal infectious disease that significantly impairs the growth and feed efficiency of chickens. There are associations between oocyst and body weight, the parameters of resistance to coccidiosis and SNPs in 2 genes [Myeloid leukemia factor 2 (*MLF2*), T cell receptor- $\beta$  (*TCR- $\beta$* )] located on chromosome 1. A total of 5 SNPs of *MLF2* and *TCR- $\beta$*  genes were assessed for genetic effects on disease resistance. The *MLF2*-SNP\_892 (GG) is most significantly related to decreased oocyst shedding. Besides, a combination of SNPs is mostly associated with disease resistance. A total of 340 female chickens from 7 pure-bred chicken lines [Rhode Island Red (RIR) D line, Korean Native Chicken (KNC) gray, KNC black, KNC, Leghorn F, Leghorn K, and Ogey line] were used to genotype with real-time PCR (Light-Cycler 960). The SNP genotyping was carried out for each line using the High Resolution Melting Master (LightCycler 480) that represent melting curves after PCR amplification. Interestingly, their haplotype is significantly matched with their economic traits. Taken together, this new information will be a parameter that disease resistance markers related with phenotypes consist of body weight, weight of first egg and further study required to prove those marker's availability in poultry breeding industry

**Key Words:** chicken, disease resistance marker, high-resolution melting

**M76 Breeding implications of heteroskedastic whole-genome prediction of genetic merit.** Zhining Ou\*<sup>1</sup>, Robert J. Tempelman<sup>2</sup>, Juan P. Steibel<sup>2</sup>, Catherine W. Ernst<sup>2</sup>, Ronald O. Bates<sup>2</sup>, and Nora M. Bello<sup>1</sup>, <sup>1</sup>*Kansas State University, Manhattan, KS*, <sup>2</sup>*Michigan State University, East Lansing, MI*.

Current breeding programs in animal production systems involve selection of candidate animals with superior genetic merit to serve as progenitors of the next generation. In this study, we predicted genomic breeding values (GEBV) for 2 quantitative traits from the Michigan State University Swine Resource Population using standard whole-genome prediction (WGP) models that assume homogeneous residual variance (i.e., RR-BLUP, BayesA, BayesB and BayesC $\pi$ ) and their heteroskedastic counterparts. We divided the data into 5 mutu-

ally exclusive folds, such that 4 folds were alternatively used to train homoskedastic and heteroskedastic versions of each WGP model and then predict GEBV on animals on the remaining validation fold. The pseudo-Bayes factors indicated that heteroskedastic error WGP models improved model fit at all 5 crossvalidation folds. Within each fold, we then computed the Spearman rank correlation between homoskedastic- and heteroskedastic-based GEBV for top and bottom 10% individuals to compare their relative rankings. We noticed a considerable degree of re-ranking of animals with 10% top and bottom homoskedastic GEBV, particularly as the amount of residual heterogeneity in the data increased. For loin muscle pH at 45 min post-mortem, the median rank correlation of GEBV for top (bottom) 10% animals between heteroskedastic and homoskedastic models ranged from 0.52 to 0.70 (0.64 to 0.70) across data folds and WGP models. Similarly, for carcass temperature at 45 min post-mortem, the median rank correlation ranged from 0.05 to 0.38 (top 10%) and from 0.43 to 0.54 (bottom 10%). These results indicated non-negligible re-ranking of individuals with extreme genetic merit when heterogeneity of residual variances across environments is accounted for, thereby supporting potential practical implications for selection purposes in breeding programs.

**Key Words:** whole-genome prediction model, residual heteroskedasticity, re-ranking

**M77 Co-association gene networks for meat quality and carcass traits in pigs and validating by transcription factors.** Darlene A. S. Duarte\*, Fabyano F. Silva, Renata Veroneze, Lucas L. Verardo, Ivan Carvalho Filho, Simone E. F. Guimarães, and Paulo S. Lopes, *Universidade Federal de Viçosa, Viçosa, MG, Brazil.*

The aim of this study was perform genome association studies for meat quality and carcass traits and thereafter build gene network to improve the knowledge of genes, pathways and the physiological mechanisms that affect meat quality and carcass traits. In addition, the identification of transcriptional factors related to those genes was used to validate this network. We performed genome association studies for 12 traits (one trait at time) in a F<sub>2</sub> population (produced by crossing naturalized Brazilian breed Piau with commercial line) and we found 144 significant SNPs (p-value < 0.05). These SNPs were selected to build the Association Weight Matrix (AWM), which was used to investigate the genetic basis of these traits and generate gene network based on the co-association of pair-wise SNPs across phenotypes. Through this methodology, we found 45 genes that were used to build a gene network based on pair-wise correlations between them. We identified 25 transcription factors (TF) strongly related (p-value < 0.001) with genes in the network. The top 3 TF (Sox5, Nkx2-5 and T) were chosen for construction of a network with their pathways and gene ontology. The genes present in the network generated from AWM and also present in TF network were involved mainly in metabolism of adipose tissue and skeletal muscle. These results suggest that genes and TF identified here are important in the control of meat quality and carcass traits. However, further efforts should be made to study in more detail the new gene-gene interactions here identified, as well as, the key transcription factors and pathways involved in these traits.

**Key Words:** genome association studies, gene network, pig

**M78 An improved approach for swine SNP genotyping using Genotyping-by-Sequencing.** Cheng Tan\*<sup>1</sup>, Jiangli Ren<sup>1</sup>, Zhuolin Huang<sup>1</sup>, Yiqiang Zhao<sup>1</sup>, Yang Da<sup>2</sup>, and Xiaoxiang Hu<sup>1</sup>, *<sup>1</sup>State Key Laboratory for Agrobiotechnology, China Agricultural University,*

*Beijing, China, <sup>2</sup>Department of Animal Science, University of Minnesota, Saint Paul, MN.*

Genotyping-by-sequencing (GBS) technology has a capacity for delivering large numbers of single nucleotide polymorphism (SNP) marker genotypes with potentially lower cost than SNP chips. To achieve stable genotyping results, we selected the *EcoRI-MspI* enzyme from a large number of candidate enzymes, and used the magnet beads-based purification method to stably and simply recapture GBS tags. Genomic DNA samples from 192 Duroc pigs were digested with *EcoRI-MspI* enzyme, ligated to adapters containing unique barcode, and sequenced on the Illumina NextSeq 500 system with 75bp single-end sequencing. The results showed that all 192 barcoded DNA samples were represented evenly, and that on average 4 million reads per animal were produced. From these samples, about 450,000 unique sequence tags per sample containing 55,843 SNPs were identified through TASSEL4.0 analysis package requiring a minimum of 10 times that a tag must be present, which covered about 1% of the whole genome. The average call rate per individual was more than 94.6%. By repeating GBS genotyping on 2 different samples with 96 pigs in each sample performed by different technicians, 88.5% of the 55,843 SNPs had the same genomic locations from both samples. The cost of the 55,843 SNPs per sample was under \$50 per animal. These results showed that our improved GBS technique is sufficiently high-throughput, economical and provides an acceptable marker density for genomic selection or genome-wide association studies.

**Key Words:** genotyping-by-sequencing, swine, SNP

**M80 Growth curve analyses of three turkey genotypes in the hot humid tropics using a Bayesian mixed model approach.** Michael O. Ozoje\*<sup>1</sup>, Sunday O. Peters<sup>2</sup>, Kyle C. Caires<sup>2</sup>, and Kadir Kizilkaya<sup>3</sup>, *<sup>1</sup>Federal University of Agriculture, Abeokuta, Ogun State, Nigeria, <sup>2</sup>Berry College, Mount Berry, GA, <sup>3</sup>Adnan Menderes University, Aydin, Turkey.*

Growth curves are critical for the understanding and formulation of breeding plans because they shift in response to selection. Nonlinear functions have been used extensively to represent changes in sizes with age, so that the genetic potentials of animals for growth can be evaluated. This study was undertaken to apply the nonlinear Bayesian mixed effect model to examine the changes in the growth pattern of 3 Turkey genotypes in Nigeria. Growth data of 435 turkey poults over a 20-wk period were evaluated using Logistic, von Bertalanffy and Gompertz growth models with normally or Student's-t distributed error. The parameter estimates were significantly different in the models by sexes and genotypes. However, the estimates of parameters were similar except for the differences observed among the local turkey genotypes whose values decreased significantly under the Student's-t distribution. The estimate of the average mature weight (A) in the von Bertalanffy model was the closest to the observed average. The estimate of constant B ranged from 15.62 to 22.17 under the Logistic model but significantly dropped to a range of 0.78 to 4.19 with Gompertz and von Bertalanffy models. The estimate of the rate of maturing (k) varied from 0.06 in von Bertalanffy to 0.26 in the Logistic model. Large estimates of A were generally associated with smaller estimates of k in the von Bertalanffy model. Evaluation of the goodness of fit based on the Deviance Information Criteria showed that the von Bertalanffy model was superior. The differences between functions with respect to average life time absolute growth rate, absolute maturing rate and relative growth rate reflect differences in rate of growth and maturing throughout the growing and maturing phases. In general, these differences are indicative of the

differences that exist among the functions with respect to their abilities to fit the actual growth curve of these turkey genotypes and their sexes.

**Key Words:** turkey, growth curve, nonlinear function

**M81 Inferring the causal effect of number of lambs born on milk yield in dairy sheep using propensity score methods.** Vera C. Ferreira\*, Bruno D. Valente, David L. Thomas, and Guilherme J. M. Rosa, *University of Wisconsin-Madison, Madison, WI.*

Assigning causal interpretation to associations obtained from observational data is challenging as they are prone to confounding. Number of lambs born (prolificacy) in sheep may be considered as a potential factor contributing to milk yield (MY). However, inferring this effect using traditional regression or ANOVA techniques can generate spurious results whenever there are confounder variables that influence both the outcome (i.e., MY) and treatment (i.e., prolificacy). Propensity score (PS) methods tackle this issue by balancing baseline covariate distributions between treatment levels, allowing unbiased inference of marginal effects. This method belongs to the framework of causal models dealing with potential outcomes. It intends to mimic aspects of randomized trials, for which comparison of treatment groups is causally meaningful. Our goal was to infer the causal effect of ewe prolificacy on her subsequent MY using PS based on Matched Samples. Data comprised 4,319 records from 1,534 crossbred dairy ewes. The set of potential confounder variables was composed by lactation number (1st, 2<sup>nd</sup>, and 3rd – 6th) and dairy breed composition (<0.5, 0.5-0.75 and >0.75 of East Friesian or Lacaune). For the treatment variable, single lamb birth was assigned to Group 0, while multiple birth (2, 3 or 4 lambs) was assigned to Group 1. MY represented the volume of milk produced for the whole lactation (mean = 268.5 L and SD = 116.4 L). The analysis was conducted using the R package “nonrandom.” A total of 1,166 pairs of treated/nontreated individuals with similar PS values were formed. The criterion for similarity was defined by a caliper size equal to 20% of the sd in the PS logit (0.13) and a ratio of treated/untreated = 1. Standardized differences were chosen as the statistical test for the hypothesis of PS balance, and all covariates were deemed balanced after matching (cutoff for standardized bias = 0.2). The estimated causal effect of prolificacy on MY was 20.52 L, SE = 3.77 L, 95% CI = 13.13–27.91 L. Hence, results indicate that ewes that gave birth to a single lamb would be expected to have MY increased by 20.52 L if they had given birth to multiple lambs and all other variables were held constant.

**Key Words:** causal inference, milk yield, prolificacy

**M82 Comparative whole-genome analysis of CpG islands in camelid and selected mammalian genomes.** Arsalan Barzandeh<sup>1,2</sup>, Mohammadreza Mohammadabadi<sup>2</sup>, Ikhide G. Imumorin<sup>3</sup>, Sunday O. Peters<sup>4</sup>, Bolaji N. Thomas<sup>5</sup>, Mostafa Ghaderi-Zefrehei<sup>6</sup>, and Hossein Nezamabadi-Pour<sup>1</sup>, <sup>1</sup>*Shahid Bahonar University of Kerman, Keerman, Iran*, <sup>2</sup>*Jiroft University, Jiroft, Iran*, <sup>3</sup>*Cornell University, Ithaca, NY*, <sup>4</sup>*Berry College, Rome, GA*, <sup>5</sup>*Rochester Institute of Technology, Rochester, NY*, <sup>6</sup>*University of Yasousj, Yasousj, Iran.*

Camels are dromedaries found in extreme desert environments of Africa and Asia with adaptations to arid conditions of temperatures exceeding 40°C, water losses greater than 25% of total body weight, and the ability to survive up to 14 d without water. Camels are important as beasts of burden for transport and as sources of meat, milk and wool in many desert countries. Therefore, camel could be very useful in the weather extremes precipitated by global climate change. Recent sequences of camelidae genomes [wild bactrian camel (*Camelus bactrianus*)

*ferus*], bactrian camel (*Camelus bactrianus*), dromedary (*Camelus dromedaries*) and alpaca (*Vicugna pacos*)] provides the opportunity to better understand the genomic architecture of this unique group of animals. CpG island (CGIs) sequence patterns in complex genomes typically consists of high-frequency of CpG dinucleotides associated with promoter regions of about 70% of all genes, found in almost all housekeeping genes and in about half of tissue-specific genes. CGIs play important roles in the regulation of gene expression including X-chromosome inactivation, imprinting, silencing of intragenic parasites and are extensively linked to the epigenetic causes of cancer. We used newly available genomic sequence to perform comparative large-scale whole-genome analysis of CGIs for the first time in Camelidae using 5 different CGI detection algorithms. These algorithms detected different numbers of CGIs, CGI density and CGI length distribution in Camelidae. All algorithms agreed on the alpaca genome as having the largest number of CGIs, CGI density and average length of CGIs. When compared with other mammalian genomes of human, mouse, dog, horse and cow, CGIs features in cow genome was the most similar with camelid genomes. These results contribute to better understanding of the evolutionary genetics of camelid genomes in comparison with other livestock species. Further analysis of camelid genomes may shed more light on molecular origins and mechanisms of heat adaptation in these extreme heat-adapted animals.

**Key Words:** Camelidae, genome, CpG island

**M83 Genome-wide association study of mandibular inferior in multiple breeds of sheep.** Michelle R. Mousel<sup>\*1</sup>, Stephen N. White<sup>1,2</sup>, J. Bret Taylor<sup>3</sup>, and Donald P. Knowles<sup>1,2</sup>, <sup>1</sup>*USDA-ARS, Animal Disease Research Unit, Pullman, WA*, <sup>2</sup>*Department of Veterinary Microbiology and Pathology, Pullman, WA*, <sup>3</sup>*USDA-ARS, Range Sheep Production Efficiency Research Unit, Dubois, ID.*

Misalignment of the jaw in sheep, which has long been considered a genetic condition, occurs in most sheep breeds, and culling of affected animals is recommended. Severe mandibular inferior, where the mandible is shorter than the maxillary, can lead to premature death due to starvation. A German breeding trial of affected sheep produced greater than 60% affected lambs and they estimated jaw misalignments were due to dominant and recessive gene effects. A genome wide association study was conducted using the OvineSNP50 marker set to genotype ewes that either produced unaffected lambs (n = 712) or at least 1 affected (n = 252) lamb during their lifetime. Large numbers of animals from multiple breeds improves the odds for detecting true positively associated genomic regions in multiple genetic backgrounds. Jaw misalignment was categorized as severe, greater than 1.3 cm longer maxillary, moderate, between 0.3 and 1.3 cm longer maxillary, mild, between 0.05 and 0.29 cm longer maxillary, and normal, no misalignment within 24 h of birth. Ewes were Rambouillet, Polypay, and Columbia breeds which produced a total of 8321 purebred and crossbred lambs over 13 y with 3.9% of lambs presented with mandibular inferior. Year of lamb birth was significant ( $P < 0.01$ ) with lambs born in 2009 having more misaligned jaws compared with all other years. Sex of lamb was significant ( $P < 0.01$ ) with a greater frequency of misaligned jaws in male lambs. Breed of lamb sire ( $P > 0.52$ ), ewe age ( $P > 0.32$ ), and breed of ewe ( $P > 0.08$ ) did not affect lamb jaw alignment. A recessive model accounting for breed, population cluster, and minor allele frequency identified 2 genomic regions that were significantly (Bonferroni-corrected  $P < 0.03$ ) and 2 genomic regions that were suggestively (unadjusted  $P < 2.3 \times 10^{-6}$ ) associated with mandibular inferior. The 2 significant markers were located on chromosome 2, near a gene which affects growth, and chromosome 22 near a gene that plays a role in membrane trafficking.



Further evaluation of these genomic regions is required to identify underlying causal mutations, which would be useful in marker-assisted selection for sheep producers.

**Key Words:** sheep, mandibular inferior, genome-wide association study

**M84 Integration of haplotype analysis of functional genomic information with single SNP analysis improved accuracy of genomic prediction.** Cheng Tan<sup>\*1,2</sup>, Dzianis Prakapenka<sup>1</sup>, Chunkao Wang<sup>1</sup>, Li Ma<sup>3</sup>, John R. Garbe<sup>4</sup>, Xiaoxiang Hu<sup>2</sup>, and Yang Da<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Minnesota, Saint Paul, Minnesota, <sup>2</sup>State Key Laboratory of Agrobiotechnology, China Agricultural University, Beijing, China, <sup>3</sup>Department of Animal and Avian Sciences, University of Maryland, College Park, MD, <sup>4</sup>Minnesota Supercomputer Institute, University of Minnesota, Minneapolis, MN.

We compared 3 methods of genomic prediction using additive and dominance effects of single SNPs and haplotype blocks. Method I was single SNP analysis of 423,131 SNPs covering all human autosomes from the Framingham Heart Study with over 6000 individuals. The next 2 methods add haplotype analysis of functional information to the single SNP analysis, i.e., Method II adds haplotype analysis of 595 “cholesterol-related genes” with 8,674 SNPs (2% of autosomes); Method III adds haplotype analysis of 9821 genes with 184,686 SNPs (36% of autosomes) after removing tiny genes without at least 2 SNPs. The results from 4 to 8 validation samples showed that adding haplotype analysis to single SNP analysis improved the prediction accuracy in most cases. Method II with cholesterol-related genes had the best prediction accuracy for total cholesterol with 4.78% increase in accuracy over single SNP analysis, and had stable accuracy increases across validation samples for all cholesterol phenotypes. Method III using all autosomal genes had the best accuracy for triglyceride with 17.75% increase in accuracy over single SNP analysis and tended to have the best performance across different phenotypes, but had larger variations than Method II across validation samples for cholesterol phenotypes. Results were also obtained from one validation sample for adding 3 other haplotype analyses to single SNP analysis: ChIPseq sites with 375,924 SNPs and average block size of 115.8Kb; non-hotspot blocks with each block between 2 crossover hotspots with 422,695 SNPs and average block

size of 65Kb, and evenly divided blocks with block size of 100Kb of 422,814 SNPs. All 3 methods improved the prediction accuracy for most phenotypes but ChIPseq blocks mostly had better prediction accuracy than the other 2 methods, indicating that ChIPseq sites likely contained useful functional information not present in anonymous blocks. The results in this study tend to conclude that the integration of haplotype analysis of functional genomic information with single SNP analysis may improve the accuracy of genomic prediction for some phenotypes.

**Key Words:** haplotype, genomic selection, SNP

**M85 Preliminary study of *DMRT3* variation and association with performance gait for American Saddlebred horses.** Inaê C. Regatieri<sup>\*1</sup>, John E. Eberth<sup>2</sup>, Fred Sarver<sup>3</sup>, and Ernest Bailey<sup>2</sup>, <sup>1</sup>Faculdade de Ciências Agrárias e Veterinárias, UNESP – Univ Estadual Paulista, Jaboticabal, São Paulo, Brazil, <sup>2</sup>Department of Veterinary Science, University of Kentucky, Lexington, KY, <sup>3</sup>Cornerstone Farm, Carlisle, KY.

The Doublesex and Mab-3 Related Transcription Factor 3 (*DMRT3*) gene encodes an important transcription factor in the setting of spinal cord circuits controlling movement in vertebrates. The SNP g.22999655C>A of the *DMRT3* gene was significantly associated with performance of 4-beat gaits in horses, such as the running walk and the amble American Saddlebreds ridden and shown under saddle seat in both 3-gaited (walk, trot, and canter) or 5-gaited classes (walk, trot, slow gate, rack, and canter). We investigated whether SNP g.22999655C>A of *DMRT3* was more prevalent among 5-gaited horses than among the random population of Saddlebred horses. The genotyping of the SNP of the *DMRT3* gene was performed by PCR-RFLP using the restriction enzyme *DdeI*. The frequency of the *A* allele among 37 randomly selected Saddlebreds was 0.28. This is in agreement with results from a previous study in which the prevalence of the *A* allele in American Saddlebred horses was 27.5. Only 3 of the horses in our study had been shown as 5-gaited horses and all 3 had the genotype *A/C*. Although consistent with the hypothesis that the *A* allele is more prevalent among for 5-gaited horses, the numbers are too small for any conclusion. Additional Saddlebred horses, and especially 5-gaited horses, will be tested and results reported allowing determination of statistical significance.

**Key Words:** equine, genomics, genetics



## Breeding and Genetics: Molecular genetics

**M86 Heat storage and HSP expression of Holstein females – an *in vivo* study.** Ana C. A. P. M. Geraldo<sup>1,2</sup>, Thays M. C. Leme<sup>1</sup>, Reissa A. Vilela<sup>1</sup>, Cristiane G. Titto<sup>1</sup>, Evaldo A. L. Titto<sup>\*1</sup>, Paulo Infante<sup>3</sup>, Fernando J. Moreira da Silva<sup>4</sup>, and Alfredo M. F. Pereira<sup>2</sup>, <sup>1</sup>*Animal Science and Food Engineering Faculty, University of São Paulo, Pirassununga, São Paulo, Brazil*, <sup>2</sup>*Institute of Mediterranean Agricultural and Environmental Sciences, University of Évora, Évora, Portugal*, <sup>3</sup>*Mathematics Department, University of Évora, Évora, Portugal*, <sup>4</sup>*Department of Agrarian Sciences, University of Azores, Angra do Heroísmo, Azores, Portugal*.

One of the main factors that affect animals' performance is high temperature, causing several changes, including at the cellular level. These changes lead to an increased expression of heat shock proteins. This experiment aimed to study the *HSPA1A* and *HSP90AA1* gene expressions of Holstein cows after exposure to direct solar radiation. The heat tolerance test was performed and rectal temperature and respiratory rate measured, and blood samples collected. After the erythrocytes lysis to obtain the buffy-coat, the RNA was isolated by the TRIzol method and RT-PCR performed with SuperScript III after digestion with DNase I. The qPCR apparatus took place in 7500 Real Fast Time, using TaqMan Gene Expression Assays for *HSPA1A* and *HSP90AA1* target genes, *ACTB* and *PPIA* as endogenous genes. The  $\Delta Ct$  ( $Ct_{\text{target}} - Ct_{\text{endogenous}}$ ) were calculated as well as gene expression through the  $2^{-\Delta\Delta Ct}$  method. The treatments considered for statistical analysis were  $t < 44^\circ\text{C}$  and  $T \geq 44^\circ\text{C}$  (with T being the black globe temperature). We used linear mixed models in the program R Project Software (version 3.0.1). There weren't significant differences between treatments for any of the variables. This way we can say that animals were in a moderate stress condition, which did not allow the identification of differences in *HSPA1A* and *HSP90AA1* gene expression.

**Key Words:** dairy cattle, *HSPA1A*, *HSP90AA1*

**M87 Gene and pathway analysis of metabolic traits in dairy cows.** Ngoc-Thuy Ha<sup>1,2</sup>, Josef J. Gross<sup>\*1</sup>, Jens Tetens<sup>3</sup>, Martin Schlather<sup>4</sup>, Rupert M. Bruckmaier<sup>1</sup>, and Henner Simianer<sup>2</sup>, <sup>1</sup>*Veterinary Physiology, Vetsuisse Faculty University of Bern, Bern, Switzerland*, <sup>2</sup>*Animal Breeding and Genetics Group, Department of Animal Sciences, Georg-August-University Goettingen, Goettingen, Germany*, <sup>3</sup>*Institute of Animal Breeding and Husbandry, Christian-Albrechts-University Kiel, Kiel, Germany*, <sup>4</sup>*Chair of Mathematical Statistics, University of Mannheim, Mannheim, Germany*.

During excessive body fat mobilization in early lactation, dairy cows experience a severe metabolic load. Here, a failure in metabolic adaptation often results in an increased susceptibility to health problems. In this study, we analyzed the genetic basis of the metabolic adaptability during the transition period. To this end, blood samples were taken from 178 cows (Holstein, Red Holstein, Fleckvieh and Braunvieh) at 3 critical stages: T1 = wk 3 before expected calving (not lactating and no metabolic load); T2 = wk 4 postpartum (lactating and high metabolic load), and T3 = wk 13 after parturition (lactating and no noteworthy metabolic load). Plasma concentrations of nonesterified fatty acids (NEFA),  $\beta$ -hydroxybutyrate (BHBA) and glucose, 3 metabolites characterizing the metabolic status and adaptability, were measured at T1, T2, and T3. All cows were genotyped with the Illumina next-generation High-Density Bovine BeadChip resulting in a data set of 777,692 SNPs. After quality control and filtering, the SNPs were annotated to known

genes (Ensembl Genes Database) and pathways (KEGG Database). For each gene G with g SNPs, we performed a score test based on the linear regression model  $y = \gamma_1 C_1 + \dots + \gamma_m C_m + \beta_1 S_1 + \dots + \beta_g S_g + \epsilon$ , where  $C_i$  are environmental or breed effects and  $y$  are either one of the 3 metabolites measured at T1, T2 or T3 or the ratio of their concentrations measured at the different points of time, to assess the association of the gene G to the phenotype  $y$ . The results were then used to identify pathways enriched with significant genes using a weighted Kolmogorov-Smirnov test. As a result, we found 99 significant genes associated with at least one of the 3 metabolites. For each metabolite, we found genes that are significant at T2 but not at T1 and T3 or vice versa. This strongly suggests the genes to be potential candidates for the adaptive regulation. We further find 3 pathways (steroid hormone biosynthesis, ether lipid metabolism and glycerophospholipid metabolism) to jointly affect the 3 metabolites. In conclusion, this may be regarded as evidence for the genetic basis for the adaptation performance of dairy cows and, at the same time, reveals its complexity.

**Key Words:** genome-wide association study, gene-based score test, dairy cow

**M88 A novel intronic SNP marker candidate which associated with fatty acids profile in Korean cattle (Hanwoo).** Yoonseok Lee<sup>\*1</sup>, Dongyep Oh<sup>3</sup>, Jaeyung Ha<sup>3</sup>, Jinduk Bok<sup>1</sup>, Sangkee Kang<sup>1,2</sup>, YunJaie Choi<sup>1,2</sup>, and Myunggi Baik<sup>1,2</sup>, <sup>1</sup>*Institute of Green Bio Science & Technology, Seoul National University, Pyeongchang-gun, Gangwon-do, South Korea*, <sup>2</sup>*Department of Agricultural Biotechnology, Seoul National University, Gwanak-gu, Seoul, South Korea*, <sup>3</sup>*Gyeongsangbuk-do Livestock Research Institute, Yeongju, Gyeongsangbuk-do, South Korea*.

Recently, consumers have become increasingly health-conscious and demand not only palatable but also health-beneficial meat in the market. In Korea, degree of the intramuscular fat deposit (marbling) is one of the crucial indicators to judge beef quality. Thus, the fatty acids profile in beef is important because it affects both flavor and health-benefit. Various trials have been conducted to change fatty acids profile into health-beneficial composition, such as poly-unsaturated fatty acids-abundant meat, in Korean beef. The fatty acid binding protein 4 (*FABP4*) gene is a cytosolic protein abundantly expressed in adipocyte and plays an important role on lipid synthesis and intracellular fatty acid trafficking. An SNP located on the splice site variation between the third exon and the third intron of bovine *FABP4* showed a statistically significant effect on intramuscular fatty acid deposition. The aim of this study was to investigate the relationship between SNP within the third intron region of *FABP4* and fatty acids profile of longissimus dorsi area in Korean cattle. We sequenced the third intron region of bovine *FABP4*. Among this SNPs, the g.3996 C>T SNP show statistically significant effect on oleic acid and polyunsaturated fatty acids ( $P < 0.001$ ). Especially, the animals with CC genotype have a lower ratio of omega-6/-3 fatty acid (5.4:1) than the ratio of animals with TT genotypes (9.9:1). Therefore, our result suggest that g.3996 C>T intronic SNP will be used as a useful marker-assisted selection for breeding on fatty acids profile of Korean cattle.

**Key Words:** Korean cattle, intronic SNP, fatty acid binding protein 4 (*FABP4*)

**M89 A QTL on BTA16 is associated with *Mycobacterium avium* ssp. *paratuberculosis* (*Map*) tissue infection.** Jennifer N. Kiser\* and Holly L. Neibergs, *Washington State University, Pullman, WA.*

Johne's disease is a contagious bacterial infection in cattle caused by *Mycobacterium avium* ssp. *paratuberculosis* (*Map*) infection. A previous genome wide association study (GWAS) in Holstein cattle identified QTL on bovine chromosome 3 (BTA3) and BTA9 that were highly associated ( $P < 5 \times 10^{-7}$ ) and on BTA1, BTA16, and BTA21 that were moderately associated ( $P < 5 \times 10^{-5}$ ) with *Map* tissue infection. The objective of this study was to validate these GWAS results in Holsteins from the Pacific Northwest (PNW,  $n = 191$ ) and a combined population from the PNW and the Northeast (PNW+NE,  $n = 432$ ). DNA was genotyped using the Illumina BovineSNP50 BeadChip. Cases were ileo-cecal node positive for *Map* by PCR, and controls were *Map* tissue negative. Individuals were removed if the SNP call rate was  $< 90\%$ , and SNPs were removed if the genotype call rate was  $< 95\%$  or they had a MAF of  $< 0.01$ . After filtering, 162 cases, and 247 controls and 44,445 SNPs remained for analysis. A GWAS for the PNW and PNW+NE was conducted using an efficient mixed-model association eXpedited (EMMAX) method using 3 gene effect models. No loci were associated with the recessive model in PNW or PNW+NE. For the PNW cows, 4 dominant QTL were identified; 3 on BTA21 ( $P = 1.18 \times 10^{-6}$ ,  $8.15 \times 10^{-6}$ ,  $2.13 \times 10^{-5}$ ) and 1 on BTA3 ( $9.88 \times 10^{-6}$ ). In the additive model, a new QTL on BTA21 ( $5.14 \times 10^{-7}$ ) and on BTA1 ( $P = 5.82 \times 10^{-5}$ ) were identified. In the PNW+NE population, 2 QTL were identified with the dominant model: one on BTA14 ( $P < 3.17 \times 10^{-5}$ ) and one on BTA16 ( $P < 3.17 \times 10^{-5}$ ). The BTA16 QTL was also identified by the additive model ( $P < 4.59 \times 10^{-5}$ ) and identified previously by our group using the NE cows alone ( $P < 2.57 \times 10^{-5}$ ). SNPs associated with *Map* tissue infection on BTA16 lie within introns of *CDC42BPA*, a Serine/Threonine-protein kinase that has downstream effects on *CDC42*. Through the regulation of *CDC42*, *CDC42BPA* has been linked to several immunological pathways in humans including B-cell receptor and chemokine signaling pathways. Although none of the previous QTL in the NE GWAS were validated in the PNW population, a QTL on BTA16 was associated with susceptibility to *Map* tissue in both the NE and PNW+NE populations.

**Key Words:** bovine paratuberculosis, genomics

**M90 Effect of the STAT5A *BstEII* polymorphism on reproductive parameters of Holstein dairy cows.** Pedro A. S. Silveira<sup>1</sup>, Walter R. Butler<sup>2</sup>, Carlos C. Barros<sup>1</sup>, Marcio N. Corrêa<sup>1</sup>, and Augusto Schneider\*<sup>1</sup>, <sup>1</sup>Federal University of Pelotas, Pelotas, RS, Brazil, <sup>2</sup>Cornell University, Ithaca, NY.

The signal transducer and activator of transcription 5A (STAT5A) is a transcription factor that mediates the function of various hormones and cytokines, including growth hormone (GH). Mutations in the STAT5A gene, such as the substitution of a G by a C in exon 8, have been associated with differences in production and reproductive performance of dairy cows. The aim of this study was to evaluate the effect of STAT5A *BstEII* polymorphism on the days from calving to first ovulation and calving-conception interval (CCI). For identification of the polymorphisms DNA was extracted from blood and a fragment of the STAT5A gene was amplified by PCR. The presence of the G and C alleles was determined after digestion of the PCR products with the *BstEII* enzyme and gel electrophoresis. For this study 73 Holstein cows were followed from 21 d prepartum to 210 d in milk (DIM). At 55 DIM the cows were submitted to an OvSynch-TAI protocol, which was repeated in cows diagnosed as not pregnant. From calving, milk production was recorded, milk samples for progesterone measurement were collected twice a week

to determine ovulation day until 60 DIM, and the CCI was evaluated until 210 DIM. Serum samples for insulin-like growth factor I (IGF-I) measurement were collected at -21, 0, 7, 21 and 60 DIM. Data were analyzed using the GLM procedure of SAS. In total, 19 cows (26%) were of the CC genotype, 32 cows (43.8%) of the GC genotype and 22 cows (30.2%) of the GG genotype. The calving to ovulation interval was not different between genotypes ( $P > 0.05$ ):  $28.4 \pm 3.1$ ,  $29.5 \pm 2.5$  and  $29.4 \pm 2.9$  DIM for the CC, CG and GG genotypes. The CCI was  $101.3 \pm 9.4$ ,  $102.7 \pm 7.6$  and  $93.9 \pm 9.4$  DIM for the CC, CG and GG genotypes ( $P > 0.05$ ). Milk production was similar between the 3 genotypes ( $P > 0.05$ ). Serum IGF-I was also not different between genotypes, being  $66.3 \pm 7.2$ ,  $61.8 \pm 5.8$ ,  $65.6 \pm 6.7$  ng/mL for CC, CG and GG genotypes ( $P > 0.05$ ). Therefore, the STAT5A *BstEII* polymorphism did not affect the calving to ovulation interval, CCI, milk production or IGF-I concentrations in Holstein dairy cows. It should be noticed that this study used a small number of cows and larger studies are necessary to confirm current results.

**Key Words:** SNP, growth hormone (GH), insulin-like growth factor I (IGF-I)

**M91 Differentially expressed genes for beef fatty acid profile in Nellore cattle.** Mariana P. Berton\*<sup>1</sup>, Marcos V. A. Lemos<sup>1</sup>, Hermenegildo L. J. Chiaia<sup>1</sup>, Fabieli L. B. Feitosa<sup>1</sup>, Carolyn Aboujaoude<sup>1</sup>, Larissa F. S. Fonseca<sup>1</sup>, Bianca F. Olivieri<sup>1</sup>, Daniela F. R. J. Gimenez<sup>1</sup>, Bruno L. Utembergue<sup>2</sup>, Lucia G. de Albuquerque<sup>1</sup>, Aline S. M. Cesar<sup>2</sup>, Angélica S. C. Pereira<sup>2</sup>, and Fernando Baldi<sup>1</sup>, <sup>1</sup>State University of Sao Paulo, Jaboticabal, Sao Paulo, Brazil, <sup>2</sup>University of Sao Paulo, Pirassununga, Sao Paulo, Brazil.

The aim of this study was to use the RNaseq technique to identify differentially expressed (DE) genes in the *Longissimus thoracis* muscle of Nellore cattle finished in feedlot with extreme phenotypes for beef fatty acid profile. After slaughter, a muscle tissue sample was collected of each animal for the extraction of RNA and in the deboning a sample was taken for determining the fatty acid profile. The fatty acids were quantified by gas chromatography (CG-2010 Plus; Shimadzu), using capillary column SP-2560. The following fatty acids were quantified: myristic (C14:0), palmitic (C16:0), stearic (C18:0), oleic (C18:1 *cis*-9), linoleic (C18:2 *cis*-*cis*-9-12), CLA (C18:2 *cis*-9 *trans*-11) and linolenic (C18:3), total saturated fatty acids (SFA), total monounsaturated fatty acids (MUFA), total polyunsaturated fatty acids (PUFA), the ratio of polyunsaturated fatty acids on saturated (PUFA/SFA), n-3 (omega-3) fatty acids and n-6 and ratio of n-6 fatty acids on n-3 (n-6/n-3). Two groups of animals with extreme phenotypes for the composition of meat fatty acid were formed, being considered 10 animals with the highest (H) and 10 animals with the lowest (L) concentrations for each fatty acid. The RNA sequencing data were generated on the Illumina HiSeq System platform. The differential expression of RNA was determined using the iPlant Collaborative platform, containing the FastQC package (version 0.10.1), TopHat2 (version 2.0.9) and Cuffdiff (2.1.1). The analysis of the metabolic pathways of differentially expressed genes was performed with the DAVID tool. The ACS1 was upregulated ( $q < 0.05$ ) for saturated fatty acids (palmitic, stearic, oleic, total saturated), and downregulated ( $q < 0.05$ ) for unsaturated acids, (omega-3). This gene assists in the conversion of acetate, into acetyl-CoA to incorporate it into the fatty acids by the action of acetyl CoA synthetase. Other DE genes involved in metabolic pathways of fatty acids synthesis were found, such as: BDH1\_BOVIN, ACSM3, CBPE\_BOVIN, F1N650\_BOVIN. Through the RNaseq technique was identified possible genes acting

in the synthesis of beef fatty acids, which can be beneficial for human health.

**Key Words:** *Bos indicus*, transcriptomic, fatty composition

**M92 Polymorphisms in the promoter of interleukin-12 $\beta$ 2 and interleukin-23 receptor genes influence milk production traits in Chinese Holstein cows.** Yongjiang Mao\*, Xiaorui Zhu, Shiyu Xin, Huiming Zhang, and Zhangping Yang, *College of Animal Science and Technology, Yangzhou University, Yangzhou, Jiansu, China.*

Interleukin-12 (IL-12) and interleukin-23 (IL-23) are proinflammatory cytokines produced by macrophages and dendritic cells in response to infection with intracellular pathogens. Given the importance of IL-12 and IL-23 for modulating inflammation and the host immune response, the IL-12 and IL-23 receptor genes may be suitable candidate genes for studying disease resistance in dairy cattle. Twenty Chinese Holstein cows were selected randomly for PCR amplification and sequencing, and used for SNP discovery in the bovine IL-12R $\beta$ 2 and IL-23R promoter region. One SNP (c.-246G>T) in IL-12R $\beta$ 2 gene and 2 SNPs (c.-856A>G and c.-207T>C) in IL-23R gene were identified. Chinese Holstein cows (n = 866) were then genotyped using Sequenom MassARRAY (Sequenom Inc., San Diego, CA) based on the 3 identified SNPs, and the associations between SNPs or haplotype of the genes and milk production traits, SCS were analyzed by the least squares method in the GLM procedure of SAS. The IL-23R c.-856A>G and IL-23R c.-207T>C showed close linkage disequilibrium ( $r^2 = 0.89$ ). No association was found with SCS, but associations were found between 3 of these SNP with milk protein content and lactose content. The software MatInspector revealed that these SNPs were located within several potential transcription factor binding sites, and may alter gene expression, but further investigation will be required to elucidate the biological and practical relevance of these SNP.

**Key Words:** interleukin-12 receptor  $\beta$ 2 (IL-12R $\beta$ 2), interleukin-23 receptor (IL-23R), SNP

**M93 Evolution of mutational variance associated with age and sex of the parent for weaning weight in C57BL/6J mice.** Mayela Castillo<sup>1</sup>, Juan F. Medrano<sup>2</sup>, and Joaquim Casellas\*<sup>1</sup>, <sup>1</sup>Universitat Autònoma de Barcelona, Bellaterra, Spain, <sup>2</sup>University of California, Davis, CA.

New mutations are a very relevant component of polygenic variability and they must be viewed as the raw material for the maintenance of genetic diversity. Nevertheless, the biological phenomena that originate new mutational variants are poorly understood, particularly in regard to the effects that factors such as a parent's age may have. This research focuses on the analysis of weaning weight ( $10.48 \pm 0.02$  g) in 12,644 C57BL/6J mice from 46 non-overlapping generations. Data (y) were analyzed under the following model:  $y = Xb + Z_1p + Z_2a + Z_2m_p + Z_2m_m + e$ , where systematic (b), permanent environmental (p), and additive genetic effects (a,  $m_p$  and  $m_m$ ) were linked by appropriate incidence matrices (X,  $Z_1$  and  $Z_2$ ). Note that mutational effects were modeled depending on their origin (i.e., paternal,  $m_p$ ; maternal,  $m_m$ ), and an independent set of parent-specific mutational effects were estimated on the basis of new mutations arising to each individual in the pedigree file (i.e.,  $m_p = m_{p,1} + m_{p,2} + \dots + m_{p,12644}$ ). Within this context,  $m_{p,i}$  was assumed multivariate normal (MVN) distributed,  $p(m_{p,i}|M_i, \sigma_{mp}^2, \lambda_p) = MVN(0, 0.5M_i\sigma_{mp}^2 [1 + \varepsilon_{p,i}\lambda_p])$   $M_i$  being the matrix of mutational relationships specific to new mutations arising in the  $i$ th individual,  $\sigma_{mp}^2$

( $\sigma_{mm}^2$ ) being the paternal (maternal) mutational variance,  $\varepsilon_{p,i}$  being sire's age when conceiving  $i$ th individual, and  $\lambda_p$  being a linear regression coefficient linking mutational variance and sire's age. This model was solved under a standard Bayesian approach. Mode (and credibility interval) for  $\sigma_{mp}^2$  and  $\sigma_{mm}^2$  were 0.113 (0.069 to 0.163) and 0.052 (0.020 to 0.097), respectively. Whereas linear regression coefficient for dam's age collapsed to 0 and discarded additional changes in  $\sigma_{mm}^2$ ,  $\lambda_p$  reached a modal estimate of 0.001 ( $10^{-5}$  to 0.051); e.g., this linearly increased  $\sigma_{mp}^2$  from  $\sim 0.11$  g<sup>2</sup> (puberal sires) to 0.154 g<sup>2</sup> (one-yr-old sires). This parameterization allowed us to clearly characterize different mutational patterns associated with the sex of the parent, as well as the effect of the accumulation of new mutations along breeding stock's lifetime.

**Key Words:** age-related mutation, C57WL/6J, weaning weight

**M94 A molecular evaluation of bovine respiratory disease and carcass traits in feedlot steers.** Samantha Miller<sup>1</sup>, Ryon Walker<sup>3</sup>, Timothy Page<sup>1,2</sup>, and Matthew Garcia\*<sup>1,2</sup>, <sup>1</sup>Louisiana State University, Baton Rouge, LA, <sup>2</sup>Louisiana State AgCenter, Baton Rouge, LA, <sup>3</sup>Louisiana State AgCenter Hill Farm, Homer, LA.

Bovine respiratory disease (BRD) is the most common disease affecting feedlot cattle and economic losses incurred by affected animals is estimated to be \$640 million annually. The objective of the current study was to evaluate single nucleotide polymorphisms (SNP) and their potential associations with bovine respiratory disease susceptibility and carcass traits in feedlot steers. A population of 314 crossbred steers born from 2010 to 2013 and raised at LSU AG Center Central Research Station (CRS) in Baton Rouge, LA and LSU Ag Center Hill Farm Research Station (HFRS) in Homer, Louisiana, were utilized in the current study. Prior to shipping to a commercial feedlot, the measurements of birth weight, weaning weight and hip height were collected from each steer. After weaning, and the completion of a 45-d preconditioning period, steers were shipped to a commercial feedlot. A total of 16 steers over the entire 4-year evaluation period were affected by BRD. A total of 309 steers were harvested at a commercial packing plant and the traits of marbling score, rib eye area, back fat thickness and yield grade were collected. A total of 74 SNP were selected from a previously described BRD QTL region spanning between 40 and 80 MB on BTA 6. This same region has also been identified to have QTL associated with kidney pelvic heart fat percentage. A total of 33 SNP were selected from a previously described BRD QTL region spanning 0–30 MB on BTA 20. This region has been identified to have QTL's associated with kidney pelvic heart fat percentage, marbling score, intramuscular fat, sheer force and carcass weight. A mixed model design was fit with individual carcass traits, BRD status, and individual SNP genotype as dependent variables and sire breed and year fit as independent variables in the model to identify SNP that were significantly associated with the traits of interest. Although multiple SNP were identified as being significantly associated with carcass traits and BRD susceptibility, these SNP must be validated in larger, and more diverse populations before implementation into selection strategies.

**Key Words:** bovine respiratory disease, feedlot, single nucleotide polymorphism



**M95 Association of a polymorphism in the paraoxonase 1 (PON1) gene with reproductive performance, health and production of Holstein cows.** Pedro A. S. Silveira<sup>1</sup>, Walter R. Butler<sup>2</sup>, Carlos C. Barros<sup>1</sup>, Marcio N. Corrêa<sup>1</sup>, and Augusto Schneider\*<sup>1</sup>, <sup>1</sup>Federal University of Pelotas, Pelotas, RS, Brazil, <sup>2</sup>Cornell University, Ithaca, NY.

Paraoxonase 1 (PON1) is a negative acute phase protein associated with uterine health conditions in postpartum dairy cows and may affect reproductive performance. Recently, a single nucleotide polymorphism (SNP) was found in the promoter of the *PON1* gene in dairy cows associated with serum PON1 activity. The aim of this study was to evaluate the association of the *PON1*(A/G)-221 SNP with reproductive performance, disease incidence and milk production in Holstein cows. For the study, 85 Holstein cows were followed from 21 d prepartum to 210 d in milk (DIM). For SNP identification DNA was extracted from blood and the tetra primer ARMS-PCR technique was used. The primers produced a control 700 bp product, and smaller specific products of 500 bp (allele A) or 200 bp (allele G). After gel electrophoresis it was possible to genotype all cows and some were confirmed by sequencing of the products. At 55 DIM the cows were submitted to an OvSynch-TAI protocol, which was repeated in cows diagnosed as not pregnant. From calving, milk production was recorded, milk samples for progesterone measurement were collected twice a week to determine ovulation day until 60 DIM, and disease incidence, the number of inseminations/pregnancy (AI/P) and the calving- conception interval (CCI) was evaluated. Data were analyzed using the GLM procedure of SAS and by survival analysis and Chi-squared on GraphPad Prism. After genotyping, we detected 57 cows (67.0%) of the AA genotype, 20 cows (23.6%) of the AG genotype and 8 cows (9.4%) of the GG genotype. Cows of the GG and AG genotype ovulated earlier than AA cows ( $27.6 \pm 2.9$  and  $32.1 \pm 2.2$  DIM, respectively;  $P = 0.02$ ). There was no difference between genotypes for milk production, number of AI/P or CCI ( $P > 0.05$ ). Also, there was no difference for the occurrence of disease (metritis and mastitis) between the 3 genetic groups ( $P > 0.05$ ). Therefore, the presence of at least one G allele at the position -221 of the *PON1* gene is associated with an earlier postpartum ovulation, although more studies on the mechanism for this effect are needed.

**Key Words:** *PON1*, SNP, dairy cow

**M96 Effect of *POU1F1* gene polymorphism and dairy traits in Holstein cattle from Antioquia, Colombia.** Jose V. Isaza\*, Albeiro Lopez-Herrera, and Jose J. Echeverri, *Universidad Nacional de Colombia Sede Medellin, Medellin, Antioquia, Colombia.*

Genetic improvement has allowed great advances in selecting individuals with interest for determine characteristics. Knowledge about bovine genome and genetic markers along each chromosome allow the search for genetic variants that affect important productive and reproductive traits, making possible the genetic improvement. The goal of this study was to determinate the association between the *POU1F1* gene with productive and reproductive traits in a population of Holstein cows from Antioquia, Colombia. The *POU1F1* gene belongs to transcription factors POU, which regulates growth and development in animals. This gene participates on pituitary development and hormonal expression in mammals, regulating the production of growth hormone, thyroid stimulating hormone and prolactin. DNA was extracted from 523 samples, and 2 alleles were identified using the PCR-RFLP technique. The frequency for the alleles A and B was 21.26 and 78.74%, respectively, and the 3 genotypes AA, AB and BB showed frequencies of 2.87, 36.78, and 60.34%, respectively. The association between the genotypes and the studied

traits was analyzed with a linear mixed model including the fixed effect of *POU1F1* genotype, parity, year and month of birth, and the random effect of the animal. The analyzed SNP had no significant effect on the evaluated traits milk yield, protein percentage, fat percentage, calving interval and services per conception. The absence of association between the SNP and evaluated traits could be due to 2 reasons; the interaction of this gene with other genes involved in the *POU1F1* pathway which may hide the individual effect of this SNP on the evaluated traits, and the genotype-environment interaction, the latter has a large effect on gene expression. This way, the *POU1F1* gene is not a good candidate to be used in breeding programs assisted by molecular markers (MAS) for the evaluated population.

**Key Words:** dairy herd, molecular marker, polymorphism

**M97 Identification of genes and networks for the response to thermal stress.** Hoyoung Chung\*, *National Institute of Animal Science, Suwon, KY, Korea.*

This study has been aimed to investigate genetic responses for the genes related to heat stress when exposing Holstein calves that were selected based on no significant relationship (inbreeding coefficient  $< 0.01$ ). A total of 10 animals aged from 4 to 6 mo were selected, and heat stress was imposed on animals directly in an environmentally controlled house that was managed for 33°C and 90% of humidity based on THI values. After exposing heat stress for 8 h a day, animals were placed in a normal condition at least 12 h to recover from the heat stress. The blood samples, which were collected at the starting point (09:00 a.m.) and at the end of heat stress (21:00 p.m.), were immediately placed in liquid nitrogen. The RNA was extracted from blood samples between control and treatment, and cDNA library was constructed for each individual. After sequencing analysis that produced a minimum 3 G byte, expression analysis confirmed that 53 genes were differentially expressed according to the severe heat stress. The analysis also verified 4 major pathways (MAPK signaling; T-cell receptor signaling; B-cell receptor signaling, and Chemokine signaling) that were related to heat stress. As expected that heat-shock protein (*HSP*) is related to thermal stress and response, the analysis verified that *HSP70* presented 656.4 fold changes between control and treatments. To verify fold changes, 100 primer pairs from 51 genes with accession numbers were tested using real-time PCR, and the results confirmed 17 genes in statistical significances. The identified genes should be served as reference genes for the selection of superior animals against thermal stress.

**Key Words:** thermal stress, differentially expressed gene

**M98 Transcriptome analysis of muscular tissue in Nellore cattle divergently ranked for meat tenderness.** Larissa Fernanda Simielli Fonseca\*<sup>1</sup>, Daniele Fernanda Jovino Gimenez<sup>1</sup>, Fernando Baldi<sup>1</sup>, Jesus Aparecido Ferro<sup>2</sup>, Rafael Espigolan<sup>1</sup>, and Lucia Galvão Albuquerque<sup>1</sup>, <sup>1</sup>Departamento de Zootecnia, Faculdade de Ciências Agrárias e Veterinárias, UNESP, Jaboticabal, SP, Brazil, <sup>2</sup>Departamento de Tecnologia, Faculdade de Ciências Agrárias e Veterinárias, UNESP, Jaboticabal, SP, Brazil.

The objective of the present study was to identify differential gene expression related to meat tenderness in Nellore cattle. Meat samples from 132 animals belonging to the same contemporary group were used. Meat tenderness was measured by shear force through Warner Bratzler method and 20 divergently ranked animals for this trait (10 with tough meat and 10 with tender meat) were selected. Means and the respective



standard deviations for tough and tender meat groups were  $7.4 \pm 0.78$  kg and  $4.41 \pm 0.40$  kg, respectively. Total RNA was extracted from the selected samples and sequenced (RNA-Seq) using the HiSeq 2500 System (Illumina). The results were analyzed in the iPlant Collaborative platform. The workflow included: FastQC; TopHat2 and Cuffdiff. A total of 17 differentially expressed genes ( $q$ -value  $< 0.05$ ) were identified, and among them, 4 are highlighted. The genes *Q3ZCJ1* (transmembrane protein 37), *C1QTNF7* (C1q and tumor necrosis factor related protein 7) and *BDH1* (3-hydroxybutyrate dehydrogenase, type 1), were more expressed in tough meat samples, whereas the gene *ATP1A1* (ATPase, Na<sup>+</sup>/K<sup>+</sup> transporting,  $\alpha$  1 polypeptide) was more expressed in tender meat samples. The Q3ZCJ1 protein is involved in the same metabolic pathway of actin and myosin proteins that constitute the myofibrils, organelle which acts on muscle contraction. C1QTNF7 protein acts in complement and coagulation cascade metabolic pathway. This pathway is activated after an injury and the coagulation system is activated by exposed collagen after an injury. There is a direct link between collagen content and meat tenderness. The BDH1 enzyme is involved in the synthesis and degradation of ketone bodies that, if present, causes a decrease in muscle pH. The ATP1A1 enzyme is related to protons synthesis, which causes a decrease in intracellular pH during ATP degradation to ADP. Muscle pH decrease is related to meat tenderness during the post-mortem process. These genes seem to be involved in the meat tenderness process. This study was supported by São Paulo Research Foundation FAPESP (grants 2009/16118-5 and 2013/09190-7)

**Key Words:** RNAseq, differentially expressed gene, quality meat

**M99 Environmental risk assessment by genetically engineered mice as transgenic animal model.** Dailu Guan<sup>1</sup>, Qian Yu<sup>1</sup>, Erhu Zhao<sup>1</sup>, Yong Wang<sup>2</sup>, and Yongju Zhao\*<sup>1</sup>, <sup>1</sup>College of Animal Science and Technology, Southwest University; Chongqing Key Laboratory of Forage & Herbivore; Chongqing Engineering Research Center for Herbivores Resource Protection and Utilization, Beibei, Chongqing, China, <sup>2</sup>Department of Laboratory Animal Science, College of Basic Medicine, Third Military Medical University, Sapingba, Chongqing, China.

Environmental safety on transgenic animals is often a controversial issue within the researchers and the public. Here, a total of 12 full-sib transgenic mice expressing enhanced green fluorescent protein (eGFP) were used for assessing foreign gene expression by single-line passage method and evaluating effects on GE gene with the environment with a total of 12 full-sib mice (positive: control = 6:6, with the same size of male and female mice). PCR method and fluorescent protein observation system (FPOs) were used for detecting the inserted exogenous genes in these mice. The results showed that the expression level of eGFP and WRPE gene (vector sequence) were not significantly different, but it significantly reduced by F1 to F4 generation (F1, F2, F3 and F4) ( $P < 0.05$ ) by Real-time quantitative PCR (RT-qPCR). The effect of GE mice on the environment with 4- or 8-week-old full-sib transgenic mice offspring was evaluated. The exogenous genes were not detected by PCR in the mice manure. Microbial flora of transgenic mice were not significantly different with those of the control group ( $P > 0.05$ ), neither male or female, at 4 or at 8 week-old. In addition, there was no significant difference of the microbial communities in mice gut as assessed by PCR-DGGE or by 16S rDNA sequencing between positive

and control transgenic mice offspring. Furthermore, the phylogenetic analyses showed that the manure bacteria sampled during each of the 2 stages belonged primarily to 3 groups, *Firmicutes*, *Bacteroidetes* and *Actinobacteria*. No unknown microbial flora were found in the mice manure. [Supported by the National Natural Science Foundation of China (No. 31172195), the Fundamental Research Funds for the Central Universities(No: XDJK2014A010) and the 2013 Innovation Team Building Program in Chongqing universities (KJTD201334).]

**Key Words:** transgenic mouse, environmental risk, gene drift

**M100 Associations between HEL5, AFZ1, ILSTS002, BMS3004, IDVGA-51, LHR, and FSHR alleles on reproductive evaluation of bulls.** Gabriel R. Pereira\*<sup>1</sup>, Silvio R. O. Menegassi<sup>1</sup>, Paulo R. Aguiar<sup>2</sup>, Katiana S. Pereira<sup>2</sup>, Celso Koetz<sup>3</sup>, Flavio G. Lopes<sup>3</sup>, Vanerlei M. Roso<sup>4</sup>, Vanessa Peripolli<sup>1</sup>, Fernanda G. Moojen<sup>1</sup>, and Julio O. J. Barcellos<sup>1</sup>, <sup>1</sup>Federal University of Rio Grande do Sul - UFRGS, Porto Alegre, RS, Brazil, <sup>2</sup>Lutheran University of Brazil - ULBRA, Canoas, RS, Brazil, <sup>3</sup>University of Northern Paraná - UNOPAR, Arapongas, PR, Brazil, <sup>4</sup>GenSys Association, Porto Alegre, RS, Brazil.

This study emphasized the importance to develop new molecular tools to accurately identify candidate genes in predicting semen quality in bulls that can be used in livestock production. We assessed the frequency distribution of molecular markers linked to insulin growth factor I, follicle stimulating hormone (FSH), luteinizing hormone (LH) and leptin genes in Braford and Hereford population. All bulls (Braford and Hereford;  $n = 188$ ) showed good body condition scores throughout the experiment. Blood sampling and measurements of scrotal circumference were performed on weaned bulls at 7 and 24 mo of age during the bull breeding soundness evaluations. Semen collection was performed using an electroejaculator on bulls at 24, 28, 32 and 36 mo. Blood samples for DNA extraction were collected by puncture of the coccygeal vein. Five microsatellites or short tandem repeats (HEL5, AFZ1, ILSTS002, BMS3004, and IDVGA-51) and 2 SNP markers (LHR and FSHR) were evaluated by the amplification of DNA products, which then underwent electrophoresis in 10.5% polyacrylamide gel. Molecular markers were analyzed with PROC GLM ANOVA using SAS and significant were considered when  $P < 0.05$ . Hereford bulls that expressed the variation in the IDVGA51 allele was associated ( $P < 0.05$ ) with sperm motility and vigor traits in Hereford bulls. Hereford animals showed polymorphic information content (PIC) of 0.36 to 0.75% along with expected heterozygosity (H) of 0.49 to 0.78%. However, Braford bulls that expressed the ILSTS002 and AFZ1 alleles were associated ( $P < 0.05$ ) with major and minor defects, respectively. Braford PIC ranged from 0.28 to 0.78% with an expected H of 0.35 to 0.81%. We found no relation between HEL5, BMS3004, LHR and FSHR to predict semen quality in bulls ( $P > 0.05$ ). Therefore the present study revealed that 3 microsatellite alleles are important male reproductive biomarkers for improving semen parameters in bulls. We concluded that the ILSTS002 and AFZ1 alleles in the Braford and the IDVGA51 allele in the Hereford may be used for improving the reproductive traits in bulls.

**Key Words:** molecular marker, microsatellite, genetic variability

# Dairy Foods: Chemistry

**M101 Inhibition of lipid oxidation in fish oil-in-water emulsions by the combination of bovine and caprine caseins with phospholipids.** Adela Mora-Gutierrez\*, Rahmat Attaie, Sela Wolde-senbet, and Jeneanne Kirven, *Prairie View A&M University, Prairie View, TX.*

The nutritional benefits of polyunsaturated fatty acids from fish oil in terms of their role in growth and development and in health and disease are limited by their oxidative degradation. The objective of this study was to investigate the effect of bovine and caprine caseins in combination with phospholipids on the oxidative stability of 5% fish oil-in-water emulsions at pH 7.0. Menhaden oil was emulsified with phospholipids such as phosphatidylcholine, phosphatidylethanolamine, and phosphatidylserine in a high-pressure valve homogenizer. The effect of bovine and caprine caseins in combination with phospholipids on oxidation rate was monitored by measurement of thiobarbituric acid reactive substances (TBARS) and lipid peroxides. Oxidation of 0.5% phospholipid-stabilized fish oil-in-water emulsions were performed at 25°C for 48 h, in the dark, in the presence of 0–0.3% bovine and caprine caseins and 0.1 mM Fe<sup>2+</sup>. It was found that the emulsion stabilized with phosphatidylserine showed the best oxidative stability in terms of TBARS concentration and peroxide value. Formation of TBARS was inhibited 32% by addition of 0.3% caprine casein ( $P < 0.05$ ). Inhibition of lipid oxidation by addition of 0.3% bovine casein was less effective, decreasing the concentration of TBARS by 24% ( $P < 0.05$ ). The combination of casein and phosphatidylserine resulted in favorable structure and thickness of the interfacial layer, thus preventing lipid oxidation in the emulsion. These results indicated that the combination of casein and phosphatidylserine can be utilized to enhance the oxidative stability of emulsified fish oil.

**Key Words:** casein, phospholipid, fish oil

**M102 Obtaining of casein fractions by preparative ion-exchange chromatography on weak anion-exchangers.** Andrij Iukalo<sup>1</sup>, Orysyia Tsisaryk\*<sup>2</sup>, and Volodymyr Yukalo<sup>1</sup>, <sup>1</sup>*Ternopil Ivan Pulu National Technical University, Ternopil, Ukraine*, <sup>2</sup>*Lviv National University of Veterinary Medicine and Biotechnology, Lviv, Ukraine.*

The aim of study was to obtain the main casein fractions ( $\alpha_{S1}$ -CN,  $\alpha_{S2}$ -CN,  $\beta$ -CN, and  $\kappa$ -CN) in preparative amounts using weak anion-exchangers. Total casein was obtained using isoelectric precipitation from fresh skim milk for fractionation. Fractional composition of total casein and the homogeneity of its fractions at different stages of the isolation were analyzed by electrophoresis on the vertical plates of polyacrylamide gel. At this stage, an alkaline buffer system containing 25 mM tris, 27 mM diethylbarbiturate, 3 mM ethylenediaminetetraacetate, and 4.5 M urea was used. Electrophoregrams were fixed and stained by conventional methods. The concentration of protein was determined by spectrophotometer (SF-46,  $\lambda$ -280 nm). Previously installed absorption coefficients for different fractions of casein (D 1% in 1 cm) were used: 10.0 – for  $\alpha_{S1}$ -CN; 10.1 – for  $\alpha_{S2}$ -CN; 4.6 – for  $\beta$ -CN; 9.6 – for  $\kappa$ -CN, and 8.2 – for total casein. DEAE-cellulose (DEAE-52 “Serva”) and toyopearl DEAE 650M (Toyo soda) were used as anion-exchangers. Batch separation of total casein was performed in 0.02 M acetate buffer (pH = 6.5) in the presence of 3.3 M urea and 0.01 M 2-mercaptoethanol. CaCl<sub>2</sub> was used as eluent. This salt is more effective than NaCl due to the interaction with phosphoserine groups of caseins. For fractionation

we used the following concentrations of CaCl<sub>2</sub>: 0.0 M (I fraction); 0.005 M (II); 0.01 M (III); 0.02 M (IV); 0.035 M (V); 0.05 M (VI). The experiment was replicated 3 times. As a result, electrophoretic analysis shows that  $\kappa$ -casein is found in fraction I; II contains  $\beta$ -casein; III and IV –  $\alpha_{S2}$ -casein; V –  $\alpha_{S1}$ -casein with minor impurities of  $\alpha_{S2}$ -casein, and VI contains only  $\alpha_{S1}$ -casein. Obtained fractions were dialyzed, lyophilized and weighed. The total yield of caseins was 71% using DEAE-cellulose and 77% in the case of anion-exchangers toyopearl DEAE 650M ( $P < 0.05$ ). Weak anion-exchangers are more effective than strong ones in caseins preparative fractionation. It allowed us to obtain a separate group of  $\alpha_{S2}$ -caseins, which includes  $\alpha_{S2-}$ ,  $\alpha_{S3-}$ ,  $\alpha_{S4-}$ , and  $\alpha_{S6-}$  caseins. These fractions are characterized by the same sequence of amino acid residues and are precursors of many original bioactive peptides.

**Key Words:** casein, fraction, ion-exchange chromatography

**M103 Size distribution of casein micelles in raw skim milk from individual cows as studied using cryo-TEM.** Maneesha S. Mohan\* and Federico M. Harte, *Food Science Department, Pennsylvania State University, University Park, PA.*

Casein micelles have a complex structure comprising of 4 caseins:  $\alpha_{S1}$ ,  $\alpha_{S2}$ ,  $\beta$ , and  $\kappa$  caseins, assembling as many as 20,000 casein proteins. Polydispersity is the degree of disparity or variation in size of particles. Traditionally, casein micelles have been considered highly polydisperse with relative polydispersity between 0.27 to 0.5 as reported by different studies using dynamic light scattering techniques. A recent study has defined casein micelles to be more monodisperse. Hence, we decided to get a visual confirmation of the polydispersity of casein micelles by studying the casein micelle size distribution in milk from 4 cows by cryo-transmission electron microscopy (cryo-TEM). The raw milk from 4 cows of first parity, mid lactation and low somatic cell count, was skimmed by centrifugation ( $6414 \times g$  for 20 min). The diluted skim milk (10  $\mu$ L in 500  $\mu$ L of protein-free serum obtained by ultrafiltration of skim milk using a 3-kDa filter) was subjected to cryo-TEM at 200kV. The diameter of casein micelles was measured manually using ImageJ software. The relative polydispersity (relative standard deviation of size distribution) of casein micelles was a Gaussian distribution (x-axis = number of micelles as a fraction of the total micelles measured, y-axis = diameter of casein micelles) spanning between 10 to 341 nm (1545 micelles), 24 to 693 nm (1073 66 micelles), 11 to 514 nm (2621 micelles) and 11 to 532 nm (1891 micelles) for milk from 4 cows, respectively. The positively skewed distribution curve had a tapering tail of large casein micelles of diameter greater than 350 nm. The maximum number of casein micelles existed in the size range of 90 to 110 nm for milk from all the 4 cows. The relative polydispersity ranged between 0.39 and 0.47 for casein micelles with mean diameters ( $D_{10}$ ) between 112 to 134 nm for the milk from the 4 cows. The results indicate high level of polydispersity in the casein micelles in milk of every cow. Hence it was possible to effectively utilize cryo-TEM to elucidate the size distribution of casein micelles and visually reconfirm the polydispersity of casein micelles in bovine milk from individual cows.

**Key Words:** polydispersity, electron microscopy, Gaussian

**M104 Effect of casein micelle dissociation and casein modification on plasmin-induced hydrolysis.** Hemang Bhatt<sup>1,2</sup>, Aurelie Cucheval<sup>1</sup>, Christina Coker<sup>1</sup>, Hasmukh Patel\*<sup>3</sup>, Alistair Carr<sup>2</sup>, and Rod Bennett<sup>2</sup>, <sup>1</sup>Fonterra Research and Development Centre, Palmerston North, New Zealand, <sup>2</sup>Institute of Food, Nutrition and Human Health, Massey University, Palmerston North, New Zealand, <sup>3</sup>Dairy Science Department, South Dakota State University, Brookings, SD.

Plasmin-induced hydrolysis of casein in milk can lead to many defects including proteolysis, age gelation, and bitterness. We hypothesized that the susceptibility of casein micelles to plasmin can be affected by its micellar structure and modification of the lysine residues on caseins. Different levels of dissociation of the casein micelle structure were achieved by calcium chelation using EDTA and by succinylation by attaching a succinate group at the  $\epsilon$ -amino group of lysine residues, leading to the formation of succinyl-lysine. The target sites for succinylation were identified using liquid chromatography-tandem mass spectrometry. Changes in the particle size and levels of casein micelle dissociation in skim milk were determined using a Zetasizer and sodium dodecyl sulfate PAGE, respectively. The subsequent plasmin hydrolysis was monitored by quantifying the hydrolyzed product using reverse phase high performance liquid chromatography. The EDTA-induced calcium chelation and dissociation of the casein micelle increased plasmin-induced hydrolysis. In contrast, succinylation had an inhibitory effect on plasmin-induced hydrolysis. Calcium chelation and succinylation of the skim milk resulted in dissociation of caseins from the casein micelle, a decrease in turbidity, extensive unfolding, and expansion of the polypeptide chain; all of these changes collectively reduced steric hindrance and made the protein more readily hydrolyzed by plasmin. However, the formation of succinyl-lysine rendered  $\beta$ -casein unrecognizable to the substrate-binding pocket of plasmin, resulting in a nonlinear decrease in the hydrolysis because of the competitive effect of micelle dissociation. These results clearly indicate the importance of the casein micelle structure and its susceptibility to plasmin action. They will be useful in understanding and controlling the plasmin-induced hydrolysis of milk proteins in food systems.

**Key Words:** plasmin, succinylation, liquid chromatography-tandem mass spectrometry

**M105 The role of lactose and whey proteins in plasmin resistance of heat-treated milk.** Hemang Bhatt<sup>1,2</sup>, Aurelie Cucheval<sup>1</sup>, Christina Coker<sup>1</sup>, Hasmukh Patel\*<sup>3</sup>, Alistair Carr<sup>2</sup>, and Rod Bennett<sup>2</sup>, <sup>1</sup>Fonterra Research and Development Centre, Palmerston North, New Zealand, <sup>2</sup>Institute of Food, Nutrition and Human Health, Massey University, Palmerston North, New Zealand, <sup>3</sup>Dairy Science Department, South Dakota State University, Brookings, SD.

Sterilization and UHT treatment are commonly used to increase the shelf life of milk and dairy beverages. However, the high heat resistance of the plasmin system enables it to partially survive UHT treatment, which could lead to defects in the texture (gelation, sedimentation) and the taste (bitterness) of these products. The present study investigated the effect of high heat treatment on plasmin-induced hydrolysis using a sequential approach. A high-heat-treated (120°C/15 min) skim milk had been found to have higher resistance to plasmin-induced hydrolysis, compared with a control skim milk. The effect of heat treatment on the resistance of skim milk was studied with a sequential approach. Different milk systems were prepared by removing whey protein and then lactose from skim milk and reconstituting each milk separately, to observe the effect of whey protein and lactose both individually and in combination. The milk systems were heated at 120°C for different times to achieve different levels of heat treatment. All treated milk systems

were then hydrolyzed by adding plasmin and the reaction was monitored using reverse-phase HPLC. The extents of whey protein denaturation, dissociation of casein from the micelles and association of whey protein with the micelles were monitored using sodium dodecyl sulfate-PAGE. Particle size and levels of lactosylation were also measured. Plasmin-induced hydrolysis was negatively affected by an increase in the heat treatment. Both whey protein association with the casein micelles and lactosylation decreased the availability of protein to plasmin. These effects were compared and mechanisms for them have been proposed. Whey-protein-free milk was the most resistant to plasmin, followed by skim milk and lactose-free milk, suggesting that lactosylation plays a more major role than whey protein association with the casein micelles in making protein resistant to plasmin-induced hydrolysis.

**Key Words:** plasmin, lactosylation, sterilization

**M106 Oxidative stability of an Iranian ghee (butter fat) versus soybean oil during storage at different temperatures.** Mahshid Azizi\*<sup>1</sup>, Maryam Enteshari<sup>2</sup>, and Mohammadreza Dolatkahnejad<sup>3</sup>, <sup>1</sup>Islamic Azad University of Birjand, Chemical Engineering and Food Industries Department, Tehran, Iran, <sup>2</sup>Department of Food Science and Technology, Shahid Beheshti University of Medical Sciences, Tehran, Iran, <sup>3</sup>Islamic Azad University of Ayatollah Amoli, Food Industries and Scientific Engineering Department, Tehran, Iran.

The oxidative stability of an Iranian ghee (butter fat) and soybean oil, which were stored at 4 different storage temperatures (4, 25, 45, and 60°C) and during one-month storage was investigated. Oxidation changes were monitored every 2 weeks by analytical parameters including acid value (AV), peroxide value (PV), *p*-anisidin value (*p*-AV), thiobarbituric acid value (TBA) and oxidative stability index (OSI). In addition, iodine value (IV) and fatty acids profile were determined to elucidate intrinsic differences between ghee and soybean oil. The PV, *p*-AV and TBA were gradually increased during storage time, while the OSI decreased. The ghee in contrast to soybean oil, showed significantly ( $P < 0.05$ ) lower amounts of PV, *p*-AV and TBA and higher OSI. However higher amounts of AV were obtained for ghee samples at all of mentioned storage temperatures which indicated higher free fatty acids (FFA %) as a result of production procedure for this kind of fat. Moreover, obtained data revealed that the effects of applied different temperatures on the mentioned oxidation parameters were significant during studied storage time ( $P < 0.05$ ); additionally, at higher temperatures (45 and 60°C) these effects were more intensive particularly for the soybean oil samples. Results confirmed that ghee displayed better oxidative stability behavior as evidenced by lower PV, *p*-AV and TBA with higher values of OSI at accelerated storage conditions.

**Key Words:** oxidative stability, Iranian ghee, soybean oil

**M107 Effect of hydrodynamic cavitation on acid gelation properties of skim milk.** Harsh Dahiya\*<sup>1</sup>, Hasmukh A. Patel<sup>1</sup>, and Thom Huppertz<sup>1,2</sup>, <sup>1</sup>South Dakota State University, Brookings, SD, <sup>2</sup>NIZO Food Research, Ede, the Netherlands.

Acid gelation is an important property of milk with relevance to yogurt manufacturing. Hydrodynamic cavitation (HC) is an emerging technology involving a process of vaporization, bubble generation followed by bubble collapse in a flowing liquid brought about by a decrease in pressure followed by a subsequent increase in pressure. In this study, we compared the effects of HC with conventional heat treatments used in dairy industry on acid gelation property of skim milk. Pasteurized skim milk (3.5% protein and 9% total solids) was preheated to 50°C and



then subjected to 2 sets of HC treatments, namely, HC at 20, 40, and 60 Hz at sufficiently high flow rate (950 L/h) to avoid any temperature increase during HC (T1) and HC at 60 Hz at low flow rates (200 L/h) to allow scale-free heating of skim milk increasing its temperature to desired temperature up to 90°C (T2) using APV Cavitor (supplied by SPX, Denmark) fitted with 4-row rotor in 6mm housing. A portion of skim milk was also heated at 90°C for 10 min. Acid milk gels were prepared from untreated (control), skim milk samples obtained from T1, T2 as well as the skim milk samples conventionally heated to 90°C for 10 min using glucono- $\delta$ -lactone to obtain final pH  $4.6 \pm 0.05$  after 4h incubation at 30°C. Rheological characteristics of acid gels were studied using small amplitude oscillatory rheology (SAOR) at 1% strain and 0.1 Hz frequency. The soluble (serum) phases obtained by centrifugation of control, cavitated and heated milk samples at 25000g/1h were characterized using sodium dodecyl sulfate PAGE (SDS-PAGE). It was observed that the storage modulus ( $G'$ ) of acid gels prepared with skim milk samples subjected to T1 was significantly lower ( $P < 0.05$ ) than those prepared with samples subjected to T2. Acid gels prepared from the samples subjected to T2 with temperatures reaching 90°C had highest final  $G'$ , which was attributed to significant denaturation of whey proteins in these samples. It was also observed that this value of  $G'$  was similar to the one obtained from the conventional heating of milk, suggesting that the cavitation treatment of milk do not adversely affect the acid gelation property of the milk.

**Key Words:** acid gel, cavitation, rheology

#### M108 Molar mass of a crude extract of *Streptococcus thermophilus* St-143 exopolysaccharide during fermentation of milk.

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Some lactic acid bacteria produce high molecular weight polysaccharides (EPS) during milk fermentation. It is unclear if there is any change in the properties of EPS during the fermentation. The molar mass of EPS was investigated during fermentation of milk with *Streptococcus thermophilus* (St-143). We used crude extracts, instead of highly purified samples, to observe more native EPS systems. Reconstituted nonfat milk samples were fermented by *S. thermophilus* at 40°C until pH 4.5. Crude extracts of EPS were obtained at 4 different fermentation times starting from 150 min (pH ~5.3) to 330 min (pH~4.5). Whey from fermented milk was ultrafiltered using 100 kDa membrane to remove soluble milk sugars and proteins. The UF retentate was centrifuged and supernatant was further concentrated using single pass pressure UF cell with 100 kDa membrane. The concentrated EPS solution was analyzed for protein and sugar contents. Molar mass of EPS was estimated using size exclusion chromatography-multi angle laser light scattering (SEC-MALLS). Polysaccharide content of the EPS concentrates varied from ~0.3 to 0.4 mg/mL. Protein impurities in the EPS concentrate were high when sampled at pH 5.3, and significantly decreased ( $P < 0.05$ ) until pH 4.5. Extracts separated using SEC gave an initial peak that appeared to be mostly high  $M_w$  protein aggregates and a second peak that corresponded to EPS. The molar mass of EPS ranged from  $1.57 \times 10^6$  to  $2.86 \times 10^6$  g/mol. EPS sampled at pH 5.3 had significantly higher ( $P < 0.05$ ) molar mass than samples obtained at pH 4.5. EPS samples also

appeared to be monodisperse (i.e.,  $M_w/M_n = 1$ ). The radius of gyration of EPS was ~54 nm and was not significantly different between the different pH samples. It appears that the properties of EPS may vary during milk fermentation.

**Key Words:** exopolysaccharide, molar mass, ultrafiltration

#### M109 Formation of hydroxymethylfurfural and other caramelization products during extrusion of lactose blends.

Catrin E. Tyl, Elizabeth M. Reid, and Tonya C. Schoenfuss\*, University of Minnesota, St. Paul, MN.

As part of our ongoing efforts to finding value-added uses for lactose, we carried out reactive extrusion on blends of lactose, glucose, and citric acid to polymerize lactose to indigestible carbohydrates. This treatment can also induce the formation of color and hydroxymethylfurfural (HMF). The objective of this study was to investigate the effect of formula and extrusion conditions on color and HMF formation. Data analysis was carried out with ANOVA followed by Tukey's HSD test. Three citric acid (CA) concentrations (2, 4, and 6% w/w) were tested. Glucose comprised 20% w/w and lactose made up the remainder. All blends were extruded in duplicate at 2 feed rates: 15 and 30 kg/h. Color was measured as Hunter  $L$ ,  $a$ , and  $b$  values and absorption at 420 nm, indicative for yellow-brown pigments. Absorption was higher for samples of all CA levels extruded at 15 kg/h and these extrudates also had lower  $L$  and higher  $b$  values (i.e., samples were darker and more yellow). Thus, the extent of caramelization increased when mixtures spent more time in the extruder at the low feed rate. Higher CA levels lead to more pigment formation at 15 kg/h, but not at 30 kg/h. Along with other caramelization products, HMF is formed in sugars heated with acids. HMF has shown toxicity in in vitro and animal studies. Thus, reducing its formation through optimum processing conditions and/or sample treatments is needed. HMF content was measured by RP-HPLC/UV. Increasing CA in the raw mix produced extrudates with lower amounts of HMF. The lower feed rate resulted in significantly higher levels of HMF. We evaluated sample treatments to remove HMF using Amberlite (an ion exchanger) and activated charcoal. Filtration over Amberlite lead to 27–57% less HMF. Preliminary results indicate that charcoal treatment may further reduce HMF (>95%). However, because it also lead to losses in oligo- and polysaccharide yields, further optimization of this method is necessary.

**Table 1 (Abstr. 109).** Effect of formula (citric acid at 2, 4, and 6% wt/wt) and feed rate (15 and 30 kg/h) on color and hydroxymethylfurfural (HMF) formation

Item	15 kg/h			30 kg/h		
	2	4	6	2	4	6
A <sub>420</sub>	0.554	0.654	0.934	0.194	0.195	0.200
L	83.84	83.59	80.99	87.41	88.50	89.17
b	27.23	30.01	35.03	14.83	15.21	15.79
HMF (mg/g)	1.20	0.91	0.73	0.71	0.55	0.52
HMF after Amberlite (mg/g)	0.51	0.46	0.53	0.42	0.38	0.31

**Key Words:** hydroxymethylfurfural, lactose, extrusion



## Forages and Pastures: Silages and forages in dairy production systems

**M110 Cool season grass-legume mixtures in north-central Florida.** Erick R. S. Santos<sup>\*1</sup>, José C. B. Dubeux Jr.<sup>1</sup>, Lynn E. Sollenberger<sup>2</sup>, Marta M. Kohmann<sup>2</sup>, Stephanie Pope<sup>2</sup>, Hiran M. S. Silva<sup>1</sup>, and Ana C. C. Melo<sup>1</sup>, <sup>1</sup>University of Florida–North Florida Research and Education Center (NFREC), Marianna, FL, <sup>2</sup>University of Florida – Agronomy Department, Gainesville, FL.

Grass-legume mixtures may increase dry matter (DM) yield and forage N concentration due to biological N<sub>2</sub> fixation. A study was conducted to evaluate yield and botanical composition in different cool season grass-legume mixtures in Gainesville, FL from 4 February to 14 May 2014 (4 harvests). Treatments were allocated in a split-plot arrangement in a completely randomized block design with 4 replicates. Main plots were formed by annual ryegrass (*Lolium multiflorum* Lam.) or a rye (*Secale cereal* L.)-annual ryegrass mixture. The subplot was the combination of cool-season legumes or presence/absence of N fertilizer in grass monocultures, as follows: (1) hairy vetch (*Vicia villosa* ssp.), (2) crimson clover (*Trifolium incarnatum* L.), (3) red clover (*Trifolium pratense* L.), (4) ball clover (*Trifolium nigrescens* Viv.), (5) hairy vetch-red clover-ball clover, (6) crimson clover-red clover-ball clover, (7) N-fertilized grass (50 kg N·ha<sup>-1</sup>·cut<sup>-1</sup>), and (8) unfertilized grass. Total DM yield was greater for N-fertilized grass in the 2nd and 3rd harvests (660 and 1360 kg DM·ha<sup>-1</sup>). Mixtures containing subplot-Treatments 3 and 6 increased total DM yield in the 4th harvest (1250 and 1280 kg DM·ha<sup>-1</sup>, respectively) and did not differ from N-fertilized grass in this harvest. Legume DM yields were minimal in Harvests 1 and 2. In the 3rd and 4th harvests, the crimson clover-red clover-ball clover mixture had the greatest legume DM yield (330 and 730 kg DM·ha<sup>-1</sup>), followed by crimson clover (320 kg DM·ha<sup>-1</sup>) in the 3rd harvest, and by red clover (660 kg DM·ha<sup>-1</sup>) in the 4th harvest, whereas hairy vetch presented the lowest productivity in both harvests (33 and 31 kg DM·ha<sup>-1</sup>). Dry matter yield of annual ryegrass and rye-annual ryegrass did not differ among harvests. Grass DM yield was greater for N-fertilized grass in the 2nd and subsequent harvests (660, 1360, and 1220 kg DM·ha<sup>-1</sup>). Red clover and the crimson clover-red clover-ball clover mixture were not different in DM yield than N-fertilized grass in the last harvest, while the other mixtures did not differ from the unfertilized grass.

**Key Words:** annual ryegrass, rye, crimson clover

**M111 Length of ensiling effects on fermentation characteristics, DM recovery and aerobic stability of corn Shredlage.** Luis C. Solórzano<sup>\*1</sup>, Luis L. Solórzano<sup>2</sup>, and Abner A. Rodríguez<sup>1</sup>, <sup>1</sup>Universidad de Puerto Rico, Mayagüez, PR, <sup>2</sup>Lankin, Fitchburg, WI.

The effect of length of ensiling (LOE) on fresh whole-plant corn Shredlage was evaluated after it was fermented for 0, 45, 90, or 180 d at a temperature of 20 to 23°C using 3-L capacity PVC mini-silos fitted with mechanics to vent gas. Mini-silos were filled with about 1.9 kg of the crop at about 33% DM, >3.1% soluble carbohydrates (fresh matter). Four mini-silos were opened at each LOE to determine fermentation characteristics and DM recovery. Statistical analysis was performed according to a completely randomized design with a 4 LOE. Aerobic stability (AS) was determined by placing the ensiled material from each mini-silo in Styrofoam containers placed inside a Styrofoam chamber. Temperature was measured at 6-h intervals for 7 d. Statistical analysis used a split-plot design with a 3 LOE by 29-timepoint factorial arrangement using silo as the repetitive measurement. Significance ( $P < 0.05$ ) is denoted by <sup>a, b, c</sup>. Ensiling increased the % of lactic acid, from fresh

(0.12<sup>a</sup>), to 45 d (5.26<sup>b</sup>), 90 d (6.32<sup>b</sup>), and 180 d (5.31<sup>b</sup>). A similar trend occurred for acetic (0.07<sup>a</sup>) and propionic acid (<0.01<sup>a</sup>) from 0d to 45d (0.51<sup>b</sup>, 0.16<sup>b</sup>), 90 d (0.72<sup>b</sup>, 0.19<sup>b</sup>) and 180 d (0.62<sup>b</sup>, 0.16<sup>b</sup>). Low TVFA at 0 d (0.19<sup>a</sup>), increased at 45 d (5.93<sup>b</sup>), 90 d (7.23<sup>b</sup>) and 180 d (6.09<sup>b</sup>). Butyric acid was <0.01 and not different among LOE. Recovery of DM did not differ among LOE and averaged 91.33%. Starting pH at 0d was highest (5.75<sup>a</sup>) compared with 45d (3.71<sup>c</sup>) and 90 d (3.64<sup>c</sup>), and both of these were lower than pH at 180 d (3.87<sup>b</sup>). Ammonia-N (% of CP) has an increasing trend from 0 d (0.65<sup>c</sup>) to 45 d (2.54<sup>b</sup>) and 90 d (2.37<sup>b</sup>), which were lower than at 180 d (4.5<sup>a</sup>). Loss of AS occurred on 45 d by 36 h post-aerobic exposure (PAE), 90 d by 48 h PAE, and 180 d by 42 h. PAE. Ensiling resulted in increases of lactic, acetic, propionic acids and TVFA, resulting in a lower pH. There were no differences in these parameters after 45 d of ensiling. Ammonia-N increased due to ensiling and was not different between 45 and 90 d, and increased after 180 d. The shorter LOE resulted in lower AS. Shredlage continues to cure for at least 180 d of fermentation.

**Key Words:** corn silage, fermentation, aerobic stability

**M112 Fermentation of frozen whole-plant corn silage after defrosting.** Luiz F. Ferraretto<sup>\*1</sup>, Gilson S. Dias Junior<sup>1,2</sup>, John P. Goeser<sup>1,3</sup>, and Randy D. Shaver<sup>1</sup>, <sup>1</sup>University of Wisconsin, Madison, WI, <sup>2</sup>Universidade Federal de Lavras, Lavras, MG, Brazil, <sup>3</sup>Rock River Laboratory Inc., Watertown, WI.

Late harvest of whole-plant corn silage (WPCS) into late fall and winter months during 2014/2015 raised concerns among central and northern Wisconsin dairy farmers and their nutritionists about fermentation of frozen WPCS. The objective of the present study was to evaluate fermentation profile of defrosted WPCS after several months stored frozen. An unfermented WPCS sample that had been obtained from the University of Wisconsin–Madison Agricultural Research Station (Arlington, WI) on September 23, 2014, was immediately frozen and stored at -20°C until January 26, 2015. Sample was defrosted, homogenized, and divided into 24 sub-samples of 250 g each. Sub-samples were vacuum-sealed in plastic bags and randomly assigned to 8 treatments so that each treatment had 3 replications. Treatments were 0, 0.5, 1, 2, 3, 7, 14, and 28 d fermentations. Bags were stored in the dark at room temperature (approximately 20°C) until reaching the targeted ensiling time. All samples were analyzed for DM, pH, organic acids and ammonia-N (%DM). Data were analyzed using Proc Mixed of SAS with the Fixed effect of ensiling time and the random effect of bag. Content of DM did not differ ( $P = 0.31$ ; 36.1% on average). Measurements of pH were affected by ensiling time ( $P = 0.001$ ) with a decline observed after only 1 d (5.23) of fermentation and a gradual decrease until 28 d (3.84). This is related to the gradual increase ( $P = 0.001$ ) in lactate and acetate concentrations from 1 (0.88% and 0.39%, respectively) to 14 d of fermentation (5.48% and 1.22%, respectively). Total acid concentrations follow the same pattern ( $P = 0.001$ ). Propionate concentration did not differ ( $P = 0.77$ ) whereas butyrate was not detected. Concentration of succinate increased ( $P = 0.001$ ) after 1 d of fermentation, peaked on 3 d and decreased on 14 d. Ethanol concentration was greater ( $P = 0.001$ ) for 2, 3, 7, 14 and 28 d (0.32% on average) compared with 0, 0.5 and 1 d (0.03% on average). Ammonia-N increased ( $P = 0.001$ ) 3-fold from 0 to 28 d of fermentation (0.02% vs. 0.06%, respectively). These findings

suggest that WPCS maintains fermentation capacity upon defrosting even after frozen for a prolonged period in storage.

**Key Words:** corn silage, fermentation

**M113 Length of ensiling effects on starch characteristics of corn Shredlage.** Luis C. Solórzano\*<sup>1</sup>, Luis L. Solórzano<sup>2</sup>, Beatriz A. Quintana<sup>1</sup>, and Abner A. Rodríguez<sup>1</sup>, <sup>1</sup>Universidad de Puerto Rico, Mayagüez, PR, <sup>2</sup>Lankin, Fitchburg, WI.

The effect of length of ensiling (LOE) on fresh whole-plant corn Shredlage was evaluated after it was fermented for 0, 45 or 180 d at a temperature of 20–23°C using 3 L capacity PVC mini-silos fitted with mechanics to vent gas. Mini-silos were filled with about 1.9 kg of the crop at about 33% DM, > 3.1% soluble carbohydrates (fresh matter). Four mini-silos were opened at each LOE to determine starch characteristics and in vitro starch digestibility (IVSd) during 2 in vitro fermentation endpoints (FEP, 3 or 7 h.). Statistical analysis for nutritional characteristics was performed using a completely randomized design (CRD) for 3 LOE. Data for IVSd was analyzed using a CRD with a 3 LOE by 2 FEP factorial arrangement. Different superscripts denote statistical significance ( $P < 0.05$ ). The structure of the starch granule was documented using a scanning electron microscope (SEM) at each LOE. Ammonia-N (% of CP) and IVSd increased as LOE increased, while sugar decreased (Table 1). There were no changes for starch content across LOE. Increasing the FEP from 3 to 7 h. increased IVSd from 72.4<sup>a</sup> to 84.1<sup>b</sup>%. The 2,000× micrograph at ensiling (0 d) shows starch granules with a smooth surface and a spherical shape surrounded by protein bodies. After 45 d of ensiling, the starch granules lost their sphericity and some of the protein bodies start to break down into small pieces (supported by increases in ammonia-N). After 180 d of ensiling, starch agglutination may be observed as well as some pitting of the starch granule due to hydrolytic enzyme activity. In summary, structural changes in the starch-protein matrix and in the starch granule during ensiling result in increased starch digestibility.

**Table 1 (Abstr. 113).** Effects of length of ensiling (0, 45, or 180 d) on nutrient characteristics and starch digestibility of corn Shredlage

Item	Length of ensiling, d					
	0		45		180	
	Mean	SD	Mean	SD	Mean	SD
N	4		8		8	
Ammonia-N, % of CP	0.65 <sup>c</sup>	0.1	2.54 <sup>b</sup>	0.04	4.5 <sup>a</sup>	0.39
Starch, % of DM	34.2	2.39	30.4	1.58	32.2	1.84
Sugar, % of DM	8.7 <sup>a</sup>	0.21	1.9 <sup>b</sup>	0.26	2.8 <sup>c</sup>	0.22
In vitro starch digestibility, % of starch	65.4 <sup>a</sup>	11.2	76.5 <sup>ab</sup>	10.8	84.1 <sup>b</sup>	8.1

**Key Words:** length of ensiling, corn silage, starch digestibility

**M114 Relationship between corn silage processing score and kernel fraction geometric mean particle size in whole-plant corn silage.** Gilson S. Dias Junior<sup>1,2</sup>, Luiz F. Ferraretto\*<sup>1</sup>, Gustavo G. S. Salvati<sup>1</sup>, Lucas C. de Resende<sup>1,2</sup>, Pat C. Hoffman<sup>1</sup>, and Randy D. Shaver<sup>1</sup>, <sup>1</sup>University of Wisconsin, Madison, WI, <sup>2</sup>Universidade Federal de Lavras, Lavras, MG, Brazil.

Greater kernel processing increases starch digestibility in whole-plant corn silage (WPCS). Corn silage processing score (CSPS), the percentage of starch passing through a 4.75 mm sieve, is widely used to assess degree of kernel breakage in WPCS. However, the geometric mean particle size (GMPS) of the kernel fraction that passes the 4.75 mm sieve has not been well described. Therefore, the objective of this study was to evaluate the relationship between CSPS and GMPS of the kernel fraction. Samples of WPCS ( $n = 80$ ) from 3 field trials representing varied theoretical length of cut settings and processor types and settings were evaluated. Each sample was divided in 2 and then dried at 60°C for 48 h. The CSPS was determined in duplicate on one of the split samples, while on the other split sample the kernel and stover fractions were separated using a hydrodynamic separation procedure. After separation, the kernel fraction was re-dried at 60°C for 48 h in a forced-air oven and dry sieved to determine GMPS ( $\mu\text{m}$ ) and surface area using 9 sieves with nominal square apertures of 9.5, 6.7, 4.75, 3.35, 2.36, 1.70, 1.18, 0.589 mm and pan. Regressions to determine linear and quadratic relationships were performed using Proc Reg of SAS (SAS Institute, 2004). Linear relationships between CSPS and kernel fraction GMPS ( $R^2 = 0.11$ ;  $P = 0.01$ ), surface area ( $R^2 = 0.06$ ;  $P = 0.03$ ) and proportion passing through the 4.75 mm screen ( $R^2 = 0.34$ ;  $P = 0.001$ ) were poor. Strong quadratic relationships between proportion of kernel fraction passing through the 4.75 mm screen and kernel fraction GMPS ( $R^2 = 0.70$ ;  $P = 0.001$ ) and surface area ( $R^2 = 0.56$ ;  $P = 0.001$ ) were observed. Corn silage processing score was poorly correlated with kernel fraction GMPS and surface area. These findings suggest that hydrodynamic separation and dry sieving of kernel fraction may provide better assessment of kernel breakage in WPCS.

**Key Words:** mean particle size, corn silage, processing score

**M115 The effects of different silo plastics on the fermentation, aerobic stability, and dry matter recovery of corn silage.** Michelle Windle\*, Vita Plus, Madsion, WI.

The objective of this experiment was to evaluate the efficacy of 4 different silo plastics to mitigate silage surface spoilage. Bag silos (60 cm × 30 cm) were created by heat-sealing single layers of 4 different types of silo plastics: black and white plastic (BW, KSI Supply Inc., Sheboygan Falls, WI), Silostop oxygen barrier plastic (SS, Bruno Rimini Ltd., London, United Kingdom), KSI oxygen barrier plastic (KSI), and Hitec oxygen barrier plastic (HT, Shanghai Hitec Plastics Co. Ltd., Shanghai, China). There were 5 silos per each types of plastic, yielding 20 experimental silos. Approximately 2.75 kg of chopped whole plant corn (40.9% DM) were placed in each silo, manually compressed and sealed. One sample per plastic type was analyzed for oxygen transfer rate (OTR). Silages were stored for 112 d at 15–21°C and data were analyzed using the Fit Model procedure of JMP. Means were compared using the Tukey's test. Relationships were evaluated between OTR and fermentation parameters. The OTR for BW, KSI, SS and HT were 4039, 154, 23.2 and 8769 cc/m<sup>2</sup>/d, respectively. Dry matter, CP, soluble CP, NH<sub>3</sub>-N, and water-soluble carbohydrates of corn silage were not affected by plastic type ( $P > 0.05$ ). Silages ensiled with HT had a lower DM recovery, higher pH, less lactic and acetic acids, and less ethanol, as compared with silages stored in BW, SS or KSI ( $P < 0.05$ ). Aerobic stability was lowest for silages ensiled within HT (8 h), intermediate for BW (37 h) and greatest for KSI (76 h) and SS (67 h) ( $P < 0.05$ ). Colony forming units of yeasts for KSI, SS, BW and HT were 3.94, 4.56, 6.19, and 8.05, respectively. Relationships between OTR and fermentation parameters indicated that as OTR increased, corn silage was less stable, had a higher pH, more yeasts, ethanol, and a lower lactic-to-acetic ratio ( $P < 0.05$ ).

Results indicate that ensiling corn silage with differing “oxygen barrier” plastics results in large differences in silage quality.

**Key Words:** spoilage, silo plastic, yeast

**M116 Length of ensiling effects on nutritional characteristics and in vitro NDF digestibility of corn Shredlage.** Luis C. Solórzano\*<sup>1</sup>, Luis L. Solórzano<sup>2</sup>, and Abner A. Rodríguez<sup>1</sup>, <sup>1</sup>Universidad de Puerto Rico, Mayagüez, PR, <sup>2</sup>Lankin, Fitchburg, WI.

We evaluated the effect of length of ensiling (LOE) on fresh whole plant corn shredlage (CS) that was fermented for 0, 45, 90, or 180 d at a temperature of 20 to 23°C using 3-L capacity PVC mini-silos fitted with mechanics to vent gas. Mini-silos were filled with approximately 1.9 kg of the crop at about 33% DM, >3.1% soluble carbohydrates (fresh matter). Four mini-silos were opened at each LOE to determine nutritional characteristics and in vitro NDF digestibility (NDFd) during 3 in vitro fermentation endpoints (FEP, 0, 30 or 120 h.) at a commercial laboratory. Statistical analysis for nutritional characteristics was performed using a completely randomized design (CRD) for 4 LOE. Data for NDFd was analyzed using a CRD with a 4 LOE by 3 FEP factorial arrangement. Significance ( $P < 0.05$ ) is denoted by <sup>a, b, c</sup>. There were no LOE differences ( $P > 0.5$ ) on the % content of ADF, NDF, lignin and ash, which averaged, 24.19, 36.71, 1.76, and 3.96, respectively. There were significant differences in the % content of DM, CP, ADF bound protein (ADFbP), fat and cell wall protein (CWP). Increasing LOE led to a decrease in the DM content from 0 d (36.68<sup>a</sup>), to 90 d (32.25<sup>b</sup>), and 180 d (32.51<sup>b</sup>), but did not differ from 45 d (34.04<sup>ab</sup>). The content of CP increased for LOE 90 d (8.78<sup>a</sup>) and 180 d (8.16<sup>a</sup>) compared with LOE at 0 d (7.19<sup>b</sup>) or 45 d (7.32<sup>b</sup>). The amount of ADFbP was 0.52<sup>b</sup> at 0 d, similar to ADFbP at 90 d (0.62<sup>b,c</sup>). The ADFbP content at 90 d did not differ from 45 d (0.74<sup>a</sup>) and 180 d (0.67<sup>a,c</sup>). Fat content increased at 90 d (3.40<sup>a</sup>) and 180 d (3.45<sup>a</sup>) compared with 0 d (1.95<sup>b</sup>) or 45 d (2.36<sup>b</sup>). At 0 d (0.18<sup>b</sup>) CWP did not differ from 45 d (0.47<sup>b</sup>), or 180 d (0.28<sup>b</sup>), but all 3 did differ from 90 d (1.20<sup>a</sup>). Increasing the LOE led to an increase in IVNDFd from 0 d (40.39<sup>a</sup>) and 45 d (34.91<sup>a</sup>) to 90 d (50.46<sup>b</sup>) or 180 d (60.83<sup>c</sup>). Ensiling leads to various nutritional changes over time, which can affect ration balancing and on-farm ration preparation. This calls for continuous silage nutritional monitoring over time to proper balance rations and feed cows.

**Key Words:** corn silage, fiber digestibility, nutritional characteristics

**M117 Ensiling practices of corn on California dairies.** Jennifer M. Heguy\*<sup>1</sup> and Noelia Silva-del-Río<sup>2</sup>, <sup>1</sup>University of California, Ag & Natural Resources, Modesto, CA, <sup>2</sup>University of California, VMTRC, Tulare, CA.

The aim of this study was to obtain information on current corn ensiling practices in California’s San Joaquin Valley. In summer 2014, 20 dairies were visited to create a snapshot of ensiling practices. Producers answered a short survey, and observations at the silage structure were made. Herd size ranged from 350 to 5250 cows (median = 1800). Five consecutive truckloads of corn silage delivered to dairies were sampled and composited for wet chemistry nutrient analysis (Table 1). Descriptive statistics were conducted with PROC MEANS of SAS and correlations were evaluated with PROC CORR of SAS. Structures were primarily wedge piles (n = 16). Delivery rate varied; the 5 truckloads of corn were delivered in as little as 8 min and in as many as 64 min. Most dairies utilized one packing tractor (n = 12), with delivery rate of the 5 loads ranging from 8 to 40 min. Fewer dairies used 2 packing tractors (n = 7), with delivery rate ranging from 10 to 64 min, and one

dairy utilized 3 packing tractors with a delivery rate of 22 min. Fourteen custom harvesting companies were utilized on the 18 dairies that did not harvest their own corn, with 4 of the companies harvesting on more than one dairy. Corn silage processing score (CSPS) was analyzed; 9 samples were optimally processed (CSPS > 70%) and 11 samples were inadequately processed (CSPS between 50% and 70%). No samples were inadequately processed. No correlation was found between DM and starch content ( $r = 0.290$ ,  $P = 0.214$ ) or DM and CSPS ( $r = -0.001$ ,  $P = 0.995$ ). Harvesting parameters varied, but kernel processing was a practice utilized on each dairy; CSPS showed that all samples were either adequately or optimally processed.

**Table 1 (Abstr. M117).** Nutrient composition (% of DM unless otherwise noted) of chopped corn (n = 20) taken at harvest in California’s San Joaquin Valley

	DM, %	CP	ADF	NDF	Starch	NFC	Ash	CSPS (%)
Average	35.9	7.7	24.4	41.0	30.2	43.6	5.4	70.7
Median	35.9	7.8	24.9	42.3	29.0	43.2	5.4	69.4
Minimum	31.2	6.2	20.2	35.2	23.3	36.6	4.2	50.7
Maximum	40.3	8.8	28.3	46.7	36.7	50.7	6.8	82.2
SD	2.5	0.6	2.1	2.8	3.6	3.1	0.7	9.8

**Key Words:** California, corn silage, corn silage processing score

**M118 Investigation into the accuracy of a commercially available activity meter for measuring grazing duration.** Emer Kennedy\*, James Moloney, Donagh P. Berry, Michelle Liddane, and Frank Buckley, Teagasc, Animal & Grassland Research and Innovation Centre, Moorepark, Fermoy, Co. Cork, Ireland.

Grass intake is influenced by grazing duration. Current methods of measuring grazing behavior are best suited to research environments. Activity meters, used primarily for oestrus detection, are now incorporating behavioral measures. The hypothesis was that an activity meter would provide similar information on grazing duration to that generated by a behavior recorder. One hundred 24-h records were obtained from grazing Holstein-Friesian cows between August 26 and September 30, 2014. Grazing time was recorded using IGER behavior recorders (IGER), which were processed using the ‘Graze’ analysis software. All cows were fitted with MooMonitor+ activity meters (MOO; Dairymaster, Causeway, Co. Kerry). Total grazing time data from the MOO was obtained from the Dairymaster database. Total 24-h grazing duration determined by the IGER was linearly regressed on the MOO grazing duration to determine the association between each method; no intercept was fitted in the model. A fixed effects linear model was used to quantify the association between parity, measurement day and IGER recorder number on the IGER recorded grazing duration. Two-way interactions between the MOO grazing duration and the main fixed effects were considered in the model to determine if the association between the MOO and IGER grazing duration differed by any of the main effects. Mean 24-h grazing duration was 543 (±6.39 SEM) min for IGER and 540 (±5.9 SEM) min for MOO. A correlation of  $r = 0.72$  existed between the IGER and MOO grazing durations. The regression coefficient of IGER grazing duration on MOO grazing duration was 1.003 (±0.008 standard error) which was not different from the expectation of unity should the measured differences in grazing duration between the 2 devices be equivalent. Neither parity nor calendar day was associated with the difference in grazing duration estimated by the 2 devices. The regression coefficient of the IGER on the MOO grazing duration varied from 0.87 to 1.1. The results from this study, albeit from a limited data



set, indicate good concordance between 24-h grazing duration estimated from the IGER and MOO recorders.

**Key Words:** grazing behavior, dairy cow, activity meter

**M119 Forage quality of two different pasture systems incorporating warm and cool season forages for grazing organic dairy cattle.** Kathryn E. Ruh\*<sup>1,2</sup>, Bradley J. Heins<sup>1,2</sup>, and Jim C. Paulson<sup>3</sup>, <sup>1</sup>West Central Research and Outreach Center, Morris, MN, <sup>2</sup>University of Minnesota, St. Paul, MN, <sup>3</sup>University of Minnesota Extension, Rochester, MN.

Two pasture systems (cool and warm season grass species) with enhanced in-field and landscape level species diversity were analyzed for forage quality characteristics across the grazing season at the West Central Outreach and Research Center organic dairy in Morris, Minnesota, for 2 years. System 1 was a diverse-mixture of cool season grasses and legumes (perennial ryegrass, white clover, red clover, chicory, orchardgrass, meadow brome, alfalfa, meadow fescue). System 2 was a combination of perennial polycultures and annual-warm season grasses (BMR sorghum-sudangrass and teff grass). Grazing of lactating cows was initiated when forages were 20–30 cm tall and strip size was adjusted to leave 7 to 13 cm of refusals. Random samples of pasture forage were sampled every other day when a group of cows moved to a new paddock. Pasture clippings were randomly collected in a 0.76-m<sup>2</sup> square of pasture. Forage samples were sent to Rock River Laboratory, Inc., Watertown, WI and were analyzed with NIR spectrophotometry for DM, CP, and total-tract NDF digestibility (TTNDFD). Data were analyzed using the MIXED procedure of SAS. Independent variables for analyses were the fixed effects of system [cool (1) or cool-warm (2)], month (May to October), forage (grass pasture, turnips, BMR sorghum-sudangrass or teff), year (2013 or 2014) and their interactions, and date of harvest was a random variable. The DM averaged 20.7% and 21.2% for systems 1 and 2, respectively ( $P = 0.75$ ). The CP was 21.2% and 18.3% for systems 1 and 2, respectively ( $P < 0.05$ ). The CP for system 1 averaged 19.9% in 2013 and 22.5% in 2014 ( $P < 0.01$ ). The CP for system 2 averaged 16.1% in 2013 and 20.4% in 2014 ( $P < 0.01$ ). The TTNDFD averaged 69.9% and 53.1% for system 1 and system 2, respectively ( $P < 0.01$ ). The TTNDFD was 78.1% in 2013 and 61.7% in 2014 for cool-season grasses, and 59.8% in 2013 and 46.4% in 2014 for warm season grasses ( $P < 0.0001$ ). In summary, CP and TTNDFD were greater in cool-season pasture systems; however, DM did not differ between pasture systems. Yearly effects and weather may affect forage quality in both pasture production systems.

**Key Words:** teff, organic, grazing

**M120 Effect of starchy or fibrous carbohydrate supplementation of an herbage diet on ruminal fermentation and methane output in continuous culture.** Kathy J. Soder\*<sup>1</sup>, Aimee N. Hafila<sup>1</sup>, Andre F. Brito<sup>2</sup>, Melissa D. Rubano<sup>1</sup>, and Curtis J. Dell<sup>1</sup>, <sup>1</sup>USDA-ARS, Pasture Systems and Watershed Management Research Unit, University Park, PA, <sup>2</sup>University of New Hampshire, Durham, NH.

A dual-flow continuous culture fermentor system was used to assess the effect of supplementing 2 levels (5 or 10% of diet DM) of starchy (barley: BAR) or fibrous (beet pulp: BP) carbohydrate (CHO) to an orchardgrass diet on nutrient digestibility, VFA production, bacterial protein synthesis, and methane output. Treatments were randomly assigned to fermentors in a 4 × 4 Latin square design with a 2 × 2 factorial arrangement using 7 d for diet adaptation and 3 d for sample collection. Treatments included: 1) 57 g DM herbage + 3 g DM BAR; 2) 54 g DM herbage +

6 g DM BAR; 3) 57 g DM herbage + 3 g DM BP; 4) 54 g DM herbage + 6 g DM BP. Feeding and pH sampling occurred at 0730, 1030, 1400 and 1900 h. Gas samples for methane analysis were collected at 0725, 0900, 1000, 1355, 1530, and 1630 h. Effluent samples were analyzed for OM, CP, NDF, nutrient digestibilities, estimation of bacterial protein synthesis, ammonia-N and VFA. Data were analyzed using the MIXED procedure of SAS with period and treatment as fixed effects and fermentor as random. Orthogonal contrasts were tested for CHO type and level. No significant interactions were detected. Apparent and true OM digestibilities were not affected ( $P > 0.10$ ) by CHO source (72.4 and 81.9%, respectively). True CP digestibility was greater ( $P < 0.05$ ) for BP (75.3%) than BAR (52.5%) diets. Apparent NDF digestibility was lower ( $P < 0.05$ ) for BP (79.5%) than BAR (85.1%) diets. Barley diets produced lower ( $P < 0.05$ ) molar proportions of acetate (43.5 vs. 49.4 mol/100 mol, respectively), lower concentrations of total VFA (67.2 vs. 72.2 mmol/L, respectively) and tended ( $P = 0.08$ ) to have greater mean pH (6.75 vs. 6.72) compared with BP diets. Methane production was not affected ( $P > 0.10$ ) by CHO source. The 10% supplement produced greater ( $P < 0.05$ ) concentrations of methane (35 vs. 27 mmol/d) and tended ( $P = 0.07$ ) to increase apparent DM digestibility. Diet had no effect on bacterial efficiency or ammonia-N. Supplementation of an herbage-based diet with BP improved CP digestibility compared with barley but did not affect OM digestibility, methane production, or microbial efficiency.

**Key Words:** barley, beet pulp, ruminal fermentation

**M121 Heifer growth performance from fall-oat pastures.** Wayne K. Coblenz\*<sup>1</sup>, Geoff E. Brink<sup>2</sup>, Nancy M. Esser<sup>3</sup>, Jason S. Cavadini<sup>3</sup>, and Patrick C. Hoffman<sup>4</sup>, <sup>1</sup>US Dairy Forage Research Center, Marshfield, WI, <sup>2</sup>US Dairy Forage Research Center, Madison, WI, <sup>3</sup>University of Wisconsin, Marshfield, WI, <sup>4</sup>University of Wisconsin, Madison, WI.

Fall-grown oat has shown promise as an emergency fall forage option, or to extend the grazing season in Wisconsin. Our objectives for this project were: (1) to assess the pasture productivity and forage characteristics of 2 fall-grown oat cultivars (Ogle and ForagePlus; OG and FP, respectively) using grazing initiation dates timed to late-September (EARLY) or mid-October (LATE); and (2) to evaluate growth performance by heifers grazing these oat forages compared with performance of heifers reared under controlled conditions with traditional confinement management (CONTROL). A total of 160 gravid Holstein heifers (80 heifers/yr) were stratified by weight, and assigned to 10 research groups (8 heifers/group). Initial BW was 509 ± 40.5 kg in 2013 and 517 ± 30.2 kg in 2014. Heifer-groups were maintained as units, and assigned to specific pastures arranged as a 2 × 2 factorial of oat cultivars and grazing initiation dates. Grazing heifer groups were allowed to strip-graze oat pastures for 6 h daily before returning to the barn, where they were offered a forage-based basal TMR. During both years, oat forage mass increased until early-November before declining in response to freezing weather conditions, exhibiting linear ( $P < 0.01$ ) and quadratic ( $P < 0.01$ ) effects of calendar date, regardless of oat cultivar. For 2013 and 2014, the respective maximum forage mass was 5329 and 4501 kg/ha for FP, and 5046 and 5111 kg/ha for OG. ForagePlus oat did not reach the boot stage of growth during either year; in contrast, OG matured more rapidly, and reached a late-heading stage during 2013, but only the early-boot stage in 2014. For 2013, ADG for CONTROL did not differ from grazing heifer groups (overall mean = 0.63 kg/d;  $P = 0.619$ ); however, ADG from FP was greater than OG (0.68 vs. 0.57 kg/d;  $P = 0.02$ ), and greater from EARLY than LATE (0.82 vs. 0.43 kg/d;  $P < 0.01$ ). During 2014, ADG from CONTROL exceeded grazing heifer groups

(0.81 vs. 0.57 kg/d;  $P = 0.01$ ), and ADG from EARLY again exceeded LATE (0.70 vs. 0.44 kg/d;  $P < 0.01$ ). These results suggest that delaying grazing until mid-October to allow more oat forage to accumulate will consistently suppress heifer growth performance.

**Key Words:** heifer, grazing, oat

**M122 Fertilization of fall-grown oat with dairy slurry or urea.**

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Oat has shown promise as a fall-forage option for dairy producers in Wisconsin. Our objectives were to assess the effects of summer applications of urea fertilizer or dairy slurry on the DM yield, N uptake, and apparent N recovery from fall-grown oat forages. 'ForagePlus' oat was established in early-August of 2013 and 2014, and fertilized with 0, 20, 40, 60, 80, or 100 kg N/ha as urea (46-0-0), or dairy slurry applied at rates of 43,250 (LM) or 86,500 L/ha (HM). All plots were harvested in early-November of each year. Yields of DM increased in response to fertilization with urea, exhibiting linear ( $P < 0.01$ ) and quadratic ( $P = 0.03$ ) effects of fertilization rate. The 2-yr mean DM yield at the 100 kg N/ha fertilization rate was 3968 kg DM/ha, which was nearly twice that of the unfertilized (0 kg N/ha) check plots (2105 kg DM/ha). Yields of DM from LM and HM plots differed from unfertilized check plots (3164 vs. 2105 kg DM/ha;  $P < 0.01$ ), but did not differ from each other (3029 vs. 3298 kg DM/ha;  $P = 0.15$ ). Collectively, total N uptake from plots fertilized with urea differed from unfertilized check plots (89 vs. 44 kg N/ha;  $P < 0.01$ ), and also increased linearly ( $P < 0.01$ ) with N fertilization rate from 44 kg N/ha for the unfertilized checks to 110 kg N/ha at the greatest urea fertilization rate. Uptake of N for plots receiving dairy slurry also differed from unfertilized check plots (77 vs. 44 kg N/ha;  $P < 0.01$ ), but the HM and LM application rates only tended to differ (82 vs. 71 kg N/ha;  $P = 0.06$ ). The apparent N recoveries from plots receiving urea differed from those receiving dairy slurry (83.0 vs. 23.1%;  $P < 0.01$ ); however, apparent N recoveries for plots fertilized with urea only tended ( $P = 0.10$ ) to decrease with N fertilization rate (range = 101.2 to 67.8%), and apparent N recoveries for LM and HM plots did not differ (27.2 vs. 19.0%;  $P = 0.65$ ). When expressed as a percentage of the  $\text{NH}_4\text{-N}$  applied within dairy slurry, apparent N recoveries for LM and HM accounted for approximately half of the  $\text{NH}_4\text{-N}$  applied, but there was no statistical difference between slurry rates (66.7 vs. 46.9%;  $P = 0.22$ ). Overall, fall-grown oat exhibited excellent ability to recover readily available fertilizer and manure N during a short fall growing season.

**Key Words:** apparent N recovery, fall-grown oat, N uptake

**M123 Effect of different ruminal incubation orders on in situ degradability of maize silage and alfalfa haylage in lactating dairy cows.**

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A study using 4 ruminally cannulated Holstein cows was conducted to evaluate the degradability of maize silage and alfalfa haylage by different ruminal incubation orders. Maize silage and alfalfa haylage samples were dried and ground to pass through a 2.5-mm screen. All nylon bags containing feed samples for each cow were either placed in rumen simultaneously (OI) or removed from rumen simultaneously (OII). Samples were incubated in the rumen for 2, 6, 12, 24, 36,

48, and 72 h. For maize silage, OI had higher neutral detergent fiber (NDF) and acid detergent fiber (ADF) degradability at 6 h and 12 h ( $P < 0.05$ ) compared with OII. Contrarily, slowly degraded fraction (b) of OI was significantly lower than that of OII ( $P < 0.05$ ). For alfalfa haylage, different ruminal incubation orders had significant effect ( $P < 0.05$ ) on the NDF degradability at 6 h and 12 h, constant rate of slowly NDF degraded fraction ( $K_d$ ), and rapidly ADF degraded fraction (a) (Table 1). Overall, the effects of different ruminal incubation orders concentrated during first 12 h of incubation period, but did not affect ruminally degradable part.

**Table 1 (Abstr. 123).** Degradability and degradation variations of NDF and ADF of maize silage and alfalfa haylage incubated in situ in different ruminal incubation orders<sup>1</sup>

Item	Maize silage				Alfalfa haylage			
	OI	OII	t	P-value	OI	OII	t	P-value
<b>NDF</b>								
2 h	7.14	5.59	0.98	0.39	3.70	5.59	-0.53	0.19
6 h	1.41 <sup>a</sup>	6.23 <sup>b</sup>	4.01	0.01	11.62 <sup>b</sup>	7.75 <sup>b</sup>	0.20	<0.01
12 h	17.92 <sup>a</sup>	8.38 <sup>b</sup>	4.03	0.01	21.31 <sup>a</sup>	15.76 <sup>b</sup>	0.10	0.05
a	1.84	-1.33	2.25	0.07	-0.52	0.96	-1.19	0.28
b	59.91 <sup>b</sup>	81.87 <sup>a</sup>	-4.16	0.01	38.20	51.02	-1.34	0.27
$K_d$	3.14	1.86	2.76	0.07	6.70 <sup>a</sup>	3.70 <sup>b</sup>	2.58	0.04
RDNDF	31.10	29.33	1.20	0.31	25.40	25.39	0.01	0.99
<b>ADF</b>								
2 h	4.02	4.18	-0.12	0.91	0.61	2.63	-1.36	0.25
6 h	7.15 <sup>a</sup>	3.65 <sup>b</sup>	2.49	0.05	7.73	4.74	0.65	0.15
12 h	14.14 <sup>a</sup>	7.34 <sup>b</sup>	2.77	0.03	17.60	13.03	1.71	0.14
a	-1.49	-3.30	1.05	0.34	-5.04 <sup>a</sup>	-1.94 <sup>b</sup>	-3.65	0.02
b	65.25 <sup>b</sup>	81.54 <sup>a</sup>	-3.15	0.03	42.35	44.43	-1.23	0.27
$K_d$	2.84	2.00	2.00	0.14	6.21	4.17	1.82	0.13
RDADF	28.85	28.45	0.28	0.79	22.84	23.09	-0.16	0.88

<sup>a,b</sup>Means with different superscript letters with the same row represent a significant difference between treatments ( $P < 0.05$ ).

<sup>1</sup>OI = placed simultaneously, OII = removed simultaneously, t = t-test, a = soluble fraction, b = slowly degradable fraction,  $K_d$  = rate of slowly degradability fraction, RDNDF/RDADF = ruminally degradable NDF/ADF.

**Key Words:** in situ incubation order, degradability, lactating cow

**M124 Productive performance and morphological composition of two genotypes of *Brachiaria* grazed by dairy calves in Mexico's Central Highlands.**

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The objective of this study was to evaluate the production and morphological characteristics of Insurgente (*Brachiaria brizantha*), and Mulato II (hybrid *Brachiaria*) forages, when grazed by dairy calves during the rainy season in Mexico's Central Highlands. The variables net herbage accumulation (NHA), leaf proportion and appearance of stems was evaluated in 3 periods consisting of 21 d each, between July and September 2014. The pasture used was composed of 16 plots (15 m × 4 m), with 4 plots per species and were fertilized with vermicompost (10 t·ha<sup>-1</sup>). The pasture was grazed continually by 4 dairy calves, aged

12 ± 4 mo, weight of 250 ± 35 kg. Sixteen exclusion cages (1m × 1m) and a metal quadrant measuring 0.16 m<sup>2</sup> (0.40 m × 0.40 m) were used. Leaf proportion was measured for each period, with a 100 g sample taken randomly from each plot. In the appearance of stems was identified the number of plants for each plot by m<sup>2</sup>, and selected a single plant randomly. At the beginning of each period the initial population of stems was measured for each plant, and marked with a plastic yellow ring. Then, new stems were counted every 15 d and marked with a ring of another color. The design was based on plots divided at random, with larger plots (species) and smaller plots (time periods). A variance analysis was conducted, using MINITAB, and measurements were compared using the Tukey's test ( $P < 0.05$ ). Significant differences ( $P < 0.015$ ) in the NHA were noted in the Insurgente and Mulato II species and in the interaction ( $P < 0.001$ ). Higher NHA was found in Mulato II forage (10,584 kg DM·ha<sup>-1</sup>). In both species the highest NHA was found in P2 and P3 2,662 and 3,858 kg DM·ha<sup>-1</sup>, respectively ( $P < 0.05$ ). Also, 11% more leaf appearance was found in Mulato II. The greatest appearance of stems was found in Mulato II forage (240 stems/m<sup>2</sup>;  $P < 0.001$ ). Insurgente and Mulato II demonstrated different growth behaviors under continuous grazing during the rainy season. Mulato II showed better performance, had higher net herbage accumulation, leaf proportion and appearance of stems.

**Key Words:** *Brachiaria*, pasture

**M125 Effect of processing methods on in situ degradability of maize silage and alfalfa haylage in lactating dairy cows.** Yang Zou\*, Shuangzhao Dong, Yun Du, Yajing Wang, Shengli Li, and Zhijun Cao, *State Key Laboratory of Animal Nutrition, College of Animal Science and Technology, China Agricultural University, Beijing, China.*

A study using 4 ruminally cannulated Holstein cows was conducted to evaluate the degradability of maize silage and alfalfa haylage prepared by different processing methods. Maize silage (20-mm length) and alfalfa haylage (40-mm length) were either wet (MSW; AHW) or air-dried (MSD; AHD). Air-dried silage and haylage were also ground to pass through a 2.5-mm screen (MSG; AHG). Samples were incubated in the rumen for 2, 6, 12, 24, 36, 48, and 72 h. Cows were fed ad libitum with free access to water. Treatment MSW had a lower acid detergent fiber (ADF) degradability at 2 h ( $P < 0.05$ ) compared with air-dried samples (MSD and MSG). Processing methods had significant effects ( $P < 0.05$ ) on NDF degradability at 72 h, ADF degradability at 36, 48, and 72 h, and ruminal degradable ADF. All of the highest values were observed in AHG treatment (Table 1). Based on this study, processing methods of drying and grinding should be taken into consideration when evaluating nutritive values of forages.

*Contd.*

**Table 1 (Abstr. M125).** Degradabilities of NDF and ADF in differently processed maize silage and alfalfa haylage incubated in situ<sup>1</sup>

Item	Maize silage				Alfalfa haylage			
	MSW	MSD	MSG	P-value	AHW	AHD	AHG	P-value
<b>NDF</b>								
2 h	13.72	16.68	14.48	0.31	24.62	27.76	27.81	0.30
36 h	40.96	41.91	39.91	0.94	50.79	46.23	51.89	0.27
48 h	43.40	42.69	40.80	0.80	51.08	52.75	55.96	0.18
72 h	48.58	55.20	47.31	0.28	53.68 <sup>b</sup>	56.54 <sup>ab</sup>	59.42 <sup>a</sup>	0.03
RDNDF	31.16	34.18	29.81	0.39	41.96	42.82	44.57	0.31
<b>ADF</b>								
2 h	12.16 <sup>b</sup>	14.58 <sup>ab</sup>	16.17 <sup>a</sup>	0.09	20.61	21.50	23.91	0.54
36 h	41.54	41.39	51.57	0.22	41.71 <sup>ab</sup>	39.26 <sup>b</sup>	46.68 <sup>a</sup>	0.04
48 h	44.54	45.21	43.30	0.97	46.19 <sup>b</sup>	47.66 <sup>b</sup>	53.10 <sup>a</sup>	0.03
72 h	50.06	54.43	50.95	0.68	49.20 <sup>b</sup>	51.75 <sup>b</sup>	57.05 <sup>a</sup>	<0.01
RDADF	31.41	33.14	33.04	0.85	35.75 <sup>b</sup>	36.62 <sup>b</sup>	40.04 <sup>a</sup>	0.04

<sup>a,b</sup>Means with different superscript letters with the same row represent a significant difference between treatments ( $P < 0.05$ ).

<sup>1</sup>MSW = wet maize silage, MSD = air-dried maize silage, MSG = air-dried ground maize silage, AHW = wet alfalfa haylage, AHD = air-dried alfalfa haylage, AHG = air-dried ground alfalfa haylage, RDNDF = ruminally degradable NDF, RDADF = ruminally degradable ADF.

**Key Words:** processing method, degradability, lactating cow

**M126 Effectiveness of plastic underlays with or without oxygen barrier properties in maintaining corn silage quality.**

Peter H. Robinson\*<sup>1</sup>, Nadia Swanepoel<sup>1</sup>, and Jim Ralles<sup>2</sup>, <sup>1</sup>*Department of Animal Science, University of California, Davis, CA*, <sup>2</sup>*ARI, Belmont, CA.*

Use of only a single layer of plastic to cover silage piles can lead to substantial surface spoilage of silage (i.e., increased pH and temperature, discoloration, molding). The extent of spoilage losses can be curtailed if a thin plastic underlay film is utilized under the outer white/black plastic cover. Our objective was to determine if a clear plastic polyethylene film (Poly) has an impact on the nutritional quality of the outer 50 cm layer of corn silage vs. an oxygen barrier (OB) film when used as an underlay. Two wedge type corn silage piles with east/west, and 2 with north/south, orientation were constructed in fall 2014. Each pile was alternately covered with a clear, pliable polyethylene film of 1.6 mil HiTec Underlay (ARI, Belmont, CA) or an OB plastic film of 1.8 mil from Industria Plastica, Mongralese, Italy (trade name 'Silostop') with a 1.8 m overlap between sections and then covered with a conventional plastic cover. After 90 d, all piles were sampled to a 50 cm depth with a silage probe, divided into 2 samples of 25 cm (i.e., 0–25, 25–50 cm depth) each at 4 sample points in each of the 4 sections. Temperatures and pH were determined at core extraction. Each core sample was preserved individually (n = 32) for mold and yeast counts and part was pooled by section location (upper vs. lower; n = 16) and analyzed for volatile fatty acids, lactic acid and ethanol. There were no differences between the silages under the Poly and OB films in temperature (24.7 vs. 24.2 for N/S and 23.1 vs. 22.4°C for E/W piles), pH (3.79 vs. 3.84 for N/S and 3.77 vs. 3.84 for E/W piles) or mold and yeast counts, which were all below 0.5 million cfu/g. Volatile fatty acids were similar between treatments, with lactic, acetic and propionic acid averaging 2.93 and 3.57, 1.73 and 1.93, 0.38 and 0.24% DM for N/S and E/W piles respectively, with none being statistically different. Butyric acid occurred at a very low level, 0.05% DM, and ethanol was 0.53% DM, with no differences. Overall, it was judged that there were no differences in the silage quality of the outer 50 cm layer of corn silage after ~90 d of ensiling when covered



with a clear polyethylene film vs. an oxygen barrier film under the outer black/white plastic cover.

**Key Words:** volatile fatty acids, silage quality

**M127 Defining and measuring losses (shrink) from well-managed corn silage silos, and identifying stages in silo life where losses occur.** Peter H. Robinson\*<sup>1</sup>, Nadia Swanepoel<sup>1</sup>, Jennifer Heguy<sup>2</sup>, and Deanne Meyer<sup>1</sup>, <sup>1</sup>*Department of Animal Science, University of California, Davis, CA* <sup>2</sup>*UCCE Stanislaus, San Joaquin & Merced Counties, University of California, Davis, CA.*

Silage shrink (weight lost between ensiling and feedout) represent loss of nutrients to dairy producers, and the potential to degrade air quality if that loss is as volatile carbon compounds, or degrade water quality due to seepage to surface water and seepage to subsurface aquifers. No research has documented silage shrink in large commercial silage structures (silos) common in the SW US. ‘Shrink’ can be expressed as loss of wet weight (WW), oven dry weight (oDM) and oDM corrected for volatiles lost in the oven (vcoDM). Shrink losses, and the phase of the process where losses occurred, were measured using 7 corn silage silos (2 rollover, 1 bunker, 4 wedge) from the 2013 crop year on 4 dairy farms in 2 San Joaquin Valley areas, all covered within 48 h with an oxygen barrier inner film and black/white outer plastic weighted with tire chains. Total WW, oDM and vcoDM losses (not including wastage) calculated from weights of fresh chop delivered to the silo and silage placed in a feed mixer (n = 7) were  $9.0 \pm 1.69$ ,  $6.8 \pm 1.82$  and  $2.8 \pm 2.08\%$ , suggesting that much of the WW shrink was water and much of the oDM shrink was volatiles driven off during oven drying. The largest part of shrink occurred in the silage mass (measured using in/out weights of 9–15 buried bags in each of 4 silos) before face exposure (WW, oDW and vcoDW losses from the mass were  $3.9 \pm 2.40$ ,  $7.2 \pm 1.12$  and  $3.5 \pm 1.27\%$  respectively), with losses from the exposed face (measured as loss in core weight between freshly exposed faces and ~21 h exposed faces from 4 cores of 50 cm depth on 2 occasions in each silo), as well as between face removal and the mixer (measured between compositional changes between freshly exposed faces and silage placed in the mixer on 2 occasions in 4 silos), being negligible. Silo bulk density, face management, rate of face use and face orientation had no obvious effects on shrink. Real shrink losses (i.e., vcoDM) of well managed corn silages piles are much lower than has been generally assumed, the exposed face is a very small portion of those losses, and many of the proposed mitigations may not be effective in reducing shrink, possibly because it is quantitatively so small in large well managed silos.

**Key Words:** volatile compound, air quality

**M128 Effects of sealing time post-filling and sealing material on fermentation, nutritional quality, and organic matter loss of whole-plant corn ensiled in a drive-over pile.** Katie Natcher<sup>1</sup>, Estela Uriarte<sup>2</sup>, Keith K. Bolsen\*<sup>2</sup>, Ron Kuber<sup>3</sup>, and Connie Kuber<sup>3</sup>, <sup>1</sup>*California Polytechnic State University, San Luis Obispo, CA*, <sup>2</sup>*Kansas State University, Manhattan, KS*, <sup>3</sup>*Connor Agriscience, Clovis, CA.*

Sealing time, immediate and 24-h delay, and sealing material, standard plastic (std.) and oxygen barrier (OB) film, was compared using whole-plant corn. The crop was harvested on August 21 and 22, 2013 in the 2-thirds milk line stage, contained 32% DM, and was inoculated at the forage harvester. The forage was ensiled in a drive-over pile, which was 18 m wide x 60 m long x 1.8 m apex height with an east to west orientation. One-half of the forage was harvested the first day, packed with a payloader, and not sealed. On the second day, the remainder of the corn was harvested and packed with a payloader. One-half of each day’s forage surface was sealed with a sheet of std. plastic, and the other half sealed with a sheet of OB film, which was protected from UV light with a sheet of std. plastic. The sealing materials were removed from the south half of the pile after 90 d. Samples were collected at 0 to 15, 15 to 30, and 30 to 45 cm depths from the surface at 3 north to south locations, which were equal distance from the east and west boundaries of each of the 4 sealing treatments. The results are presented for the mean of the 3 sampling depths. The immediate sealed silages had a lower ( $P < 0.05$ ) pH value than the delay sealed silages. The silage that was delay sealed with std. plastic had higher ( $P < 0.05$ ) ash and NDF contents and lower ( $P < 0.05$ ) NDF digestibility than the silage that was delay sealed with OB film and the silages that were sealed immediately with std. plastic or OB film. The OB film was more effective than std. plastic in preventing the entry of oxygen into the surface of the corn silage during the 90-d storage period. Delay sealing increased OM loss in the original top 0 to 45 cm of corn silage by 27.2% compared with immediate sealing (15.64 vs. 12.30%). However, delay sealing with OB film decreased OM loss in the original top 0 to 45 cm of corn silage by 20.6% compared with immediate sealing with std. plastic (12.33 vs. 15.54%).

**Key Words:** silage, sealing, delay

# Graduate Student Competition: ADSA Production Division Graduate Student Poster Competition, MS

**M129 Transcriptome profiling of pituitary gland from pre- and post-pubertal beef heifers.** Robmay Garcia\*, Dianelys Gonzalez-Pena, Bruce R. Southey, and Sandra L. Rodriguez-Zas, *University of Illinois at Urbana Champaign, Urbana, IL.*

Onset of puberty is a key component of animal production and reproductive efficiency. The pituitary gland plays a major role in reproductive readiness. A comprehensive understanding of the associated transcriptome changes during peripuberty will aid in the development of treatments and management practices to improve reproductive performance. The objective of this study was to gain understanding of the transcriptome associated with changes pre- and post-puberty. The transcriptome of pituitary tissue from 4 pre-pubertal beef heifers was compared with that of 4 different post-pubertal beef heifers using individual RNA-seq libraries. Single-end reads were mapped to the *Bos taurus* reference genome (Btau\_4.6.1) using Tophat v2.0.12. In total, 10,489 isoform transcripts pertaining to 10,234 genes were identified and 1,162 isoform transcripts pertaining to 1,149 genes were differentially expressed between post-puberty and pre-puberty pituitary tissue (False Discovery Rate-adjusted  $P$ -value  $< 0.05$ ) using Cufflinks v2.2.1. Among the most relevant and significant (false discovery rate adjusted  $P$ -value  $< 0.03$ ) genes associated with reproductive maturation were leptin receptor (*LEPR*) and vasoactive intestinal peptide (*VIP*), and endothelin 1 (*EDN1*). Increments in leptin receptors encoded by *LEPR* are associated with gonad maturation, the advent of reproductive maturity and fertility. *VIP* encodes the VIP neuropeptide that among other roles, participates in ovarian development and stimulation of prolactin release under the influence of estrogen. *EDN1* encodes the EDN1 neuropeptide that is involved in mechanisms controlling the gonadotropins and prolactin secretion, also inhibiting synthesis of progesterone and premature luteinization of granulosa cells. Functional analysis of the differentially expressed genes using DAVID identified 4 enriched functional category clusters (enrichment score  $> 2$ ): blood vessels and vasculature development, collagen fibrin and extracellular organization, carbohydrate and hyaluronic acid binding, and transforming growth factor  $\beta$  receptor signaling pathway. These categories confirm a wide range of changes associated with beef heifers transitioning from pre- to post-puberty and suggest potential pharmacological targets.

**Key Words:** transcriptome, puberty, pituitary

**M130 Prepartum supplementation of niacin increases colostral immunoglobulin G content in dairy cows.** Kayla M. Aragona\*, Colleen E. Chapman, André B.D. Pereira, and Peter S. Erickson, *University of New Hampshire, Durham, NH.*

In the US, 60% of colostrum fails to meet the minimum standard of 50g/L of immunoglobulin G (IgG). High quality colostrum is essential for the survival and future production of dairy heifers. Previous studies have shown that niacin (N) may increase vasodilation and in dairy cows, prepartum N supplementation may increase blood flow and potentially Ig flow, to the mammary gland. The objective of this study was to determine if feeding 48g/d of supplemental N to prepartum dairy cows effects colostrum quality. The experiment used 26 multiparous Holstein cows housed in a tie-stall barn. Cows were blocked by expected calving date and randomly assigned to 1 of 2 treatments 4wk prepartum: (1) 48g/d N (Lonza) or (2) 0g/d N (C), both with 52g/d of corn meal as a carrier. Total mixed ration amounts fed and refused were measured

daily at 0800h to determine dry matter intake (DMI). Within 90min of parturition, colostrum was collected and weighed. IgG concentration was analyzed using a radial immunodiffusion (RID) assay (Triple J Farms). Calves were removed from their dams immediately and weighed. Calves received 3 bags of a lacteal based colostrum replacer (180g IgG, Saskatoon Colostrum Co.), fed within 2 h of birth. Blood samples were collected from calves via jugular vein at 0 and 24 h of age and analyzed for IgG concentration and apparent efficiency of absorption (AEA). No differences were observed for DMI [15.7 kg/d (C), 14.3 kg/d (N);  $P = 0.13$ ], IgG yield [748.87 g (C), 773.67 g (N);  $P = 0.83$ ], colostrum yield [10.64 L (C), 9.45 L (N);  $P = 0.19$ ], fat % [5.5% (C), 4.72% (N);  $P = 0.45$ ] and solids nonfat % [23.9% (C), 22.9% (N);  $P = 0.7$ ]. No differences were observed for 24-h calf serum IgG concentration [15.9 g/L (C), 15.5 g/L (N);  $P = 0.8$ ] or AEA [32.46% (C), 30.92% (N);  $P = 0.5$ ]. Niacin tended to increase colostrum protein percent (14.6% (C), 17.9% (N);  $P = 0.09$ ). Niacin increased IgG concentration in colostrum from 73.8 g/L (C) to 86.8 g/L (N) ( $P < 0.05$ ). These results indicate that supplementing N (48 g/d) during the prepartum period improves colostrum quality.

**Key Words:** niacin, IgG, colostrum

**M131 Innate immune cells may be involved in bovine mammary development.** Kirsten. L. Beaudry\*<sup>1</sup>, Cathy. L. Parsons<sup>1</sup>, Adam. J. Geiger<sup>1</sup>, Steven. E. Ellis<sup>2</sup>, and Robert M. Akers<sup>1</sup>, <sup>1</sup>*Department of Dairy Science, Virginia Polytechnic Institute and State University, Blacksburg, VA,* <sup>2</sup>*Department of Animal and Veterinary Sciences, Clemson University, Clemson, SC.*

Bovine mammary development begins before puberty with the elongation and branching of ducts and stromal remodeling. This process is highly regulated, and in mice impacted by the presence of innate immune cells which cluster near developing ductal structures. For the first time, we studied the presence, location and changes in numbers of eosinophils, mast cells and macrophages in prepubertal bovine mammary tissue. Chemical stains and immunofluorescence were used to identify the 3 cell types in formalin fixed, paraffin embedded mammary tissue from prepubertal female calves in archived sets. The first set (ONT) included samples ( $n = 4$  per week) from birth to 6 wk of age. A second set (OVX) allowed for examination of the potential influence of ovaries on innate immune cells. It included samples from 19 animals who were either intact or ovariectomized 30 d before sample collection. These animals were 90, 120 or 150 d old at examination. The third set (EST) allowed for examination of the potential influence of exogenous estrogen on innate immune cells in the mammary gland. This set included samples from 10 calves given estrogen implants ( $n = 6$ ) or placebo ( $n = 4$ ) at 56 d of age. Calves were sampled at 70 d of age. Eosinophils were identified via LUNA staining, mast cells by May-Grunewald Giemsa staining, and macrophages with immunofluorescence. Stroma was examined in 20 images from each animal of the NEAR ( $< 100 \mu\text{m}$ ) and FAR ( $> 100 \mu\text{m}$ ) locations relative to the epithelial ducts. Results were expressed as cells/mm<sup>2</sup>. More eosinophils and mast cells were observed in NEAR compared with FAR stroma in the ONT ( $P < 0.001$ ) and OVX ( $P < 0.05$ ) but not EST. More macrophages were also observed in NEAR compared with FAR in ONT ( $P < 0.001$ ). Treatment (age, ovariectomy or estrogen) had no impact on stromal eosinophils. Age tended to affect mast cell numbers in ONT, with the most mast cells observed at 14 d

of age. In summary, we show for the first time that innate immune cells are present in prepubertal bovine mammary tissue and that abundance is related to the epithelial structure and physiological state of development. We suggest there may be a role for these cells in control of bovine mammary development.

**Key Words:** mammary, immune cell, development

**M132 An evaluation of precision dairy farming technologies monitoring rumination, feeding, and lying behaviors.** Matthew R. Borchers\*, I-Ching Tsai, Barbara A. Wadsworth, and Jeffrey M. Bewley, *University of Kentucky, Lexington, KY.*

The objective of this study was to evaluate commercially available precision dairy farming technologies against direct visual observations for feeding, rumination, and lying behaviors. Primiparous ( $n = 24$ ) and multiparous ( $n = 24$ ) lactating Holstein dairy cattle (mean  $\pm$  SD; 223.4  $\pm$  117.8 d in milk, producing 29.2  $\pm$  8.2 kg milk/d) were fitted with 6 triaxial accelerometer technologies to evaluate cow behavior. Technologies recording feeding behavior were the CowManager SensOor (Agis, Harmelen, Netherlands) and the Track a Cow (ENGS, Israel). Technologies recording rumination behavior were the CowManager SensOor and the Smartbow (MKW electronics GmbH, Jutogasse, Austria). Lying behavior was recorded by the AfiAct Pedometer Plus (Afirmilk, Kibbutz Afikim, Israel), HOBO Data Logger (HOBO Pendant G Acceleration Data Logger, Onset Computer Corporation, Pocasset, MA), CowAlert IceQube (IceRobotics Ltd., Edinburgh, UK), and Track a Cow. Over 8 d in October 2014, 6 cows per day were visually observed for feeding, rumination, and lying behavior for 2 h after morning and evening milking. The CORR procedure of SAS Version 9.3 (SAS Institute Inc., Cary, NC) was used to evaluate agreement between visual observations and technology-recorded behaviors. Visually recorded feeding behavior was strongly correlated with CowManager SensOor ( $r = 0.97$ ;  $P = 0.03$ ) and Track a Cow ( $r = 0.91$ ;  $P = 0.09$ ). Between the CowManager SensOor and Track a Cow, a strong correlation ( $r = 0.91$ ;  $P = 0.09$ ) was found. Visually recorded rumination was strongly correlated with the Smartbow ( $r = 0.99$ ;  $P < 0.01$ ) and CowManager SensOor ( $r = 0.96$ ;  $P < 0.01$ ). Between the Smartbow and CowManager SensOor, a strong correlation ( $r = 0.94$ ;  $P = 0.06$ ) was found. Visually recorded lying behavior was highly correlated with the AfiAct Pedometer Plus ( $r = 0.97$ ;  $P < 0.01$ ), HOBO Data Loggers ( $r = 0.98$ ;  $P < 0.01$ ), CowAlert IceQube ( $r = 0.99$ ;  $P < 0.01$ ), and Track a Cow ( $r = 0.99$ ;  $P < 0.01$ ). Results indicate these precision dairy farming technologies to be accurate dairy cattle behavioral monitors.

**Key Words:** precision dairy farming technologies, dairy cow behavioral monitoring, accuracy evaluation

**M133 The effect of estrus expression on target genes in the endometrium, conceptus and corpus luteum of cows.** Saeideh Davoodi\*<sup>1</sup>, Reinaldo F. Cooke<sup>2</sup>, Arthur C. C. Fernandes<sup>1</sup>, Bruno I. Cappellozza<sup>2</sup>, Jose L. M. Vasconcelos<sup>3</sup>, and Ronaldo L. A. Cerri<sup>1</sup>, <sup>1</sup>*Faculty of Land and Food Systems, University of British Columbia, Vancouver, BC, Canada*, <sup>2</sup>*Eastern Oregon Agricultural Research Center, Oregon State University, Burns, OR*, <sup>3</sup>*Faculdade de Medicina Veterinária e Zootecnia, UNESP – Botucatu, SP, Brazil.*

The aim of this study was to test the effect of estrus expression at artificial insemination (AI) on the endometrium, conceptus and corpus luteum (CL) gene expression. Twenty-three multiparous nonlactating Nelore cows were enrolled on a estradiol and progesterone (P4) based timed-AI protocol (AI = d 0), then slaughtered for endometrium, CL

and conceptus collection on d 19. Body condition score (BCS), blood samples for analysis of P4 and ultrasound examination was performed on d 0, 7 and 18 of the experiment. RNA extraction and qRT-PCR analysis of 58 target genes were analyzed on tissues. Data were checked for normality and analyzed by ANOVA for repeated measures using proc GLM, MIXED and UNIVARIATE. Estrous expression had no correlation with parameters such as BCS, pre-ovulatory follicle and CL diameter, P4 concentration in plasma on d 7 and 18 after AI and IFN-tau concentration in the uterine flushing ( $P > 0.05$ ); however, a significant increase was observed in conceptus size ( $P = 0.02$ ; 38.3  $\pm$  2.8 vs. 28.2  $\pm$  2.9). The majority of transcripts affected by estrous expression in the endometrium belong to the immune system and adhesion molecule family (MX1, MX2, MYL12A, MMP19, CXCL10, IGLL10 and SLPI;  $P \leq 0.05$ ). Genes related to apoptosis, P4 synthesis and prostaglandin receptor were downregulated (CYP11A, BAX and PGR;  $P < 0.05$ ) in the CL tissue of cows in estrus. In addition, 4 genes were identified as differentially expressed in the 19 old conceptus from cows observed in estrus (ISG15, PLA2, BMP15 and EEF1A1;  $P < 0.05$ ). There was an interaction ( $P < 0.05$ ) between estrus expression and P4 concentration on d 7 which mainly affected genes involved in immune system (MX1, MX2, TRD, SLPI and IGLL1). This study demonstrated that estrous expression at the time of AI, in spite of ovulation being induced by estradiol, can alter the gene expression profile in reproductive tissues during the preimplantation phase. These effects seem to be more pronounced in endometrium during the time of dynamic remodeling to prepare a receptive uterus for embryo implantation.

**Key Words:** conceptus, corpus luteum, endometrium

**M134 Accuracy and intensity of heat detection with activity monitoring systems for lactating dairy cows.** Craig LeRoy\* and Stephen LeBlanc, *Department of Population Medicine, University of Guelph, Guelph, ON.*

Activity monitors are becoming a widely used tool for estrus detection in dairy cows. The objective of this study is to assess the intensity and accuracy of estrus detection with automated activity monitoring systems. Three commercial dairy farms with an activity monitoring system (Heatime SCR Inc., or Afi Pedometer Plus, Afikim) that was used for essentially all inseminations between 50 and 80 d in milk (DIM) were enrolled in this observational study. Herds were visited once weekly. Blood samples were collected at wk 5, 7, and 9 postpartum and from a subset of cows on the day of insemination, to measure serum progesterone concentration. Cows were examined at wk 5 for purulent vaginal discharge (PVD). Lameness and body condition were scored at wk 7. Cows were classified as anovular if all 3 blood samples from wk 5, 7, and 9 had progesterone  $< 1$  ng/mL. Overall, 6.7% of cows were anovular (range of 3.6% to 8.2% among farms). Of these cows, 39% had a lameness score of 3 or above (1 to 5 scale) and 29% had PVD. Among cows that reached 80 DIM ( $n = 419$ ), 14% had not been inseminated by 80 DIM; of these, 12% were anovular, 23% were lame, 21% had PVD and 4% had a BCS of  $< 2.5$ . Among 352 blood samples taken on the day of insemination (all services; 212 signaled by the activity system and 140 by Ovsynch), 8% and 3% had progesterone  $> 1$  ng/mL; that is, the cows were not in estrus. In this sample, the accuracy of detection of estrus by AAM was high, but a subset of cows would likely require intervention for timely first AI and only a fraction of these were truly not cyclic.

**Key Words:** activity monitor, anovular, progesterone



**M135 Extruded flaxseed products improve the fatty acid profile of bovine milk.** Janna Moats<sup>\*1,2</sup>, Timothy Mutsvangwa<sup>2</sup>, and David Christensen<sup>2</sup>, <sup>1</sup>O&T Farms Ltd., Regina, SK, Canada, <sup>2</sup>University of Saskatchewan, Saskatoon, SK, Canada.

There is interest in increasing the content of omega-3 (n-3) fatty acids in bovine milk, primarily because of their beneficial effects on human health. This study investigated the effects of extrusion and the inclusion of condensed tannins (CT) in a flaxseed product on rumen fermentation characteristics, animal performance and milk fatty acid composition in dairy cows. Eight Holstein cows were used in a replicated 4 × 4 Latin Square with 28-d periods (20 d for dietary adaptation and 8 d for collection). Four cows in one Latin square were ruminally-cannulated. Cows were fed either a control diet (CTL) or one of 3 treatment diets that consisted of the daily substitution of 3 kg of the CTL concentrate pellet with 3 kg of either a non-extruded flaxseed and pea product (55% flaxseed, 36% peas, 8% alfalfa, 1% antioxidant; designated RAW), an extruded flaxseed and pea product (55% flaxseed, 36% peas, 8% alfalfa, 1% antioxidant; designated LPR), or an extruded flaxseed and high-tannin fava bean product (55% flaxseed, 36% fava beans, 8% alfalfa, 1% antioxidant; designated LPF). Diets were fed twice daily as total mixed rations. Dry matter intake was lower for cows fed LPR compared with those fed CTL ( $P < 0.05$ ). Milk yield was unaffected by dietary treatment ( $P > 0.05$ ). Rumen pH was not affected by dietary treatment ( $P > 0.05$ ). Rumen ammonia concentration was lower in animals fed CTL compared with those fed RAW, LPR or LPF ( $P = 0.04$ ). Propionate concentrations were higher for animals fed LPR compared with those fed RAW ( $P = 0.04$ ), while acetate concentrations did not differ between dietary treatments ( $P > 0.05$ ). Total n-3 and CLA milk contents were higher in cows fed LPR and LPF compared with those fed RAW and CTL ( $P < 0.01$ ). Milk fat content was lower for cows fed LPF compared with those fed CTL ( $P = 0.03$ ); however, milk fat yield was unaffected by dietary treatment ( $P > 0.05$ ). The use of extruded flaxseed products is an effective strategy for increasing the n-3 and CLA concentrations of bovine milk without negatively affecting rumen fermentation or animal production; however, the inclusion of CT in the extruded product had no additional benefit.

**Key Words:** omega-3, flaxseed, milk

**M136 Probiotic supplementation influences feed intake, body weight gain, and adaptive immunity in Holstein heifer calves during commingling.** Caleigh E. Payne<sup>\*</sup>, Luis G. D. Mendonça, Sonia J. Moisés, Sophia C. Trombetta, Lucas D. S. Rocha, Suzy Q. Fowler, Juan C. Gordienko, and Lindsey E. Hulbert, *Kansas State University, Manhattan, KS.*

After weaning, the transition from individual housing to group housing (commingling) is stressful for calves and increases the risk of respiratory disease during this period. Probiotics can include mannan-oligosaccharide (MOS) and b-glucan (BG) which may help improve calf health and performance. Therefore, the objectives of this study were to determine if a daily bolus dose of oral probiotics (20% MOS; 10% BG) influence feed intake, body weight gain, or adaptive immunity in Holstein heifer calves ( $n = 60$ ) during the transition from individual hutches to pens of 3. One week before commingling, calves (age  $52 \pm 4.0$  SD d; body weight  $83 \pm 14.92$  SD kg) were randomly assigned to daily treatments of either 4 g of probiotic (Preb) dissolved in 15 mL of molasses or control (Con; 15 mL of molasses only) for 7 weeks. Calves were fed ad libitum TMR top-dressed with 9 pounds of 18% CP calf starter once per day with refusals collected and measured daily. Calves were weighed and measured weekly. All calves were administered an innocuous protein

injection, ovalbumin (OVA; subQ; 0.5 mg/mL), at commingling and 4 weeks after commingling. Blood was collected 2 weeks after OVA to measure primary and secondary OVA-specific IgG and IgA responses. Prebiotic-fed calves ate more than control-fed calves 2 weeks after commingling ( $3.18$  vs.  $3.06 \pm 0.034$  kg/d;  $P = 0.015$ ). Prebiotic calves also gained more weight 3 weeks (ADG  $0.90$  vs.  $0.86 \pm 0.060$  kg/d;  $P = 0.037$ ) and 6 weeks post-commingling (ADG  $1.08$  vs.  $0.96 \pm 0.060$  kg/d;  $P = 0.025$ ). In addition, the interaction between treatment and week affected F:G ( $P = 0.044$ ) with preb-calves having lower F:G 1, 3, and 6 weeks post-commingling. The Preb-calves had a greater primary IgA response to OVA than Con-calves ( $P = 0.0315$ ), but no differences were observed for IgG ( $P = 0.8289$ ). Prebiotic supplementation may improve calf performance and adaptive immunity in the transition to group-housing situations post weaning.

**Key Words:** prebiotic, bovine, performance

**M137 Supplementation of conjugated linoleic acid during the transition to lactation period increased milk production in a commercial dairy.** Tawny L. Chandler<sup>\*1</sup>, Robert T. Fugate<sup>1</sup>, Arnulf H. A. Troescher<sup>3</sup>, Joshua A. Jendza<sup>2</sup>, and Heather M. White<sup>1</sup>, <sup>1</sup>University of Wisconsin-Madison, Madison, WI, <sup>2</sup>BASF Corporation, Florham Park, NJ, <sup>3</sup>BASF Corporation, Lampertheim, Germany.

*Trans*-10, *cis*-12 conjugated linoleic acid (CLA), a bioactive fatty acid, has the potential to alter energy partitioning in lactating cows by marginally reducing milk fat synthesis in the mammary gland. The objective of this study was to determine the effects of pre- and postpartum CLA supplementation on lactation performance in a commercial dairy setting. Holstein cows in a robotic milking system were blocked by multiparous (mp) or primiparous (pp), and expected calving date, and randomly assigned to either a CLA group (mp  $n = 100$ ; pp  $n = 39$ ) or control group (mp  $n = 98$ ; pp  $n = 38$ ). Cows were supplemented with 100g of lipid encapsulated CLA methyl esters (Lutrell Pure, BASF, Germany) mixed 50:50 with soybean meal to provide 10g each of *trans*-10, *cis*-12 CLA and *cis*-9, *trans*-11 CLA via a robot mineral supplement unit. Supplementation was from -21d precalving through 30d in milk (DIM) for mp or 70 DIM for pp. Milk yield, fat, and protein concentration were recorded daily and averaged by wk for the first 100 DIM. Data were analyzed using the MIXED procedure of SAS 9.4 with repeated measures. Treatment, wk, and treatment × wk were fixed effects with random effects of (cow)group. Means were considered different when  $P \leq 0.1$  and tended to differ when  $P \leq 0.15$ . Daily milk yield over 100d was increased with CLA supplementation (100d mean mp:  $49.7$  vs.  $51.3 \pm 0.6$  kg/d,  $P = 0.07$ ; pp:  $30.3$  vs.  $32.0 \pm 0.7$  kg/d  $P = 0.09$ ) with the greatest increase in milk production being 3.0 kg/d for mp (wk 5;  $P = 0.007$ ) and 3.9 kg/d for pp (wk 14;  $P = 0.001$ ) cows. Supplementation with CLA did not alter milk fat concentration ( $P > 0.15$ ) or yield ( $P > 0.15$ ) over the 100 d period. Milk protein concentration was reduced ( $P = 0.05$ ) with CLA supplementation in mp ( $3.18$  vs.  $3.12 \pm 0.02\%$ , control vs. CLA) but not changed ( $P > 0.15$ ) in pp ( $3.27$  vs.  $3.22 \pm 0.04\%$ , control vs. CLA) cows; however milk protein yield was unaffected ( $P > 0.15$ ). These results suggest that CLA supplementation during the transition to lactation period altered energy partitioning to increase milk production during the first 100 DIM.

**Key Words:** conjugated linoleic acid, transition cow, energy partitioning

**M138 Effects of plane of nutrition on glucose tolerance test kinetics pre- and postweaning in Holstein calves fed twice daily.**

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Feeding an elevated plane of nutrition to dairy calves preweaning is becoming more popular as it has been linked to higher growth rates, earlier onset of puberty, and increased milk production. Providing these feeding schemes in 2 meals is thought to impair glucose clearance and insulin responsiveness in young calves, but whether or not there is a carryover effect postweaning remains unclear. The objective of this study was to investigate this using a glucose tolerance test (GTT). A total of 12 female Holstein calves were selected and blocked by cow parity and colostrum intake, then assigned to an elevated plane (EP; 8 L/d) or a low plane (LP; 4 L/d) of nutrition. Calves received 4 L of colostrum provided in 2 meals (1h and 6h after birth), and were stepped-down from milk by 50% during wk 7 then weaned on wk 8. All calves had ad libitum access to pelleted calf starter, chopped wheat straw, and water. Milk replacer consisted of 24% CP and 18% CF and was fed at 150g/L. The GTT (glucose 30%) was conducted at 4, 7 and 10 weeks of age by infusing 540 mg of glucose per kg of metabolic bodyweight at the time of the morning feeding after a 12 h fast. Blood samples were collected at -15, 0, 5, 10, 20, 30, 45, 60, 120, 180, and 240 min via a jugular catheter. Changes in glucose concentration (mmol/L), clearance rate (%/min), and area under the curve (AUC<sub>240</sub>) were calculated. Preliminary data from a Hemocue blood glucose analyzer indicates that the increase in glucose (C<sub>max</sub>; mmol/L) was similar ( $P = 0.31$ ) between treatment groups and no differences between ages (pre- and postweaning) were detected. The time to reach the peak concentration (T<sub>max</sub>; minutes) was similar across treatments and age ( $P = 0.29$ ). Area under the curve was significantly affected by calf age ( $P = 0.005$ ) with AUC<sub>240</sub> increasing with age, and a trend for interaction with treatment was observed, where EP calves tended ( $P = 0.07$ ) to have a greater AUC at 10 weeks of age. The results show that the rate of glucose clearance following a tolerance test was not affected ( $P = 0.14$ ) by plane of nutrition or age when calves were fed milk replacer in 2 meals per day.

**Key Words:** glucose, glucose tolerance test, calf nutrition

**M139 Metabolic characteristics of protein, nutrient supply and feed milk value of the newly developed AAFC yellow and brown carinata lines for dairy cattle in comparison with commercial canola seeds.** Yajing Ban\*, David A. Christensen, John J. McKinnon, and Peiqiang Yu, *Department of Animal and Poultry Science, College of Agricultural and Bioresources, University of Saskatchewan, Canada.*

New lines of *Brassica carinata*, which were developed recently as oil crops for bio-fuel production in the dry western Canadian prairie, could also be regarded as a potential new feed source for animals. To our knowledge, there is little research on metabolic characteristics of protein in the new yellow and brown lines of carinata seeds. The objective of this study was to determine the differences in metabolic characteristics of protein from the new lines of carinata seeds in yellow and brown coats for dairy cows compared with commercial canola seeds. The new carinata lines were bred by Agriculture and Agri-Food Canada (AAFC). The experiment of rumen and intestinal digestion was carried out in U of S dairy research facility. Four cannulated lactating dairy cows were used for rumen degradation trial. The nutrient supply to dairy cattle and feed milk value (FMV) were determined based on the DVE/OEB system

with data from rumen and intestinal digestion experiments. Statistical analyses were performed using PROC MIXED procedure of SAS 9.3 with significance declared at  $P < 0.05$ . The results showed that yellow carinata seed line had higher ( $P < 0.05$ ) rumen undegraded feed crude protein (RUP) and effective degradability of crude protein (EDCP) compared with canola seeds. From in vitro intestinal digestion results, carinata seeds had higher total digestible protein (TDP) than canola seeds ( $P < 0.05$ ). Based on the DVE/OEB system, carinata seeds had higher truly absorbed rumen-synthesized microbial protein, total truly digested protein (DVE) in small intestine and feed milk value (FMV) than canola seeds ( $P < 0.05$ ). Yellow carinata seed line had higher protein supply than the brown seed line. In conclusion, the new lines of carinata seeds could be regarded as a potential feed protein supplement for dairy cows, and yellow carinata seed line had greater protein digestibility than brown carinata seeds.

**Key Words:** carinata seed, metabolic characteristics of protein, DVE/OEB system

**M140 Responses of neonatal goats (kids) to different concentrations of carbon dioxide gas.** Isabelle C. Withrock\*, Paul J. Plummer, Timothy A. Shepherd, Anna Johnson, Hongwei Xin, Johann F. Coetzee, and Suzanne T. Millman, *Iowa State University, Ames, IA.*

The dairy goat industry faces challenges for kid euthanasia. The methods approved by the AVMA are barbiturate overdose, gunshot, and captive bolt. The disadvantages of these methods range from cost to operator safety. Currently, there is no published research on inhalant euthanasia methods for neonatal ruminants. The objective of this study was to evaluate carbon dioxide (CO<sub>2</sub>) for kid euthanasia. A preference testing device was custom made to hold 2 connected chambers at static atmospheric concentrations. One chamber maintained ambient conditions, and the other designated CO<sub>2</sub> levels (treatment). A total of 7 mixed-breed dairy kids were enrolled in the study. Six males and 1 female were enrolled, and body weights ranged from 3.18 to 4.17 kg. Kids were individually trained for 5 consecutive days to enter the treatment chamber from the ambient chamber to access a milk ration (32oz). During training both chambers were set at ambient air concentrations. Kids were held in the ambient chamber for 5 min, after which the sliding door was opened to provide access to the treatment chamber. Kids were provided 10 min access to the treatment chamber after which they were removed and returned to their home pen. After training, testing began with the treatment chamber set at one of 3 CO<sub>2</sub> levels: 10%, 20% or 25%, while the ambient chamber was maintained at 1% CO<sub>2</sub>. Kids were randomly assigned 10% or 20% as the first treatment and received 25% CO<sub>2</sub> last. Kids experienced all 3 CO<sub>2</sub> treatments, with a 2-d wash-out (ambient CO<sub>2</sub>) between tests. Outcomes were collected using continuous sampling, and included latency to enter and exit treatment chamber to measure aversion, loss of muscle control (ataxia), and loss of posture. Five kids (71%) tolerated 10% CO<sub>2</sub> for 10 min. One kid left at 8.5 min after consuming his full ration. One kid lost posture at 289 s. At 20 and 25% CO<sub>2</sub>, a total of 100% of the kids became ataxic, and posture loss ranged from 93 to 215 s. All kids continued to consume milk before and during ataxia and reentered the treatment chamber on wash out days. In conclusion, kids did not display aversion to CO<sub>2</sub> and therefore, CO<sub>2</sub> has merit for goat euthanasia.

**Key Words:** carbon dioxide, euthanasia, goat

**M141 Analyzing udder efficiency of Brown Swiss cattle using data envelopment analysis.** Keith D. Gibson\* and Chad D. Dechow, *The Pennsylvania State University, State College, PA.*

Udder conformation traits such as udder depth are favorably associated with somatic cell score (SCS) and mastitis resistance, but unfavorably associated with yield, resulting in unclear selection aims. Data envelopment analysis (DEA) is an approach to characterize the efficiency of different units based on inputs and outputs. The objective of this research was to use DEA to create an efficiency trait for udders of Brown Swiss cattle, which we termed udder efficiency, and to evaluate the genetic relationship of udder efficiency with yield, fitness, and udder conformation. The lone input was identified as milking speed and the outputs were milk, fat, and protein yield, and the inverse of SCS. A SAS DEA macro was used to determine udder efficiency relative to herd mates, with the most desirable cows milking quickly, producing large volumes of milk, fat, and protein with a low SCS. Values for the trait ranged from 0.03 (very inefficient cows) to 1.0 (efficient cows). There were 105,164 first through fifth lactation yield and SCS records from 45,464 cows born in 2000 or later. There were 41,704 milking speed records with a high

score corresponding to faster milking speed. From these, 40,960 udder efficiency records were derived. These data were analyzed using a series of 3 trait models in ASREML that included fixed effects for lactation, age, and herd-calving cluster. Random effects were animal, permanent environment, and residual. The heritability of udder efficiency was  $0.23 \pm 0.0133$ . Genetic correlations with udder efficiency were moderate and positive for yield (0.40 to 0.42), favorable with SCS (-0.21), and strongly positive for milking speed (0.86). Udder efficiency was also strongly correlated with productive life (0.69). Of the udder traits, rear udder height (0.30), teat length (-0.28), fore udder attachment (0.25), and rear udder attachment (0.21) were most favorably correlated to udder efficiency. The correlation of udder depth with udder efficiency was 0.16. Based on these findings, DEA could be a useful and novel method for evaluating udder efficiency, helping to identify traits that enable selection for mastitis resistance without compromising yield.

**Key Words:** Brown Swiss, udder efficiency, data envelopment analysis



# Lactation Biology I

**M142 Distribution and analysis of milk fat globule and crescent in Murrah buffalo and crossbred cow.** Ratan K. Choudhary\*, Harmanjot Kaur, Shanti Choudhary, and Ramneek Verma, *School of Animal Biotechnology, Guru Angad Dev Veterinary and Animal Science University, Ludhiana, Punjab, India.*

India is the largest buffalo milk producer in the world. Buffalo milk is rich in minerals like calcium and magnesium, contains 100% more fat and less cholesterol than cow milk. Physiochemical properties of dairy products depend upon the size and distribution of milk fat globules (MFG). Individual MFG from Murrah buffalo were analyzed as a function of size and distribution and comparisons were made with MFG of crossbred (Holstein Friesian × Sahiwal) cows. Main objectives of this study was to measure areas of buffalo MFG and analyze percentage of crescents associated with MFG using a simple microscopic technique. Crescents of MFG can provide an alternative source of RNA of mammary epithelial cells origin. Acridine orange staining of fresh milk was visualized by light microscopy under red and green channel filters. MFG and MFG with crescents were analyzed using ImageJ image analysis software. The percentage of large sized MFG (area >20  $\mu\text{m}^2$ ) was greater and the percentage of small sized MFG (area <10  $\mu\text{m}^2$ ) was lower in buffalo than cow milk. The percentage of crescents was < 1% of total MFG in both the species. These results indicate that MFG in buffalo milk is larger than that MFG of cow milk. RNA isolation from buffalo milk fat and its suitability for microarray analysis remains to be determined.

**Key Words:** buffalo, milk fat globule, crescent

**M143 Fatty acid synthase (FASN) gene polymorphism and early lactation milk fat composition in Xinong Saanen goats.** Abiel b Haile\*, Zhang Wei, Wang Wei, Yang Dikun, Yi Yongqing, and Luo Jun, *Northwest A&F University, Yangling, Shaanxi, China.*

The fatty acid synthase (*FASN*) gene codes for a multifunctional protein, catalyzing the de novo fatty acid synthesis. Its high mRNA expression in GMEC (goat mammary epithelial cells), coinciding with medium to high heritability of milk fat, substantiated its role as a candidate gene for association analysis with milk fat profile. The main objectives of the study were to develop markers of *Capra hircus* *FASN* gene to improve healthfulness of goat milk FAs (fatty acids) and to investigate early lactation milk fat profile. A Linear mixed model association analysis of 46 dependent variables, analyzed from 300 milk samples 30 d post-partum, with 3 intragenic SNPs and 2 haplotype groups was conducted. The results had revealed associations of *FASN* SNPs and haplotype groups with myristic acid (C14:0), palmitic acid (C16:0) and linoleic acid (C18:2 n-6, cis). Herd was the predominant factor affecting fatty acid levels followed by herd-test-day (HTD) and parity, indicating the niche of nutrition and management in modulating early lactation milk fat profile. De novo FAs (C6:0 to C14:0) showed suppressed levels with strong negative correlations with C18 FAs. Contradicting to the presumed source of origin, butyric acid (C4:0) was impervious to the observed de novo FA depression, it had shown unique correlation patterns. Oleic acid (C18:1 *cis*-9) scored the highest mean value followed by palmitic acid. Palmitic acid had shown normal levels with weak negative correlations with C6:0 and C8:0. Here, its dual source of origin supposedly had played a role, in replenishing the diminished levels of de novo contribution, by the high C16 FAs bioavailability of body fat mobilization, which masked the inhibitory effect of long chain FAs. The correlation patterns of odd chain FAs with CLA and rumen derived

biohydrogenation intermediates reinforce the supposition that odd chain fatty acids could serve as a diagnostic tool of rumen condition and biohydrogenation patterns. The SNP markers developed in the present study will assist in marker assisted selection, and the comprehensive analysis of early lactation milk fat profile will help to decipher main factors of variation, thereby aiding fatty acid profile modulation.

**Key Words:** dairy goat, SNP marker, milk fat

**M144 Pattern of immunoglobulin A in milk from gorillas throughout lactation.** M. Garcia\*<sup>1</sup>, M. L. Power<sup>2</sup>, and K. M. Moyes<sup>1</sup>, <sup>1</sup>*Department of Animal and Avian Sciences, University of Maryland, College Park, MD,* <sup>2</sup>*Smithsonian Conservation Biology Institute, Washington, DC.*

In cases when mother's milk is not available to a great ape infant (e.g., rejection), zoo nutritionists/veterinarians are responsible for formulating a milk replacer to maximize the chances of survival of the neonate. To accomplish this, they generally use milk from commercially available species (e.g., cow and goat) as well as nutritional supplements and other additives. As is the case in humans, placental transfer of immunoglobulin (Ig)A occurs throughout pregnancy in the gorilla. After birth, neonates rely solely on nutrients and Igs via the mother's milk until approximately 5 mo of age and are weaned at 3 to 4 years of age. Unfortunately, there is currently no useful information regarding the IgA profile of milk and how stage of lactation alters IgA concentration in gorilla milk. The objective of this study was to identify the pattern of IgA concentrations in milk from gorillas throughout lactation. Milk samples from 4 gorillas were collected between January 2009 and October 2014 and stored at -80°C until further analysis. Concentrations of IgA in milk were measured using a Human IgA ELISA kit. Data were grouped according to month in milk (MIM; 0 < MIM = 5, 5 < MIM = 10, 10 < MIM = 15, 15 < MIM = 36, and MIM >36). The data were analyzed as a complete randomized design with gorilla nested within MIM as a random term and MIM as a fixed effect. Month in milk had a significant ( $P < 0.01$ ) affect IgA concentrations. Concentrations of IgA were highest for MIM >36 (81.3  $\mu\text{g}/\text{mL}$ ) followed by 0 < MIM = 5 (53.9  $\mu\text{g}/\text{mL}$ ). The increase in concentrations of IgA in later lactation may reflect reductions in milk production and this warrants further investigation. Results from this study provided a better understanding of the IgA profile of milk from gorillas. Additional knowledge regarding fluctuations of other immunoglobulins as well as nutritional profiles in gorilla milk will assist professionals charged with their care in captivity to develop improved hand-rearing protocols.

**Key Words:** gorilla, immunoglobulin A, milk

**M145 Peroxisome proliferator-activated receptor gamma (PPAR $\gamma$ ) agonist does not stimulate mammary lipogenic gene expression or overcome the effect of *trans*-10,*cis*-12 conjugated linoleic acid (CLA) in lactating ewes.** Eveline C. Sandri<sup>1</sup>, Elvis Ticiani<sup>1</sup>, Monica Urio<sup>1</sup>, Mauricio Camera<sup>1</sup>, Ana P. Povaluk<sup>1</sup>, Kevin J. Harvatine<sup>2</sup>, and Dimas E. Oliveira\*<sup>1</sup>, <sup>1</sup>*Santa Catarina State University/CAV, Lages, Santa Catarina, Brazil,* <sup>2</sup>*Penn State University, State College, PA.*

Milk fat synthesis involves biochemical processes, including fatty acid synthesis, uptake, transport and desaturation. *Trans*-10,*cis*-12 CLA inhibits milk fat synthesis by decreasing the expression of genes

and transcription factors. Peroxisome proliferator-activated receptor gamma (PPAR $\gamma$ ) is a key regulator of lipid synthesis in many tissues and is affected by *trans*-10,*cis*-12 CLA but, the mechanisms by which *trans*-10,*cis*-12 CLA suppresses the expression or activity of PPAR $\gamma$  and expression of its targeted genes are not clear. This study used a chemical PPAR $\gamma$  agonist to evaluate the effect of PPAR $\gamma$  on mammary lipid synthesis and its interaction with *trans*-10,*cis*-12 CLA in lactating ewes. Twenty-four crossbred lactating ewes [70  $\pm$  3 DIM; 60  $\pm$  0.45 kg body weight (BW)] were randomly assigned to one of the 4 treatments (n = 6/treatment) for 7 d. Treatments were (1) Control (intravenous infusion of 100 mL/d of saline); (2) TZD (intravenous infusion of 4 mg/kg of BW per d in 100 mL of saline); (3) CLA (27 g/d orally dosed methyl ester containing 29.9% *trans*-10,*cis*-12 CLA); and (4) TZD+CLA. Mammary and adipose tissue biopsies were taken, RNA was extracted, cDNA synthesized and qRT-PCR analysis conducted for PPAR $\gamma$ , SREBP1 and SCD1. Compared with control, fat content was 22.3% lower in CLA ( $P = 0.05$ ), tended to be 20.7% lower in TZD+CLA ( $P = 0.06$ ). In the mammary gland, CLA decreased expression of PPAR $\gamma$ , SREBP1 and SCD1 by 64.4, 60 and 19% compared with control ( $P = 0.02$ ,  $P = 0.01$  and  $P = 0.005$ , respectively), confirming its negative effects on the expression of lipogenic genes. However, TZD did not stimulate the expression of these genes or overcome the effect of CLA in mammary tissue. In adipose tissue, expression of PPAR $\gamma$  were not affected by treatment, whereas SREBP1 expression was increased by TZD, CLA and TZD+CLA compared with control and SCD1 expression was higher in TZD+CLA compared with the other treatments. Overall, CLA negatively affected mammary expression of genes involved in lipid synthesis and TZD was unable to overcome those effects demonstrating that the mechanism of CLA is not dependent on inhibition of PPAR $\gamma$ .

**Key Words:** lipogenesis, milk fat depression, milk synthesis

**M146 Effect of *trans*-10,*cis*-12 CLA on the expression of genes involved in milk fat synthesis in ovine mammary gland tissue cultured in vitro.** Monica Urio, Ana P. Povaluk, Humberto T. Borges, June A. Favaretto, Maurício Camera, and Dimas E. Oliveira\*, *Santa Catarina State University/CAV, Lages, Santa Catarina, Brazil.*

Culturing lactating mammary explants allows evaluating the effect of bioactive molecules in a controlled environment while maintaining the same characteristics found in vivo (Keys et al., 1997; *In Vitro Cell. Dev. Biol. Animal*, 33:206–21). However, studies using ovine mammary tissue are still scarce. The objective of this study was to evaluate the effect of *trans*-10,*cis*-12 CLA on the expression of key genes involved in milk fat synthesis in mammary explants cultured in vitro. Mammary samples were obtained through biopsies in lactating ewes (120 DIM) and were grown on plates with growth area of 1.9 cm<sup>2</sup> in mammary epithelial cell growth media supplemented with fetal bovine serum, antibiotics, insulin and growth factors at 37°C with 5% CO<sub>2</sub> and humidity saturated. Tissues were cultured for 3 and 24h in triplicates using the following treatments: 75  $\mu$ mol/L of *trans*-10,*cis*-12 CLA or Control (no CLA). After cultures were stopped RNA was extracted, cDNA synthesized and qRT-PCR performed. The genes studied were SREBP1 (sterol regulatory element-binding protein), PPAR $\gamma$  (peroxisome proliferator-activated receptor gamma) and SCD1 (stearoyl-CoA desaturase). Statistical analysis was performed using the MIXED procedure of SAS (2002) and ribosomal protein S18 housekeeping gene was used as a covariate in the model. CLA treatment had no effect on gene expression of SREBP1, PPAR $\gamma$ , and SCD1 at 3 h ( $P = 0.19$ ,  $P = 0.52$  and  $P = 0.20$ , respectively). At 24 h, there was a trend for CLA-treated explants to have reduced gene expression of SREBP1 and PPAR $\gamma$  ( $P = 0.07$  and  $P = 0.08$ , respectively), and a 12-fold decrease in SCD1 gene expres-

sion ( $P = 0.009$ ). Overall, our results suggest that *trans*-10,*cis*-12 CLA downregulates expression lipogenic genes in ovine mammary tissue, however explants may need to be cultured for at least 24 h

**Key Words:** dairy ewe, gene expression, milk fat depression

**M147 Gene expression of transcription factors and genes involved in milk fat depression in lactating ewes of different body weights fed the same dose of CLA *trans*-10,*cis*-12.** Monica Urio, Eveline C. Sandri, Ana P. Povaluk, Elvis Ticiani, Camila Renneberg, Maurício Camera, June A. Favaretto, and Dimas E. Oliveira\*, *Santa Catarina State University/CAV, Lages, Santa Catarina, Brazil.*

During milk fat depression (MFD) induced by the feeding of *trans*-10,*cis*-12 CLA the magnitude in the reduction of milk fat content and/or yield varies considerably among animals. The objective this study was to evaluate the gene expression of transcription factors and genes downregulated during MFD in lactating ewes fed a uniform dose of *trans*-10,*cis*-12 CLA but differing in body weight. Fourteen lactating ewes were selected according to BW and separated in 2 groups that averaged 51.0  $\pm$  1.0 and 69.0  $\pm$  4.0 kg of BW. Ewes received for 10 d a daily oral dose of 27g/d of CLA (rumen-unprotected, 29.9% *trans*-10,*cis*-12 isomer). Mammary and adipose tissue biopsies were performed on d 10. RNA was extracted, cDNA synthesized and qt-RT-PCR carried out. Data were analyzed using the MIXED procedure of SAS (2002) and ribosomal protein S18 housekeeping gene was used as a covariate. Gene expression of SREBP1 (sterol regulatory element binding protein), S14 (thyroid hormone responsive) and SCD1 (stearoyl-CoA desaturase) were evaluated in mammary tissue samples and leptin in adipose tissue. Gene expression of SREBP1, S14 and SCD1 did not differ between the BW groups ( $P = 0.29$ ,  $P = 0.17$  and  $P = 0.57$ , respectively). Leptin gene expression was higher in the heavier BW group ( $P = 0.02$ ). Overall, our results suggest that animals of different body weights receiving a uniform dose of *trans*-10,*cis*-12 CLA showed change in gene expression of leptin.

**Key Words:** fat synthesis, mammary biopsy, ovine

**M148 Expression of acetyl-CoA carboxylase alpha (ACC $\alpha$ ) transcripts from different promoters in mammary and adipose tissue from lactating ewes at different stages of lactation.** Grégory J. Cardoso<sup>1</sup>, Elvis Ticiani<sup>1</sup>, Monica Urio<sup>1</sup>, Ana P. Povaluk<sup>1</sup>, Maurício Camera<sup>1</sup>, Rogério Ferreira<sup>2</sup>, Kevin J. Harvatine<sup>3</sup>, and Dimas E. Oliveira\*<sup>1</sup>, <sup>1</sup>*Santa Catarina State University/CAV, Lages, Santa Catarina, Brazil*, <sup>2</sup>*Santa Catarina State University/CEO, Chapecó, Santa Catarina, Brazil*, <sup>3</sup>*Penn State University, State College, PA.*

Initiation and maintenance of lactation are complex phenomena involving cellular and enzymatic changes including those in mammary gland and adipose tissue. Fat is the main energy component of the milk, originating from the diet, mobilization from adipocytes, and from de novo synthesis in the mammary gland. Milk fat synthesis is controlled in various ways and one example is via regulation of acetyl-CoA carboxylase  $\alpha$  (ACC $\alpha$ ), a key enzyme in the de novo fatty acid synthesis pathway. ACC $\alpha$  is encoded by mRNAs transcribed from 3 promoters (PI, PII and PIII) characterized as tissue-specific in the ovine genome. PI transcripts are restricted to the adipose tissue and liver, whereas PII is found in all tissues and PIII has been isolated from mammary tissue from lactating ewes. This study evaluated the transcription of ACC $\alpha$  from PI, PII and PIII in mammary and adipose tissue from dairy ewes at different stages of lactation. Mammary gland and adipose tissue biopsies were taken from 6 lactating ewes at 3 periods (15, 70 and 120 d in milk) to represent early, mid and late lactation. Subsequently RNA

was extracted, cDNA synthesized and qRT-PCR analysis conducted. Data were analyzed by SAS using ribosomal protein S18 housekeeping gene as a covariate. As expected, no expression of PI was detected in the mammary gland. Transcripts from PII in mammary gland were higher in early lactation and decreased as lactation advanced (early vs. mid  $P = 0.02$ ; early vs. late  $P = 0.002$ ; mid vs. late  $P = 0.23$ ). Similarly, the PIII transcripts decreased throughout lactation in mammary gland (early vs. mid  $P = 0.001$ ; early vs. late  $P = 0.001$ ; mid vs. late  $P = 0.005$ ). On the contrary, in adipose tissue the PI gene expression was higher in mid and late lactation ( $P = 0.001$ ), and transcripts from PII and PIII did not differ among periods. Results suggest that PII transcripts in mammary glands are upregulated in early and mid lactation according to a higher requirement for fat synthesis and PI transcripts have a greater expression level in adipose tissue in late lactation likely due to a positive energy balance replenishing body fat reserves.

**Key Words:** adipose tissue, gene expression, mammary gland

**M149 Effects of sodium salicylate on early lactation milk production parameters.** Caroline M. Ylloja\*, Abigail J. Carpenter, Laman K. Mamedova, and Barry J. Bradford, *Department of Animal Sciences and Industry, Kansas State University, Manhattan, KS.*

Treatment with non-steroidal anti-inflammatory drugs (NSAID) after calving can have a positive effect on whole-lactation milk production in older dairy cows. Ongoing research aims to determine whether this response is due to increased mammary epithelial cell function and whether this is also tied to changes to inflammation in early lactation. For this study, cows in 3rd or greater lactation ( $n = 8$ /treatment) were enrolled at parturition to receive a daily drench of sodium salicylate (SS; 125g/d) or water (CON) for 3 d beginning approximately 24 h postpartum. Mammary biopsies were conducted on d 1, 4, and 45 after calving. Daily milk weights were recorded and milk composition was determined 2×/week. Blood samples were taken 2×/wk and analyzed for metabolites. Results up to 45 DIM show no difference between treatments for plasma glucose, insulin,  $\beta$ -hydroxybutyrate, or nonesterified fatty acid levels. Weekly means of milk production parameters revealed no detectable differences for fat content or yield, milk urea nitrogen, somatic cell score, energy-corrected milk yield, or fat-corrected milk yield. Treatment interacted with week for average protein yield ( $P = 0.02$ ), with SS cows increasing more (1.07 to  $1.49 \pm 0.09$  kg/d) over the 7 weeks of the study than CON (1.19 to  $1.37 \pm 0.09$  kg/d). Lactose content tended to be greater for SS (4.89%) compared with CON (4.74  $\pm 0.05\%$ ;  $P = 0.06$ ). Milk yield tended to increase more for SS (from 26.6 to  $55.8 \pm 2.6$  kg/d) than CON (from 30.7 to  $52.3 \pm 2.3$  kg/d) over time (interaction  $P = 0.10$ ). Results agree with previous studies where milk production increased for salicylate-treated cows, although the small sample size in the present study limited power to observe production responses, and production data included only the first 45 d of lactation. Ongoing analysis of morphological and transcriptional changes in mammary tissue may reveal mechanisms that contribute to a sustained increase in milk production following early-lactation NSAID administration.

**Key Words:** NSAID, inflammation, mammary function

**M150 Effect of Leu and His on casein protein synthesis via mTOR signaling pathway in bovine mammary epithelial cells.** Haina Gao<sup>1,2</sup>, Han Hu<sup>1,3</sup>, Nan Zheng<sup>1,3</sup>, and Jiaqi Wang<sup>\*1,3</sup>, <sup>1</sup>Ministry of Agriculture-Milk Risk Assessment Laboratory, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>2</sup>College of Animal Science and Technology, Gansu Agricultural

University, Lanzhou, Gansu, China, <sup>3</sup>State Key Laboratory of Animal Nutrition, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China.

Assessing the regulatory effect of individual amino acids (AA) on milk protein synthesis is vital to AA requirement models for lactation. This study employed the immortalized bovine mammary epithelial cells (CMEC-H) as a model to investigate the effects of Leu or His on mTOR signaling and casein synthesis by Western blotting. Cells were cultured in the Earle's balanced salts (EBSS) with Leu (0.45, 1.35, 5.4, 10.8 mmol/L) or His (0.15, 1.2, 4.8, 9.6 mmol/L) addition for 6h, respectively, and the EBSS without AA was set as negative control, the EBSS supplemented with 10% FBS was used as positive control. The protein band values from the AA-supplemented cells were related to their AA-deprived controls. The experimental data were analyzed using the Duncan's test for post-hoc multiple comparisons of treatment means by SAS. Meaningful relationships among phosphorylation of the signaling proteins and casein expression were quantified with simple linear regression models using the REG procedure of SAS. Differences between experimental groups were considered significant at a  $P < 0.05$ . The results showed that, compared with the negative control, Leu or His significantly increased the phosphorylation of mammalian target of rapamycin (mTOR, Ser2481), a binding partner of target of rapamycin (raptor, Ser792), the ribosomal protein S6 kinase 1 (S6k1, Thr389), eukaryotic initiation factor 4E (eIF4E, Ser209), eukaryotic elongation factor 2 (eEF2, Thr56) and casein synthesis ( $P < 0.05$ ). These results suggest that the supplement of Leu or His could activate the mTOR pathway and in turn catalyze the phosphorylation of signaling protein and increase milk protein synthesis. Our linear regression model assay declared that the expression of  $\alpha$ s1-casein was positively correlated with P-mTOR ( $R^2 = 0.7820$ ,  $P < 0.01$ ), P-S6k1, ( $R^2 = 0.7881$ ,  $P < 0.01$ ) and eEF2 ( $R^2 = 0.7835$ ,  $P < 0.01$ ) with a dose-dependent effect of Leu. While the expression of  $\beta$ -casein ( $R^2 = 0.9638$ ,  $P < 0.01$ ) and  $\kappa$ -casein ( $R^2 = 0.9048$ ,  $P < 0.01$ ) were positively correlated with P-eEF2 with a dose-dependent effect of His. In conclusion, our results can provide certain basic information for the further study of the regulation mechanism of Leu or His on casein expression via mTOR pathway in CMEC-H.

**Key Words:** leucine, histidine, mammalian target of rapamycin (mTOR)

**M151 MicroRNA miR-200b regulates lactation and cell proliferation by concurrently targeting *Dnmt3a* and *Dnmt3b* in bovine mammary epithelial cells.** Yanjie Bian, Chunmei Wang, Na Zhang\*, Yingjun Cui, and Qingzhang Li, *Key Laboratory of Dairy Science, Ministry of Education, Northeast Agricultural University, Harbin, P.R. China.*

With the rapid development of dairy industry and improvement of human living standards, the consumption market of dairy products constantly expanding. How to increase the milk yield and optimize milk quality has become the focus of dairy industry. Although numerous microRNA (miRNA) have been verified to play critical roles in the regulation of milk protein synthesis and mammary gland development in cows, but the mechanisms underlying their effects remain unknown. Here, 6 healthy, multiparous Chinese Holstein cows with similar genetic backgrounds were divided into 2 groups according to milk quality ( $n = 3$  per group), statistical analyses were conducted using Student's test, and  $P < 0.05$  was considered statistically significant. We found that the expression of miR-200b was significantly different between high-milk-quality cows and low-milk-quality ones by using small RNA sequencing and qRT-PCR techniques. Increasing evidences suggested that epigenetic mechanism was involved in the regulation of mammary function in the



dairy cow. In recent studies, DNA methylation, as the major epigenetic regulators, has been shown to regulate the expressions of milk proteins in mammary epithelial cells. In vitro, subcultured bovine mammary epithelial cells (BMEC) were cultured in an incubator at 37°C in the presence of 5% CO<sub>2</sub> and were maintained in DMEM/DF-12 with 10% FBS. Our results showed that miR-200b regulated the DNA methylation level of BMECs by inversely targeting both DNMT3A and DNMT3B. We also found that the overexpression of miR-200b promoted BMECs proliferation and significantly decreased the expression of  $\alpha_{S1}$ -casein and  $\beta$ -casein. The treatment of BMECs with 5-aza-2'-deoxycytidine (5-Aza-dC) increased the expression of  $\alpha_{S1}$ -casein and  $\beta$ -casein. This study provided new insight into the molecular mechanisms of miR-200b in regulating lactation performance through DNA methylation, and provided a potential to optimize milk quality not only of the cows but also of their next generations.

**Key Words:** miR-200b, DNA methylation, bovine mammary epithelial cells (BMEC)

**M152 MicroRNAs miR-181a and miR-194 can regulate the biosynthesis of milk fat and protein by targeting ACSL1 and STAT5a.** S. Lian<sup>1,2</sup>, X. M. Nan<sup>2,3</sup>, D. P. Bu<sup>\*2,4</sup>, F. Wang<sup>2</sup>, and J. R. Guo<sup>1</sup>, <sup>1</sup>College of Animal Science and Veterinary Medicine, Heilongjiang Bayi Agricultural University, Daqing, Heilongjiang, China, <sup>2</sup>State Key Laboratory of Animal Nutrition, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>3</sup>Synergetic Innovation Center of Food Safety and Nutrition, Harbin, China, <sup>4</sup>CAAS-ICRAF Joint Lab on Agroforestry and Sustainable Animal Husbandry, Beijing, China.

MicroRNAs (miRNA) are a class of small noncoding RNAs and act as important post-transcriptional regulators of gene expression. *ACSL1* is an important gene in milk lipid synthesis and *STAT5a* is one of the key genes in milk protein synthesis. In a previous study dealing with incubation of stearic acid in bovine mammary epithelial cells, an opposite expression pattern was observed between *ACSL1* and miR-181a, *STAT5a* and miR-194. *ACSL1* and *STAT5a* were also predicted as potential target genes of miR-181a and miR-194 respectively by TargetScan and PicTar, which are the most popular target gene prediction softwares. The objective of this work was to determine the potential function of miR-181a and miR-194 on milk fat and protein synthesis by defining the regulatory relationship between miR-181a and *ACSL1*, miR-194 and *STAT5A*. In the study, primary bovine mammary epithelial cells harvested from mid-lactation cows were cultured in DMEM/F12 medium added with 10% FBS and lactational hormones. Cells were transfected with miR-181a mimic, miR-181a inhibitor, miR-194 mimic and miR-194 inhibitor, and non-transfected cells were set as control. After an 48 h treatment, expression of miR-181a, miR-194, *ACSL1*, *STAT5a* were determined by real-time reverse-transcription PCR. The results showed that the overexpression of miR-181a and miR-194 inhibited the expression of *ACSL1* and *STAT5a* respectively ( $P < 0.05$ ), while the downregulation of the 2 miRNAs increased *ACSL1* and *STAT5a* expression ( $P < 0.05$ ). Therefore suggesting that miR-181a and miR-194 may play important roles in milk fat and protein synthesis.

**Key Words:** miR-181a, miR-194, bovine mammary epithelial cells

**M153 Establishment of a mammary epithelial cell line from a Chinese Holstein dairy cow and effects of different concentrations of insulin-like growth factor-I on expression of genes related to milk synthesis.** T. Qin<sup>1</sup>, H. Y. Wang<sup>1</sup>, D. P. Bu<sup>2</sup>, and H. B. Zhu<sup>\*1</sup>, <sup>1</sup>Embryo Biotechnology and Reproduction Laboratory, Institute of

Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>2</sup>State Key Laboratory of Animal Nutrition, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China.

This experiment was conducted to establish an efficient culture method for bovine mammary epithelial cells (bMEC) in vitro, and to research the effect of different concentrations (0, 0.1, 10 and 100ng/mL) insulin-like growth factor-I (IGF-1) on the mRNA expression of genes related to milk protein and fat synthesis in bMEC. Bovine mammary epithelial cells were cultured by tissue mass culture method, and cells were purified according to different sensitivity to trypsin of fibroblasts and bovine mammary epithelial cells. The growth curve of purified cells was detected by cell counting assay method. The expression of keratin 18 was detected by immunofluorescent histochemistry staining method. mRNA expression of 8 genes were measured by real time quantified PCR(RT-qPCR). The results showed that mRNA abundance of insulin-like growth factor-I receptor (*IGF1R*), insulin-like binding protein 3 (*IGFBP3*),  $\alpha_{S1}$ -casein (*CSN1S1*) and  $\kappa$ -casein (*CSN3*) did not differ ( $P > 0.05$ ) within different concentrations of IGF-1. However, within the increasing concentrations of IGF-1, the mRNA abundance of  $\beta$ -casein (*CSN2*), acetyl-CoA carboxylase (*ACACA*), fatty acid synthase (*FASN*) and fatty acid binding protein 3 (*FABP3*) increased significantly ( $P < 0.05$ ). These results indicated that IGF-1, as an important cytokine, might involve in regulating the mRNA expression of genes related to milk protein and fat synthesis in bovine mammary epithelial cells in vitro.

**Key Words:** insulin-like growth factor-I, bovine mammary epithelial cells, gene expression.

**M154 Influence of heat stress and amino acid supplementation on microRNA expression in bovine mammary epithelial cells.** M. Duque<sup>\*1</sup>, A. A. K. Salama<sup>2</sup>, and J. J. Loo<sup>3</sup>, <sup>1</sup>Grupo de Investigación Biogénesis and GRICA, Facultad de Ciencias Agrarias, Universidad de Antioquia, Medellín, Colombia, <sup>2</sup>Grup de Recerca en Remugants (G2R), Departament de Ciència Animal i dels Aliments, Universitat Autònoma de Barcelona, Bellaterra, Spain, <sup>3</sup>Department of Animal Sciences, University of Illinois, Urbana, IL.

Heat stress and essential amino acid profiles are important factors that can affect milk fat and protein synthesis in the mammary gland of dairy cows. MicroRNAs (miRNA) are non-coding RNAs that play a critical role in post-transcriptional regulation of bovine mammary gene expression. Regulatory microRNAs affect a wide variety of cellular processes such as cellular differentiation, proliferation, apoptosis, genome stability and milk protein and fat synthesis pathways. Although miRNA expression profiling in various bovine tissues or cells have been performed, studies to elucidate the potential role of miRNAs under stress conditions are limited or nonexistent. The objective of this study was to determine using MACT cells the effects of heat stress and amino acid (AA) supplementation on microRNA expression. A thermo neutral (TN, 37°C) and heat stress (HS, 42°C) environment with one of 3 AA supplementation groups were studied: control (Con), control + methionine (Met) and control + arginine (Arg). A total of 6 treatments were evaluated: TN-Con, TN-Met, TN-Arg, HS-Con, HS-Met and HS-Arg. After 6 h of incubation cells were harvested, and miRNAs were extracted for quantitative RT-PCR analysis. Targets included miR-141, miR-221, miR-34a, miR-200a, miR-27a/b, miR-92a, miR-99a, miR-23a, miR-26a, miR-103, and miR-184. Both HS and AA increased ( $P < 0.05$ ) the expression of miR-141 and miR-221, which were reported to decrease STAT5 protein expression and induce angiogenesis. An upregulation ( $P < 0.05$ ) of miR-34a, miR-200a, miR-27a/b, miR-92a, miR-99a, and miR-184 was observed in the HS group, but not with AA supplementation. These miRNAs are reported to be involved in the regulation of

heat-shock proteins, milk fat synthesis, prolactin receptor expression, cell cycle regulation, and mTOR and insulin signaling. Both miR-23a (MAPK pathway) and miR-26a (cell proliferation) were not affected ( $P > 0.05$ ) by temperature, but Met and/or Arg decreased ( $P < 0.05$ ) their expression. The sole miRNA unaffected by temperature or AA was miR-103. The results suggest that AA supplementation and HS can alter miRNA expression and, as such, could serve as another regulatory factor of genes that play important functions in the mammary gland.

**Key Words:** bovine mammary cells, miRNA, amino acid

**M155 Characterization of prolactin and adrenergic and serotonergic receptors in bovine apocrine sweat glands.** Courtney A. Burger, Jayne L. Collier, and Robert J. Collier\*, *University of Arizona, Tucson, AZ.*

Apocrine sweat glands are involved in thermoregulation via evaporative heat loss in the bovine and are most active at temperatures above thermoneutral. We have previously shown that serotonin (5-HT) receptors 5-HT 1b, 2b, 4, and 7 are found in whole skin, apocrine sweat glands, and epithelial cells, while 5-HT 2a is present in whole skin and epithelial cells. It has been shown that prolactin (PRL) plays a role in lactational homeostasis, because mammary glands are modified apocrine sweat glands, we hypothesized that PRL receptors would be present in sweat glands. Additionally, we looked at adrenergic (ADR) receptors, as epinephrine is known to influence sweating response. The present study was conducted to determine if the 5-HT, PRL, and ADR systems are involved in thermoregulation by examining the cDNA expression of receptors through conventional PCR in bovine whole skin, apocrine sweat glands, and epithelial cells. For the 5-HT system, brain was used as control and additional isoforms classified were 5-HT 1a, 1d, 1f, 5a, and 6. We identified isoforms 5-HT 1d, 1f, and 5a in whole skin, apocrine sweat glands, and epithelial cells, whereas 5-HT 1a is present in whole skin and epithelial cells, and 5-HT6 is present in whole skin and apocrine sweat glands. For the PRL system, bovine mammary epithelial cells (BMEC) were the control and 2 isoforms were identified: PRL receptor long (PRLr-L) and PRL receptor short (PRLr-S) which differ in their intracellular domain length and sequence. We found PRLr-S in whole skin, apocrine sweat glands, and epithelial cells, while PRLr-L is in whole skin only. For ADR receptors, BMEC and liver were used as control, and we found  $\alpha 1$  ADR 1a,  $\alpha 2$  ADR,  $\beta 1$  ADR, and  $\beta 2$  ADR receptors in whole skin, apocrine sweat glands, and epithelial cells.  $\alpha 1$  ADR 1b was only present in BMEC and liver, while  $\alpha 1$  ADR 1d was only in BMECs. The presence of these receptors in the apocrine glands and epithelial cells indicates that the 5-HT, PRL, and ADR systems are involved in regulation of apocrine sweat gland function.

**Key Words:** apocrine, bovine, serotonin

**M156 RNA-sequencing analysis of milk somatic cells in heat-stressed dairy goats.** A. A. K. Salama\*<sup>1</sup>, B. Badaoui<sup>2</sup>, S. Hamzaoui<sup>1</sup>, and G. Caja<sup>1</sup>, <sup>1</sup>*Grup de Recerca en Remugants (G2R), Departament de Ciència Animal i dels Aliments, Universitat Autònoma de Barcelona, Bellaterra, Spain,* <sup>2</sup>*Integrative Biology Group, Parco Tecnologico Padano - CERSA, Lodi, Italy.*

Milk transcriptome characterization could help in understanding how milk synthesis and mammary cell turnover are affected by heat stress. Eight multiparous Murciano-Granadina dairy goats ( $44.1 \pm 1.7$  kg BW;  $2 \pm 0.04$  L/d;  $90 \pm 3$  DIM) were kept in metabolic cages and randomly assigned to 2 climatic treatments according to a crossover design (2 28-d periods). Treatments were: 1) thermal neutral (TN; 15 to 20°C, 40 to 45% humidity, THI = 59 to 65), and 2) heat stress (HS, 12 h/d at 37°C and 40%, and 12 h/d at 30°C and 40%, THI = 86 and 77, respectively). Milk samples were collected at d 28 of each period, somatic cells were separated, and RNA was extracted. The RNA of 4 goats for each treatment at each period was mixed, resulting in final 4 RNA samples (2 TN and 2 HS) that were analyzed by Illumina RNA-sequencing (RNA-Seq). The RNA-Seq reads were mapped to the bovine genome by “tophat” and the expression level was quantified by “cufflinks.” The RNA-Seq produced a total of ~170 million reads with an average of 85 million reads for each treatment (range of reads for the 4 samples was 38 to 46 million). We detected 140,39 transcripts expressed in milk cells, 700 of them were differentially ( $P < 0.01$ ) expressed between HS and TN. The HS reduced ( $P < 0.01$ ) the expression of genes related to de novo fat synthesis (*ACACA* and *FASN*), fatty acid desaturation (*SCD*), milk fat globule formation (*BTN1A1*, *XDH*, and *GLYCAM1*), and protein and lactose synthesis (*CSN1S1*, *CSN1S2*, *CSN3*, and *LALBA*). The HS upregulated ( $P < 0.01$ ) cathepsin genes (*CTSB*, *CTSD*, *CTSZ*, *CTSS*, *CTSC*, and *CTSL1*) as well as genes related to plasminogen pathway (*PLAU*, *PLAUR*) and ubiquitin-proteasome pathway (*FBXW2*, *UBAPI*, *RPS27A*). Furthermore, HS increased ( $P < 0.05$ ) the expression of the pro-apoptotic tumor necrosis factor receptors superfamily (TNFRSF1A and TNFRSF1B). Seven miRNA (miR-23a, miR-24-2, miR-27a, miR-29c, miR-29d, miR-142, and miR-221) had a greater ( $P < 0.01$ ) expression with HS. In conclusion, heat stress reduces the synthetic capacity of mammary cells and increases the expression of proteases in milk. This increase in proteases expression could negatively affect milk coagulation properties and cheese making using milk produced from heat-stressed goats.

**Key Words:** RNA-sequencing, heat stress, dairy goat

# Meat Science and Muscle Biology

**M157 Molecular background of differential expression of *THRSP* in bovine longissimus muscle.** Lisa Schering\*, Elke Albrecht, Yinuo Liu, Christa Kühn, Klaus Wimmers, and Steffen Maak, *FBN Dummerstorf, Dummerstorf, Germany*.

Increased expression of thyroid hormone responsive protein (*THRSP*) has been related to higher intramuscular fat (IMF) content in previous studies in cattle. Higher expression of the *THRSP* gene (1.7-fold,  $P < 0.05$ ) could be confirmed in *M. longissimus* of bulls of a Charolais × Holstein F<sub>2</sub>-cross with high IMF ( $6.8 \pm 2.4\%$ ,  $n = 10$ ) compared with those with low IMF ( $1.9 \pm 0.5\%$ ,  $n = 10$ ). The presented study aimed at elucidation of the background of the observed expression differences. First, we screened the *THRSP* locus including 700 bp located 5' of the transcription start and 1,000 bp containing the 3'-untranslated region (UTR) for single nucleotide polymorphisms (SNPs) with potential effects on gene expression. Only 3 out of 23 previously annotated SNPs were polymorphic in our sample. One new SNP (c.\*695T>C) was found in the 3'UTR. None of the SNPs was related to expression of *THRSP*. To account for the possibility that an elevated amount of *THRSP* mRNA is an effect of more adipocytes in muscle samples with high IMF, we analyzed adipocytes and myocytes separately. Cells of the respective types were isolated by laser capture microdissection from unstained sections of *M. longissimus*. Total RNA was extracted from both cell types and cDNA was reversely transcribed from mRNA. Besides *THRSP*, fatty acid binding proteins (FABP) 3 and 4 were determined as specific markers for muscle and fat cells, respectively. In 2 independent experiments, we found exclusive expression of FABP3 in myocytes and of FABP4 in adipocytes indicating purity of the cell type preparations. *THRSP* expression was observed in adipocytes but not in myocytes. Moreover, expression of *THRSP* was not different in subcutaneous fat between both groups of bulls (1.0-fold,  $P = 0.93$ ) despite a high level of expression. Our results suggest that differences of *THRSP* mRNA abundance in muscle of cattle with differing IMF result from variable numbers of *THRSP*-expressing adipocytes in the samples rather than from different activity of the gene.

**Key Words:** cattle, *THRSP*, intramuscular fat

**M158 Effects of feeding different sources of vegetable oils on meat quality traits of Nellore steers.** Fabiane de Souza Costa\*<sup>1</sup>, Anderson Roberto Cabral<sup>1</sup>, Marília Aparecida Izepe da Silva<sup>1</sup>, Beatriz de Conti Fiorese<sup>1</sup>, Danielle Leal Mataram<sup>1</sup>, Saulo da Luz e Silva<sup>1</sup>, and Angélica Simone Cravo Pereira<sup>2</sup>, <sup>1</sup>*University of Sao Paulo/Faculty of Animal Science and Food Engineering, Pirassununga, Sao Paulo, Brazil*, <sup>2</sup>*University of Sao Paulo/School of Veterinary Medicine and Animal Science, Pirassununga, Sao Paulo, Brazil*.

The objective of this study was to examine the effect of feeding soybean, sunflower or linseed oils on beef quality traits of feedlot finished Nellore steers. Ninety-six steers were individually fed one of the following diets: CON = a control diet with 79% of concentrate and 21% of corn silage, without oil; SOY = diet with soybean oil; SUN = diet with sunflower oil; LIN = diet with linseed oil. Different oil sources were included at 3.5% of DM in substitution of corn at the same percentage. After 84 d of feeding steers were harvested and after 48 h of chilling 2 samples of *Longissimus* muscle (2.5 cm thick) were collected between 12th and 13th ribs, vacuum packed and aged for 0 and 14 d. Samples were analyzed for pH, color (L\*, a\*, b\*), cooking loss (CL) and Warner-Bratzler shear force (WBSF). Data were analyzed using the MIXED procedure of SAS

software as a randomized complete block (initial body weight) design, considering treatment, aging period, treatment × aging interaction as fixed effects, and block as a random effect. There was no treatment × aging period interaction for any trait. Treatments did not affect any trait evaluated in this study ( $P > 0.05$ ). The average pH values ( $5.6 \pm 0.02$ ) were within the normal range for all treatments. The WBSF also did not differ among treatments but the average values ( $8.2 \pm 0.46$  kg) were considered very high, indicating a tough meat. The average cooking loss was  $23.9 \pm 1.71\%$  and also is within the range normally reported in the literature for Nellore cattle. Color attributes averaged 32.7, 14.7 and 12.6 for L\*, a\* and b\*, respectively. The pH showed a small increase with aging period, from 5.5 to 5.6 ( $P < 0.001$ ) but was within the normal range. The WBSF and CL decreased with increase of aging period ( $P < 0.001$ ) from 10.5 to 6.0 kg for WBSF and from 25.6 to 22.2% for CL for 0 and 14 d of aging, respectively. The L\* values increased with aging period ( $P < 0.001$ ) from 29.4 to 35.9, however the a\* ( $14.6 \pm 0.70$ ) and b\* ( $12.6 \pm 1.80$ ) values were not affected by aging. Inclusion of different oil sources does not affect meat tenderness and color attributes of feedlot finished Nellore steers.

**Key Words:** tenderness, feedlot, linseed oil

**M159 Effects of beta agonist and immunocastration on fatty acid profile of cattle.** Madeline Rezende Mazon\*, Saulo da Luz Silva, Daniel Silva Antonelo, Keni Nubiato, Juan Fernando Morales Gomez, Roberta Ferreira Carvalho, and Paulo Roberto Leme, *University of São Paulo, Pirassununga, São Paulo, Brazil*.

Beta agonists (BAA) are known for decreasing lipogenic activity and increasing lipolysis on beef and immunocastration (IM) could be an alternative tool to improve fat deposition. Therefore, this work was developed to evaluate the effect of BAA and IM on meat quality traits of feedlot finished Nellore cattle. Ninety-six bulls (409 ± 50 kg LW; 20 mo old) were divided into 2 groups and half of the bulls remaining bulls and the other half receiving 2 doses of IM vaccine (Bopriva) over a 30 d period before arrival at the feedlot. Cattle were fed for a period of 70 d on common diet (76% concentrate and 24% roughage). Each of these groups were then split in 3 groups and fed during 30 d one of the following treatments: control diet without BAA (CON); CON diet plus 80 mg/day zilpaterol hydrochloride (Zilmax; ZIL); CON diet plus 300 mg/day of ractopamine hydrochloride (Optaflexx; RAC). The cattle fed ZIL were 2 d withdrawal period before harvested. After 100 d on feed, cattle were harvested and one samples of LM was collected between 12/13th ribs for fatty acid profile determinations. Data was analyzed by ANOVA as randomized complete block (initial LW) design in 2 × 3 factorial arrangement and means were compared by Student *t*-test. There was no significant interaction of gender condition and BAA for any traits evaluated. Immunocastrated cattle had a greater percentage of MUFA (45.9 vs. 43.0;  $P = 0.002$ ), palmitoleic (3.0 vs. 2.6;  $P = 0.002$ ), Oleic (36.1 vs. 33.3;  $P = 0.002$ ) and total Lipids (3.2 vs. 2.3 g;  $P = 0.002$ ) compared with non-castrated, respectively, whereas non-castrated had greater stearic concentration (13.8 vs. 12.6;  $P = 0.006$ ). Treatments CON and RAC showed greater values than ZIL for myristic (2.5 and 2.4 vs. 2.1;  $P = 0.017$ ) and palmitic (23.7 and 23.0 vs. 21.9;  $P = 0.009$ ), whereas ZIL treatment had greater values than CON and RAC for linoleic (8.5 vs. 6.7 and 6.5;  $P = 0.030$ ), arachidic (2.6 vs. 1.8 and 1.8;  $P = 0.046$ ), Omega 6 (12.1 vs. 9.2 and 9.4;  $P = 0.039$ ). Beef from cattle fed ZIL presented greater CLA and PUFA concentrations, thus having more healthy beef. Immunocastrated cattle have more fat (lipids), MUFA and



saturated fatty acids. Sex condition and BAA can be used to change the fatty acid profile of beef from Nellore cattle.

**Key Words:** zilpaterol, ractopamine

**M160 Growth and serial real-time ultrasound measurements for backfat and loin muscle area in three breeds of swine.** L. L. Lo\* and C. C. Tsai, *Chinese Culture University, Taipei, Taiwan.*

A serial real-time ultrasonic measured of backfat and loin muscle area was recorded to examine the rate of change per unit of live weight during the finishing stage of production. Sixty-nine sets of 3 littermates (boar, gilt, and barrow one each) involving the 3 major breeds Duroc, Landrace, and Yorkshire in Taiwan were used in this study. Real-time ultrasonic measurements of backfat and loin muscle area were collected at average body weight (BW) of 57.2, 78.8, 94.2, 113.5, and 128.1 kg. Backfat was measured off the midline at the 3/4 point (TRBF) over the tenth rib. Loin muscle area (LMA) images were collected from the left side of each pig at the 10th rib. Deposition rates were calculated for the dependent ultrasound traits TRBF and LMA in models that included the independent variables BW and BW<sup>2</sup>. Body weight was regressed on age and age<sup>2</sup> for growth rate (GR). The serial measurements data were analyzed using a mixed model (PROC MIXED, SAS) that included the fixed effects of test group, breed, sex, and random effect of animal. Significant breed effects were detected for deposition rate of TRBF ( $P = 0.0405$ ), LMA ( $P = 0.0210$ ), and GR ( $P = 0.0082$ ). Backfat thickness was deposited at a mean of 0.109 mm/kg, whereas LMA was averaged 0.299 cm<sup>2</sup>/kg deposited across 3 breeds. Average GR across breeds was 677.3 g. Barrows deposited fat at a greater rate ( $P = 0.0220$ ) and grew at a greater rate ( $P = 0.0462$ ), when compared with gilts. However, barrows grew at a lesser rate ( $P = 0.0380$ ) than boars. The results of this study indicated that effects of breed and sex play important roles for rates of backfat and loin muscle deposition and weight per day of age in swine, and therefore, breed- and sex-specific adjustment factor are required.

**Key Words:** backfat, loin muscle area, pig

**M161 Effects of gender status on beef quality from crossbred Angus x Nellore cattle.** Lenise F. Mueller\*<sup>1</sup>, Júlio C. C. Balieiro<sup>1</sup>, Adrielle M. Ferrinho<sup>1</sup>, Joyce J. M. Furlan<sup>1</sup>, Laura R. R. Mantilla<sup>3</sup>, Fernando S. Baldi<sup>2</sup>, and Angélica S. C. Pereira<sup>1</sup>, <sup>1</sup>University of Sao Paulo, Pirassununga, Sao Paulo, Brazil, <sup>2</sup>Sao Paulo State University, Jaboticabal, Sao Paulo, Brazil, <sup>3</sup>University Cooperative of Colombia, Medellin, Colombia.

The goal of this study was to evaluate the influence of gender status on beef quality from feedlot crossbred Angus x Nellore cattle. Twenty months old cattle (n = 454) presenting initial mean weight of 265 kg ± 24.24 were grouped into lots, confined and assigned into the following treatments: bulls, steers, immunocastrated, and heifers. Cattle were fed ad libitum high-grain diet containing 80% concentrate. At the end of the experimental period (190 d), 40 cattle from each group were chosen based on body weight (bulls 555 kg ± 43.36, steers 478 kg ± 32.40, immunocastrated 509 kg ± 30.75, and heifers 468 kg ± 34.21) and harvested. Steaks (2.54 cm thick) were collected from the *Longissimus dorsi* and aged for 0, 7, and 14 d (d) at 2°C; after aging, all steaks were stored at -18°C until analyzed for tenderness and pH. Statistical analyzes were performed using the proc MIXED SAS program (version 9.2), including the fixed effect of gender status (bulls, steers, immunocastrated, and heifers), and the covariate d on feed, and the random effect of cattle within of gender groups. Significant effects for gender status were evaluated by *t*-test using PDIF option. On d 0, immunocastrated

cattle and steers beef presented decreased shear force values ( $P > 0.05$ ), when compared with heifers and bulls; heifers presented decreased shear values when compared with bulls ( $P < 0.05$ ). After 7 d of aging bulls presented increased shear values (4.74kg ± 0.20) when compared with the other treatments ( $P < 0.01$ ). Also, no difference was detected between immunocastrated cattle (3.40kg ± 0.20), steers (3.32kg ± 0.20), and heifers (3.94kg ± 0.20), however heifers presented increased shear force values when compared with steers ( $P < 0.05$ ). At 14 d of aging bulls presented increased shear values (3.84kg ± 0.15) when compared with the other groups ( $P < 0.01$ ); there was no difference ( $P < 0.05$ ) between the other categories. Gender status influenced pH values, bulls showed higher pH values when compared with the other groups ( $P < 0.05$ ). Despite the effect of gender status on beef quality, it is important to highlight that the use of steers, immunocastrated cattle and heifers is necessary to attend demanding consumers seeking for tender beef.

**Key Words:** immunocastration, tenderness, beef

**M162 Influence of aging on intramuscular color variations in beef semimembranosus.** Mahesh Narayanan Nair\*, Shuting Li, Ryan Chaplin, Gregg Rentfrow, and Surendranath P. Suman, *University of Kentucky, Lexington, KY.*

Previous research documented intramuscular variations in the color and color stability of beef semimembranosus, which can be separated into color-labile inside (ISM) and color-stable outside (OSM) regions. Post-mortem aging is employed to improve beef quality attributes such as tenderness and palatability. Aging can also influence fresh beef color, whereas its effect on intramuscular color variations in beef semimembranosus is yet to be examined. Therefore, the objective of the present study was to evaluate the influence of aging on color and color stability of ISM and OSM and to examine if aging can mitigate intramuscular color variations. Semimembranosus muscles from 8 (n = 8) beef carcasses (USDA Choice, 24 h post-mortem) were vacuum packaged and aged at 2°C for 7, 14, and 21 d. On each aging period, muscles were fabricated into ISM and OSM steaks. Aerobically packaged steaks were stored in the dark at 2°C for 6 d. On d 6 of storage, lightness ( $L^*$ ), redness ( $a^*$ ), yellowness ( $b^*$ ), and color stability (ratio of reflectance at 630 nm and at 580 nm) were evaluated on the steak surfaces using a HunterLab LabScan XE colorimeter. The data were analyzed using PROC MIXED procedure in SAS. OSM exhibited greater ( $P < 0.05$ ) surface redness ( $a^*$  value) than ISM after 7 and 14 d of aging. However, after 21 d of aging, ISM and OSM demonstrated similar  $a^*$  values ( $P > 0.05$ ). While OSM was darker ( $P < 0.05$ ; lower  $L^*$  value) than ISM at all aging periods, there was no difference ( $P > 0.05$ ) in yellowness throughout the aging. Color stability was greater ( $P < 0.05$ ) in OSM than in ISM on d 7 and 21 of aging, whereas ISM and OSM exhibited similar ( $P > 0.05$ ) color stability on d 14. These results suggested that 21 d aging could be employed to potentially minimize the intramuscular variation in redness of beef semimembranosus.

**Key Words:** semimembranosus, beef color, aging

**M163 Effect of oil source and storage time on burger sensorial traits from Nellore cattle.** Anderson R. Cabral\*<sup>1</sup>, Fabiane de Souza Costa<sup>1</sup>, Maria E. Groto<sup>1</sup>, Saulo da Luz e Silva<sup>1</sup>, and Angélica S. Cravo Pereira<sup>2</sup>, <sup>1</sup>University of Sao Paulo/FZEA, Pirassununga, SP, Brazil, <sup>2</sup>University of Sao Paulo/FMVZ, Sao Paulo, SP, Brazil.

The aim of this study was to evaluate the effect of fed plant based oil source over 2 different storage period in sensorial burger traits. Ninety-six Nellore steers were confined for 82 d. The experimental design used

was a split plot in a  $4 \times 2$  factorial arrangement (3 oil sources plus control  $\times$  2 storage time). The oils sources were sunflower oil (SU); linseed oil (LO) and soybean oil (SO). Plant oils were added to the feedlot diet at a rate of 3.5% of concentration in each diet (based upon DM). Utilizing the *Longissimus dorsi* and subcutaneous fat from the carcass. 40 (100g/patties) hamburger patties were prepared per treatment utilizing a commercial formulation (85.4% of meat, 12% of fat, 2% of salt, 0.3% of garlic and 0.3% of emulsifier) with a weight around 100g and packed in plastic bags permeable to oxygen and then immediately frozen at  $-18^{\circ}\text{C}$  and stored for 2 time periods, 0 and 90 d. The burgers were grilled in  $170^{\circ}\text{C}$  for 4 min on each side (internal temperature  $75^{\circ}\text{C}$ ). Each panel member received 4 pieces (one for each treatment) randomly. Sensory analysis was performed utilizing an acceptance test with 9 point hedonic scale (9 = like extremely, 1 = dislike extremely) by a consumer panel (n = 100). They evaluated aroma (A), texture (T), juiciness (J), flavor (F) and overall acceptability (OA). The data was analyzed using a mixed model (MIXED procedure of SAS), including the fixed effects of oil source, storage time, the interaction between storage time with oil source and the random animal effect. There was no interaction between storage times with oil source ( $P = 0.56$  to A,  $P = 0.51$  to T,  $P = 0.85$  to J,  $P = 0.41$  to F,  $P = 0.89$  to OA) for any sensorial trait evaluated, neither the oil source for A ( $P = 0.08$ ) and J ( $P = 0.26$ ) nor storage time for A ( $P = 0.36$ ) and J ( $P = 0.07$ ). There was an effect ( $P = 0.01$ ) of storage time for T (0d = 6.29; 90d = 5.90). There was a reduction OA ( $P = 0.02$ ) at time zero (6.87) compared with the time 90d (6.58). The hamburgers patties from SO, SU and control were tastier than those fed LO (7.12, 7.10, 7.04 vs. 6.65) ( $P = 0.001$ ). In conclusion, the storage time and oil source have an effect on the burger sensorial traits, and the use of linseed oil or the burger storage for a period of 90d would reduce the acceptance by the consumers.

**Key Words:** lipid, texture, flavor

**M164 Association of SNPs in the *THRSP* gene with morphological and biochemical traits of longissimus muscle in  $F_2$  offspring of a Charolais  $\times$  Holstein cross.** Elke Albrecht\*, Lisa Schering, Dirk Dannenberger, Gerd Nuernberg, Christa Kuehn, and Steffen Maak, *Leibniz Institute for Farm Animal Biology, Dummerstorf, Germany*.

Expression and genetic variants of the thyroid hormone responsive protein (*THRSP*) gene has been reported to be associated with marbling and fatty acid composition in cattle. The study was undertaken to investigate the effect of 2 single-nucleotide polymorphisms (SNP) in the *THRSP* gene, namely c.88 G>A and c.194 C>T, on morphological and biochemical traits of the longissimus muscle. Genotyping of 247  $F_2$ -generation bulls of a Charolais  $\times$  Holstein cross revealed that both SNPs were in complete linkage disequilibrium in our data set with 37 bulls of the GG/CC, 110 of the AG/CT, and 100 of the AA/TT diplotype. Muscle fiber composition, intramuscular fat cell and marbling traits were analyzed using image analysis. Fatty acid concentrations/proportions were determined with gas chromatography using internal standard procedure as fatty acid methyl esters. The  $F_2$  design yielded substantially increased variation within the population compared with

both founder breeds regarding muscle structure, fat cell, and marbling data within  $F_2$  bulls ranged from 4.5 standard deviations (cross sectional area of intermediate muscle fibers) to 7.4 standard deviations (area percentage of slow muscle fibers). Furthermore, absolute and relative fatty acid composition exceeded variation of muscle structural traits with differences between individuals up to 11.3 standard deviations (C21) indicating extreme divergence in fat deposition within the population. However, association analysis indicated no clear effect of *THRSP*-SNP diplotype on longissimus muscle structure and intramuscular fat deposition, but showed an indication on association of the *THRSP*-SNPs with muscle fatty acid composition ( $P < 0.05$ ).

**Key Words:** cattle, fatty acid, muscle structure

**M165 Meat quality of *M. Longissimus dorsi* of lambs fed with sunflower cake.** Anny Graycy Vasconcelos de Oliveira Lima, Ronaldo Lopes de Oliveira\*, Thadeu Mariniello Silva, Patrícia Gonçalves Cirqueira, and Marcondes Dias de Freitas Neto, *Federal University of Bahia, Salvador, Bahia, Brazil*.

This trial was conducted to determine the effect of sunflower cake on the physicochemical properties of meat lamb. Forty crossbreed Santa Ines rams were fed 50% hay (Tifton-85 chopped) to 50% concentrate (corn, soybean meal, urea, ammonium sulfate and inclusion of sunflower cake) on diet. The 4 treatment diets were 0, 10, 20, and 30% sunflower cake. To account for the increasing concentration of sunflower cake, corn and soybean meal were removed from the diet. The rams were fed the treatment diets twice daily for a period of 71 d in feedlot, and were slaughtered after that. After a period 24 h postmortem, the *Longissimus dorsi* muscle were removed of carcass, separated into proximal and distal portions. The muscles from the distal portion's left side were used to determine the pH and color measurements with Minolta colorimeter and pH probe. The muscles from proximal portion the right side were used to measure the cooking loss and Warner-Bratzler shear force analysis was conducted according to the guidelines of AMSA. The experiment followed a completely randomized design with 4 treatments and 10 replicates per treatment. The data were analyzed using PROC GLM procedure of SAS (SAS Institute, Cary NC) with the treatments as the fixed effect. Regardless of inclusion rate, sunflower cake did not have an effect on pH ( $P = 0.15$ ),  $L^*$  ( $P = 0.28$ ), cooking loss ( $P = 0.23$ ), shear force ( $P = 0.47$ ). There was an effect on color  $a^*$  ( $P = 0.03$ ) and  $b^*$  ( $P = 0.04$ ) with increasing concentration of sunflower cake, that characterizing bright cherry red color meats. Many factors (endogenous and exogenous) can contribute to meat color stability as pH, muscle source and mitochondrial activity, as well as live animal related factors, such as management, diet, and genetics. The competition between mitochondria and myoglobin (Mb) is a key component in the development of bright-red color. Probably occurred saturation Mb with oxygen forming oxymyoglobin or saturation Mb with carbon monoxide, forming carboxymyoglobin that offer bright cherry-red color meat. The sunflower cake inclusion has provided bright redder color meat without changing the cooking loss and tenderness of meat.

**Key Words:** color, cooking loss, tenderness

## Nonruminant Nutrition: Amino acids

### **M166 Effects of arginine supplementation during early gestation (d 1 to 30) on litter size and plasma metabolites in gilts and sows.**

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Two experiments were conducted, using typical commercial swine production conditions, to determine effects of dietary arginine supplementation during early gestation on the performance of gilts and sows. In Experiment 1, 62 Landrace gilts and 113 sows were used to evaluate the effect of dietary arginine supplementation during early gestation (d 1 to 30) on gilt and sow performance. On d 1 of gestation, all gilts and sows were randomly assigned to a corn-soybean meal basal diet supplemented with either 1.3% L-arginine-HCl or 2.2% L-alanine (isonitrogenous control). The total number of piglets born per litter for arginine-supplemented was increased ( $P < 0.01$ ), compared with the control group. In Experiment 2, 155 Landrace sows were used to evaluate the effect of dietary arginine supplementation between d 1 and 14, d 15 and 30 or d 1 and 30 on sow performance. Among these sows, 24 with an average parity of 2.54 were used to determine the effect of dietary arginine supplementation on plasma amino acid composition and the concentration of plasma metabolites. The total number of piglets born per litter for arginine-supplemented sows tended to increase ( $P = 0.08$ ), compared with the control group, with the greatest improvement in sows treated from d 1 to 14. The concentrations of nitric-oxide metabolites, arginine, proline, ornithine and spermidine were increased ( $P < 0.05$ ) in the plasma of arginine-supplemented sows on d 14 and 28 of gestation, as were agmatine and putrescine on d 14 of gestation. The concentrations of urea and glutamine were lower in the plasma of arginine-supplemented than in the control group. These results indicate that dietary arginine supplementation in early gestation improved the reproductive performance and plasma biochemical metabolites in sows.

**Key Words:** arginine, early gestation, sow performance

### **M167 Limiting dietary lysine increased plasma concentration of total cholesterol in finishing pigs.**

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Lysine is the first limiting amino acid (AA) in typical swine diets, and plays important roles in nutrient metabolism and growth performance of pigs. This research was conducted to study the effects of dietary lysine on blood plasma concentrations of protein, carbohydrate and lipid metabolites of pigs. Eighteen crossbred (Yorkshire × Landrace) finishing barrows and gilts (9/sex; BW 92.3 ± 6.9 kg) were individually penned in an environment-controlled barn. Pigs were assigned to 3 dietary treatments according to a randomized complete block design with sex as block and pig as experimental unit (6 pigs/treatment). Three corn and soybean-meal based diets were formulated to contain total lysine at 0.43, 0.71, and 0.98% (as-fed basis) for diets 1 (D1; lysine-deficient), 2 (D2; lysine-adequate), and 3 (D3; lysine-excess), respectively. After 4 weeks on trial, jugular vein blood was collected and plasma separated. The

plasma concentrations of glucose, total protein, albumin, triglyceride, urea nitrogen (PUN), and total cholesterol were determined using an ACE Clinical Chemistry System (Alfa Wassermann, Inc.). Data were analyzed using the GLM Proc with pdiff (adjust = T) option of SAS. No differences were found between barrows and gilts in any of the measured metabolites. The plasma concentration of PUN was greater for pigs fed D1 than D3 ( $P < 0.05$ ), and greater for pigs fed D3 than D2 ( $P < 0.05$ ). The plasma albumin concentration was lower for pigs fed D1 than D2 or D3 ( $P < 0.05$ ), and no difference between D2 and D3. The plasma total cholesterol concentration was greater in pigs fed D1 than D2 or D3 ( $P < 0.05$ ), and no difference between D2 and D3. The plasma concentration of total protein, glucose or triglyceride did not differ among the 3 treatments. These results confirm that a diet deficient in one essential AA can lead to catabolism of other AAs and impair body protein syntheses, and that the excess amount of one AA over the requirement will be catabolized. However, the metabolic mechanism for the increased plasma total cholesterol concentration of finishing pigs by a lysine-deficient diet warrants further investigation. This research was supported by USDA-NIFA Hatch/Multistate Project 233803.

**Key Words:** lysine, nutrient metabolite, pig

### **M168 The effect of dietary protein and amino acid concentration on certain production parameters of growing ostrich chicks (*Struthio camelus*).**

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The effect of 3 different dietary protein (with a specific associated amino acid profile) concentrations on certain production parameters in growing ostriches were investigated. Measured parameters included feed intake, feed conversion ratio, and growth rate. Basic abattoir mass, postmortem measurements of the commercial cuts of meat were also done. The crude protein and amino acid requirements of ostrich chicks for the different production phases (pre-starter; 19.0% CP, starter; 15.5% CP, grower; 14.25% CP and finisher; 12.6% CP) were predicted by a newly developed mathematical growth and optimization model for ostriches (Gous and Brand, 2008; Aust. J. Exp. Agric. 48:1266-1269). Three basic diets per production phase were then formulated to be 20% lower and 20% above these predicted levels for lysine, sulfur-containing amino acids, threonine, tryptophan and arginine (named diets with a low, medium or high protein content). In the study 18 groups of birds, with 10 birds per group were used as experimental animals and were allotted to the different treatments according to live weight. The 3 dietary treatments were evaluated with 6 groups of birds per treatment. The 3 diets with the different levels of dietary protein (with a specific amino acid composition related to the dietary protein content) were fed to the ostriches during each of the 4 production phases from hatching up to slaughtering. Feed and water was available ad libitum. Results were analyzed by ANOVA with dietary protein level as main effect. Differences were determined to be statistically significant at  $P < 0.05$ . Significantly lower values with the low protein diet for the final live mass of birds at slaughter at 300 d of age (respectively 89.6, 98.8 and 102.2 kg), cold carcass mass (respectively 41.2, 45.1 and 45.1kg) and thigh weight as well as for most of the weighed muscles were found, while values for the medium and high protein diet did not differ. Both ADG (respectively 338, 361 and 377 g/bird/day) and feed intake (respectively 1.63, 1.67 and 1.76



kg/bird/day) for the low protein diet was significantly lower than that of the high protein diet with the medium diet intermediate. Results indicated that birds that consumed the diet with the medium protein performed optimal in most cases, except during the starter phase where the high protein diet outperformed the other 2 diets. This study showed that feeding diets with a higher protein and amino acid content than that predicted by the model was unable to increase performance levels of growing ostriches in most cases.

**Key Words:** ostrich, amino acid requirement, dietary protein level

#### **M169 Oral supplementation of tryptophan and pyridoxine to nursing piglets on performance and behavior after weaning.**

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The objective of this study was to evaluate the oral supplementation of Trp and pyridoxine to nursing piglets on performance and behavior after weaning. Forty 8 piglets (24 males and 24 females) from 8 sows were used. At d 14, 6 piglets per sow were divided into 3 treatments (2 animals/treatment) and were allotted in a randomized block design with 8 replicates. Treatments consisted of a Control (alanine + glucose + distilled water); Trp (3.5 g Trp/day + distilled water); Trp + pyridoxine (3.5 g Trp/day + 0.0095 g pyridoxine/day + distilled water). Each treatment was orally supplied 5 times/d and each dose contained 2.5 mL, totalling 12.5 mL/d. The supplementation was carried out during a 7 d period (d 14 to d 21 of age), and at d 21 piglets were weaned. After weaning, piglets from the same treatment were housed together in nursery pens and all animals, from each treatment, received the same pre-starter diet. During nursery period (d 21 to d 34) piglets were monitored in 3 different periods (at d 22, from d 28 to d 29 and from d 33 to d 34 of age) by a set of cameras. Behavior (standing, lying, eating, socializing, sitting and drinking) was assessed by an instantaneous sampling that was performed with intervals of 5 min to obtain the frequencies (%) of each behavior. Eating behavior was considered when pigs were eating at the feeders. Piglets' performance was evaluated in the nursing and nursery period. Piglets receiving oral Trp + pyridoxine showed higher body weight ( $P = 0.06$ ) on the first day after weaning (d 21 to d 22) compared with the Control which probably affected the weight loss ( $P = 0.06$ ) of piglets on the first day after weaning, which was lower for piglets that received oral Trp + pyridoxine. No changes in performance were observed in the remaining periods. From d 28 to d 29 piglets that received Trp supplementation in nursing period, with or without pyridoxine, showed a higher eating behavior ( $P = 0.01$ ) than the Control, which probably affected eating behavior ( $P < 0.01$ ) in the total period (from d 21 to d 34). It can be concluded that oral supplementation of Trp and its association with pyridoxine, one week before weaning, reduces the weight loss in the first day after weaning and increases eating behavior until 1 wk after weaning.

**Key Words:** postweaning stress, B6 vitamin, tryptophan

#### **M170 Methionine + cysteine and its ratios for barrows from 15 to 30 kg.**

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Aiming to determine the requirements of SID methionine + cysteine (SID Met + Cys) for barrows (15–30kg) 70 pigs (15.216 ± 0.540kg) were used, being distributed in 7 treatments with 5 replicates. A basal diet was formulated to contain 0.52% SID Met + Cys (0.26% SID Met + 0.26% SID Cys). Three treatments consisted in levels of SID Met + Cys (0.58, 0.64, 0.70%) that was maintained the 1:1 ratio between sulfur amino acids (using DL-methionine and L-cysteine) and other 3 treatments consisted in the same levels (0.58, 0.64, 0.70%), but not maintaining the 1:1 ratio (using only DL-methionine), assuring 0.55:0.45, 0.59:0.41 and 0.63:0.37 ratios. Performance, blood parameters, longissimus dorsi muscle depth (LD) and backfat thickness (BF) were evaluated. The sulfur linkages (S-S) in animals' bristles were determined by using a confocal microscope Senterra Raman. First- and second-degree models were fitted for the levels of SID Met + Cys (0.52, 0.58, 0.64, 0.70%) maintaining or not the 1:1 ratio and the low SID Met + Cys level (basal diet, 0.52%) was used to fit models maintaining or not the 1:1 ratio. Initial body weight was also included as a covariate. A likelihood ratio test was used to determine the best fitting model (specific models for maintaining or not the 1:1 ratio or a common model for both situations). Linear Response Plateau was used to determine the better SID Met + Cys level when the model of second degree was fitted. The significance level adopted was  $P < 0.05$ . The average daily gain (ADG) was affected ( $P < 0.01$ ) only when the 1:1 ratio was not maintained (using only DL-methionine), reaching a plateau at 0.661% of SID Met + Cys. The LD and BF were not affected. Plasma glucose increased ( $P < 0.01$ ) and cholesterol decreased ( $P < 0.01$ ) as SID Met + Cys increased in the diets when supplementing only DL-methionine in the diet. Homocysteine in the blood serum was not affected. In conclusion, barrows from 15 to 30 kg needs 0.661% of SID Met + Cys in the diet (7.88g/day) for a maximum ADG, being not necessary to maintain the 1:1 ratio between SID Met and SID Cys.

**Key Words:** carcass trait, homocysteine, sulfur amino acid

#### **M171 Bioavailability of L-methionine relative to DL-methionine in weaned pigs.**

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Information about the relative bioavailability (RBV) of L-methionine (LM; 100% L-isomers, 99% purity) compared with DL-methionine (DLM; a 50:50 mixture of D- and L-isomers; 99% purity) for pigs is limited. A 21-d experiment was conducted to determine the RBV of LM compared with DLM to maximize the performance of 7 to 13 kg weaned pigs. A total of 168 weaned pigs (Topigs; initial BW of 7.01 ± 0.04 kg) were assigned to 7 dietary treatments with 6 pen replicates (2 barrows and 2 gilts/pen). A corn-wheat-soybean meal-whey powder and fish meal-based basal diet (BD) was formulated using analyzed ingredient AA contents and published SID coefficients to contain 0.27% standardized ileal digestible (SID) Met and 0.54% SID Met + Cys but adequate in all other AA (1.35% SID Lys) and contained 2,498 kcal/kg NE. Dietary treatments included (1) BD, (2) BD + 0.05% DLM, (3) BD + 0.10% DLM, (4) BD + 0.15% DLM, (5) BD + 0.05% LM, (6) BD + 0.10% LM, and (7) BD + 0.15% LM. Data were analyzed by ANOVA using the GLM procedure of SAS. Orthogonal-polynomial contrasts were used to determine linear and quadratic effects of increasing levels of DLM and LM on response criteria, and the effect of Met sources. The final BW at d 21 (11.73, 12.49, 12.74, 12.68, 12.42, 12.65, and 12.82 kg for diets 1 to 7) and overall ADG (224, 258, 271, 269, 255, 268, and 272 g/d for diets 1 to 7) increased linearly ( $P < 0.05$ ) by graded additions with both Met sources. The ADFI increased linearly ( $P = 0.03$ ) by graded additions with DLM but was not affected by LM additions.

The overall FCR (1.42, 1.32, 1.29, 1.31, 1.35, 1.32, and 1.29 for diets 1 to 7) improved ( $P < 0.01$ ) linearly and quadratically by DLM additions and linearly by additions with LM. Performance responses (ADG, ADFI and FCR) of pigs fed diets supplemented with the same inclusion levels of DLM and LM were not different ( $P \geq 0.487$ ). Based on ADG as a response of Met intake, the nonlinear asymptotic model estimated the RBV of 89% (95% confidence interval: 49–128%) for LM relative to DLM. In conclusion, the bioavailability of LM relative to DLM is not different to optimize the performance of 7 to 13 kg weaned pigs.

**Key Words:** bioavailability, DL-methionine, L-methionine

**M172 Bioavailability of L-lysine sulfate compared with L-lysine·HCl to optimize performance of 12 to 25 kg pigs.** J.P.

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L-Lysine sulfate (Biolys), containing 54.6% L-Lys as well as other amino acids (AA) and phosphorus as fermentation co-products, is available as an alternative to L-lysine·HCl. A 21-d experiment was conducted to evaluate the relative bioavailability (RBV) on equimolar basis of Biolys relative to L-Lys·HCl (set at 100% availability). A total of 135 barrows (PIC; initial BW of 12.46 ± 0.55kg) were assigned to 5 treatments with 9 pen replicates (3pigs/pen) for 21 d. A Lys-deficient basal diet (BD) was formulated to contain 0.57% standardized ileal digestible (SID) Lys but adequate in all other AA. The diets included (1) BD, (2) BD + 0.128% L-Lys·HCl, (3) BD + 0.256% L-Lys·HCl, (4) BD + 0.183% Biolys, and (5) BD + 0.366% Biolys. The final BW on d 21 and the overall ADG increased linearly ( $P < 0.01$ ) by adding both Lys sources. Feed intake was not different but F:G improved ( $P < 0.01$ ) linearly by supplementing both Lys sources. Performance responses (ADG, ADFI, F:G) of pigs fed diets supplemented with the same inclusion levels of L-Lys·HCl and Biolys were not different ( $P > 0.05$ ). Using multiple linear regression and the slope ratio the RBV estimates were 104.1 and 113.3% for Biolys relative to L-Lys·HCl to optimize ADG and F:G, respectively. In conclusion, Biolys and L-Lys·HCl are equally bioavailable on equimolar basis as Lys sources for 12 to 25 kg pigs.

**Table 1 (Abstr. M172).** Performance and bioavailability (BV) of L-Lys sulfate for piglets from 12 to 25 kg<sup>1</sup>

Item	BD		L-Lys HCl		Biolys			CV (%)
	0.0%	0.128%	0.256%	Linear	0.183%	0.366%	Linear	
BW, kg <sup>2</sup>	23.58 <sup>B</sup>	24.35 <sup>AB</sup>	25.23 <sup>A</sup>	0.01	24.39 <sup>AB</sup>	25.28 <sup>A</sup>	0.03	5.1
ADG <sup>2</sup>	0.530 <sup>A</sup>	0.566 <sup>A</sup>	0.608 <sup>A</sup>	0.01	0.568 <sup>A</sup>	0.611 <sup>A</sup>	0.03	10.4
F:G <sup>2</sup>	1.921 <sup>A</sup>	1.730 <sup>B</sup>	1.685 <sup>B</sup>	0.01	1.721 <sup>B</sup>	1.647 <sup>B</sup>	0.01	7.1
BV of Biolys vs. L-Lys·HCl	Intercept	b <sup>LH</sup>	b <sup>BL</sup>	Bioavailability				
ADG	0.634	0.392	0.408	104.1				
F:G	1.938	-1.103	-1.250	113.3				

<sup>A,B</sup>Means within a row with different letters differ ( $P < 0.05$ ) by the SNK test. <sup>1</sup>Linear regression analysis; b<sup>LH</sup> = L-Lys·HCl regression coefficients; b<sup>BL</sup> = Biolys regression coefficients.

**Key Words:** bioavailability, L-Lys·HCl, Lys sulfate

**M173 Pig's biochemical plasmatic variables response to methionine ingestion in a phase-feeding program or an individual daily feeding program.** Aline Remus\*<sup>1,4</sup>, Dani Perondi<sup>1</sup>, Jaqueline P. Gobi<sup>1</sup>, Ines Andretta<sup>2</sup>, Luciano Hauschild<sup>1</sup>, Marie-Pierre L. Montminy<sup>3</sup>, and Candido Pomar<sup>4</sup>, <sup>1</sup>FCAV-UNESP, Jaboticabal, SP, Brazil, <sup>2</sup>UFRGS, Porto Alegre, RS, Brazil, <sup>3</sup>Université Laval, Quebec, QC, Canada, <sup>4</sup>AAFC, Sherbrooke, QC, Canada.

The effect of different methionine intakes and pig's individual response to this amino acid in precision feeding program is not completely known. The aim of this study was especially to evaluate the existence of methionine (met) toxicity in traditional group-phase-feeding program and in precision feeding program in which pigs are fed individually with daily tailored diets. Sixty pigs with initial BW of 25 kg (MSE = ± 2.23) were used in a 28-d trial, and were arranged in a 2 × 3 factorial randomized design testing: (1) 2 feeding programs (individual daily program (IDP) or a conventional 1 phase program by group (1PP) and (2) 3 methionine levels (70, 100, or 130% of recommended level). In the IDP pigs had the daily and individually SID lysine requirements (SIDLys) estimated using the Precision Feed model (Hauschild et al., 2012) and in the 1PP the phase SIDLys was established using the average pig of the population at day one (Rostagno et al., 2011). In both programs the amino acids profile was kept constant with exception of met. Blood samples were collected weekly at jugular vena after an 8-h fasting period and analyzed for alanine aminotransferase (ALT), aspartate aminotransferase (AST), creatinine (CK), plasmatic total protein (PTP), and protein C-reactive (CRP) levels. The variables were analyzed as a 2 × 3 factorial arrangement (confidence interval = 95%) using GLM procedure of SAS. The main effects included feeding program, methionine level, and their interaction ( $P \times L$ ). The met intake inside the IDP was 2.7, 3.7 and 6.0 g/day for the 70, 100 and 130% treatments, in the same order for the 1PP, the met intake was 4.6, 6.1 and 8.5 g/day. The PTP showed no differences among the treatments. The ALT and AST presented quadratic tendency for  $P \times L$  interaction ( $P < 0.10$ ). The CK showed a  $P \times L$  interaction with quadratic effect ( $P = 0.02$ ) and linear effect inside IDP program ( $P = 0.02$ ). As the met requirement could increase in sanitary challenge the protein C-reactive was analyzed, and concentration was not influenced ( $P > 0.05$ ) by programs or by methionine levels. Current results showed no evidence that supports toxicity of increasing the methionine until a lysine ratio up to 30% in both feeding-programs.

**Key Words:** amino acid requirement

**M174 Effects of dietary supplementation of branched-chain amino acids on growth performance, nitrogen balance, and whole-body protein turnover in piglets.** Liufeng Zheng, Hongkui Wei\*, Chuanshang Chen, Quanhang Xiang, and Jian Peng, Department of Animal Nutrition and Feed Science, College of Animal Science and Technology, Huazhong Agricultural University, Wuhan, Hubei, China.

The aim of the study was to investigate the effects of dietary BCAA supplementation on growth performance, N balance, and whole-body protein turnover in piglets from 8 to 21 kg. Twenty-eight individually caged Large White × Landrace barrows were randomly assigned to 1 of 4 dietary groups (n = 7). All 4 diets were fortified with Lys, Met, Thr and Trp to satisfy the SID AA requirements recommended by NRC (2012). Diet PC was a positive control diet (19.5% CP); diet NC was a reduced-CP negative control diet (16.7% CP); diets T1 and T2 were reduced-CP diets to which 0.17% Ile, 0.16% Val, and 0.24% Leu or 2-fold dose of each BCAA were added, respectively. Nitrogen balance was determined

over 5 subsequent days at the beginning of the 3rd week of the experiment. Whole-body protein turnover was determined by using the end product method after a single oral dose of  $^{15}\text{N}$ -glycine. All data were analyzed using the One-way ANOVA and Duncan's procedures. The results showed that NC group had decreased ADG, ADFI and absolute N retention compared with the PC group, which were restored in T1 and T2 groups ( $P < 0.05$ ). Total N excretions (g/d) were reduced ( $P < 0.05$ ) in NC, T1 and T2 groups compared with PC group. There was no difference between PC and NC groups for protein synthesis, whereas T1 and T2 groups had higher ( $P < 0.05$ ) protein synthesis than NC group. No difference in protein degradation were detected among 4 groups. There was no difference between T1 and T2 groups except for a higher N excretions in T2 group. These results indicate dietary BCAA supplementation improves growth performance and dietary N utilization for protein accretion in piglets.

**Table 1 (Abstr. M174).** Effect of dietary BCAA supplementation on growth performance in piglets

Item	PC	NC	T1	T1	SEM	P-value
ADFI, g/d	714 <sup>a</sup>	558 <sup>b</sup>	707 <sup>a</sup>	721 <sup>a</sup>	20	<0.01
ADG, g/d	422 <sup>a</sup>	322 <sup>b</sup>	449 <sup>a</sup>	452 <sup>a</sup>	15	<0.01
G:F	0.59 <sup>ab</sup>	0.58 <sup>b</sup>	0.64 <sup>a</sup>	0.63 <sup>a</sup>	0.01	0.03
N excretion, g/d	6.14 <sup>a</sup>	3.74 <sup>c</sup>	3.59 <sup>c</sup>	4.83 <sup>b</sup>	0.23	<0.01
N retention, g/d	12.95 <sup>a</sup>	8.72 <sup>b</sup>	11.48 <sup>a</sup>	12.15 <sup>a</sup>	0.45	<0.01
Protein synthesis, g of N/(kg <sup>0.75</sup> ·d)	36.29 <sup>ab</sup>	26.21 <sup>b</sup>	37.82 <sup>a</sup>	36.52 <sup>a</sup>	2.30	0.05
Protein degradation, g of N/(kg <sup>0.75</sup> ·d)	34.44	24.81	36.15	34.71	1.84	0.14

<sup>a,b</sup>Within a row, means without a common superscript letter differ ( $P < 0.05$ ).

**Key Words:** branched-chain amino acid, piglet, growth performance

**M175 Standardized ileal digestibility of amino acids in single cell protein and tuna fish meal fed to growing pigs.** A. R. Son\* and B. G. Kim, *Konkuk University, Seoul, Republic of Korea.*

The objective was to determine the apparent ileal digestibility (AID) and standardized ileal digestibility (SID) of CP and AA in 2 sources of single cell protein (SCP) and fish meal (FM) fed to growing pigs. The SCP1 (produced in Korea) contained 88.0% DM, 65.1% CP, 2.8% ether extract, and 4.2% ash; the SCP2 (produced in China) contained 90.4% DM, 75.6% CP, 1.3% ether extract, and 4.3% ash; and the FM (tuna fish meal produced in Korea) contained 85.2% DM, 54.9% CP, 8.8% ether extract, and 19.8% ash. Eight barrows (initial BW = 41.3 ± 1.2 kg) surgically fitted with a T-cannula in the distal ileum were allotted to a replicated 4 × 4 Latin square design. Three experimental diets were formulated to contain 25% SCP1, SCP2, and FM, respectively, which were the sole source of nitrogen in each experimental diet. A nitrogen-free diet was also prepared. All experimental diets contained 0.5% chromic oxide as an indigestible index. Values for the AID and SID of most indispensable AA in the SCP1 and FM were greater ( $P < 0.05$ ) than those in the SCP2. There were no differences in the AID and SID of most indispensable AA between the SCP1 and FM. In conclusion, the digestibility of most indispensable AA in the SCP1 and FM were greater than those in the SCP2.

*Contd.*

**Table 1 (Abstr. M175).** Apparent ileal digestibility (AID) and standardized ileal digestibility (SID) of AA in SCP (n = 7 and 7) and FM (n = 8)

Item	AID				SID			
	SCP1	SCP2	FM	SEM	SCP1	SCP2	FM	SEM
Arg	41.7 <sup>b</sup>	22.0 <sup>c</sup>	53.0 <sup>a</sup>	3.6	55.7 <sup>b</sup>	38.5 <sup>c</sup>	70.3 <sup>a</sup>	3.6
His	41.1 <sup>a</sup>	25.3 <sup>b</sup>	41.2 <sup>a</sup>	4.1	51.9 <sup>a</sup>	36.1 <sup>b</sup>	53.5 <sup>a</sup>	4.1
Ile	43.0 <sup>a</sup>	21.9 <sup>b</sup>	45.8 <sup>a</sup>	5.1	52.1 <sup>a</sup>	32.7 <sup>b</sup>	58.4 <sup>a</sup>	5.1
Leu	46.5 <sup>a</sup>	28.1 <sup>b</sup>	53.5 <sup>a</sup>	4.4	54.1 <sup>a</sup>	36.8 <sup>b</sup>	63.9 <sup>a</sup>	4.4
Lys	81.5 <sup>a</sup>	22.4 <sup>c</sup>	48.5 <sup>b</sup>	3.8	84.3 <sup>a</sup>	36.6 <sup>c</sup>	59.2 <sup>b</sup>	3.8
Met	63.0	63.0	53.3	7.0	66.4	66.9	59.7	7.0
Phe	40.3 <sup>a</sup>	25.0 <sup>b</sup>	46.6 <sup>a</sup>	4.4	49.9 <sup>a</sup>	35.5 <sup>b</sup>	57.8 <sup>a</sup>	4.4
Thr	30.0 <sup>a</sup>	13.7 <sup>b</sup>	28.2 <sup>ab</sup>	5.2	43.6 <sup>ab</sup>	29.6 <sup>b</sup>	48.5 <sup>a</sup>	5.2
Trp	25.1 <sup>ab</sup>	-11.7 <sup>b</sup>	30.7 <sup>a</sup>	11.2	40.7 <sup>ab</sup>	10.1 <sup>b</sup>	52.5 <sup>a</sup>	11.2
Val	34.4 <sup>a</sup>	18.2 <sup>b</sup>	39.2 <sup>a</sup>	4.7	45.5 <sup>a</sup>	31.4 <sup>b</sup>	56.2 <sup>a</sup>	4.7

**Key Words:** single cell protein, digestibility, swine

**M176 Apparent and standardized ileal digestibility of amino acids in soybean meal and canola meal in finished pigs.** Juan Edrei Sanchez\*<sup>1</sup>, Ignacio Arturo Dominguez<sup>1</sup>, Ernesto Morales<sup>1</sup>, Jose Luis Yañez<sup>2</sup>, and Miguel Cervantes<sup>3</sup>, <sup>1</sup>Universidad Autonoma del Estado de Mexico, Toluca, Estado de Mexico, Mexico, <sup>2</sup>Universidad Autonoma de Tlaxcala, Tlaxcala, Tlaxcala, Mexico, <sup>3</sup>Universidad Autonoma de Baja California, Mexicali, Baja California, Mexico.

In México the production of soybeans and canola seeds are limited; the countries exporters of these ingredients to Mexico are United States and Canada for oil extraction and animal feed. The remainder after oil extraction in these ingredients is used in swine nutrition. Digestibility studies in soybean meal and canola meal have been conducted in other countries, however, the digestibility of these ingredients in Mexico is unknown. A study was conducted to determinate apparent ileal digestibility (AID) and standardized ileal digestibility (SID) of amino acids (AA) for soybean meal and canola meal. Six Ileal-cannulated barrows (75.1 kg BW) were fed 3 diets in a replicated 3 × 3 Latin square design in finished pigs in 3 periods, which consisted at 2.8 maintenance over 3 periods consisting of a 5-d diet adaptation, 2-d collection of feces, and a 2-d collection of ileal digesta. In the total AID of AA was similar ( $P > 0.05$ ) between soybean meal and canola meal, and had 79.62 and 78.60%, respectively. With respect to lysine and threonine the soybean meal had higher ( $P < 0.05$ ) AID values in 84.7 vs 78.4 and 74.4 vs. 70.8%, respectively, than canola meal. The AID in methionine was higher ( $P < 0.05$ ) 3.88% in canola meal than soybean meal. for SID, all indispensable AA was higher ( $P < 0.05$ ) in the soybean meal than canola meal except for methionine that was higher ( $P < 0.05$ ) in canola meal (3.61%-units) than soybean meal. The digestibility of AA in the present study was lower compared with other studies. The difference may be due to the varieties of soybean meal and canola meal used for feed pigs, furthermore, The temperature used in the process of oil extraction in soybean and canola seeds is the main factor that could affect the quality of AA, decreasing the digestibility of CP and AA.

**Key Words:** soybean meal, canola meal, digestibility



**M177 Nutritional value of soybean products for growing pigs.** Tofuko A. Woyengo\*<sup>1,2</sup>, Jorge Yañez<sup>1,3</sup>, and Ruurd T. Zijlstra<sup>1</sup>, <sup>1</sup>University of Alberta, Edmonton, AB, Canada, <sup>2</sup>South Dakota State University, Brookings, SD, <sup>3</sup>Universidad Autónoma de Tlaxcala, Tlaxcala, México.

A study was conducted to determine the standardized ileal digestibility (SID) of AA and calculate NE value for 9 soybean products. Ten ileal-cannulated barrows (30 kg BW) were fed 10 diets in a 10 × 6 Youden square at 2.8 × maintenance requirement for energy. The 10 diets were 9 cornstarch-based with 1 soybean co-product as the sole source of protein, and cornstarch, sucrose, canola oil and soybean product as energy source; and an N-free diet with cornstarch, sucrose, and canola oil as energy source. The ratio of cornstarch to sucrose and canola oil in soybean product diets was identical to the N-free diet to allow calculation of energy digestibility of soybean products by difference. The SID of Lys ranged from 86 to 97% (Table 1). Apparent total-tract digestibility of GE digestibility was lower ( $P < 0.05$ ) for high-ether extract than low-ether extract soybean products. The NE value ranged from 2.3 to 4.5 Mcal/kg of DM, and did not differ among the soybean products. In conclusion, the soybean products had high SID of AA, and hence they

can serve as a good source of AA in swine diets. However, ether extract may hinder energy digestibility in the soybean products.

**Table 1 (Abstr. M177).** Lysine, ether extract (EE), and NDF content; Lys and GE digestibility; and NE value for soybean products fed to growing pigs<sup>1</sup>

Item	Soybean products								
	1	2	3	4	5	6	7	8	9
CP, % DM	51.6	58.1	65.2	60.7	71.8	55.0	50.2	40.8	38.5
Lys, % DM	3.31	3.56	4.19	3.58	4.49	3.33	3.09	2.52	2.32
EE, % DM	1.55	1.18	1.51	2.11	0.98	1.31	1.99	23.3	20.7
NDF, % DM	11.1	13.7	22.7	30.6	22.6	13.1	16.7	12.7	16.6
SID of Lys, %	89.4 <sup>bc</sup>	96.5 <sup>a</sup>	95.8 <sup>ab</sup>	90.6 <sup>abc</sup>	92.1 <sup>abc</sup>	86.2 <sup>c</sup>	95.4 <sup>ab</sup>	91.0 <sup>abc</sup>	86.7 <sup>c</sup>
ATTD <sup>2</sup> of GE, %	91.0 <sup>ab</sup>	90.1 <sup>ab</sup>	81.9 <sup>bc</sup>	90.3 <sup>ab</sup>	88.7 <sup>ab</sup>	83.9 <sup>abc</sup>	91.7 <sup>a</sup>	76.4 <sup>c</sup>	76.8 <sup>c</sup>
NE, Mcal/kg of DM	2.92	2.92	2.71	3.01	2.93	2.73	2.97	2.96	2.88

<sup>abc</sup>Means within a row without a common superscript differ ( $P < 0.01$ ).

<sup>1</sup>SEM and  $P$ -values are respectively: 2.7 and 0.02 for SID of Lys, 1.2 and 0.01 for ATTD of GE, and 0.13 and 0.58 for NE.

<sup>2</sup>ATTD = apparent total-tract digestibility.

**Key Words:** soybean, digestibility, pig

## Nonruminant Nutrition: Enzymes and minerals

**M178 Porcine in vitro degradation and fermentation characteristics of regular and low-fat corn distillers dried grains with soluble without or with carbohydrase.** Tofuko A. Woyengo<sup>\*1,2</sup>, Natasha Miceli<sup>1</sup>, and Ruurd T. Zijlstra<sup>1</sup>, <sup>1</sup>University of Alberta, Edmonton, AB, Canada, <sup>2</sup>South Dakota State University, Brookings, SD.

Low-fat corn DDGS (L-DDGS) is available for pig feeding. However, information is lacking on fermentation characteristics of L-DDGS in the pig intestine and on the effect of fiber-grading enzymes (carbohydrases) on fermentation characteristics of L-DDGS. Thus, we determined porcine in vitro digestion and fermentation characteristics of regular (R-DDGS) and L-DDGS without or with a carbohydrase that supplied 4,880 units of xylanase and 608 units of  $\beta$ -glucanase per treated feedstuff in a completely randomized design. Samples were hydrolyzed in 2 steps using pepsin and pancreatin. Subsequently, residues were incubated in a buffer solution with minerals and fresh pig feces as inoculum. Gas production was measured for 72 h, and modeled to estimate kinetics of gas production. Concentration of VFA per unit weight of residue incubated or feedstuff was measured in fermented solutions. Carbohydrase did not affect in vitro digestibility of DM (IVDDM) and total gas production. However, IVDDM for L-DDGS (61.1%) was greater ( $P < 0.05$ ) than that for R-DDGS (54.7%). Also, total gas production for L-DDGS (129.9 mL/g DM) was greater ( $P < 0.05$ ) than that for R-DDGS (119.0 mL/g DM). Total VFA production per unit weight of residue incubated for L-DDGS (2.66 mmol/g DM) was lower ( $P < 0.05$ ) than for R-DDGS (2.93 mmol/g DM). Also, total VFA production per unit weight of feedstuff for L-DDGS (1.19 mmol/g DM) was lower ( $P < 0.05$ ) than for R-DDGS (1.51 mmol/g DM). Carbohydrase and feedstuff interacted ( $P < 0.05$ ) on total VFA production per unit weight of feedstuff such that the carbohydrase tended to increase ( $P = 0.09$ ) total VFA production per unit weight of feedstuff for R-DDGS, but did not affect total VFA production per unit weight of feedstuff for R-DDGS. In conclusion, L-DDGS may be more digestible in the small intestine of pigs than the R-DDGS, whereas R-DDGS may be more fermentable in hindgut of pigs than the L-DDGS. The carbohydrase used in the present study may have limited effect on porcine small intestine digestibility and hindgut fermentation of R-DDGS and L-DDGS.

**Key Words:** corn DDGS, in vitro fermentation, pig

**M179 Nutrient digestibility of sorghum, with or without exogenous enzymes, for young broiler chickens.** C. Gallardo, J. C. Dadalt\*, J. C. C. Balieiro, and M. A. Trindade Neto, University of São Paulo, Pirassungua, SP, Brazil.

A total of 245 male Cobb broilers (10 to 18 d old) were used to determine the total-tract nutrient digestibility (TTND) of dry matter (DM), mineral matter (MM), metabolizable energy (ME), nitrogen retention (NR), calcium (Ca), phosphorus (P) and neutral detergent fiber (NDF) of sorghum, with or without multi-carbohydrase (Carb) and phytase (Phy) enzyme supplementation. The birds were allotted in a completely randomized design with 5 treatments and 7 replications (7 birds per replication). The experimental period was 8 d, 5 d of adaptation and 3 d of total feces collection. The treatments were: Control diet (CD); CD + 30% sorghum (Sorghum); CD + 30% of sorghum + 200 mg/kg of Carb (Sorghum + Carb); CD + 30% of sorghum + 50 mg/kg of Phy (Sorghum + Phy); CD + 30% of sorghum + Phy + Carb (Sorghum + Phy + Carb). The diets were formulated to meet or exceed the nutrient requirements

(Rostagno et al., 2011) for broiler chickens. The TTND coefficients of sorghum nutrients are presented in the table below. The Carb and Phy, isolated or in combination, improved ( $P < 0.05$ ) the TTND of sorghum. Positive effects of Carb and Phy supplementation in sorghum-based diets for broiler chicks are associated with reduction in the antinutritional factors from ingredient. In general, the enzymes improved the TTND of sorghum for young broiler chickens, however, when isolated, Carb and Phy were effective to improve only NR and ME.

**Table 1 (Abstr. M179).** Apparent digestibility coefficients and metabolizable energy of sorghum for broiler chickens

Item	DM (%)	NR (%)	ME (kcal·kg <sup>-1</sup> )	MM (%)	Ca (%)	P (%)	NDF (%)
Diet							
Sorghum	84.16	63.28	3,887	38.85	56.29	55.12	54.19
Sorghum + Carb	89.49	65.80	3,913	43.46	60.95	57.01	55.28
Sorghum + Phy	90.65	73.50	4,117	42.65	60.52	57.10	56.17
Sorghum + Carb + Phy	90.33	72.54	3,995	38.67	64.63	59.69	57.57
SD by Diet	1.55	2.55	60.99	2.92	3.45	2.66	4.19
P-values for contrasts <sup>1</sup>							
C1	<0.001	<0.001	<0.001	0.044	0.001	0.024	0.252
C2	0.729	0.023	0.476	0.004	0.023	0.044	0.352
C3	0.178	<0.001	<0.001	0.613	0.819	0.949	0.696

<sup>1</sup>C1 = Sorghum vs. Sorghum + Carb; Sorghum + Phy; Sorghum + Carb + Phy; C2 = Sorghum + Carb; Sorghum + Phy vs. Sorghum + Carb + Phy; C3 = Sorghum + Carb vs. Sorghum + Phy.

**Key Words:** antinutritional factor, carbohydrase, phytase

**M180 Performance of commercial market hogs supplemented with a new generation *Buttiauxella* phytase.** Janet C. Remus<sup>\*1</sup>, Yueming Dersjant-Li<sup>2</sup>, Peter Plumstead<sup>3</sup>, and Ajay Awati<sup>2</sup>, <sup>1</sup>Danisco Animal Nutrition/DuPont Industrial Bioscience, St. Louis, MO, <sup>2</sup>Danisco Animal Nutrition/DuPont Industrial Bioscience, Marlborough, Wiltshire, UK, <sup>3</sup>University of Pretoria, Pretoria, South Africa.

This experiment was conducted to determine if a new generation phytase from *Buttiauxella* sp. can replace all inorganic phosphate (PO<sub>4</sub>) in diets also reduced in Ca and ME and fed to commercial pigs from 12 kg to slaughter while maintaining performance and carcass quality. Four treatments were tested in a randomized block design with 9 replicate pens, each with 31 mixed sex pigs of Newsholm Choice genetics. The corn-soybean meal-wheat middlings-based treatment diets are (1) positive control (PC) that met nutrient requirements; (2) negative control (NC) without inorganic PO<sub>4</sub> and with incremental phase nutrient reductions of 0.131% Ca and 35 kcal ME/kg; (3) NC+500 FTU/kg phytase (NC+500) and (4) NC+1000 FTU/kg phytase (NC+1000). The mash diets were fed ad lib in 5 feed phases. Data were tested using the Fit Model procedure in JMP, mean separation utilized Tukey's test. Significance was  $P \leq 0.05$  unless noted. Due to poor growth at the end of Finisher 1, the NC was placed on PC feed in Finisher 2 (F2) to see if they could get to target weight. Performance from Nursery 2 (N2) thru F2 shows the PC and NC+1000 have similar ADG and improved results versus NC. The

NC+500 had better ADG and FCR than the NC and had similar FCR with PC and NC+1000 in N2 thru F2. From N2 to processing, the PC and both phytase treatments were similar on performance (no NC due to insufficient pens at target weight by F2 end). No differences noted for carcass weight, lean carcass or yield. Carcass data for PC, NC+500, and NC+1000 shows that the NC+500 had higher 10th rib fat but had a tendency ( $P = 0.07$ ) toward lower yield while NC+1000 had lower muscle depth. Overall, the data indicate that 500 and 1000 FTU phytase/kg can replace all inorganic  $PO_4$  and maintain performance compared with PC in pigs fed a corn-soybean meal-wheat middlings based diet.

**Table 1 (Abstr. M180).** Performance and carcass data

Treatment	N2 to F2 (12-120 kg)			Carcass	
	ADG, kg	ADFI, kg	FCRmc	Muscle depth, mm	10th rib backfat, mm
PC	0.78 <sup>ab</sup>	2.17 <sup>a</sup>	2.80 <sup>ab</sup>	63.78 <sup>a</sup>	18.94 <sup>b</sup>
NC	0.69 <sup>c</sup>	1.98 <sup>b</sup>	2.86 <sup>a</sup>		
NC+500	0.75 <sup>b</sup>	2.09 <sup>ab</sup>	2.77 <sup>b</sup>	63.91 <sup>a</sup>	19.72 <sup>a</sup>
NC+1000	0.79 <sup>a</sup>	2.20 <sup>a</sup>	2.79 <sup>ab</sup>	62.63 <sup>b</sup>	18.99 <sup>b</sup>
P-value	<0.01	<0.01	0.05	0.02	0.05

**Key Words:** *Buttiauxella* phytase, pig, performance

**M181 Ileal amino acid digestibility in high protein sunflower meal and pea protein isolate fed to growing pigs with or without multi-carbohydase supplementation.** J. C. Dadalt<sup>\*1</sup>, D. E. Velayudhan<sup>1</sup>, M. A. Trindade Neto<sup>2</sup>, and C. M. Nyachoti<sup>1</sup>, <sup>1</sup>University of Manitoba, Winnipeg, MB, Canada, <sup>2</sup>University of São Paulo, São Paulo, SP, Brazil.

Eight ileal-cannulated barrows (initial BW = 23.5 ± 0.85 kg) were used to determine the apparent (AID) and standardized (SID) ileal AA digestibilities in high protein sunflower meal (HiPSF) and pea protein isolate (PPI) with or without multi-carbohydase enzyme (MC) supplementation. Pigs were randomly assigned to 1 of 5 treatments in a replicated 4 × 5 incomplete Latin square design to give 8 observations per treatment. The experimental diets consisted of HiPSF or PPI as the sole source of protein with or without MC and a low-protein diet (5% casein) used to quantify endogenous AA losses. All diets contained titanium dioxide (0.3%) as indigestible marker. Pigs were given their daily feed allowance at a rate of 4.5% of BW determined at the beginning of each experimental period. Each experimental period lasted 7 d and the ileal digesta were collected on d 6 and 7. All data were analyzed using the MIXED procedure of SAS (SAS Inst. Inc., Cary, NC). The model included diet as the fixed variable and animal and period as the random variables. In general, AA digestibilities were higher in PPI than in HiPSF, with the exception of Met and Cys ( $P > 0.05$ ). There was no effect of MC ( $P > 0.05$ ) on AA digestibility. The AID and SID of essential AA in HiPSF and PPI (without MC) were, respectively: Arg, 83.2, 87.8 and 90.0, 94.2; His, 29.6, 40.3 and 50.0, 60.5; Ile, 73.9, 79.8 and 83.1, 88.6; Leu, 72.9, 78.0 and 83.4, 87.7; Lys, 66.1, 72.6 and 88.3, 91.4; Met, 82.1, 86.0 and 80.8, 87.3; Phe, 75.5, 79.0 and 83.0, 86.1; Thr, 62.3, 72.9 and 72.6, 82.6; Val, 70.1, 76.1 and 79.2, 85.0. The MC increased ( $P < 0.01$ ) the AID and SID of Proline (75.37 vs 82.14% and 86.82 vs 93.66%, respectively) in PPI. Compared with HiPSF, PPI had better digestible AA profile for growing pigs. However, no differences were detected for the digestibility of most AA when diets were supplemented with MC.

**Key Words:** enzyme, high-protein sunflower meal digestibility, pea protein isolate

**M182 Ileal amino acid digestibility in rice polished and broken fed to weaned pigs with or without multi-carbohydase and phytase supplementation.** J. C. Dadalt<sup>\*1</sup>, G. V. Polycarpo<sup>1</sup>, C. Gallardo<sup>1</sup>, T. W. Almeida<sup>1</sup>, J. C. S. M. Souza<sup>1</sup>, F. E. L. Budiño<sup>2</sup>, and M. A. Trindade Neto<sup>1</sup>, <sup>1</sup>University of São Paulo, São Paulo, SP, Brazil, <sup>2</sup>Institute of Animal Science, Nova Odessa, SP, Brazil.

Twenty-five barrows (initial BW = 8.81 kg ± 1.52 kg) were used to determine the apparent (AID) and standardized (SID) ileal AA digestibilities in rice polished and broken (RB) with or without multi-carbohydase (Carb) and phytase (Phy) enzyme supplementation. Pigs were allotted in a completely randomized design to 5 treatments with 5 replicates. The experimental diets consisted of RB as the sole source of protein without enzymes, with Carb, Phy or Carb+Phy and a low-protein diet (5% casein) used to quantify endogenous AA losses. All diets contained chromic oxide (0.3%) as indigestible marker. Pigs were given their daily feed allowance at a rate of 4.5% of BW determined at the beginning of each experimental period. Animals were slaughtered at d 43 and ileal contents collected. The data were analyzed using the GLM (SAS Inst. Inc., Cary, NC), and means were compared by Tukey's test. The phytase, singly or in combination with multi-carbohydase, improved AID and SID ( $P < 0.05$ ) of histidine, leucine and lysine of polished and broken rice, while singly carbohydase improved only AID and SID ( $P < 0.05$ ) of histidine. Coefficients of AID and SID of AA in RB (without enzymes) were, respectively: Arg, 0.662 and 0.841; His, 0.626 and 0.769; Ile, 0.596 and 0.792; Leu, 0.700 and 0.790; Lys, 0.663 and 0.775; Met, 0.710 and 0.808; Phe, 0.721 and 0.843; Thr, 0.492 and 0.650; Val, 0.630 and 0.811; Ala, 0.509 and 0.613; Asp, 0.618 and 0.735; Glu, 0.667 and 0.778; Pro, 0.623 and 0.853; Ser, 0.404 and 0.716; Tyr, 0.744 and 0.865. In general, these results were lower than those in NRC (2012). Underestimated digestibility coefficients may be related to animal ages in the present study, since most of the studies in literature were obtained from pigs with higher weights. The phytase, singly or combined with carbohydase, improves the digestibility of His, Leu and Lys in rice polished and broken.

**Key Words:** alternative ingredient, enzyme, piglet

**M183 Ileal amino acid digestibility in micronized full fat soybean fed to weaned pigs with or without multi-carbohydase and phytase supplementation.** J. C. Dadalt<sup>\*1</sup>, C. Gallardo<sup>1</sup>, T. W. Almeida<sup>1</sup>, P. A. P. Ribeiro<sup>1</sup>, G. V. Polycarpo<sup>1</sup>, G. Galvão<sup>1</sup>, B. A. Malheiros<sup>1</sup>, F. E. L. Budiño<sup>2</sup>, and M. A. Trindade Neto<sup>1</sup>, <sup>1</sup>University of São Paulo, São Paulo, SP, Brazil, <sup>2</sup>Institute of Animal Science, Nova Odessa, SP, Brazil.

Twenty-five barrows (initial BW = 8.53 kg ± 1.48 kg) were used to determine the apparent (AID) and standardized (SID) ileal AA digestibilities in micronized full fat soybean (MFFS) with or without multi-carbohydase (Carb) and phytase (Phy) enzyme supplementation. Pigs were allotted in a completely randomized design under 5 treatments and 5 replicates. The experimental diets consisted of MFFS as the sole source of protein without enzymes, with Carb, Phy or Carb+Phy. A low-protein diet (5% casein) was used only to calculate the digestibility of the MFFS. All diets contained Chromium (0.3%) as indigestible marker. Pigs were given their daily feed allowance at a rate of 4.5% of BW determined at the beginning of each experimental period. Animals were slaughtered at d 43 and ileal contents collected. The data were analyzed using the GLM (SAS Inst. Inc., Cary, NC), and means were compared by Tukey's test. The AID and SID of AA in MFFS (without enzymes) were lower (average 16.9 and 14.7%, respectively) compared with NRC (2012). Underestimated digestibility coefficients may be related with animal ages in the present study, since most of the studies



related in literature were obtained from pigs with higher weights. There was no effect of Carb, Phy or Carb+Phy ( $P > 0.05$ ) on AA digestibility. According with literature, sometimes the use of exogenous enzymes to degrade indigestible dietary components has inconsistent results mainly because of the presence of complex substrates in feedstuffs and the use of enzyme activities often not suitable for effective hydrolysis of such components. The AID and SID of AA in MFFS (without enzymes) were, respectively: Arg, 71.6 and 76.5; His, 69.8 and 77.0; Ile, 59.9 and 66.0; Leu, 68.1 and 74.2; Lys, 75.8 and 79.6; Met, 72.3 and 81.1; Phe, 68.8 and 74.2; Thr, 61.4 and 74.6; Val, 65.2 and 72.1; Ala, 66.7 and 78.0; Asp, 66.9 and 69.8; Glu, 71.1 and 75.0; Pro, 70.1 and 80.7; Ser, 70.0 and 81.5; Tyr, 68.9 and 74.0. Carbohydrase and phytase, alone or in combination, don't affect AA digestibilities of MFFS for weaned pigs.

**Key Words:** antinutritional factor, enzyme, piglet

**M184 Nutrient digestibility of rice bran, with or without exogenous enzymes, for young broiler chickens.** C. Gallardo, J. C. Dadalt\*, J. C. C. Balieiro, and M. A. Trindade Neto, *University of São Paulo, Pirassununga, SP, Brazil.*

A total of 245 male Cobb broilers (10 to 18 d old) were used to determine the total-tract nutrient digestibility (TTND) of dry matter (DM), mineral matter (MM), metabolizable energy (ME), nitrogen retention (NR), calcium (Ca), phosphorus (P) and neutral detergent fiber (NDF) of rice bran (RB), with or without multi-carbohydrase (Carb) and phytase (Phy) enzyme supplementation. The birds were allotted in a completely randomized design with 5 treatments and 7 replications (7 birds per replication). The experimental period was 8 d, 5 d of adaptation and 3 d of total feces collection. The treatments were: Control diet (CD); CD + 30% of RB; CD + 30% of RB + 200 mg/kg of Carb (RB+Carb); CD + 30% of RB + 50 mg/kg of Phy (RB+Phy); CD + 30% of RB + Carb + Phy (RB+Carb+Phy). CD was used only to calculate the digestibility of the RB. The diets were formulated to meet or exceed the nutrient requirements (Rostagno et al., 2011) for broiler chickens. The TTND coefficients of RB are presented in Table 1. Alone or combined, Carb and Phy improved ( $P < 0.05$ ) the TTND of RB. These results are associated with enzymatic activity to enhance nutrient digestibility, focusing primarily on removing the antinutritive effects of nonstarch polysaccharides (NSP), such as arabinoxylans and  $\beta$ -glucans from broiler diets based on viscous grains. In general, enzymes improved the TTND of RB for young broiler chickens.

**Table 1 (Abstr. M184).** Apparent digestibility coefficients and metabolizable energy of rice bran for broiler chickens

Item	DM (%)	MM (%)	ME (kcal·kg <sup>-1</sup> )	NR (%)	Ca (%)	P (%)	NDF (%)
Diet							
RB	66.42	75.38	3765	62.04	49.31	69.60	68.09
RB + Carb	69.82	80.59	3916	66.42	56.64	74.11	74.80
RB + Phy	70.36	75.85	3999	70.25	58.34	71.11	74.59
RB + Carb + Phy	75.07	78.14	4252	70.07	57.76	74.88	68.61
SD by Diet	1.74	2.25	53.52	2.28	3.94	1.88	2.76
P-values for contrasts <sup>1</sup>							
C1	<0.001	0.009	<0.001	<0.001	<0.001	<0.001	<0.001
C2	<0.001	0.939	<0.001	0.117	0.886	0.016	<0.001
C3	0.582	<0.001	0.007	0.005	0.432	0.007	0.892

<sup>1</sup>C1 = RB vs. RB + Carb; RB + Phy; RB + Carb + Phy; C2 = RB + Carb; RB + Phy vs. RB + Carb + Phy; C3 = RB + Carb vs. RB + Phy.

**Key Words:** antinutritional factor, carbohydrase, phytase

**M185 Effects of phosphorus source and content on bone mineralization and performance of broiler chickens.** Roseline Kahindi\*<sup>1</sup>, Phil Thacker<sup>2</sup>, and Martin Nyachoti<sup>1</sup>, <sup>1</sup>*University of Manitoba,* <sup>2</sup>*University of Saskatchewan.*

Two hundred and ten 1-d-old, (Ross 308 strain) broiler chicks were housed in a temperature controlled room with 5 birds per cage and randomly allocated to 1 of 7 dietary treatments for a 21 d study. The objective of the study was to determine the effect of feeding diets containing low phytate barley and pea vs. the normal phytate cultivars and the effects of inorganic P reduction on performance and bone characteristics of broiler chicks. The main feed ingredients used were: low phytate barley (LPB), low phytate pea (LPP), normal hull barley (NHB), normal phytate pea (NPP), and soybean meal (SBM). The dietary treatments included (1) Control that was SBM-NHB based with 100% inorganic P; (2) control with 50% inorganic P; (3) low phytate cultivars based on SBM-LPB-LPP with 100% inorganic P; (4) treatment 3 with 50% inorganic P; (5) treatment 3 with no addition of inorganic P; (6) normal phytate cultivars based on SBM-NHB-NPP with 100% inorganic P; (7) normal phytate cultivars with 50% inorganic P. Feed and water were offered ad-libitum throughout the study and average daily gain (ADG) and feed intake (ADFI) were determined weekly and used for calculating feed conversion ratio (FCR). On d 21, 2 birds per cage were killed to collect left tibias for bone length and ash determination. Data were subjected to ANOVA as a completely randomized design using Proc Mixed of SAS 9.2. In addition, orthogonal contrasts were used to compare diets 1 and 2 vs. the rest of the diets, to compare 100 vs. 50% inorganic P diets, and to compare diets with low phytate vs. the normal phytate grain cultivars. The reduction in dietary inorganic P content significantly decreased ADG and ADFI but had no effect on FCR. The ADG was 18 g/chick and lowest ( $P < 0.05$ ) for treatment 7, whereas ADG was similar for treatments 1, 3, and 6 that is, 38, 42, and 41 g/chick, respectively. The overall ADG tended ( $P < 0.10$ ) to be higher for the low phytate compared with normal phytate diets. The tibia ash and length were significantly decreased with inorganic P reduction. The results indicate that feeding low phytate barley and pea based diets can replace 29% of supplemental inorganic P required in normal phytate cultivars.

**Key Words:** broiler chicken, low phytate cultivar, performance

**M186 A comparison of total, apparent total-tract digestible, and standardized total-tract digestible phosphorus on excretion and digestibility of phosphorus in pigs.** W. B. Kwon\*<sup>1</sup>, S. K. Park<sup>2</sup>, and B. G. Kim<sup>1</sup>, <sup>1</sup>*Konkuk University, Seoul, Republic of Korea,* <sup>2</sup>*Sejong University, Seoul, Republic of Korea.*

An experiment was conducted to test a hypothesis that 3 diet formulation methods of using total, apparent total-tract digestible (ATTD), and standardized total-tract digestible (STTD) phosphorus (P) differ in excretion and digestibility of P in pigs. Twenty-four barrows with an initial BW of 18.1  $\pm$  0.7 kg were randomly allotted to 3 dietary treatments with 8 replicate pigs per diet in a completely randomized design. Experimental diets were formulated to contain constant amounts of corn, soybean meal, whey powder, and dicalcium phosphate. The 3 diets were formulated based on a total, ATTD, or STTD basis to meet the dietary P requirement by changing the inclusion rate of rice bran at the expense of cornstarch and cellulose. Each experimental period consisted of a 4-d adaptation period and a 5-d fecal collection period. During the collection period, total feed intake did not differ among the treatments. The daily P intake and daily P output of pigs fed the diet based on ATTD P was greater (10.87 vs. 7.65 or 8.96 g/d and 4.27 vs. 2.60 or 3.30 g/d, respectively;  $P < 0.05$ ) compared with the total P- or STTD P-based group. The ATTD of P was not different among the treatment groups.

Taken together, formulating a diet based on total P or STTD P resulted in less P excretion mainly due to the less concentration of dietary P compared with a diet based on ATTD P. Further research is warranted to investigate performance and P retention in pigs fed diets formulated based on total P, ATTD P, and STTD P.

**Key Words:** diet formulation, phosphorus, swine

**M187 Standardized total tract digestibility of phosphorus in various inorganic phosphates fed to growing pigs.** W. B. Kwon\* and B. G. Kim, *Konkuk University, Seoul, Republic of Korea.*

An experiment was conducted to determine the standardized total-tract digestibility (STTD) of phosphorus (P) in 5 sources of inorganic phosphate fed to pigs. The 5 sources of inorganic phosphate were dicalcium phosphate (DCP, 19.3% P), monocalcium phosphate (MCP, 22.7% P), monodicalcium phosphate (MDCP, 22.0% P) which is the mixture of MCP and DCP at a ratio of 3:1, monosodium phosphate (MSP, 24.5% P), and tricalcium phosphate (TCP, 19.4% P). Six barrows with an initial body weight of 42.4 ± 1.1 kg were individually housed in metabolism cages equipped with a feeder. Pigs were randomly allotted to a 6 × 6 Latin square design with 6 dietary treatments and 6 periods. Each experimental period consisted of a 4-d adaptation and a 5-d collection period. Five experimental diets contained 0.25% P from each inorganic phosphate as a sole source of P, and were formulated to contain calcium in amounts that were 1.3 times greater than the concentration of P except the TCP diet. A P-free diet mainly based on cornstarch, gelatin, and sucrose was also prepared to estimate the basal endogenous loss of P. Feces were collected using the marker-to-marker procedure. Data were analyzed using the MIXED procedure of SAS with a model including dietary treatment as a fixed variable and animal and period as random variables. Values for the apparent total-tract digestibility of P in DCP, MCP, MDCP, and MSP (83.0, 86.5, 81.4, and 88.7% respectively) were greater ( $P < 0.05$ ) than in TCP (67.0%). Values for the STTD of P in DCP, MCP, MDCP, and MSP (89.8, 93.3, 88.2, and 95.5%) were also greater ( $P < 0.05$ ) than in TCP (72.8%). In conclusion, digestibility of P in TCP was less than other inorganic P sources.

**Key Words:** inorganic phosphate, phosphorus digestibility, swine

**M188 Effect of phosphorus and calcium depletion-repletion sequences on femoral mechanical properties in growing pigs.**

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Consequences of a P-Ca deficient diet (depletion) or a control diet (repletion) on the mechanical properties of growing pig femurs were studied. Two different groups of pigs were fed with different P-Ca depletion-repletion sequences, that involved a control diet (C) or a deficient diet (L), supplying 100% or 60% of P-Ca requirements, respectively, in an incomplete factorial design. The first group (n = 30; initial BW 24 ± 3.3 kg) received during 2 28-d phases CC, CL or LC dietary treatments. The second group (n = 50; initial BW 24 ± 2.8 kg) received during three 28-d phases CCC, CLC, LCC, LLC or LLL diets. At the end of the 2nd and 3rd phases, respectively, pigs were killed and right femurs collected for computer tomography (CT) scans and mechanical tests. Data were analyzed using Proc Mixed procedures and orthogonal contrasts for treatments interferences. A 28-d repletion period in the 2nd phase (CC, LC) increased (43%,  $P = 0.03$ ) bone-bending moment (BM, kg-mm)

compared with depleted pigs (CL). Repletion P-Ca diets in the 3rd phase (CCC, CLC, LCC and LLC) increased BM ( $P < 0.01$ ) and strain ( $P = 0.03$ ) compared with LLL pigs. Moment of inertia (MI, mm<sup>4</sup>) tended to be lower ( $P = 0.08$ ) in depleted animals (LLL) compared with repleted animals, resulting in a reduced ( $P = 0.02$ ) bone stress (kg/mm<sup>2</sup>) in LLL pigs. However, neither bone MI nor stress differed among treatments. P-Ca depletion at the end of the 2nd or 3rd phase (CL, LLL) reduced bone mechanical properties. No differences were found between CC and LC treatments regarding mechanical properties, which implies bone recovery from P-Ca deficiencies of the 1st phase. Mechanical properties after three 28-d growing phases (final BW 107 ± 11.3 kg) are shown in the table. Superscripts indicate treatment differences ( $P < 0.05$ ). A 28-d repletion in the 3rd phase (CCC, CLC, LCC or LLC) is sufficient for recovery from previous deficiencies as similar mechanical properties were detected among treatments.

**Table 1 (Abstr. M188).**

Item	Trait					SEM
	CCC	CLC	LCC	LLC	LLL	
BM (kg·mm)	3049 <sup>A</sup>	2501 <sup>A</sup>	2527 <sup>A</sup>	2354 <sup>AB</sup>	1726 <sup>B</sup>	259
Strain	0.026 <sup>AB</sup>	0.027 <sup>AB</sup>	0.030 <sup>A</sup>	0.025 <sup>AB</sup>	0.023 <sup>B</sup>	0.002
Stress (kg/mm <sup>2</sup> )	1.346	1.129	1.170	1.105	0.886	0.123

**Key Words:** phosphorus-calcium, depletion-repletion, mechanical properties

**M189 Effects of dietary calcium concentrations on growth performance, litter quality, and tibia characteristics in starter broiler chickens.**

Jong Hyuk Kim\*<sup>1</sup>, Byung Bo Lee<sup>1</sup>, Moon Chan Kim<sup>1</sup>, Jae Sang Um<sup>2</sup>, and Dong Yong Kil<sup>1</sup>, <sup>1</sup>Chung-Ang University, Anseong-si, Gyeonggi-do, Republic of Korea, <sup>2</sup>Nonghyup Feed, Gangdong-gu, Seoul, Republic of Korea.

The objective of this experiment was to investigate the effects of dietary calcium (Ca) concentrations on growth performance, litter quality, and tibia characteristics in starter broiler chickens. A total of 2,520 one-day-old broiler chicks were used and housed in floor pens for 3 weeks. Birds were randomly allotted to 1 of 6 dietary treatments with 6 replicates. Each replicate consisted of 70 chicks. Dietary Ca concentrations were set to 1.0, 0.9, 0.8, 0.7, 0.6, and 0.5%. Non-phytate P (NPP) was maintained at 0.35% and 0.02% of a 5,000 FTU/g phytase (Phyzyme XP, Danisco Animal Nutrition, Marlborough, UK) was included in all diets. At the end of experiment, 2 birds from each replicate were euthanized and tibia samples were collected. Litter samples were also collected from each pen floor. Pre-planned orthogonal polynomial contrast test was performed to determine linear and quadratic effects of Ca concentrations. Results indicated that decreasing concentrations of Ca in diets from 1.0 to 0.5% increased (linear,  $P < 0.05$ ) BW, BWG, FI, and feed efficiency. Litter quality including litter pH, litter moisture, and litter nitrogen and tibia characteristics including breaking strength were not affected by dietary Ca concentrations. In conclusion, decreasing concentrations of Ca from 1.0 to 0.5% in diets containing 0.35% NPP and phytase (1,000 FTU/kg) have positive effects on growth performance of starter broiler chickens, but little effects on litter quality and tibia characteristics.

**Key Words:** starter broiler chicken, dietary calcium, performance

**M190 Effects of dietary calcium concentrations on growth performance, litter quality, and tibia characteristics in growing broiler chickens.** Jong Hyuk Kim<sup>1</sup>, Byung Bo Lee\*<sup>1</sup>, Moon Chan Kim<sup>1</sup>, Jae Sang Um<sup>2</sup>, and Dong Yong Kil<sup>1</sup>, <sup>1</sup>*Chung-Ang University, Anseong-si, Gyeonggi-do, Republic of Korea*, <sup>2</sup>*Nonghyup Feed, Gangdong-gu, Seoul, Republic of Korea*.

This study was conducted to investigate the effects of dietary calcium (Ca) concentrations on growth performance, litter quality, and tibia characteristics in growing broiler chickens. A total of 1,800 21-d-old broiler chickens were used and housed in floor pens for 2 weeks. Birds were randomly allotted to 1 of 6 dietary treatments with 6 replicates. Each replicate consisted of 50 chickens. Dietary Ca concentrations were set to 0.9, 0.8, 0.7, 0.6, 0.5, and 0.4%. Non-phytate P (NPP) was maintained at 0.3% and 0.02% of a 5,000 FTU/g phytase (Phyzyme XP, Danisco Animal Nutrition, Marlborough, UK) in all diets. After 2 weeks, 2 birds from each replicate were euthanized and tibia samples were collected. Litter samples were also collected from each pen floor. Pre-planned orthogonal polynomial contrast test was performed to determine linear and quadratic effects of dietary Ca concentrations. Results indicated that growth performance including BWG, feed intake (FI), and feed efficiency (FE) was not affected by dietary Ca concentrations. Litter quality including litter pH, litter moisture, and litter nitrogen was not influenced by dietary Ca concentrations. However, decreasing concentrations of Ca in diets from 0.9 to 0.4% decreased (linear and quadratic,  $P < 0.01$ ) the concentrations of Ca and P in tibia. In conclusion, decreasing concentrations of Ca from 0.9 to 0.4% in diets containing 0.3% NPP and phytase (1,000 FTU/kg) decrease the concentrations of Ca and P in tibia, but no adverse effects on growth performance and litter quality in growing broiler chickens.

**Key Words:** growing broiler chicken, dietary calcium, performance

**M191 Determination of additivity of apparent and standardized total tract digestibility of phosphorus in mixed diet fed to growing pigs.** W. B. Kwon\*<sup>1</sup>, S. K. Park<sup>2</sup>, and B. G. Kim<sup>1</sup>, <sup>1</sup>*Konkuk University, Seoul, Republic of Korea*, <sup>2</sup>*Sejong University, Seoul, Republic of Korea*.

This study was conducted to test the hypothesis that the values of standardized total-tract digestibility (STTD) of phosphorus (P) measured in individual feed ingredients are additive when used in diet formulation for swine. Eight crossbred barrows with an initial BW of  $30.5 \pm 1.5$  kg were individually housed in metabolism cages equipped with a feeder. The pigs were randomly allotted to a replicated  $4 \times 4$  Latin square design with 4 dietary treatments and 4 periods. Three diets were formulated by mixing cornstarch, sucrose, and soybean oil with SBM (50%), wheat (70%), or both (30.6% of SBM and 55.7% of wheat), as the source of P in each diet. A P-free diet was also formulated to estimate the basal endogenous loss of P. All experimental diets were formulated to maintain constant total calcium to total P ratio of 1.2:1 except the P-free diet. Each experimental period consisted of a 4-d adaptation period and a 5-d collection period. Feces were collected using the marker-to-marker procedure. In the mixed diet, a relative contribution of P from SBM and wheat was 51.9 and 48.1%, respectively. The ATTD of P for SBM and wheat was measured to be 40.9 and 41.8%, respectively, and the measured ATTD of P in the mixed diet was greater (45.1 vs. 41.3%;  $P < 0.05$ ) than the predicted ATTD of P. The determined STTD of P for SBM and wheat was 46.4 and 49.3%, respectively, and the measured STTD of P in the mixed diet was not different (49.7 vs. 47.8%;  $P = 0.26$ ) from the predicted STTD of P. In conclusion, the ATTD values

of P are not additive whereas STTD values of P may be additive when formulating swine diets.

**Key Words:** additivity, phosphorus digestibility, swine

**M192 Effect of dietary calcium and phosphorus concentration on apparent ileal and total-tract digestibility of calcium and phosphorus of pigs.** Fengrui Zhang\*<sup>1</sup>, Darryl Ragland<sup>2</sup>, and Layi Adeola<sup>1</sup>, <sup>1</sup>*Department of Animal Science, Purdue University, West Lafayette, IN*, <sup>2</sup>*Department of Veterinary Clinical Sciences, Purdue University, West Lafayette, IN*.

The objective of this study was to test the hypothesis that the apparent total-tract digestibility (ATTD) of calcium (Ca) is not different from the apparent ileal digestibility (AID) in growing-finishing pigs fed different Ca and Phosphorus (P) concentration diets with a fixed Ca:total P (tP) ratio. Diets with 4 Ca concentrations (0.39%, 0.59%, 0.79% and 0.99%) were formulated by adjusting the dietary level of calcium carbonate. Potassium phosphate was supplemented to adjust the dietary P level and maintain a 1.5:1 Ca:tP ratio. Sixteen pigs (initial BW =  $73 \pm 4$  kg), fitted with a T-cannula at the distal ileum, were sorted by BW and assigned in a randomized complete block design to 4 treatments and 2 experimental periods. Each period consisted of a 3-d acclimation period, a 2-d fecal collection, and a 2-d ileal collection period. Data from the study was analyzed as a split-plot in a randomized complete block design using the GLM procedure of SAS (SAS Inst. Inc., Cary, NC). Diets with different Ca and P levels were the main-plot factor, and digestibility sites (AID and ATTD) were the split-plot factor. Results indicated that the ATTD of Ca or P is not different from AID in all treatments. Furthermore, the results also showed the digestibility of Ca was not affected by the dietary treatments, but the apparent digestibility of Ca had a tendency to increase as the dietary Ca concentration increased (linear,  $P = 0.095$ ). The AID and ATTD of P linearly increased ( $P < 0.05$ ) from 34.45 to 63.07% and 35.96 to 62.35% respectively, as the Ca and P concentration increased. Therefore, current study showed that with the same dietary Ca:tP ratio, there is no difference between the AID and ATTD of Ca and P. Thus, the net absorption of Ca and P in large intestine of growing-finishing pigs is negligible and both AID and ATTD can be used to describe the digestibility of Ca for growing-finishing pigs. It is also concluded that the effects of adding different levels of calcium carbonate and potassium phosphate on digestibility of Ca is not statistically significant, but it increased the apparent ileal and total-tract digestibility of P in the diets based on Corn-SBM.

**Key Words:** calcium, digestibility, phosphorus

**M193 Influence of calcium source and fat level on broiler performance and nutrient utilization.** O. El-Husseiny<sup>1</sup> and M. A. Michael\*<sup>1,2</sup>, <sup>1</sup>*Department of Animal Sciences, Cairo University, Egypt*, <sup>2</sup>*Affor. and Environment, Ministry of Agriculture, Egypt*.

The objective of this experiment was to evaluate the effect of the dietary inclusion of calcium lactate (CL), calcium bio lactate (CBL) or calcium carbonate (CC) at NRC calcium level, in the presence of 0, 3%, 6% or 9% beef tallow on improving the performance, intestinal digestion and absorption of broiler chicks. Where, CBL derived from reacted between calcium hydroxide and biologically lactic acid secreted from lactic acid bacteria based on probiotic. Three hundred and six 1-d-old Cobb 500 broiler chicks were fed 12 treatments containing 3 calcium sources and 4 fat levels in a  $3 \times 4$  factorial design with 3 replicates, 10 chicks each. Weight gain (WG), feed intake (FI), feed conversion rate (FCR), blood composition, nutrient digestibility and soap formation



were recorded up to 7 weeks of age. The weight gain of broilers fed diets containing CBL with 6% tallow fortified diet was higher than those fed CL plus 6% tallow. Improvement ( $P < 0.05$ ) in FCR was observed when broilers fed diets contained CL or CBL compared with those fed diets contained CC with different fat levels. Abdominal fat was increased ( $P < 0.05$ ) when broilers fed diets containing CBL with 9% fat than those fed the other calcium sources and fat levels. The digestibilities of all nutrients were higher ( $P < 0.05$ ) with broilers fed diets contained CBL or CL plus 6% tallow than those fed diets contained CC with different fat levels. The highest amount of excreted fat as soap formation were noticed for broilers fed diets contained CC with 9% tallow, while the lowest values observed with those fed diets contained CBL with 6% or 9% tallow. Total blood lipid of broilers fed diets contained CL or CBL with 6 or 9% tallow were greater ( $P < 0.05$ ) compared with those fed the other treatments. Broilers fed diets contained CBL recorded the highest nitrogen and calcium retained compared with those fed diets contained CL or CC. It is concluded that the calcium lactate or calcium bio lactate based on probiotic were effective improving fat absorption and calcium retention with positive effect on broiler performance.

**Key Words:** calcium bio lactate, beef tallow, soap formation

**M194 Dietary nitrogen level affects ileal phosphorus digestion in growing pigs.** Pengcheng Xue<sup>\*1</sup>, Darryl Ragland<sup>2</sup>, Kolapo Ajuwon<sup>1</sup>, and Olayiwola Adeola<sup>1</sup>, <sup>1</sup>Departments of Animal Sciences, Purdue University, West Lafayette, IN, <sup>2</sup>Departments of Veterinary Clinical Sciences, West Lafayette, IN.

A study was conducted to investigate the effect of dietary CP levels on ileal phosphorus (P) digestion in growing pigs. A total of 18 ileal-cannulated pigs (initial BW  $44.2 \pm 3.2$  kg) were used in a duplicated  $9 \times 3$  incomplete Latin Square design, with 9 treatments and 3 7-d experimental periods giving 6 replicates per treatment. The 9 treatments consisted of 1 nitrogen free diet to estimate basal endogenous loss of AA, and 8 corn-soybean meal based diets in a  $2 \times 4$  factorial arrangement, which included 2 CP levels (6.9 or 13.4%) and 4 apparent total-tract digestible P (ATTDP) levels (0.09, 0.16, 0.24, or 0.32%). Soybean meal and mono-calcium phosphate were used to adjust the CP level and ATTDP level, respectively. Limestone was included in diets to maintain the Ca: ATTDP ratio across treatments. Ileal digesta was collected continuously for 10 h on the last 2 d of each 7-d experimental period. The ileal digesta samples were lyophilized and analyzed to calculate ileal digested N and P. Data were analyzed using PROC MIXED procedure of SAS (9.3) and contrasts were used to test the linear and quadratic effects of increasing levels of P within each CP level. In the model, CP and P levels were considered as fixed effects and block (BW) was the random effect. In the low CP groups (6.9%), the ileal digested P were 0.71, 1.16, 1.64, and 2.03 g/kg of DMI for diets that contained 0.09, 0.16, 0.24, and 0.32% ATTDP, and their counterparts in high CP groups (13.4%) were 0.70, 1.54, 2.03, and 1.99 g/kg of DMI. There was a main effect of CP level ( $P < 0.05$ ) on ileal digested P (g/kg of DMI). The ileal digested P (g/kg of DMI) increased linearly ( $P < 0.01$ ) with increasing ATTDP levels in low CP groups, but the pattern was linear ( $P < 0.01$ ) and quadratic ( $P < 0.01$ ) in high CP groups. In conclusion, this research indicated that the amount of ileal digested P (g/kg of DMI) could be limited by protein deficiency and thus, dietary CP level could affect ileal P digestion.

**Key Words:** amino acid, ileal digestibility, phosphorus

**M195 Could a reduction of crude protein content avoid the use of ZnO and antibiotics in pig diets without affecting their subsequent performance?** Sergi López-Vergé, Laia Blavi<sup>\*</sup>, David Solà-Oriol, José Francisco Pérez, and Josep Gasa, *Animal Nutrition and Welfare Service, Department of Animal and Food Sciences, Universitat Autònoma de Barcelona, Bellaterra, Spain.*

It's well known that ZnO and antibiotics (Ab) are usually included in nursery diets as a therapeutic use to reduce the impact of weaning in young piglets. The objective of this study was to analyze the effect of removing ZnO and Ab associated with CP reduction in piglet's diets in terms of mortality and days required to reach marketing BW (MBW, fixed at 105 kg). A total of 400 crossbreed piglets [Pietrain  $\times$  (Landrace  $\times$  Large White)] from 40 litters were used. During the nursery period, pigs were reared in 40 pens (10 piglets/pen) and assigned to 2 treatments taking into account the BW at weaning following a RCBD: medicated, M (2,480ppm Zn as ZnO, 120 ppm colistin sulfate, 300 ppm amoxicillin, 11.0 MJ/kg NE, 20.2% CP, 1.37% Lys) and unmedicated, Ø (10.6 MJ/kg NE, 16.5% CP, 1.20% Lys) diets until 35 d post-weaning. Thereafter, pigs were moved to a growing-finishing facility (maintaining the same littermates). Pigs were fed ad libitum the same commercial growing and finishing diets (10.0 MJ/kg NE, 16.0% CP, 0.95% Lys). Mortality rate (MR) was monitored from 0 to 14 d post-weaning. All animals were individually weighed at d 2 (CF; cross fostering), d 28 (weaning), d 42 (14 d post-weaning), d 63 (35 d post-weaning) and every 3 weeks until pigs were slaughtered. Data were adjusted using the Gompertz model:  $BW = A \times \exp\{-\exp[b - (c \times t)]\}$ ; the predicted time to reach 7, 10, 20, 40, and 105 kg of BW were statistically analyzed by ANOVA by using the GLM procedure of SAS. No differences in time to reach BW of 7 (M = 34.2 d vs. Ø = 35.0 d;  $P = 0.22$ ), 10 (M = 43.5 d vs. Ø = 44.3 d;  $P = 0.24$ ), 20 (M = 65.4 d vs. Ø = 66.3 d;  $P = 0.31$ ) and 40 kg (M = 95.4 d vs. Ø = 96.5 d;  $P = 0.28$ ) were observed. In contrast, time to reach MBW was slightly higher in Ø pigs (179.3 d vs. 175.1 d;  $P = 0.01$ ) and also had higher MR than the M (control) diet (11.7% vs. 4.5%;  $P = 0.02$ ) during the 14 d post-weaning. Results suggest that removing ZnO and Ab (also reducing CP and Lys) in early diets, seems not to have a huge impact in the time to reach MBW but causes higher MR in the first 14 d post-weaning.

**Key Words:** performance, piglet, zinc oxide

**M196 Comparing zinc status, growth, and mortality in piglets fed with or without therapeutic doses of Zn oxide.** Laia Blavi<sup>\*</sup>, Sergi López-Vergé, David Solà-Oriol, and José Francisco Pérez, *Animal Nutrition and Welfare Service, Department of Animal and Food Sciences, Universitat Autònoma de Barcelona, Bellaterra, Spain.*

Zinc (Zn) is an essential micronutrient required for multiple biological functions and used at therapeutic doses to reduce diarrhea and increase feed intake and performance in piglets. However, high doses of Zn in the diet are associated with heavy metal soil contamination and antimicrobial resistance which it may indicate the need to restrict or modify the therapeutic use of ZnO in piglets. A study was performed to assess how growth, mortality and Zn serum concentration change when Zn therapeutic is removed. A total of 400 weaned piglets [Pietrain  $\times$  (Landrace  $\times$  Large White)] were assigned to 2 different dietary strategies: Therapeutic (T diet, 122ppm of Zn as nutrient and 2,480ppm Zn as ZnO, 11.0 MJ/kg NE, 20.2% CP, 1.37 Lys) and Nutritional, in which the diet was formulated with a lower CP content, (N diet with 125ppm Zn as nutrient, 10.6 MJ/kg NE, 16.5% CP, 1.20 Lys) during the nursery period

(0 to 35d). On d 35, piglets were moved to the growing barns and fed the same diet (110 ppm Zn, 10.0 MJ/kg NE, 16.0% CP, 0.95 Lys). Piglets were weighted on d 0, 14, 35 and 57 and mortality was registered. Blood samples were obtained from 15 piglets per group to analyze Zn status on d 0, 7, 35 and 49 post-weaning. All studied parameters were analyzed with one-way ANOVA taking into account the experimental treatment as main factor with  $\alpha$  level of 0.05. No differences were observed on growth during the whole period, but higher mortality was observed in the N group compared with the T group. A temporary decrease of Zn serum concentration was observed after weaning in pigs fed the N diet (0.95, 0.67 and 1.07 mg/L on d 0, 7 and 35, respectively), whereas T piglets increased Zn concentration (0.95, 1.20, 1.31 mg/L, on d 0, 7 and 35, respectively;  $P < 0.001$ ). When T piglets were moved to the growing barns showed a decrease of Zn concentration (1.05 mg/L on d 49) while the N piglets showed a recuperation of Zn status to physiological levels (1.20 mg/L on d 49). These results might indicate a drop of blood Zn levels and an increase on mortality when therapeutic doses of Zn are removed and low CP diet is used, suggesting the need of using new, more efficient Zn compounds.

**Key Words:** ZnO, Zn status, piglet

**M197 Prediction equation to estimate digestible energy content of crude glycerin for swine based on chemical composition.**

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The energy value of crude glycerin (CG) from a palm oil biodiesel facility was determined in growing pigs to estimate its digestible energy

(DE) based on chemical composition. The study was conducted using the mobile nylon bag technique (MNBT). Nylon bags were inserted into the duodenum of 8 barrows (average body weight of 57.7 kg) through simple duodenal T-cannulas. Carboxymethylcellulose was used as a carrier of CG. Treatments consisted of a control (carboximethylcellulose) and 15 mixtures containing 10%, 20% and 30% of CG from 5 different batches of biodiesel production. Each batch showed variation in the content of glycerol and it was analyzed for dry matter (DM), gross energy (GE), ether extract (EE), ash, glycerol (GLY), pH and acid index (AI). Eight bags were administered to each pig daily (2 bags being inserted at 8, 10:30, 13:00 and 15:30 h into each of the 8 barrows). Sixteen nylon bags were prepared for each mixture (one gram per bag) therefore 256 bags were inserted over a 4-d period. The GE of each mixture was determined in a bomb calorimetry. The DE values of each mixture were estimated by difference, where by the DE content of the control was subtracted from each mixture containing CG. A multiple linear regression model was used to determine the value of the DE of CG as the slope relative to the DM of the CG included. The DE values of CG were correlated with its chemical composition and the value ranged from  $3264 \pm 202$  to  $3731 \pm 202$  kcal/kg DM ( $P < 0.01$ ). The prediction model with the lowest AIC (Akaike's information criterion) was selected.  $DE$  (kcal/kg as fed) =  $[1.29 \times GE$  (kcal/kg as fed)] +  $[109.7 \times \text{ash} (\%)] - (19.4 \times GLY)$ , ( $R^2$ -adj = 0.999,  $P < 0.02$ ). According to these results, the CG from palm oil biodiesel is a valuable energy source, with DE concentration depending on the concentration of GLY, GE and ash. The DE of crude glycerin was 77.5% of the GE.

**Key Words:** crude glycerin, digestible energy, prediction

## Nonruminant Nutrition: General I

**M198 Effect of dietary gamma-aminobutyric acid on egg production, egg quality, blood profiles, cecal microbial populations and excreta gas emission in layers.** J. H. Park<sup>\*1</sup>, B. Balasubramanian<sup>1</sup>, S. Kathannan<sup>1</sup>, J. H. Cho<sup>2</sup>, and I. H. Kim<sup>1</sup>, <sup>1</sup>Department of Animal Resource & Science, Dankook University, Cheonan, Chungnam, South Korea, <sup>2</sup>Department of Animal Science, Chungbuk National University, Cheongju, Chungbuk, South Korea.

Thirty-two-week-old Hy-line brown commercial layers were used to investigate the additive effect of gamma-aminobutyric acid (GABA) derived from *E. coli* strains, on the productivity, egg quality, blood profile, cecal population and excreta noxious gas emission. Two hundred eighty-eight birds ( $46 \pm 0.5$  g) were fed 4 different levels of GABA (0, 25, 50 and 100 ppm), in a basal diet based on corn-soybean meal, for 5 wks. Daily egg production was expressed on a hen-day basis, and egg quality and blood profiles were determined using an egg multi tester and an automatic blood analyzer, respectively. The *Lactobacillus* and *E. coli* medium agar plates were incubated for 24 h at 37°C, under anaerobic and aerobic conditions, respectively. A Gastec gas sampling pump was used for noxious gas detection. Polynomial contrasts were conducted to measure the linear and quadratic effects for increasing GABA levels on all measurements. As a result of the current study, egg production (93.5 vs. 95.2, 95.4, 97.1), egg weight (62.2 vs. 64.3, 65.9, 66.8), and egg mass (58.2 vs. 61.2, 62.9, 64.9) during 32–36 wks showed significant improvement, as dietary GABA increased from 0 to 100 ppm (linear,  $P < 0.05$ ). In addition, GABA supplementation was associated with increased eggshell breaking strength (4.28 vs. 4.48, 4.46, 4.44; quadratic,  $P < 0.05$ ) and albumen height (8.16 vs. 8.58, 8.62, 8.78; linear,  $P < 0.05$ ). Blood parameters, such as white blood cells, red blood cells, lymphocyte, cortisol, epinephrine and norepinephrine concentrations, were not influenced by GABA added into the diet; however, the haptoglobin concentration (15.0 vs. 12.3, 11.0, 10.8) was significantly decreased linearly, and IgG concentration (48.8 vs. 54.0, 54.3, 68.3) was increased quadratically in GABA-fed groups ( $P < 0.05$ ). *Lactobacillus* populations (7.46 vs. 7.48, 7.58, 7.90) in the cecum were significantly increased, as dietary GABA supplementation increased (linear,  $P < 0.05$ ). The ammonia (70 vs. 62, 59, 54) and hydrogen sulfide (12.0 vs. 9.7, 9.3, 7.7) gas emissions in the excreta were significantly decreased (linear,  $P < 0.05$ ). These results suggest that diets containing GABA may beneficially affect productivity, egg quality, serum haptoglobin and IgG concentration, and excreta noxious gas emission. Besides, GABA may also improve the intestinal environment, by increasing the *Lactobacillus* population of layers.

**Key Words:** blood profile, egg production, gamma-aminobutyric acid

**M199 Egg production, egg quality, blood profiles, cecal microflora, and excreta noxious gas emission in layer hens fed with fenugreek (*Trigonella foenum-graecum* L.) seed powder.** J. H. Park<sup>\*1</sup>, P. Y. Zhao<sup>1</sup>, H. L. Li<sup>1</sup>, J. H. Cho<sup>2</sup>, and I. H. Kim<sup>1</sup>, <sup>1</sup>Department of Animal Resource & Science, Dankook University, Cheonan, Choongnam, South Korea, <sup>2</sup>Department of Animal Science, Chungbuk National University, Cheongju, Chungbuk, South Korea.

The objective of this study was to investigate the effects of dietary fenugreek seed powder (FSP) supplementation in layers. A total of 288 layers (26 weeks old, Hyline-brown) were divided into 3 groups fed a corn-soybean meal diet (corn 50.4%, soybean meal 18.7%) containing

0 [control (CON)], 0.05 or 0.1% FSP for 5 weeks. Data were statistically analyzed via ANOVA using the GLM procedure of SAS for a completely randomized design, replicated 8 times with 12 layers per replication. Egg production was recorded daily on a hen-day basis, and egg quality and blood profiles were analyzed using an egg multi tester and an automatic biochemistry blood analyzer, respectively. For the measurement of cecal *Lactobacillus* and *E. coli*, cecal digesta was serially diluted in 0.1% peptone water, and incubated onto agar plates at 37°C for 24 h, under anaerobic and aerobic conditions, respectively. A Gastec gas sampling pump was used to detect gases in layer excreta. The inclusion of FSP in the layer diet did not affect egg production, feed intake and feed conversion among treatments. However, egg weight (CON = 63.1 vs. 0.1% FSP = 66.4 g), eggshell breaking strength (CON = 4.39 vs. 0.1% FSP = 4.66 kg/cm<sup>2</sup>), eggshell thickness (CON = 35.4 vs. 0.1% FSP = 36.7 mm<sup>-2</sup>) and yolk color (CON = 7.0 vs. 0.1% FSP = 7.8) was increased in FSP-fed groups ( $P < 0.05$ ). Supplemental FSP decreased serum total cholesterol concentration (CON = 222 vs. 0.05% FSP = 172, 0.1% FSP = 184 mg/dL), whereas the HDL-cholesterol (CON = 33 vs. 0.1% = FSP 44 mg/dL) and IgG concentration (CON = 446 vs. 0.1% FSP = 469 mg/dL) was increased in the FSP fed-groups ( $P < 0.05$ ). The inclusion of FSP led to an increase in cecal *Lactobacillus* number (CON = 6.98 vs. 0.1% FSP = 7.42 cfu/g;  $P < 0.05$ ) and a decrease in *E. coli* number (CON = 6.57 vs. 0.05% FSP = 6.14 cfu/g;  $P < 0.05$ ) and excreta ammonia gas emission (CON = 48 vs. 0.1% FSP = 30 ppm;  $P < 0.05$ ). These results suggest that the addition of FSP does not increase egg production, but may affect egg quality, serum total- and HDL-cholesterol, IgG concentration and cecal microflora. The inclusion of FSP also decreased ammonia gas emission in layer excreta.

**Key Words:** blood profile, egg quality, fenugreek

**M200 Effect of astaxanthin produced by *Phaffia rhodozyma* on growth performance, meat quality, and fecal noxious gas emission in broilers.** M. M. Hossain\*, M. Begum, H. Y. Shin, J. S. Jeong, M. Mohammadi, and I. H. Kim, Department of Animal Resource & Science, Dankook University, Cheonan, Chungnam, South Korea.

*Phaffia rhodozyma* is a species of yeast that produces the carotenoid pigment astaxanthin (AST), which exhibits a wide variety of biological activities in animals. A total of 432 1-d-old male broilers (Arbor Acres) was used in a 4-wk trial. Birds were allotted to 1 of 3 corn-soybean meal-based diets supplemented with 0 mg (CON, basal diet), 1,000 mg (CON + AST production 0.1%) or 2,000 mg (CON + AST production 0.2%) *P. rhodozyma* yeast per kg feed. There were 9 replicate cages per treatment, with 16 broilers per pen. Nine broilers were randomly selected from each treatment (1 bird per cage) for sampling on d 28. Statistical analysis was performed using the mixed models analysis procedures in SAS, with AST treatment and pen serving as the fixed and random effect model terms, respectively. In addition, orthogonal comparison was conducted using polynomial regression to measure the linear and quadratic effect of increasing dietary concentrations of supplemental AST production. Alpha level of significance was set at  $P < 0.05$ . The inclusion of yeast-AST in feed improved body weight gain in a linear fashion with increasing AST concentration (969, 989, 1024 g) in the finisher period and the overall experimental period (1377, 1401, 1439 g). Furthermore, yeast supplementation reduced feed conversion ratio (1.650, 1.603, 1.562) in the finisher period, and tended ( $P < 0.10$ ) to reduce feed conversion ratio for the overall experimental period (1.588, 1.554, 1.525). The yeast-AST supplemental diet had no effect on breast



muscle color or pH, and water holding capacity values. Treatments with yeast-AST reduced ammonia emission in a linear fashion (17.98, 14.42, 14.32 ppm). This study demonstrated that yeast producing AST supplementation has a positive effect on BWG and FCR in the finisher period and the overall experimental period. It is possible that yeast-AST might maintain and promote the growth of a beneficial microbial population in fast-growing broilers. Moreover, there was an effective reduction in the fecal ammonia in response to treatment with yeast-AST. Ammonia is considered the most harmful gas to livestock, and can reduce daily weight gains and feed utilization.

**Key Words:** astaxanthin, growth performance, noxious gas emission

**M201 Effect of protected organic acid blends on growth performance, nutrient digestibility and fecal micro flora in growing pigs.** Y. H. Liu<sup>\*1</sup>, J. K. Kim<sup>1</sup>, P. Y. Zhao<sup>1</sup>, K. Y. Lee<sup>2</sup>, and I. H. Kim<sup>1</sup>, <sup>1</sup>Department of Animal Resource & Science, Dankook University, Cheonan, Chungnam, South Korea, <sup>2</sup>Morningbio Co., LTD, Cheonan, Chungnam, South Korea.

Protected organic acids are suggested to more effectively deliver acids to the distal ileum, cecum, and colon of piglets. The objective of this study was to evaluate the effects of dietary supplementation of protected organic acid blends (17% fumaric acid, 13% citric acid, 10% malic acid, 1.2% medium chain fatty acid) and carrier on growth performance, nutrient digestibility and fecal micro flora in growing pigs. A total of 100 crossed [(Landrace × Yorkshire) × Duroc] with an average initial BW of 23.4 kg was used in a 6-wk trial. Pigs were randomly allocated into 1 of 4 treatments (5 replications with 5 pigs per pen) in a randomized complete block design based on their BW. Treatments consisted of a control diet (CON) and the control plus 0.1, 0.2, and 0.4% of organic acid blends. Body weight and feed intake were measured initially and at the end of 6 wk to determine average daily gain (ADG), average daily feed intake (ADFI) and gain/feed. The digestibility of nutrients and fecal micro flora was also assessed at the end of the experiment. Data were analyzed using contrast statements to test the linear and quadratic effect of organic acid concentration in the diet. Linear effects ( $P < 0.05$ ) were observed for ADG (713 g, 724 g, 745 g, and 737g) as well as G:F (0.450, 0.456, 0.471, and 0.464) with the increase in the level of organic acid blends. Dietary supplementation with 0.1%, 0.2% and 0.4% protected organic acid did not affect DM, N and energy digestibility. Supplementation of organic acid blends in the diet linearly increased ( $P < 0.01$ ) fecal *Lactobacillus* population counts (7.26, 7.31, 7.47, and 7.38 log<sub>10</sub> cfu/g) with the increase in the dose of organic acid blends. Our result suggests that protected organic acid has the potential to enhance growth performance and improve microbial population in growing pigs.

**Key Words:** growth performance, microflora, protected organic acids

**M202 Effect of phytochemicals on egg production, egg quality, excreta microbiota, noxious gas emission and nutrient digestibility in laying hens fed with different density diets.** A. Hosseindoust<sup>\*</sup>, H. L. Li, P. Y. Zhao, J. S. Jeong, and I. H. Kim, Department of Animal Resource & Science, Dankook University, Cheonan, Chungnam, South Korea.

This study was conducted to evaluate effects of dietary supplementation with phytochemicals on production performance, feed intake, egg quality, excreta microbiota, noxious gas emission, and nutrient digestibility in laying hens. Total of 240 Hy-line Brown 50-week-old laying hens were randomly assigned to 1 of 4 treatments. The dietary treatments included (1) T1, basal diet (ingredients included Ca: 4.92%; protein:

16.9%, phosphorus: 0.64% and energy: 3920 kcal/kg); (2) T2 = T1 + 150 ppm phytochemicals (quillaja 30%, anise 20% and thyme 17%); (3) T3 = T1 + 150 ppm phytochemicals w/matrix (reduction in Ca -0.072% and P -0.068% only); (4) T4 = T1 + 150 ppm phytochemicals w/matrix (reduction in Ca -0.072%, P -0.068%, protein -0.5%, energy -32 kcal/kg and amino acids). Variability in the data was expressed as the pooled standard error (SE) and probability level of  $P < 0.05$  was considered to be significant. Egg weight was improved ( $P < 0.05$ ) by the T2 (62.95), T3(63.54) and T4 (61.02) treatments compared with the T1 treatment during wk 15–20. The feed intake was higher ( $P < 0.05$ ) in the T3 (112) and T4 (115) treatments than the T1 and T2 treatments. The yolk height in the T4 treatment (8.38) was higher ( $P < 0.05$ ) than that in the T1 treatment at wk 20. T2 (8.95) and the T4 (9.00) treatments had higher ( $P < 0.05$ ) yolk color compared with that in the T3 treatment at wk 15. *E. coli* and *Lactobacillus* counts were not affected by dietary treatments. Laying hens in T3 (14.2) and T4 (16.4) treatments excreted significantly less ( $P < 0.05$ ) ammonia than that in the T1 and T2 treatments at wk 20. At wk 15, the T1 (59.25) treatment had the greater ( $P < 0.05$ ) ATTD (apparent total-tract digestibility) of Ca compared with the T3 treatment. Meanwhile, the T2 (47.59) and T4 (48.53) treatments had greater ( $P < 0.05$ ) ATTD of P compared with the T1 and T3 treatments. At wk 20, the T3 (48.90) group had the highest ( $P < 0.05$ ) ATTD of P when compared with the T2 and T4 groups. Thus the dietary supplementation with phytochemicals has some influence on performance, feed intake, egg quality, excreta microbiota, noxious gas emission, and nutrient digestibility in laying hens.

**Key Words:** phytochemicals, performance, egg quality

**M203 Effects of gel based phytochemical feed supplement on growth performance, nutrient digestibility, blood characteristics, and intestinal morphology in weanling pigs.** P. Y. Zhao<sup>\*1</sup>, H. M. Yun<sup>1</sup>, H. L. Li<sup>1</sup>, J. D. Hancock<sup>2</sup>, and I. H. Kim<sup>1</sup>, <sup>1</sup>Department of Animal Resource & Science, Dankook University, Cheonan, Chungnam, South Korea, <sup>2</sup>Department of Animal Science & Industry, Kansas State University, Manhattan, Kansas.

Gel based phytochemical feed supplement (GBP) is designed to facilitate the transition from a liquid diet (sow milk) to dry ration. It contains both feed component and water component thereby influencing feed consumption and intestinal health. In the current study, 120 weanling pigs [(Landrace × Yorkshire) × Duroc] with an average body weight of 6.71 kg were used in a 42-d feeding trial divided into 3 phases; d 1 to d 7 (Phase 1), d 8 to 21 (Phase 2) and d 22 to d 42 (Phase 3) to evaluate effects of GBP containing essential oil from caraway seed on growth performance, nutrient digestibility, blood chemistry and intestinal morphology. Pigs were randomly distributed on the basis of body weight and sex with 5 pigs per pen and 8 pens per treatment. Dietary treatments were: CON (basal diet), CON + 50 g GBP per pig a day, (F50) and CON + 100 g GBP per pig a day, (F100). The GBP was mixed with powdered feed just before feeding and fed during phase 1 post weaning and thereafter all pigs were fed only basal diet without GBP. All experimental data were analyzed as a randomized complete block design using the GLM procedures (SAS) with pen as the experimental unit and blocks based on initial BW. The digestibility of nutrients and measurement of villi of small intestine was done on d 10. Orthogonal contrasts were used to test the effect of treatments. During phase 3, feeding GBP linearly increased ( $P < 0.05$ ) ADG (544 g, 553 g, 562 g) and G:F (0.644, 0.657, 0.674). Overall, linear increase were shown ( $P < 0.05$ ) in ADG (462 g, 470 g, 477 g) and G:F (0.679, 0.694, 0.707). However, ADFI was neither improved nor reduced in piglets fed GBP. The digestibility of DM (78.2%, 81.8%, 82.5%) and GE (78.2%, 82.2%, 81.9%) increased

linearly ( $P < 0.05$ ). The villi length of jejunum (540, 695, 755 $\mu$ m) and ileum (556, 713, 774 $\mu$ m) was improved linearly ( $P < 0.05$ ) with the increase in the level of GBP. In conclusion, phytogetic feed supplement did not influence feed intake but increased ADG, improved G:F through enhancement of dry matter and energy digestibility as well as improved jejunum and ileum villi length.

**Key Words:** gel-based phytogetic feed supplement, growth performance, phytogetic

**M204 Effect of medium chain fatty acids and probiotic (*Enterococcus faecium*) supplementation on the growth performance, nutrient digestibility, fecal score, fecal microflora, and fecal noxious gas emission in weanling pigs.** P. Y. Zhao\*, B. Balasubramanian, M. Begum, M. Mohammadi, and I. H. Kim, *Department of Animal Resource & Science, Dankook University, Cheonan, Chungnam, South Korea.*

The objective of the present study was to evaluate the effects of medium-chain fatty acids (MCFA) and probiotic (*Enterococcus faecium* DSM 7134) supplementation in weanling pigs. A total of 140 weanling pigs [(Yorkshire  $\times$  Landrace)  $\times$  Duroc] were allotted to 4 treatments, 7 replicates/treatment and 5 pigs/replicate. Diets include: CON, basal diet; TRT 1, CON + MCFA 0.2%; TRT 2, CON + probiotic 0.01%; TRT 3, CON + MCFA 0.2% + probiotic 0.01%. Titanium oxide was added to the feed at 0.2% dosage (2 kg/t of feed) and for a duration of 5 d as an indigestible marker for digestibility determination. Fresh fecal grab samples collected from 2 pigs per pen were mixed and pooled, and a representative sample was stored in a freezer at  $-20^{\circ}\text{C}$  until analysis. Data were analyzed as  $2 \times 2$  factorial arrangement by using the GLM Procedure of SAS. During wk 0–2, probiotics supplementation increased ( $P < 0.05$ ) ADG (393 vs. 352) and G:F (0.831 vs. 0.741). During the overall period (wk 0–6), increased ( $P < 0.05$ ) ADG (454 vs. 424) and G:F (0.709 vs. 0.660) were detected in probiotic treatments. The ATTD of DM, N, and energy were increased ( $P < 0.05$ ) by probiotic (80.07% vs. 77.67%; 80.06% vs. 77.09%; 80.28% vs. 78.10%), MCFA (80.22% vs. 77.52%; 79.86% vs. 77.29%; 80.46% vs. 77.92%) and their combination (80.96% vs. 75.86%; 81.17% vs. 75.63%; 81.12% vs. 76.40%). There was no significant difference in fecal score, microflora (*Lactobacillus* and *E. coli*) and noxious gas emission (ammonia, hydrogen sulfide, total mercaptans, and acetic acid) in all the treatments. In conclusion, dietary probiotic supplementation have beneficial effect on growth performance, additionally, MCFA, probiotic and their combination can improve nutrient digestibility in weanling pigs.

**Key Words:** medium-chain fatty acids, weanling pig, growth performance

**M205 Effect of dietary protected organic acids on growth performance, nutrient digestibility, blood profiles, microflora, and gas emission in weanling pigs.** P. Y. Zhao\*<sup>1</sup>, M. Mohammadi<sup>1</sup>, K. Y. Lee<sup>2</sup>, M. Begum<sup>1</sup>, and I. H. Kim<sup>1</sup>, <sup>1</sup>*Department of Animal Resource & Science, Dankook University, Cheonan, Chungnam, South Korea,* <sup>2</sup>*Morningbio Co. Ltd., Cheonan, Chungnam, South Korea.*

The protected organic acids consist of medium chain fatty acid (MCFA) for animal nutrition and metabolism made by technology of Joint Matrix coating (JMT). The objective of the present study was to evaluate the effect of protected organic acid supplementation in piglets particularly at the weaning. A total of 112 weanling pigs with an average BW of  $6.70 \pm 1.31$  kg were allotted to 4 experimental diets as: CON (basal diet); OA1 (CON + unprotected organic acid 0.2%); OA2, (CON + protected organic

acid 0.1%) and OA3 (CON + protected organic acid 0.2%). There are 7 replications/treatment and 4 pigs/replication. The organic acid such as 17% fumaric acid, 13% citric acid, and 1.2% medium chain fatty acid (capric and caprylic acid) were used in the present experiment. Effects of treatments (Control, OA1, OA2, OA3) were analyzed by ANOVA as a randomized complete block design. Results are presented as least squares means and the variability in data was expressed as standard error (SE). The probability values less than 0.05 were considered as significant. Average daily gain (ADG), average daily feed intake (ADFI), gain to feed ratio (G:F), apparent total-tract digestibility (ATTD) were determined along with WBC, RBC, lymphocytes, IgG, *Lactobacillus* and *Escherichia coli* (*E. coli*). Average daily gain (ADG) at 0–2 wks were increased linearly (CON:307 g, OA1: 311 g, OA2: 319 g, OA3: 324 g) ADFI at 2–6 weeks (CON:762 g, OA1:757 g, OA2:755 g, OA3:750 g) and the overall ADFI was increased. A linear increasing effect was shown in the ATTD of dry matter (DM) (CON: 80.40%; OA1: 80.24%; OA2: 82.87%; OA3: 83.11%) with OA2 and OA3 treatments, the serum IgG in OA2 (235 mg/dL) and OA3 (245 mg/dL) treatments and the lymphocyte in OA3 treatments (50.9%) and in supplementation coated protected organic acid of *Lactobacillus*. However, a linearly reduced effect was observed in the population of *E. coli* (CON: 5.70, OA1: 5.65, OA2: 5.60, OA3: 5.49). In conclusion, dietary protected organic acids improve health status and performance in weanling pigs.

**Key Words:** protected organic acid, weaning pig, growth performance

**M206 Effects of dietary fiber and benzoic acid on growth performance, nutrient digestibility, reduction of harmful gases, and lipid profiles in growing pigs.** H. Y. Shin\*<sup>1</sup>, T. S. Li<sup>1</sup>, J. Y. Cheong<sup>2</sup>, C. M. Nyachoti<sup>3</sup>, and I. H. Kim<sup>1</sup>, <sup>1</sup>*Department of Animal Resource & Science, Dankook University, Cheonan, Choongnam, South Korea,* <sup>2</sup>*Daehan Feed Co. Ltd., Incheon, South Korea,* <sup>3</sup>*Department of Animal Science, University of Manitoba, Winnipeg, Manitoba, Canada.*

We assessed the possible synergistic effects of fiber from sugar beet pulp and benzoic acid in growing-finishing pigs. In total, 96 growing pigs ([Landrace  $\times$  Yorkshire]  $\times$  Duroc) with an average initial BW of 22.8 kg were selected and provided the dietary supplements based on their BW in a  $2 \times 2$  factorial experiment, with the respective factors being fiber (low vs. high; 140 g/kg, 160 g/kg NSP, respectively) and BA (0, 5 g/kg benzoic acid) in 6 replicate pens consisting of 4 pigs per pen. Sugar beet pulp was used as a DF sources, at 50 g/kg of the diet. All diets were formulated to contain 14.44 ME MJ/kg and 190 g/kg CP. This experiment was conducted to evaluate the growth performance, nutrient digestibility, and reduction of harmful gases and serum metabolites. The pen was considered as the experimental unit for growth performance and each pig was considered the experimental unit for gas emission and serum metabolites. The final model included the main effects of DF level and BA as well as the interaction between DF and BA. Data are reported as means  $\pm$  standard error (SE). Differences were considered statistically significant when  $P < 0.05$ . There was no significant difference in feed intake (1.428). No difference was found in weight during treatments. Fiber levels affect the dry matter digestibility: it was higher in the high fiber group (79.91) than the low fiber group (78.80) ( $P < 0.05$ ). Addition of BA also improved the energy digestibility ( $P < 0.05$ ). No interaction was found between fiber level and benzoic acid treatment. There was no difference in  $\text{NH}_3$ , but RSH (25.0, 26.1) and  $\text{H}_2\text{S}$  (20.2, 25.5) gases emissions shows significant reduction with fiber and benzoic acid treatment. Additionally, serum metabolites, including lipoprotein (LDL: 56.25, HDL: 36.25) and cholesterol (93.25), were also appar-

ently unaffected by these treatments. Thus, the addition of 50 g sugar beet pulp per kg of growing feed as a DF source and the addition of BA had no significant effect on the growth performance of pigs during the growth period. We now consider that 50 g of sugar beet pulp per kg of diet may have been insufficient to reduce serum cholesterol in grower pigs. The findings of this study also suggests that the metabolism of benzoic acid may differ from that of niacin.

**Key Words:** fiber, benzoic acid, growing pig

**M207 Ileal digestibility of nutrients and amino acids in conventional hulled (44% CP) and dehulled (48% CP) soybean meal treated with  $\beta$ -mannanase for growing pigs.** S. Shanmugam<sup>\*1</sup>, J. K. Kim<sup>1</sup>, H. M. Yun<sup>1</sup>, J. H. Cho<sup>2</sup>, and I. H. Kim<sup>1</sup>, <sup>1</sup>*Department of Animal Resource & Science, Dankook University, Cheonan, Chungnam, South Korea*, <sup>2</sup>*Department of Animal Science, Chungbuk National University, Cheongju, Chungbuk, South Korea*.

This experiment was conducted to determine the efficacy of  $\beta$ -mannanase supplementation to corn SBM based diet on ileal digestibility of nutrients and amino acids in growing pigs. Twenty barrows [(Landrace  $\times$  Yorkshire)  $\times$  Duroc] with an average body weight of 25 kg were fasted for 16 h and T-cannulas were surgically attached 15 cm proximal to ileo-cecal junction upon induction of anesthesia. The experimental diet consisted of corn based hulled or de-hulled SBM supplemented with or without 400U  $\beta$ -mannanase/kg. Cannulated pigs were allotted to one of 4 dietary treatments plus a nitrogen-free diet in a completely randomized design with 4 pigs per treatment. A nitrogen-free diet was used to determine basal endogenous losses of CP and amino acids. Data were analyzed as a completely randomized design with 2  $\times$  2 factorial arrangement using GLM procedures of SAS. The main effect included dietary SBM (hulled vs dehulled) and with or without  $\beta$ -mannanase supplementation as well as the interaction between  $\beta$ -mannanase and types of SBM. Each experimental period consisted of 7 d of diet adaptation followed by total feces and ileal digesta collection. The supplementation of  $\beta$ -mannanase improved ( $P < 0.05$ ) apparent ileal digestibility (AID) of DM compared with non-supplemented in hulled SBM (80.9% vs. 77.2%) and in de-hulled SBM (83% vs. 79.5%) but it did not influence N and GE digestibility. The AID of Thr and Pro was higher ( $P < 0.05$ ) in dehulled SBM (81.3%, 82.6%) than hulled SBM (77.3%, 78.0%). Likewise, the SID (standardized ileal digestibility) of Thr and Pro was higher ( $P < 0.05$ ) in dehulled (87.3%, 83.9%) than hulled SBM (83.9%, 80.3%). The supplementation of mannanase led to higher ( $P < 0.05$ ) AID of amino acids such as Arg (78.2, 79.6% vs. 74.0, 75.5%), His (82.2, 83.2% vs. 79.0, 81.5%), Lys (84.6, 86.9% vs. 79.4, 80.1%), Val (78.2, 80.2% vs. 74.3, 76.7%) and Gly (81.2, 82.9%

vs. 77.1, 80.3%) in hulled and dehulled SBM than non-supplemented diet and higher ( $P < 0.05$ ) SID of Lys (89.0, 89.8% vs. 84.2, 87.0%) in hulled and dehulled SBM than non-supplemented diet. In conclusion the supplementation of enzyme increased the AID of Arg, His, Lys, Val and Gly and SID of Lys.

**Key Words:** mannanase, soybean meal, growing pig

**M208 Effect of flavor and sweetener on growth performance, nutrient digestibility, blood profile, and diarrhea score in weaning pigs.** Y. Lei<sup>\*1</sup>, P. Y. Zhao<sup>1</sup>, H. L. Li<sup>1</sup>, C. M. Nyachoti<sup>2</sup>, and I. H. Kim<sup>1</sup>, <sup>1</sup>*Department of Animal Resource & Science, Dankook University, Cheonan, Chungnam, South Korea*, <sup>2</sup>*Department of Animal Science, University of Manitoba, Winnipeg, Manitoba, Canada*.

A total of 120 weaning pigs [(Landrace  $\times$  Yorkshire)  $\times$  Duroc; BW = 8.95  $\pm$  0.88 kg] were allotted to 4 treatments (6 pens/treatment, 5 pigs/pen) to evaluate the effect of anise flavor (F), sweetener (ST), and compound of them in weaning pigs. Treatments were as follows: 1) control, 2) control + 500 ppm F, 3) control + 150 ppm ST, 4) control + 500 ppm F + 150 ppm ST. Individual pig BW and pen feed intake were recorded on d 1, 14 and 42 (phase 1: d 1–14, phase 2: d 15–42) to calculate ADG, ADFI, and G:F. Cr<sub>2</sub>O<sub>3</sub> (0.2%) was added to the diets from d 7 to 14 and d 35 to 42 as indigestible marker to determine apparent total-tract digestibility (ATTD) of DM, N, and GE. Blood samples were collected from the cervical vein of 2 pigs in each pen at 24, 48, and 72 h after weaning. Subjective diarrhea scores were recorded daily from d 1 to 7 and d 8 to 14. Data were analyzed as 2  $\times$  2 factorial arrangement by using the GLM Procedure of SAS. During d 1 to 14, ADFI (-F vs. +F = 412 vs. 447.5 g; -ST vs. +ST = 407.5 vs. 452 g) and ADG (-F vs. +F = 323 vs. 355 g; -ST vs. +ST = 317.5 vs. 360.5 g) respectively improved by F ( $P < 0.01$ ) and ST ( $P < 0.01$ ) supplementation. During d 1 to 42, ST increased ADFI (-ST vs. +ST = 689.5 vs. 732 g;  $P < 0.01$ ) and ADG (-ST vs. +ST = 461 vs. 508 g;  $P < 0.05$ ). Digestibility of N was improved (-ST vs. +ST = 83.5 vs. 86.1%;  $P < 0.05$ ) in ST treatments on d 14. The epinephrine level at 48 h after weaning was decreased (-ST vs. +ST = 154 vs. 148.5 pg/mL;  $P < 0.05$ ) by supplementation of ST. Diarrhea score (scoring system from 1 to 5) was decreased ( $P < 0.05$ ) by F (-F vs. +F = 4.04 vs. 3.93) and ST (-ST vs. +ST = 4.05 vs. 3.92) during d 1 to 7. No interaction effect was observed in our experiment. In conclusion, sweetener improved growth performance and nutrient digestibility, decreased serum norepinephrine concentration, epinephrine concentration and diarrhea score. Anise flavor could improve growth performance, decreased norepinephrine concentration and diarrhea score after weaning. It implies the benefit of anise flavor and sweetener might be more effective in the first week after weaning, and the effect sweetener seems better than anise flavor.

**Key Words:** flavor, sweetener, weaning pig



# Physiology and Endocrinology: Effects of nutrition and metabolism on ruminant reproduction

**M209 Consistency of metabolic responses to nutrient deficiencies in early and mid-lactation of dairy cows.** Josef J. Gross\* and Rupert M. Bruckmaier, *Veterinary Physiology, Vetsuisse Faculty University of Bern, Bern, Switzerland.*

The adaptive response to a given metabolic load during energy deficiencies varies considerably among animals. The aim of this study was to investigate if individual cows respond in a repeated manner to a negative energy balance (NEB) in early and mid-lactation. Twenty-five multiparous Holstein dairy cows ( $3.0 \pm 1.1$  parities; mean  $\pm$  SD) experienced a NEB during the first weeks of lactation, and after getting into a positive EB, cows were exposed to a 3-wk feed-restriction period providing 50% of estimated requirements starting at around 100 DIM. Dry matter intake and milk yield were recorded daily. Blood samples for analysis of plasma concentrations of glucose, NEFA, BHBA, cholesterol, and IGF-1 were obtained once weekly. Retrospectively, cows were ranked according to plasma NEFA concentration in early lactation. The cows with the 33% highest and 33% lowest NEFA concentrations (8 animals each) were selected and classified either a high response (HR) or a low response (LR) group, respectively. Data were analyzed using MIXED models in SAS including wk, group (HR or LR), parity, and the wk  $\times$  group interaction as fixed effects and the individual cow as repeated subject. Differences between HR and LR over time were detected by the Bonferroni *t*-test. Significant effects were assumed at a level of  $P < 0.05$ . Before parturition, no differences were detected between LR and HR. After parturition, milk yield and ECM were higher for HR (milk yield: +7.4 kg/d, ECM: +4.9 kg/d from wk 2 to 14 pp) compared with LR ( $P < 0.05$ ). Although plasma concentrations of glucose and cholesterol showed group differences in early lactation, but not during feed-restriction, concentrations of NEFA, BHBA, and IGF-1 showed a consistent similarly directed, but different response to a NEB at the 2 stages of lactation despite a similar EB. HR had higher NEFA (+0.8 mmol/L in wk 2 pp, +0.2 mmol/L in wk 2 of feed-restriction), BHBA (+1.3 mmol/L in wk 3 pp, +0.3 mmol/L in wk 1 of feed-restriction) and lower IGF-1 concentrations (-48.7 ng/mL in wk 2 pp, -54.9 ng/mL in wk 2 of feed-restriction) compared with LR at the 2 stages of lactation ( $P < 0.05$ ). Cows responding to high milk production and negative energy balance with higher plasma NEFA and BHBA did again respond with higher NEFA and BHBA to a negative energy balance in later lactation as compared with cows with lower NEFA and BHBA concentrations with similar production levels.

**Key Words:** adaptation, metabolic plasticity, negative energy balance

**M210 Relationship of dietary and serum phosphorus during the transition period to fertility measures.** Ellen R. Jordan<sup>1</sup>, Kevin J. Lager<sup>1,2</sup>, J. Armando Garcia Buitrago<sup>\*3</sup>, Don R. Topliff<sup>4</sup>, and Pablo J. Pinedo<sup>5</sup>, <sup>1</sup>*Texas A&M AgriLife Extension, Dallas, TX*, <sup>2</sup>*West Texas A&M University, Canyon, TX*, <sup>3</sup>*New Mexico State University, Clovis, NM*, <sup>4</sup>*Angelo State University, San Angelo, TX*, <sup>5</sup>*Texas A&M AgriLife Research, Amarillo, TX.*

The objective was to evaluate the relationship between dietary and serum phosphorus (P) during the transition period and its relationship to fertility measures. Blood samples were collected into vacuum tubes at the morning feeding from cows ( $n = 4129$ ) in 8 Holstein (H) herds in the summer (S) and winter (W), as well as 8 Jersey (J) herds in the S. Samples were placed on ice immediately. After processing, samples

were stored at  $-20^{\circ}\text{C}$  until analysis. On the day of sampling, total mixed ration samples were collected for subsequent analysis. Associations were tested by logistic regression, correlation analysis, and ANOVA. During S, P in the ration (% DM) averaged  $0.35 \pm 0.05$ , prepartum H;  $0.45 \pm 0.12$ , prepartum J;  $0.42 \pm 0.03$ , postpartum H; and  $0.47 \pm 0.07$ , postpartum J; and during W averaged  $0.35 \pm 0.05$ , prepartum H and  $0.42 \pm 0.03$ , postpartum H. Serum P values (mg/dl) for wk -3, -2, -1, 1, 2, and 3 relative to calving were  $5.23 \pm 0.09$ ,  $5.26 \pm 0.19$ ,  $5.34 \pm 0.10$ ,  $4.79 \pm 0.12$ ,  $4.97 \pm 0.12$ , and  $5.40 \pm 0.10$  for S-H;  $6.38 \pm 0.09$ ,  $6.30 \pm 0.07$ ,  $6.21 \pm 0.07$ ,  $5.88 \pm 0.09$ ,  $5.78 \pm 0.08$ ,  $5.65 \pm 0.10$  for S-J; and  $5.23 \pm 0.12$ ,  $5.29 \pm 0.20$ ,  $5.71 \pm 0.1$ ,  $5.08 \pm 0.13$ ,  $5.44 \pm 0.11$ , and  $5.53 \pm 0.10$  for W-H. The ration and serum P correlation coefficients ( $P < 0.0001$ ) ranged from 0.14 over all weeks to a high of 0.34 for wk 3. During S, serum P concentrations were significantly associated with breed for wk -3, -2, -1, 1 and 2 ( $P < 0.0001$ ), while lactation was significant for wk -1, wk 2, and wk 3 ( $P = 0.0157$ ) and tended to be significant during wk -2 ( $P < 0.0575$ ). In univariable analysis, P serum concentration appeared to be associated with days to first service (DFS) and days open (DO); however when season and dairy were added in multivariate analysis, P serum concentration was no longer significant ( $P > 0.10$ ). When estimating the odds of pregnancy in either the first 90 (P90) or 150 (P150) d postpartum, serum P concentration was not significant in the multivariate analysis; when season and lactation were included. Serum P concentrations in the peripartum period were correlated with P intake; however, the main factors influencing the reproductive factors of DFS, DO, P90, and P150 were breed, season and lactation rather than serum P concentration.

**Key Words:** phosphorus, transition cow

**M211 Reproductive performance of lactating dairy cows with an extended duration of the postpartum voluntary waiting period and injectable trace mineral supplementation.** Matias L. Stangaferro<sup>\*1</sup>, Robert Wijma<sup>1</sup>, Magdalena Masello<sup>1</sup>, Rodrigo C. Bicalho<sup>2</sup>, Mark J. Thomas<sup>3</sup>, and Julio O. Giordano<sup>1</sup>, <sup>1</sup>*Department of Animal Science, Cornell University, Ithaca, NY*, <sup>2</sup>*Department of Population Medicine and Diagnostic Sciences, Cornell University, Ithaca, NY*, <sup>3</sup>*Dairy Health and Management Services, Lowville, NY.*

Objectives were to evaluate (1) the effect of extending the duration of the voluntary waiting period (VWP) from 60 to 88 DIM; and (2) the effect of injectable trace mineral (Zn, Mn, Se, Cu) supplementation (TMS) on reproductive performance of dairy cows. Holstein cows [ $n = 1,105$ ; 441 primiparous (PP) and 664 multiparous (MP)] were blocked by parity and total milk production in their previous lactation (MP only) and assigned to a 2x2 treatment arrangement to receive: TMS or NoTMS and first timed-AI (TAI) at  $60 \pm 3$  (SVWP) or  $88 \pm 3$  DIM (LVWP) after synchronization of ovulation with the Double-Ovsynch protocol (GnRH-7d-PGF-3d-GnRH-7d- GnRH-7d-PGF-56h-GnRH-16h-TAI) resulting in the following groups: TMS-SVWP ( $n = 315$ ), NoTMS-SVWP ( $n = 325$ ), TMS-LVWP ( $n = 228$ ) and NoTMS-LVWP ( $n = 237$ ). Cows in TMS received 3 SQ injections of 5 mL of TMS (Multimin 90<sup>®</sup>) as follows: 2 injections prepartum at  $229 \pm 3$  and  $259 \pm 3$  d of gestation for MP and  $243 \pm 3$  and  $263 \pm 3$  d of gestation for PP. A third injection was given at  $20 \pm 3$  d before first TAI. The effect of VWP duration, TMS, parity, and their interaction on pregnancy per AI (P/AI) at 39 d after AI, pregnancy loss (PL), and cows pregnant by 90 DIM (PG90) were evalu-

ated by logistical regression. At 39 d after AI, P/AI was greater for cows in the LVWP (47.5%) than SVWP (39.1%) group. At 39 d after AI, TMS did not affect ( $P = 0.96$ ) P/AI (42.7% and 42.5% for TMS and NoTMS). Primiparous cows had greater ( $P < 0.01$ ) P/AI than multiparous cows (49.0% and 38.4%). Pregnancy losses were similar ( $P = 0.68$ ) for cows in LVWP (5.8%) and SVWP (4.7%) and for cows in TMS (5.7%) and NoTMS (4.9%). Also, PL were similar ( $P = 0.13$ ) for PP (3.1%) and MP (7.1%) cows. No differences ( $P = 0.59$ ) in PG90 were observed for cows in LVWP (46.2%) and SVWP (48.0%) and for cows in the TMS (48.3%) and NoTMS (46.0%) group ( $P = 0.49$ ). Conversely PG90 was greater ( $P < 0.01$ ) for PP (52.5%) than MP (43.7%) cows. We conclude that extending the duration of the VWP from 60 to 88 DIM improved P/AI of lactating dairy cows. Conversely, TMS supplementation during the prepartum period and 20 d before TAI did not improve fertility of lactating dairy cows.

**Key Words:** voluntary waiting period, trace mineral, dairy cow

#### **M212 Resumption of ovarian cycle postpartum in dairy cows is affected by metabolic load in herbage-based feeding systems.**

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Due to topographic and climatic conditions, milk production in Switzerland is mainly based on herbage feeding with only little input of concentrates. This study investigated the effects of a solely herbage-based diet on production, metabolic, and endocrine parameters of dairy cows, and determined the factors affecting the resumption of ovarian cycle postpartum (pp). Twenty-three multiparous Holstein dairy cows (were divided into 2 groups according to their previous lactation yield (4679–10808 kg): a control (C,  $n = 13$ ) and a treatment group (nC,  $n = 10$ ) from wk 3 antepartum until wk 8 pp. While C received fresh cut herbage plus additional concentrate according to their estimated energy and nutrient requirements, no concentrate was fed to nC throughout the experiment. Milk yield and DMI were recorded daily. Blood samples were taken weekly and analyzed for IGF-1, glucose, NEFA, and BHBA. Milk progesterone (P4) concentrations were measured every 3 d in morning milk samples (skim milk) by RIA, and considered as resumption of ovarian cycle if the P4 level reached  $>1$  ng/mL. Data were analyzed using mixed models. Plasma NEFA and BHBA concentrations pp were higher in nC (0.82, 1.18 mmol/L) compared with C (0.55, 0.63 mmol/L,  $P < 0.05$ ). Days to resumption of ovarian cycle was similar between 2 groups (C = 29.8 d, nC = 33.3 d), thus the cows were divided into further 2 sub-groups with earlier/later than the above mean value, and categorized as earlier ovulation (E-OV; C = 22.6 d,  $n = 7$ ; nC = 21.4 d,  $n = 6$ ) and delayed ovulation (D-OV; C = 38.3 d,  $n = 6$ ; nC = 45.2 d,  $n = 4$ ). In C group, only BCS pp revealed lower in D-OV (2.81) compared with E-OV (3.21,  $P < 0.05$ ). In nC group, however, D-OV showed lower BW (571 vs. 697 kg in E-OV) and glucose (3.5 vs. 3.6 mmol/L in E-OV + nC), higher BHBA (1.39 vs. 1.04 mmol/L in E-OV + nC) and NEFA pp (0.89 vs. 0.76 mmol/L in E-OV + nC,  $P < 0.05$ ), suggesting an energy deficiency/stress on lipid metabolism. Other metabolic and welfare-related parameters were similar between E-OV vs. D-OV. In conclusion, in herbage-based feeding system without supplementary concentrate, dairy cows experience a higher metabolic load, which is very likely to affect resumption of ovarian cycle.

**Key Words:** metabolic load, ovulation, herbage feeding

#### **M213 Blood $\beta$ -hydroxybutyrate (BHBA) concentrations during the first two weeks after calving affect pregnancy establishment in postpartum dairy cows.** Monica O. Caldeira\*<sup>1</sup>, Matthew C. Lucy<sup>1</sup>, Ricardo O. Rodrigues<sup>1</sup>, and Scott E. Poock<sup>2</sup>, <sup>1</sup>*Division of Animal Sciences, University of Missouri, Columbia, MO*, <sup>2</sup>*College of Veterinary Medicine, University of Missouri, Columbia, MO*.

Negative energy balance (NEB) occurs in early postpartum dairy cows as a normal process when the energy demand at the onset of lactation exceeds feed energy intake. A poor response to NEB leads to greater blood concentrations of nonesterified fatty acids (NEFA) and BHBA, and also lesser blood glucose concentration. The objective was to determine whether blood BHBA and glucose concentration early postpartum affected pregnancy establishment after 3 inseminations in dairy cows. Blood samples were collected from lactating Holstein dairy cows ( $n = 204$ ) twice weekly for the first 4 wk postpartum ( $n = 8$  samples per cow). Blood BHBA and glucose were measured cowside by using a hand-held meter (Precision Xtra; Abbott Diabetes Care, Alameda, CA). Body weight (BW) and body condition score (BCS) were measured at calving, 2 wk, and 4 wk. Data were analyzed using the GLIMMIX, GLM, and MIXED procedures of SAS. The mean (SD) d postpartum of sample collection was 2.1 (1.3), 5.6 (1.5), 9.2 (1.4), 12.7 (1.6), 16.3 (1.6), 19.7 (1.6), 23.4 (1.6), and 26.8 (1.7) for samples 1 to 8. There were 171 cows with pregnancy data and 127/171 (74%) were pregnant after 3 inseminations. The blood BHBA concentration for sample 1 to 3 affected whether the cow became pregnant postpartum (lesser BHBA favorable toward pregnancy). The BHBA for non-pregnant and pregnant (respectively) was  $0.91 \pm 0.07$  and  $0.73 \pm 0.04$  (sample 1;  $P < 0.029$ ),  $0.92 \pm 0.07$  and  $0.76 \pm 0.04$  (sample 2;  $P < 0.043$ ) and  $0.76 \pm 0.04$  and  $0.68 \pm 0.02$  (sample 3;  $P < 0.08$ ). The BHBA concentration for samples 4 to 8 did not affect pregnancy. Greater blood glucose concentration for sample 2 tended to be associated with pregnancy ( $51.3 \pm 2.2$  and  $55.3 \pm 1.3$  for nonpregnant and pregnant;  $P < 0.11$ ). Glucose in sample 1 and 3 to 8 were not associated with pregnancy later postpartum. BW loss and BCS loss postpartum did not affect pregnancy. In conclusion, BHBA concentrations within the first 10 d postpartum affected pregnancy outcome later postpartum.

**Key Words:**  $\beta$ -hydroxybutyrate, glucose, reproduction

#### **M214 Effects of prepartum diets supplemented with oilseeds on reproductive performance in dairy cows.** R. Salehi\*<sup>1</sup>, M. G. Colazo<sup>2</sup>, M. Oba<sup>1</sup>, and D. J. Ambrose<sup>1,2</sup>, <sup>1</sup>*University of Alberta, Edmonton, Alberta, Canada*, <sup>2</sup>*Alberta Agriculture and Rural Development, Edmonton, Alberta, Canada*.

We investigated the effects of dietary fat supplementation (oilseed vs. no-oilseed) and type of oilseed (sunflower vs. canola) during late gestation on postpartum (pp) reproductive performance. Pregnant Holsteins were, blocked by BCS and parity, assigned to 1 of 3 diets containing 8% DM rolled sunflower (SUN;  $n = 45$ ) or canola seed (CAN;  $n = 43$ ), or no oilseed (CON;  $n = 43$ ), for the last 35 d of gestation and then a common lactation diet pp. Ovaries were scanned 2x/wk until 35 DIM in 95 cows to record first appearance of 10 (DF) and 16 mm (PreOVF) follicles, and ovulation. Blood samples were collected strategically to evaluate NEFA, BHBA and glucose, and endometrial cytology performed at  $25 \pm 1$  d pp ( $n = 12$ /diet). Data were analyzed using the Mixed procedure of SAS. Multiparous CON cows consumed more DM ( $19.5 \pm 0.2$ ) than those fed oilseed ( $18.0 \pm 0.2$  kg) during pre- and pp, until wk+3 pp. Cows fed SUN consumed more than those fed CAN ( $23.5 \pm 0.2$  vs  $22.5 \pm 0.3$  kg) pp from wk+2 to wk+5. Cows fed oilseed had higher NEFA (mEq/mL) than CON at wk-3 ( $108.0 \pm 10.3$  vs.  $70.8 \pm 10.3$ ), wk+1 ( $388.7 \pm 56.2$

vs.  $271.0 \pm 56.2$ ), and wk+4 ( $242.0 \pm 36.6$  vs.  $163.5 \pm 36.6$ ), but pre and pp energy balance (EB) did not differ ( $8.1 \pm 1.6$  Mcal). Prepartum diets did not affect BHBA ( $12.1 \pm 1.6$  mg/dL) and glucose ( $54.5 \pm 2.9$  mg/dL) pre or pp. Prepartum oilseed and type of oilseed supplemented neither altered the intervals (d) from calving to DF ( $9.4 \pm 0.5$ ), PreOVF ( $14.7 \pm 0.8$ ), or ovulation ( $21.4 \pm 1.5$ ), nor conception rate to first AI (27.2%) and cumulative pregnancy rate (67.3%) up to 5 AI. The interval to DF was correlated with % of polymorphonuclear cells (PMN; R: 0.38), DMI (R: -0.28) and glucose (R: -0.32) on wk+2. Interval to PreOVF was determined by % of PMN (R: 0.38) and DMI on wk -4 (R: -0.27), -3 (R: -0.27), -2 (R: -0.33), -1 (R: -0.38), +1 (R: -0.38), +2 (R: -0.45) and +3 (R: -0.39). None of the metabolites, glucose, EB, DMI or PMN affected interval to ovulation, although the latter was associated with interval to PreOVF (R: 0.41). In summary, feeding oilseeds prepartum, particularly canola, reduced DMI and increased NEFA without affecting reproductive performance.

**Key Words:** endometrial cytology, oilseed, ovarian function

**M215 Distribution of fatty acids in reproductive tissues of cows fed flax-, canola-, or sunflower seed-based rations.** Patricia A. Dutra<sup>1,2</sup>, Mohanathas Gobikrushanth<sup>\*2</sup>, Reza Salehi<sup>2</sup>, Marcos G. Colazo<sup>3</sup>, and Divakar J. Ambrose<sup>2,3</sup>, <sup>1</sup>*Departamento de Zootecnia, Universidade Federal da Bahia, Salvador, Bahia, Brazil*, <sup>2</sup>*Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, Alberta, Canada*, <sup>3</sup>*Livestock Research Branch, Alberta Agriculture and Rural Development, Edmonton, Alberta, Canada*.

Essential fatty acids (FA) like linoleic (C18:2) and  $\alpha$ -linolenic (C18:3) acids play key roles in reproductive function. The objective of this study was to determine the distribution of fatty acids in the reproductive tissues of cows fed 3 different lipid diets. Nonlactating Holstein cows were randomly assigned to receive one of the 3 diets supplemented with rolled oilseeds: canola (CAN = 5), sunflower (SUN = 5), or flax (FLX = 5) at 8%DM. After cows had been on the experimental diets for at least 60 d, they were superovulated, inseminated and slaughtered 14 d after insemination. While embryos were used in another study, tissue samples of the uterus, oviduct and corpus luteum (CL) were collected and kept frozen until analyzed for FA. Fats from the reproductive tissues were extracted and methylated using indirect methylation method, and FA profiles in each tissue type were determined by gas chromatography. Primarily, the effects of diet, type of tissue, and their interactions on FA of our interest [oleic (C18:1), C18:2, C18:3, eicosapentaenoic (C20:5) and docosahexaenoic (C22:6)] were evaluated using Mixed procedure of SAS. As the interactions between diet and type of tissue were not significant, FA concentration was finally modeled against effects of diet and type of tissue. Diet did not affect FA concentrations. Except for oleic acid ( $P = 0.07$ ), all of the other aforementioned FA differed ( $P < 0.05$ ) among tissue types. The concentrations (mg/50mg tissue) of C18:2, C18:3 and C20:5, respectively, were higher in CL (0.282, 0.020 and 0.019) than in uterus (0.075, 0.004 and 0.008) and oviduct (0.037, 0.003 and 0.001). However, the concentration of C22:6 was higher in uterus than oviduct or CL (0.008 vs. 0.005 or 0.004). In addition, the amount of total FA, saturated FA, polyunsaturated FA, n-3 and n-6 FA were significantly higher in CL than uterus or oviduct. Results indicate that CL accumulates most of the FA of our interest than the uterus or oviduct, whereas C22:6 was more concentrated in the uterus. The biological reasons for this differential accumulation of FA among reproductive tissues need further investigation.

**Key Words:** fatty acid, reproductive tissue, diet

**M216 Fatty acid profile in follicular fluid and serum of dairy cows fed diets supplemented with rolled canola, sunflower or flax seed.** Patricia A. Dutra<sup>1,2</sup>, Mohanathas Gobikrushanth<sup>\*2</sup>, Reza Salehi<sup>2</sup>, Marcos G. Colazo<sup>3</sup>, and Divakar J. Ambrose<sup>3,2</sup>, <sup>1</sup>*Departamento de Zootecnia, Universidade Federal da Bahia, Salvador, Bahia, Brazil*, <sup>2</sup>*Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, Alberta, Canada*, <sup>3</sup>*Livestock Research Branch, Alberta Agriculture and Rural Development, Edmonton, Alberta, Canada*.

Polyunsaturated fatty acids such as n-3 and n-6 are known to influence reproductive performance of dairy cows. The objective of this study was to determine fatty acid (FA) profile in serum and follicular fluid of cows fed 3 different lipid diets as well as to determine the association between serum and follicular fluid in terms of FA concentrations. Nine nonlactating Holstein cows were randomly assigned to receive 1 of 3 diets supplemented with rolled oil seeds: canola (CAN = 3), sunflower (SUN = 3) or flax (FLX = 3) at 8%DM. After cows received their respective diets for at least 60 d, they were superovulated, inseminated and slaughtered 14 d after insemination. The experimental diets continued until 12 h before slaughter. Embryos and reproductive tissues collected were used in another study. Blood samples were collected on the day of insemination, and follicular fluid was collected from large ovarian follicles within 4 h of slaughter. Serum and follicular fluid were kept frozen until analyzed for FA concentrations. Fats from the follicular fluid and serum were extracted, methylated and FA profiles were determined by gas chromatography. Data were analyzed using the MIXED and CORR procedures of SAS. In follicular fluid, cows fed SUN had higher concentrations (mg/mL) of linoleic acid (0.60 vs. 0.30, 0.32) and total n-6 FAs (0.62 vs. 0.31, 0.33) and higher n-6/n-3 ratio (9.25 vs. 2.09, 3.84) than those fed either FLX or CAN. Similarly, cows fed FLX had higher concentrations of  $\alpha$ -linolenic acid (C18:3; 0.14 vs. 0.08, 0.06), total n-3 FAs (0.16 vs. 0.09, 0.07) and higher n-3/n-6 ratio (0.49 vs. 0.10, 0.11) in follicular fluid than those fed either CAN or SUN. None of the FA other than  $\gamma$ -linolenic acid, in FLX cows, was altered by diet in serum. No significant correlations were observed between follicular fluid and serum FA profiles. Although diets supplemented with oilseeds selectively altered FA concentrations in follicular fluid, none of the major FA of relevance to reproductive function was altered in serum.

**Key Words:** fatty acid, follicular fluid, serum

**M217 Effects of rumen-protected methionine and choline supplementation on gene expression of follicular cells of the first postpartum dominant follicle.** Diego A. Velasco Acosta<sup>\*1,2</sup>, Ines M. Rivelli<sup>2</sup>, Cassandra Skenandore<sup>2</sup>, Daniel Luchini<sup>3</sup>, Marcio Corrêa<sup>1</sup>, and Felipe Cardoso<sup>2</sup>, <sup>1</sup>*Universidade Federal de Pelotas, Pelotas, Rio Grande do Sul, Brazil*, <sup>2</sup>*University of Illinois, Urbana, IL*, <sup>3</sup>*Adisseeo NA, Alpharetta, GA*.

This study aimed to determine the effects of rumen-protected methionine and choline supplementation during the transition period on mRNA expression of follicular cells of the 1st postpartum dominant follicle in Holstein cows. Multiparous cows were assigned in a randomized complete block design into 4 treatments from 21 d before calving to 30 DIM. Treatments were: CON (n = 10, fed the close-up and fresh cow diets with a Lys:Met = 3.5:1), MET (n = 9, fed the basal diet + methionine, Smartamine M to a Lys:Met = 2.9:1), CHO (n = 9, fed the basal diets + choline 60 g/d, Reashure), and MIX (n = 12, fed the basal diets plus Smartamine M to a Lys:Met = 2.9:1 and 60 g/d Reashure). Follicular development was monitored via ultrasound every 2 d starting at 7 DIM until the first dominant follicle reached a diameter of 16 mm. Follicular fluid from each cow was aspirated and cells were retrieved immediately



by centrifugation and stored at  $-80^{\circ}\text{C}$  until RNA extraction. Statistical analysis was performed using the MIXED procedure of SAS. Gene expression of *LHCGR*, *STAR*, *3 $\beta$ -HSD*, *P450scc*, *P450c17*, *CYP19A1*, *IRS1*, *IGF*, *MAT1A*, *SAHH*, *TLR4*, *TNF*, *IL1- $\beta$* , *IL8* and *IL6* was measured by real-time PCR. Treatments did not affect mRNA expression of *LHCGR*, *STAR*, *P450scc*, *CYP19A*, *SAHH*, *MAT1A* and *IL6* ( $P > 0.05$ ) however, *3 $\beta$ -HSD* expression was higher ( $P < 0.05$ ) for MET ( $1.46 \pm 0.3$ ) and MIX ( $1.25 \pm 0.3$ ) than CON ( $0.17 \pm 0.04$ ) and CHO ( $0.26 \pm 0.1$ ). For *TNF*, *TLR4* and *IL1-B* mRNA expression was higher ( $P < 0.05$ ) for CON ( $11.70 \pm 4.6$ ,  $21.29 \pm 10.4$ ,  $6.28 \pm 1.4$ ) than CHO ( $2.77 \pm 0.9$ ,  $2.16 \pm 0.9$ ,  $2.29 \pm 0.7$ ) and MIX ( $2.23 \pm 0.7$ ,  $1.46 \pm 0.6$ ,  $2.92 \pm 0.8$ ). There was higher ( $P < 0.05$ ) *IL1- $\beta$*  expression and a tendency ( $P = 0.07$ ) for higher *TNF* expression in CON ( $6.27 \pm 1.4$ ,  $11.70 \pm 4.6$ ) than MET ( $3.28 \pm 0.6$ ,  $3.06 \pm 0.8$ ). There was no difference ( $P = 0.43$ ) between CON and MET for *TLR4*. Expression of *IL8* mRNA was lower ( $P < 0.05$ ) for CHO ( $0.98 \pm 0.3$ ) than CON ( $4.90 \pm 0.7$ ), MET ( $6.10 \pm 1.7$ ) and MIX ( $5.05 \pm 1.8$ ). In conclusion, supplementing Smartamine M and Reashure during the transition period changed mRNA expression in follicular cells of the 1st postpartum dominant follicle in Holstein cows.

**Key Words:** methionine, choline, gene expression

**M218 Effect of yeast culture plus enzymatically hydrolyzed yeast supplementation starting prepartum on acute phase protein profiles and reproductive performance in dairy cows.** Vanessa Oliveira Freitas<sup>1</sup>, Claudia Faccio Demarco<sup>1</sup>, Tatiele Mumbach<sup>1</sup>, Eduardo Gulate Xavier<sup>2</sup>, Raquel Fraga Silva Raimondo<sup>3</sup>, Fernanda Medeiros Gonçalves<sup>1</sup>, Francisco Augusto Burkert Del Pino<sup>1</sup>, Viviane Rohrig Rabassa<sup>1</sup>, Sangita Jalukar<sup>4</sup>, Marcio Nunes Corrêa<sup>1</sup>, and Cassio Cassal Brauner<sup>\*1</sup>, <sup>1</sup>Universidade Federal de Pelotas, NUPEEC, Pelotas, RS, Brazil, <sup>2</sup>Granjas 4 Irmãos S/A, Rio Grande, RS, Brazil, <sup>3</sup>Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil, <sup>4</sup>Arm & Hammer Animal Nutrition, Princeton, NJ.

The aim of this study was to evaluate the effect of yeast culture plus enzymatically hydrolyzed yeast supplementation starting prepartum on acute phase protein profile and reproductive performance in dairy cows. Twenty 9 multiparous Holstein cows were blocked by milk production on the previous lactation and randomly assigned into 2 groups which consisted in one ( $n = 15$ ) receiving 28 g/d top-dressed yeast culture plus enzymatically hydrolyzed yeast (YC-EHY; Celmanax, Arm & Hammer Animal Nutrition, Princeton, NJ), while the control group ( $n = 14$ ) did not receive the supplement. The experimental period lasted from d  $-35$  relative to calving to 150 d postpartum. Plasma samples collected on  $-21$ ,  $-14$ ,  $-7$ ,  $0$ ,  $3$ ,  $7$ ,  $14$ ,  $21$ ,  $28$ ,  $35$ , and  $42$  d relative to calving were analyzed for acute phase proteins (haptoglobin, paraoxonase and albumin). To assess estrous cyclicity resumption, blood samples were collected weekly from 14 to 35 d after calving to evaluate the concentration of progesterone. Reproductive performance was also monitored until 150 d after calving. Data were analyzed using mixed models with repeated measures over time. Interval from calving to conception was analyzed using Kaplan-Meier survival curves. The YC-EHY group had lower ( $P = 0.04$ ) interval from calving to conception than control group respectively,  $95.35 \pm 10.78$  vs.  $130.73 \pm 10.35$  d. The interval from calving to the first ovulation were similar ( $P = 0.18$ ) between groups,  $23.33 \pm 2.8$  d vs.  $29.16 \pm 2.8$  d, respectively for YC-EHY and control groups. The YC-EHY tended ( $P = 0.07$ ) to have lower activity of paraoxonase during the postpartum period ( $112.54 \pm 3.49$  U/L vs.  $106.39 \pm 3.60$  U/L) than the control group. No differences ( $P > 0.05$ ) were observed in albumin and haptoglobin concentrations during the transition period. In conclusion, dairy cows supplemented since prepartum with yeast culture plus enzymatically hydrolyzed yeast, had lower interval from calving

to conception, without affect calving to first ovulation interval and tend to have lower activity of paraoxonase during the postpartum period.

**Key Words:** nutrition, reproduction, supplement

**M219 Nutritional level of the recipient ewe, but not of the donor, determines the embryo fate when good quality embryos are transferred.** Victoria de Brun<sup>\*1</sup>, Ana Meikle<sup>1</sup>, Fernando Forcada<sup>2</sup>, Inmaculada Palacin<sup>2</sup>, Cecilia Sosa<sup>3</sup>, and José Alfonso Abecia<sup>2</sup>, <sup>1</sup>Laboratorio de Técnicas Nucleares, Montevideo, Uruguay, <sup>2</sup>Departamento de Producción Animal y Ciencia de los Alimentos, Zaragoza, Spain, <sup>3</sup>Departamento de Anatomía Patológica, Medicina Legal y Forense y Toxicología, Zaragoza, Spain.

We aimed to isolate effects of undernutrition on the embryo or the mother and whether or not the endocrine signals differ from ewes that presented late embryo mortality. Forty-five donors and 52 recipient Rasa Aragonesa ewes were fed 1.5 (control group; donor  $n = 20$ ; recipient  $n = 25$ ) or 0.5 (low group; donor  $n = 25$ ; recipient  $n = 27$ ) times the daily requirements for maintenance. These regimens were maintained up to day of embryo collection and transfer. Embryos were collected 7 d after the onset of estrus, and 2 good quality embryos per ewe were transferred into recipient ewes. Blood samples were collected at days  $-14$ ,  $-1$ ,  $7$  and  $18$ . Pregnancy and late embryonic mortality were analyzed using the GENMOD procedure, and hormone and metabolite concentrations using a MIXED model in SAS. Nutritional treatment of donor did not affect fertility. Pregnancy evaluated by ultrasound on d 40 did not differ between control and undernourished ewes (72% vs. 55%,  $P = 0.17$ ). Embryonic mortality between d 18 and 40 tended to be greater in undernourished than in control recipient ewes (35% vs. 14%,  $P = 0.11$ ). Undernourished ewes had reduced live weight and body condition score ( $P < 0.01$ ), and increased concentration of nonesterified fatty acid ( $P < 0.05$ ). Pregnancy rates at d 18 and 40 were similar between groups, but recipient undernourished ewes presented a greater late embryonic mortality (d 18 to 40) than control ewes ( $P = 0.11$ ). Pregnant ewes had greater ovulation rate than nonpregnant ewes ( $P = 0.02$ ). In recipient ewes, pregnant undernourished presented greater P4 concentrations than pregnant control ( $P < 0.05$ ) and nonpregnant underfed ewes ( $P < 0.05$ ). Recipient undernourished ewes that suffered late embryo mortality (d 18 to 40) presented or tended to present lower insulin and progesterone concentrations than low pregnant ewes ( $P = 0.05$  and  $P = 0.07$ , respectively). In conclusion, the failure in reproductive performances in underfed ewes transferred good quality embryos is caused principally by maternal factors, so a suitable maternal environment is crucial to ensure a normal embryo growth. In addition, the endocrine profiles such as progesterone during early luteal phase and insulin are associated with pregnancy outcome.

**Key Words:** sheep, undernutrition, embryo

**M220 Evaluation of the hypothalamic kisspeptin system during the attainment of puberty in gilts.** Eric S. Jolitz<sup>\*1</sup>, Waljit S. Dhillo<sup>2</sup>, and Jeffrey A. Clapper<sup>1</sup>, <sup>1</sup>South Dakota State University, Brookings, SD, <sup>2</sup>Imperial College, London, UK.

It has been demonstrated that circulating kisspeptin is increased during the attainment of puberty in humans. To determine if hypothalamic (MBH) expression of kisspeptin (Kiss1), anterior pituitary (AP) expression of GnRH receptor (GnRHR) and LH- $\beta$ , and MBH and AP kisspeptin content increase at pubertal onset in the gilt, the following experiment was performed. Twenty-four crossbred gilts of similar age (150 d) and weight ( $102.7 \pm 0.3$  kg) were relocated and exposed to a mature

boar for estrus detection on d 1 and continuing for 6 d. Gilts that stood immobile within 24 h of slaughter (d 6) were considered to have attained puberty. Plasma samples were collected on d 1, 3, and 6. All gilts were slaughtered on d 6 when MBH, AP, and blood were collected. Relative expression of MBH Kiss1 and  $\beta$ -actin and AP GnRHr, LH- $\beta$ , and  $\beta$ -actin was determined using real-time reverse transcriptase PCR. Fold changes in relative expression were determined using the Relative Expression software tool. Relative expression is based on the expression ratio of a target gene versus a reference gene. The expression ratio results of the transcripts were tested for significance by a pair-wise fixed reallocation randomized test with day compared as independent time effects. Hypothalamic and AP content of kisspeptin were determined by RIA and differences were determined using the MIXED procedure of SAS. Relative expression of Kiss1 was increased ( $P = 0.005$ ) 2.2 fold in the gilts that had attained puberty. Relative expression of GnRHr did not differ ( $P > 0.05$ ) in gilts that had attained puberty versus those that did not. Relative expression of LH- $\beta$  tended to be decreased ( $P = 0.09$ ) 0.80 fold in gilts that had attained puberty. AP concentrations of kisspeptin were not different ( $P > 0.05$ ) between treatments. Kisspeptin content in the MBH was increased ( $P = 0.03$ ) in gilts that attained puberty ( $0.72 \pm 0.14$  ng/ $\mu$ g protein) compared with gilts that did not ( $0.44 \pm 0.05$  ng/ $\mu$ g protein). These data provide preliminary evidence that hypothalamic expression of Kiss1 and content of kisspeptin are increased during the attainment of puberty in the pig, which may modulate the release of GnRH.

**Key Words:** kisspeptin, hypothalamus, pig

**M221 Ovine maternal nutrient restriction from mid to late gestation induces steroid metabolizing enzyme activity in maternal and fetal reproductive and liver tissues.** Megan P. T. Coleson<sup>\*1</sup>, Christa L. Gilfeather<sup>1</sup>, Kimberly A. Vonnahme<sup>2</sup>, and Caleb O. Lemley<sup>1</sup>, <sup>1</sup>Department of Animal and Dairy Sciences, Mississippi State University, Mississippi State, MS, <sup>2</sup>Department of Animal Sciences, North Dakota State University, Fargo, ND.

The objective was to determine the effects of nutrient restriction on steroid metabolizing enzymes within maternal, placental, and fetal tissues. Singleton pregnant ewe lambs ( $n = 30$ ) were allocated to receive either 100% [adequate (ADQ;  $n = 14$ )] or 60% [restricted (RES;  $n = 16$ )] of nutrient requirements from d 50 until d 130 of gestation. At slaughter both maternal and fetal livers and maternal (caruncle; CAR) and fetal (cotyledon; COT) placentas were collected for analysis. Activity of cytochrome P450 2C (CYP2C), cytochrome P450 1A (CYP1A), and cytochrome P450 3A (CYP3A), were determined using specific luminogenic substrates. Activities were expressed relative to mg of protein, total tissue weight, or BW. Data were analyzed using MIXED procedure of SAS and the model statement included nutritional plane. Activity of CYP2C was not detected in CAR or COT tissues. Activity of CYP1A and CYP3A were not different in CAR tissue between the 2 treatments. Activity of CYP1A relative to BW was increased ( $P = 0.03$ ) in COT tissue of RES ewes compared with ADQ fed; however, CYP3A activity in COT was not different between treatments. Maternal liver activity of CYP2C was decreased ( $P < 0.01$ ) in RES ewes compared with ADQ; however, activity of CYP1A (relative to mg of protein), and CYP3A (relative to mg of protein or maternal BW) were increased ( $P \leq 0.05$ ) in RES ewes compared with ADQ. Fetal liver activity of CYP1A (relative to liver weight or fetal BW), CYP2C (relative to liver weight), and CYP3A (relative to mg of protein, liver weight, or fetal BW) were decreased ( $P < 0.05$ ) in RES ewes compared with ADQ. In conclusion, maternal nutrient restriction increased activity of CYP1A and CYP3A in maternal liver, but decreased the activity of CYP1A and

CYP3A in fetal liver. A similar downregulation of CYP2C activity was observed in both maternal and fetal liver. The differential regulation of hepatic cytochrome P450 in maternal and fetal tissues may influence peripheral steroid concentrations during late pregnancy and deserves future investigation.

**Key Words:** cytochrome P450, nutrient restriction, steroid

**M222 Effects of maternal nutrient restriction on bovine placentome and miRNA expression during mid-gestation.** Regina K. Taylor<sup>\*</sup>, Kayla S. Mangrum, Christopher T. LeMaster, Scott L. Pratt, and Nathan M. Long, *Clemson University, Clemson, South Carolina.*

Primiparous Angus-cross cows ( $n = 22$ ) were synchronized and AIed with sexed semen from a single Angus sire. Animals were fed at  $1.3\times$  (Control [CON]) or  $0.55\times$  (Nutrient Restricted [NR]) of maintenance energy and protein requirements based on BW (NRC 1996). Animals were blocked by BCS and BW and assigned to 1 of 3 treatments: CON; ( $n = 8$ ) d 30–190; NR/CON; ( $n = 7$ ) NR d 30–110 then CON d 110–190; or CON/NR; ( $n = 7$ ) CON d 30–110 then NR d 110–190. Cows were killed on d 190 of gestation, and the fetus and placenta collected. Isolation of RNA was performed from flash frozen cotyledon samples (3 subsamples/treatment) using the *mirVana* microRNA Isolation kit and analyzed using a previously validated microarray. Placentome measures and microRNA expression was analyzed as an ANOVA analysis using appropriate procedures correcting for false discovery rate of microarray data. Total placentome weight and total caruncular weight tended to be decreased ( $P = 0.056$ ,  $P = 0.07$ , respectively) in NR/CON animals vs. CON/NR and CON/CON. Total cotyledonary weight was increased ( $P = 0.017$ ) in CON/NR animals vs. NR/CON and CON/CON. Ratio of cotyledon weight:caruncle weight was increased ( $P = 0.02$ ) in NR/CON and CON/NR vs. CON/CON animals. Total placentome surface area tended to be increased ( $P = 0.09$ ) in CON/NR animals vs. NR/CON and CON/NR. Cotyledons from CON had mdo-miR-195 upregulated ( $P < 0.01$ ) and 8 downregulated ( $P < 0.01$ ) miRNAs, including mmu-mir-5105-p3 and bta-miR-2484 vs. NR/CON and CON/NR cotyledons. Cotyledons from NR/CON had mmu-mir-5117-p3\_1ss19TA and bta-miR-99b upregulated ( $P < 0.01$ ) and mmu-mir-5105-p3\_1ss24TC and bta-miR29a downregulated ( $P < 0.01$ ) vs. CON/NR and CON cotyledons. Cotyledons from CON/NR had 11 upregulated ( $P < 0.01$ ) miRNAs including bta-miR29a and bta-miR2484, and mdo-miR-195 and bta-miR-99b downregulated ( $P < 0.01$ ) vs. CON and NR/CON cotyledons. The data show that maternal nutrient restriction during early or mid gestation causes asymmetrical fetal growth restriction and affects miRNA regulation differently depending on if the restriction is preceded or followed by a non-restriction period.

**Key Words:** fetal growth, undernutrition, fetal programming

**M223 Maternal overnutrition/obesity (MO) in the ewe has multigenerational metabolic programming effects on adult granddaughters (F<sub>2</sub>).** Megan A. Walton<sup>\*</sup>, John F. Odhiambo, Peter W. Nathanielsz, and Stephen P. Ford, *Department of Animal Science, University of Wyoming, Laramie, WY.*

We have reported on a model in which MO ewes are fed 150% of NRC recommendations from 60 d before conception to term or fed only to requirements (CON). Offspring (F<sub>1</sub>) of MO ewes exhibit hyperphagia, increased weight gain and adiposity, and insulin resistance in response to an ad libitum feeding challenge when compared with CONF<sub>1</sub> (J. Anim. Sci. 2010. 88:3546). We recently reported that male grandsons (MOF<sub>2</sub>) also experience a significant increase in body mass and insulin resistance

in response to a similar bout of ad libitum feeding as 2-year-old adults, demonstrating a multigenerational effect of MO grandmothers on their grandsons. To evaluate potential offspring sex differences, we studied the multigenerational impact of MO on the response of 2-year-old adult female F2 to a 12-week ad libitum feeding challenge, MOF2 (n = 6) or CONF2 (n = 6). A dual x-ray absorptiometry (DEXA) scan was performed before and after the feeding challenge to determine changes in % body fat and % lean. Body weights and plasma samples were obtained bi-weekly. Glucose was evaluated via a colorimetric assay and insulin was analyzed via a validated RIA. Data (M ± SEM) were analyzed using the mixed procedure in SAS. No differences in body weight, % body fat or % lean were observed for CONF2 and MOF2 females either before or after the feeding trial. By the end of the feeding trial, body weight and % body fat were markedly increased ( $P < 0.05$ ) in both groups. Plasma glucose was greater ( $P < 0.05$ ) in MOF2 than CONF2 females, while plasma insulin tended to be greater ( $P < 0.06$ ) in MOF2 than CONF2 females throughout the trial, resulting in a markedly elevated ( $P < 0.01$ ) HOMA-IR in MOF2 vs. CONF2 females ( $7.8 \pm 0.7$  vs.  $5.0 \pm 0.7$ , respectively) demonstrating severe IR in MOF2. These data demonstrate that as previously reported for male OBF2, an ad libitum feeding challenge during adulthood induced marked insulin resistance in female OBF2. In contrast to the male OBF2, however, this elevated insulin resistance developed in the total absence of any increase in weight gain over that of female CONF2 in response to ad libitum feeding.

**Key Words:** programming

#### **M224 Effect of prepartum dam supplementation, creep-feeding and post-weaning diet on age at puberty in Nellore heifers.**

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The objective of this study was to evaluate the effect of various nutritional strategies on age at puberty in Nellore heifers. Nellore cows (n = 237 / BW  $432 \pm 50$  kg / BCS  $2.85 \pm 0.4$ ; 1 to 5 scale) were used in randomized design with a  $2 \times 2 \times 2$  factorial arrangement of treatments. Factors were as follows: factor 1 = prepartum supplementation with protein (PPS; n = 122; 0.5 kg soybean meal/cow daily) during the last trimester of gestation or no supplementation (NPPS; n = 115); factor 2 = provision of creep-feed to calves for 95 d before weaning (CF; n = 117) or no creep-feeding (NCF; n = 120; 22% CP and 72% TDN); and factor 3 = fed post-weaning from 7 to 14 mo of age in a feedlot (PWF; n = 117; 15% CP and 56% TDN) or no feedlot (NPWF; n = 120; *Brachiaria* pasture). After 14 mo of age, heifers were maintained in *Brachiaria* ssp. pastures. Heifers were weighed and reproductive status assessed by ovarian US (detection of presence of CL) monthly from 12 to 30 mo old. The proportion of heifers that had attained puberty by 14, 18, 26 and 30 mo of age, and BW at puberty were assessed using PROC MIXED and GLIMMIX. At weaning ( $220.1 \pm 2.0$ ), cow supplementation did not affect BW of heifer calves ( $168.2 \pm 0.4$  vs.  $170.5 \pm 0.5$  kg for PPS and NPPS cows, respectively), but BW at weaning were greater ( $P = 0.01$ ) in the CF ( $172.7 \pm 1.4$ ) than NCF ( $165.8 \pm 1.4$  kg) treatment. At the end of the post weaning period (Factor 3) BW were greater ( $P < 0.01$ ) in the PWF ( $234.1 \pm 2.2$ ) than NPWF ( $187.2 \pm 2.1$ ) treatment and this difference was maintained through 18 mo of age. Proportions of heifers that attained puberty at 14 (7.1%), 18 (22.3%), 26 (70.0%) and 30 (93.2%) mo of age were not influenced by cow supplementation or creep

feed. However, the post-weaning feedlot treatment (PWF) increased the proportion of heifers that reached puberty at 14 (13.7 vs. 0.9%;  $P < 0.01$ ) and 18 (31.7 vs. 13.3%;  $P < 0.01$ ) mo of age and tended ( $P = 0.10$ ) to anticipate puberty at 26 mo of age (73.8 vs. 66.1%; PWS and NPWS, respectively). In conclusion, enhanced nutrition during the post weaning period increased the proportion of heifers that reached puberty between 14 and 26 mo of age and is an effective method to anticipate puberty.

**Key Words:** puberty, Nellore, supplemental management

#### **M225 Effect of prepartum dam supplementation and creep-feeding on age at puberty in Nellore heifers.**

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The objective of this study was to evaluate the effect of 2 nutritional strategies on age at puberty in Nellore heifers. Nellore cows were allocated to treatments by BW ( $438 \pm 48$  kg) and BCS ( $3.0 \pm 0.5$ ; 1 to 5 scale). The factorial experimental design ( $2 \times 2$ ) included 2 phases of supplementation: phase 1 – prepartum supplementation with protein (PPS; n = 73; 0.5 kg soybean meal/cow daily) during the last trimester of gestation or no supplementation (NPPS; n = 81); phase 2 – provision of creep-feed (creep diet was 22% CP, and 62% TDN) to calves for 118 d before weaning (CF; n = 78) or no creep-feeding (NCF; n = 75). After 7 mo of age (weaning), heifers were maintained in *Brachiaria* ssp. pastures. Heifers were weighed and reproductive status assessed by ovarian ultrasonography (detection of presence of CL) monthly from 14 to 30 mo old. The proportion of heifers that had attained puberty by 18, 26 and 30 mo of age, and BW were assessed using PROC MIXED and PROC GLIMMIX (differences with  $P < 0.05$  considered significant). At weaning (6.4 mo), cow supplementation in last trimester of gestation did not affect BW of heifer calves ( $183 \pm 1.9$  vs.  $184 \pm 1.9$  kg,  $P = 0.98$  for PPS and NPPS cows, respectively). Creep feeding supplementation also did not influence BW at weaning ( $184 \pm 1.9$  vs.  $183 \pm 1.9$  kg,  $P = 0.78$  for CF and NCF, respectively). Prepartum supplementation did not change the proportion of heifers pubertal at 18 mo (4.1 vs. 6.2%,  $P = 0.53$ ), 26 mo (18.5 vs. 16.4%,  $P = 0.57$ ) and 30 mo (82.2 vs. 76.2%,  $P = 0.36$  for PPS and NPPS, respectively) of age. Supplementation of suckled calves by Creep Feeding also did not change the proportion of heifers pubertal at 18 mo (3.8 vs. 6.7%,  $P = 0.43$ ), 26 mo (15.8 vs. 19.1%,  $P = 0.77$ ) and 30 mo (80.7 vs. 77.3%,  $P = 0.59$  to CF and NCF, respectively) of age. In conclusion, neither the cow supplementation nor creep feeding used in the present study anticipated puberty in Nellore heifers.

**Key Words:** puberty, Nellore, supplemental management

#### **M226 Effect of energy supplementation on plasmatic concentration of leptin in pre-pubertal beef heifers.**

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The objective was to evaluate nutritional influence on plasma leptin concentration in prepubertal Nellore heifers. Sixteen calves at birth



were assigned to 2 groups: 1) calves received a concentrate of ground corn (2% of BW; Treated; n = 8); and, 2) calves received mineral salt without supplementation (Control; n = 8), calves from both groups had access to the cow's diet. After weaning (5 mo of age), both groups received sugar cane bagasse, citrus pulp and concentrate, but the Treated group also received a 2% of BW ground corn supplement until first ovulation or 22 mo of age. Heifers were weighed weekly to calculate monthly weight gain and blood samples were collected twice a week. Leptin was quantified with a RIA kit; the intra-assay CV were 7.64% for high (6.84 ng/mL) and 2.09% for low controls (0.58 ng/mL); the inter-assay CV were 21% for high and 12% for low controls, and assay sensitivity was 0.229 ng/mL. Data were analyzed by ANOVA with the Mixed procedure of SAS for repeated measures. The weight gain from weaning to puberty was  $19.5 \pm 0.03$  kg/month for Treated and  $13.2 \pm 0.03$  kg/month for Control heifers. The first ovulation occurred when the Treated heifers (n = 5) weighed  $398.2 \pm 10.6$  kg ( $17.8 \pm 1.1$  mo) and the Control heifers (n = 1) 361 kg (20.1 mo). After the 7th month of age there was a difference ( $P < 0.0001$ ) in monthly weight between treatments. In both treatments, plasma leptin increased according to age ( $P < 0.0001$ ). Plasma leptin concentration was greater ( $P = 0.023$ ) in Treated compared with Control heifers at 13, 14, 16 and 17 mo of age, averaging, respectively,  $5.67 \pm 0.53$  vs.  $4.04 \pm 0.53$  ng/mL,  $5.79 \pm 0.53$  vs.  $4.34 \pm 0.53$  ng/mL,  $7.12 \pm 0.53$  vs.  $5.21 \pm 0.53$  ng/mL and  $6.74 \pm 0.67$  vs.  $5.11 \pm 0.53$  ng/mL. The greatest weight gain in Treated heifers was observed close to first ovulation. The greatest plasma leptin concentration in Treated heifers was at the 16th month of age. Thus, supplementing heifers with corn hastened age at first ovulation, which is likely mediated by increased weight gain and leptin concentrations ( $r = 0.96$ ;  $P < 0.0001$ ), permissive factors that influence GnRH/LH secretion.

**Key Words:** *Bos indicus*, heifer, feed supplementation

**M227 Form of selenium in free-choice mineral mixes affects ovarian production of progesterone but not estradiol in cycling beef cows.** Phillip J. Bridges\*<sup>1</sup>, Kathryn L. Cerny<sup>1</sup>, Michelle Rhoads<sup>2</sup>, Leslie H. Anderson<sup>1</sup>, Walter R. Burris<sup>1</sup>, and James C. Matthews<sup>1</sup>, <sup>1</sup>University of Kentucky, Lexington, KY, <sup>2</sup>Virginia Polytechnic Institute and State University, Blacksburg, VA.

Selenium (Se) affects gonadal function, and the form of Se provided to cows affects tissue-specific gene expression. The objective of this study was therefore to determine whether the form of Se consumed by cows would affect follicular growth and the production of ovarian steroids. Pasture-fed Angus crossbred cows were randomly assigned to have ad libitum access to free-choice vitamin-mineral mixes containing Se (35 ppm) in either (TRT) inorganic (ISe, n = 41), organic (OSe, n = 42), or a 50/50 mix of ISe and OSe (MIX, n = 44) forms for 175 d. A subset of these cows (n = 9–11 per TRT) with a detectable corpus luteum were maintained on TRT and administered 25 mg PGF<sub>2α</sub> to induce luteal regression, then assigned for further sampling on estrus (Day 0). Between Day 4 and 8, follicular diameter was determined by ultrasonography. On Day 6, cows were treated with 20 mg then 15 mg PGF<sub>2α</sub>, 8 to 12 h apart, to induce luteal regression and differentiation of the first-wave dominant follicle into a preovulatory follicle. On Day 8, 36 h after PGF<sub>2α</sub>, the contents of the preovulatory follicle were aspirated by ultrasound-guided follicular puncture. Progesterone and estradiol were determined in plasma collected on Day 6 and 8, and in follicular fluid collected on Day 8. TRT effects were assessed by ANOVA and Fisher's LSD. TRT affected ( $P < 0.02$ , OSe > MIX) total blood Se (OSe:  $156 \pm 5$ , ISe:  $146 \pm 4$ , MIX:  $140 \pm 6$  μg/mL). TRT affected ( $P < 0.04$ , MIX > ISe) systemic progesterone on Day 6 (MIX:  $5.1 \pm 0.6$ , OSe:  $4.6 \pm 0.5$ , ISe:  $3.4 \pm 0.2$  ng/mL) but not Day 8. TRT did not affect systemic

estradiol on Day 6 or 8. TRT tended ( $P = 0.07$ , OSe > MIX, ISe) to affect follicular fluid progesterone (OSe:  $58.9 \pm 11.5$ , MIX:  $44.4 \pm 3.8$ , ISe:  $30.5 \pm 8.1$  ng/mL) but not estradiol. TRT did not affect diameter of the dominant follicle on Days 4 to 6, and tended ( $P = 0.08$ , ISe > OSe, MIX) to affect the diameter of the preovulatory follicle on Day 8 (ISe:  $12.6 \pm 0.6$ , OSe:  $11.4 \pm 0.3$ , MIX:  $11.3 \pm 0.4$  mm). Our results indicate that form of Se fed to cows affects progesterone but not estradiol synthesis, and tends to affect follicular diameter, in a manner independent of total blood Se concentrations.

**Key Words:** selenium, follicle, corpus luteum

**M228 Relationship between plasma amino acid profile and ovarian function around the time of ovulation in beef cows.** Taylor C. Geppert\*<sup>1</sup>, Allison M. Meyer<sup>2</sup>, George A. Perry<sup>3</sup>, and Patrick J. Gunn<sup>1</sup>, <sup>1</sup>Department of Animal Science, Iowa State University, Ames, IA, <sup>2</sup>Division of Animal Sciences, University of Missouri, Columbia, MO, <sup>3</sup>Department of Animal Sciences, South Dakota State University, Brookings, SD

The objective of this experiment was to determine the relationship between plasma AA concentrations and reproductive parameters of beef cows (n = 26). Two studies evaluating the effects of excess MP on reproductive function were compiled for this analysis. Non-pregnant, nonlactating mature beef cows consuming ad libitum corn stalks were offered a once-daily supplement designed to meet NRC NE<sub>m</sub> and exceed MP requirements by 25–50%. After a 20 d adaptation period, cows were synchronized for ovulation using the 5-d CO-Synch + CIDR protocol. Ten days after synchronization, 100 μL of GnRH was administered to reset ovarian follicular growth. Daily transrectal ultrasonography was performed to diagram ovarian activity, and blood samples were taken for hormone and AA analyses. Corpus luteum (CL) volume was determined via ultrasound, and supplementation ended 7 d after estrus. Blood samples collected between d 47 and d 49 underwent AA analysis. Data were analyzed using the CORR procedure of SAS. No significant correlations were observed between AA profile and ovulatory follicle size or antral follicle count ( $P > 0.13$ ). However, a positive relationship between length of proestrus and total AA ( $r = 0.49$ ,  $P < 0.01$ ) and total essential AA ( $r = 0.48$ ,  $P < 0.01$ ) was observed. In addition, a positive relationship between length of proestrus and total glycogenic AA, branched-chain AA, urea cycle AA, arginine, threonine, valine and isoleucine ( $r \geq 0.44$ ,  $P \leq 0.04$ ) were observed. As a percent of essential AA, leucine and phenylalanine were negatively correlated ( $r \leq -0.39$ ,  $P < 0.05$ ) with length of proestrus. However, AA profile was not correlated with estradiol at estrus ( $P > 0.05$ ). As a percent of total AA and essential AA, arginine was positively correlated with circulating progesterone 7 d post-estrus and ratio of progesterone to CL vol ( $r \geq 0.41$ ,  $P \leq 0.04$ ). Based on these data, total and essential AA concentrations may be related to several reproductive parameters around the time of ovulation; however, further research is needed to establish a causal relationship between individual AA and reproductive functions at ovulation.

**Key Words:** amino acid, correlation, ovulation

**M229 Association between circulating blood or plasma urea nitrogen concentrations and reproductive efficiency in beef heifers and cows.** Patrick J. Gunn\*<sup>1</sup>, Allie L. Lundberg<sup>1</sup>, Robert A. Cushman<sup>2</sup>, Harvey C. Freely<sup>2</sup>, Olivia L. Amundson<sup>3</sup>, Julie A. Walker<sup>3</sup>, and George A. Perry<sup>3</sup>, <sup>1</sup>Department of Animal Science, Iowa State University, Ames, IA, <sup>2</sup>USDA, ARS, US Meat Animal Research Center, Clay Center, NE, <sup>3</sup>Department of Animal Sciences, South Dakota State University, Brookings, SD.

The objective was to examine the effect of circulating blood or plasma urea nitrogen concentrations (BPUNC) on reproductive efficiency in beef heifers and suckled beef cows. Data from nulliparous heifers ( $n = 284$ ) as well as primiparous ( $n = 241$ ) and multiparous ( $n = 806$ ) beef cows were compiled across 15 experiments. A single blood sample collected from each female during estrous or ovulation synchronization was analyzed for BPUNC. Only females that were maintained on the same nutritional management scheme (pasture or coproduct-based drylot ration) for at least 1 wk before synchronization through the first 21 d of the breeding season were included in the analysis. To determine if BPUNC affected first service pregnancy rate, cattle were categorized as having BPUNC above or below each integer from 10 to 25 mg/dL. The GLIMMIX procedure of SAS was used for data analysis. The model for each BPUNC classification analysis also included the fixed effects of age classification and nutritional management scheme. Interactions among fixed effects were not significant and removed ( $P \geq 0.10$ ). Days postpartum at synchronization was included in the model as a covari-

ate when applicable and experiment was included as a random effect. Average first-service pregnancy rate across the data set was 55.6%. Irrespective of age and nutritional management scheme, a BPUNC that was associated with decreased pregnancy rates could not be established. In fact, there was a tendency for improved pregnancy rate as BPUNC increased ( $P = 0.08$ ,  $r = 0.05$ ) and cattle with BPUNC above 16 mg/dL tended to have greater pregnancy rates (57.0%) than those below 16 mg/dL (54.6%;  $P = 0.07$ ). There were no differences ( $P \geq 0.16$ ) in pregnancy rates between cattle that had BPUNC above or below any other integer from 10 to 25 mg/dL. Based on these data, when cattle are allowed to adapt to a nutritional management scheme before breeding and maintained on that diet through the first 21 d of the breeding season, BPUNC is not negatively associated with first-service pregnancy rates. USDA is an equal opportunity provider and employer.

**Key Words:** crude protein, cyclicity, pregnancy

# Physiology and Endocrinology: Estrous synchronization and detection of estrus in cattle

**M230 Efficacy of PGF<sub>2α</sub> doses to induce luteolysis on day 5, 7, or 9 of estrus cycle in nonlactating Nelore cows.** Marcos V. Biehl\*<sup>1</sup>, Alexandre V. Pires<sup>1,2</sup>, Marcos V. C. Ferraz Junior<sup>2</sup>, Jose R. S. Gonçalves<sup>3</sup>, Anibal B. Nascimento<sup>1</sup>, Marcelo H. Santos<sup>2</sup>, Vinicius N. Gouvea<sup>2</sup>, Alexandre A. Miszura<sup>2</sup>, Leandro H. Cruppe<sup>4</sup>, and Michael L. Day<sup>4</sup>, <sup>1</sup>University of São Paulo, Piracicaba, São Paulo, Brazil, <sup>2</sup>University of São Paulo, Pirassununga, São Paulo, Brazil, <sup>3</sup>Experimental Station Hildegard Georgina Von Pritzelwitz, Londrina, Paraná, Brazil, <sup>4</sup>The Ohio State University, Columbus, OH.

The aim of this preliminary study was to evaluate the luteolytic competence of different PGF<sub>2α</sub> (PGF, Lutalyse) doses on d5, d7 and d9 of the estrous cycle. Nonlactating Nelore cows (n = 97) were synchronized with the 7-d estradiol benzoate + CIDR program. Cows received Estro-tract patches at CIDR removal to determine estrus response. Presence of ovulatory follicle and its disappearance were confirmed 48 and 72 h after CIDR removal, respectively. Cows detected in estrus within 48 h and with confirmed ovulation 72 h after CIDR removal remained in the study (n = 68). Cows were assigned to treatments according to BW (407.9 ± 5.1) and BCS (3.01 ± 0.02, scale 1 to 5). One of 4 PGF doses were administered either 5, 7 or 9 d after estrus and confirmed ovulation (a single 12.5, 25 or 50 mg dose or 2 25 mg doses 8 h apart of dinoprost trometamine), in a 3 × 4 factorial arrangement of treatments. Presence of a corpus luteum was determined by ovarian ultrasound scan and progesterone (P4) analyses (P4 ≥ 1ng/mL) on either d 5, 7, or 9 (0h) of the estrous cycle. Blood samples were collected at 0, 24, 48 and 72 h after PGF administration to assess the incidence of luteal regression (defined as concentration of P4 < 1ng/mL at 72 h after PGF). Serum P4 concentrations were quantified using a chemiluminescent immunoassay. Data were analyzed using GLIMMIX procedure of SAS. Differences in proportion of cows that experienced luteal regression were not detected for day of cycle (P = 0.19); PGF dose (P = 0.13) or their interaction (P = 0.99). The incidence of luteal regression by day of cycle was 45.0% (9/20; 5 d), 66.6% (16/24; 7 d) and 70.8% (17/24; 9 d) and for PGF dose was 42.1% (8/19; 12.5 mg), 56.2% (9/16; 25 mg) 76.4% (13/17; 50 mg) and 75.0% (12/16; 25 + 25 mg given 8 h apart). Animal numbers used in the present study limited the capacity to determine significance of numerical differences that were observed. It is interesting that luteal regression was induced in less than 50% of cows on d 5 of the estrous cycle and in cows that received 12.5 mg of PGF. Additional research is necessary to determine the optimal dose of PGF and timing during the estrous cycle to ensure luteal regression in cyclic Nelore cows.

**Key Words:** corpus luteum, regression, Nelore cow

**M231 Addition of gonadotropin-releasing-hormone treatment at the beginning and/or at the end of an estradiol-based protocol for timed artificial insemination in Nelore (*Bos indicus*) cows.** Carla Cristian Campos\*, Estevão Vieira de Rezende, Mayara Oliveira, Renata de Freitas Ferreira Mohallem, and Ricarda Maria dos Santos, *Federal University of Uberlândia, Uberlândia, Minas Gerais, Brazil.*

Objective was to determine the effects of adding a gonadotropin-releasing-hormone (GnRH) treatment at the beginning and/or at the end of an estradiol-based protocol for timed artificial insemination (TAI) on pregnancy per AI (P/AI) in Nelore (*Bos indicus*) cows. The experiment was conducted in 2 beef cattle farms (A and B) located in a

central-western state in Brazil. The body condition score (BCS) evaluation and the ultrasound examination to determine ovarian status (follicle diameter <10 mm; ≥10 mm; or presence of corpus luteum - CL) were performed at the beginning of TAI protocol. Cows (n = 494) were randomly assigned to 4 groups: Control (n = 126), GnRH at day zero (D0) of the protocol (n = 123), GnRH at d 10 (D10) (n = 123), and GnRH in both times (D0+D10) (n = 122). The GnRH treatment consisted of one 50 µg i.m. injection of gonadorelin. All the cows were submitted to the following TAI protocol: Day 0 = insertion of intravaginal progesterone device previously used for 8 or 16 d and 2.0 mg of estradiol benzoate (EB) i.m.; Day 8 = progesterone device withdrawal, 750 IU i.m. injection of equine chorionic gonadotropin (eCG), 1.0 mg of estradiol cypionate (ECP) i.m. and 0.265 mg of sodium cloprostenol (PGF<sub>2α</sub>) i.m.; Day 10 = TAI, performed by a single inseminator. Pregnancy was diagnosed by ultrasound 39 ± 10 d after TAI. Data were analyzed by GLIMMIX procedure of SAS. Overall P/AI was 42.7%. No effect of treatment was detected (P = 0.25) on P/AI, and it was 37.3% for Control, 45.5% to GnRH D0, 49.6% to GnRH D10 and 38.5% to GnRH D0+D10. An effect of farm was detected on P/AI, where Farm A had 47.3% and Farm B 38.2% (P = 0.02). Cows with follicles <10 mm had lower P/AI (16.4%; P < 0.01) than cows with follicles ≥10 mm (46.2%) and the ones that had a CL (48.4%). The interactions between farm and treatments (P = 0.77) or between ovarian status and treatments (P = 0.12) did not affect P/AI. In conclusion, gonadotropin-releasing-hormone treatment used at the beginning and/or at the end of an estradiol-based TAI protocol did not affect pregnancy per AI in Nelore (*Bos indicus*) cows.

**Key Words:** GnRH, Nelore, timed AI

**M232 Using estrus-detection patches to optimally time artificial insemination improved pregnancy rates in suckled beef cows in a timed AI program.** Scott L. Hill\*<sup>1</sup>, David M. Grieger<sup>1</sup>, K. C. Olson<sup>1</sup>, John R. Jaeger<sup>1</sup>, Jason K. Ahola<sup>2</sup>, Mariah C. Fischer<sup>2</sup>, Teresa L. Steckler<sup>3</sup>, G. Allen Bridges<sup>4</sup>, Jamie A. Larson<sup>5</sup>, Carl R. Dahlen<sup>6</sup>, Sarah R. Underdahl<sup>6</sup>, George A. Perry<sup>7</sup>, William D. Whittier<sup>8</sup>, John F. Currin<sup>8</sup>, Jeffrey S. Stevenson<sup>1</sup>, <sup>1</sup>Kansas State University, Manhattan, KS, <sup>2</sup>Colorado State University, Fort Collins, CO, <sup>3</sup>University of Illinois, Dixon Springs, IL, <sup>4</sup>University of Minnesota, Grand Rapids, MN, <sup>5</sup>Mississippi State University, Mississippi State, MS, <sup>6</sup>North Dakota State University, Fargo, ND, <sup>7</sup>South Dakota State University, Brookings, SD, <sup>8</sup>Virginia Tech, Blacksburg, VA.

A multiple-location study examined pregnancy rates after delaying AI in suckled beef cows from 60 to 75 h when estrus had not been detected by 60 h after a 7-d CO-Synch + progesterone insert (CIDR) timed AI (TAI) program (d 0: progesterone insert [CIDR] concurrent with injection of GnRH; d 7: prostaglandin F<sub>2α</sub> injection and removal of CIDR; and GnRH injection at TAI [60 or 75 h after CIDR removal]). A total of 1,519 suckled beef cows at 14 locations in 8 states (CO, IL, KS, MN, MS, ND, SD, and VA) were included. Before applying the TAI program, BCS were assessed. Estrus was defined to occur when an estrus-detection patch was > 50% colored (activated). Pregnancy was determined 35 d after AI via transrectal ultrasound. Cows in estrus 60 h (n = 689; 45.6%) after CIDR removal were inseminated and injected with GnRH (control). Remaining nonestrus cows were allocated to 3 treatments: (1) GnRH injection and AI at 60 h (Early-Early = EE; n = 281), (2) GnRH injection at 60 h and AI at 75 h (Early-Delayed = ED; n



= 270), or (3) GnRH injection and AI at 75 h (Delayed-Delayed = DD; n = 279). Binomial data were analyzed using procedure GLIMMIX. More cows ( $P < 0.05$ ) that showed estrus by 60 h conceived to AI at 60 h than those not showing estrus (65.9 vs. 44%). Further, more ( $P < 0.001$ ) cows showing estrus by 75 h conceived to AI (64.9 vs. 38.5%) than cows not showing estrus. Control cows had a greater ( $^{abc}P < 0.05$ ) pregnancy rates (65.9%<sup>a</sup>) than other treatments (ED = 54.6%<sup>b</sup>, DD = 53.0%<sup>b</sup>, and EE = 44.0%<sup>c</sup>). Cows not in estrus by 60 h but with activated patches by 75 h (49.8%) were more ( $P < 0.05$ ) likely to become pregnant than nonestrus herd mates when they were in the DD (71.6 vs. 39.1%), ED (70.2 vs. 46.0%), and EE (53.7 vs. 41.4%) treatments, respectively. Pregnancy rates also were greater ( $P < 0.05$ ) in early-calving (>76 d postpartum) than later-calving ( $\leq 76$  d postpartum) cows (57.5 vs. 51.4%) but were not affected by parity or BCS. Use of estrus-detection patches to delay AI in cows not in estrus by 60 h after CIDR removal optimized time of AI and improved pregnancy rates to TAI.

### **M233 Treatment of primiparous lactating dairy cows with GnRH before first insemination during summer heat stress.**

Benjamin E. Voelz\*, Lucas Rocha, Filipe Scortegagna, Jeffrey S. Stevenson, and Luís G. D. Mendonça, *Department of Animal Sciences and Industry, Kansas State University, Manhattan, KS.*

Objectives were to evaluate concentrations of progesterone (P4), ovulation incidence, fertility, and insemination pattern of primiparous dairy cows treated with GnRH before a presynchronization protocol during summer heat stress. At  $60 \pm 3$  d postpartum, primiparous cows (n = 1,352) from 3 dairies were assigned randomly to 2 treatments (d 0): receiving GnRH (Gpresynch) or no GnRH (Control). Cows were presynchronized with 2 injections of prostaglandin  $F_{2\alpha}$  (PGF) given 14 d apart beginning on d 7. Cows detected in estrus based on tail paint removal were inseminated. Cows not detected in estrus were enrolled in a Cosynch-72 protocol on d 35 (GnRH on d 35 and 45; PGF on d 42; timed AI on d 45). Incidence of uterine health disorders and mastitis recorded by farm personnel were collected. Cows were considered diseased if 1 disease event occurred before AI. Pregnancy diagnosis was conducted  $36 \pm 3$  d after AI by transrectal ultrasonography. In a subgroup of cows, examinations of ovarian structures were performed on d -14, 0, and 7 (n = 161) and blood samples were collected on d 0 and 7 to determine concentration of P4 (n = 167). Logistic regression analysis was used for dichotomous outcomes and continuous variables were analyzed by ANOVA. The rate at which cows were inseminated was analyzed using the Cox's proportional hazard model. Ovulation risk to GnRH treatment was greater ( $P < 0.01$ ) for Gpresynch than control cows. Percentage of cows that had a corpus luteum (CL) and concentration of P4  $\geq 1$  ng/mL on d 7 was ( $P < 0.01$ ) greater for Gpresynch cows. Concentration of P4 on d 0 ( $P = 0.91$ ) and d 7 ( $P = 0.31$ ) did not differ between treatments. An interaction between treatment and disease tended ( $P = 0.08$ ) to affect P4 on d 7 because diseased cows treated with GnRH had greater P4 than diseased control cows. Controls tended ( $P = 0.06$ ) to have a faster insemination rate than Gpresynch cows [adjusted hazard ratio = 1.12 (95% CI = 1.00, 1.26)]. In addition, pregnancy per AI did not differ ( $P = 0.25$ ) between treatments. Although GnRH treatment increased the percentage of cows with a CL, no difference in subsequent fertility was detected.

**Key Words:** dairy cow, gonadotropin-releasing hormone

### **M234 Treatment of multiparous lactating dairy cows with GnRH before first insemination during summer heat stress.**

Benjamin E. Voelz\*, Lucas Rocha, Filipe Scortegagna, Jeffrey S.

Stevenson, and Luís G. D. Mendonça, *Department of Animal Sciences and Industry, Kansas State University, Manhattan, KS.*

Objectives of the experiment were to evaluate concentrations of progesterone (P4), ovulation incidence, fertility, and insemination pattern of multiparous dairy cows treated with GnRH before a presynchronization protocol during summer heat stress. At  $49 \pm 3$  d postpartum, multiparous cows (n = 1,941) from 3 dairies were assigned randomly to 2 treatments (d 0): receiving GnRH (n = 965; Gpresynch) or no GnRH (n = 976; Control). Cows were presynchronized with 2 injections of prostaglandin  $F_{2\alpha}$  (PGF) given 14 d apart beginning on d 7. Cows detected in estrus based on tail paint removal were inseminated. Cows not detected in estrus were enrolled in the Cosynch-72 protocol on d 35 (GnRH on d 35 and 45; PGF on d 42; timed AI on d 45). Incidence of uterine health disorders and mastitis recorded by farm personnel were collected. Cows were considered diseased if one disease event occurred before AI. Pregnancy diagnosis was conducted  $36 \pm 3$  d after AI by transrectal ultrasonography. In a subgroup of cows, ultrasonography examinations of ovarian structures were performed on d -14, 0, and 7 (n = 351), and blood samples were collected on d 0 and 7 to determine P4 concentrations (n = 361). Logistic regression analysis was used for dichotomous outcomes and continuous variables were analyzed by ANOVA. The rate at which cows were inseminated or became pregnant was analyzed using the Cox's proportional hazard model. Ovulation risk to GnRH treatment was ( $P < 0.01$ ) greater for Gpresynch than control cows. Percentage of cows that had a corpus luteum ( $P = 0.01$ ) and P4 concentration  $\geq 1$  ng/mL at first PGF ( $P = 0.03$ ) was greater for Gpresynch than control cows. Concentrations of P4 on d 0 ( $P = 0.30$ ) and d 7 ( $P = 0.54$ ) did not differ between treatments. Furthermore, insemination rate did not ( $P = 0.52$ ) differ between treatments. In contrast, control cows tended ( $P = 0.07$ ) to become pregnant at a slower rate than Gpresynch cows [AHR = 0.81 (95% CI = 0.64, 1.02)]. Treatment with GnRH before a presynchronization protocol using PGF injections increased the percentage of cows bearing a CL at first PGF, but did not improve pregnancy per AI.

**Key Words:** dairy cow, gonadotropin-releasing hormone

### **M235 Presynchronization strategy using prostaglandin $F_{2\alpha}$ and GnRH to improve fertility in a resynchronization program based on detection of estrus.** Lucas S. Rocha, Jeffrey S. Stevenson, and Luís G. D. Mendonça\*, *Department of Animal Sciences and Industry, Kansas State University, Manhattan, KS.*

Objectives were to evaluate pregnancy per AI (P/AI) and pattern of insemination of 2 resynchronization protocols to maximize insemination rate based on estrus detection in lactating dairy cows. Holstein cows (n = 1,929) from 3 herds were assigned randomly to 2 presynchronization (Presynch) protocols based on their ear tag number. At non-pregnancy diagnosis (day of enrollment [d 0]), odd-numbered cows (P7GPG; n = 967) received a Presynch treatment of  $PGF_{2\alpha}$  (PGF) and were enrolled in a resynchronization protocol on d 7 (GnRH on d 7 and 17; PGF on d 14; timed AI [TAI] on d 17). Even-numbered cows (P7GGPG; n = 962) received a Presynch treatment of PGF on d 0 and GnRH on d 7, and were enrolled in a resynchronization protocol on d 14 (GnRH on d 14 and 24; PGF on d 21; TAI on d 24). Detected estrus based on tail paint removal was conducted once daily during the study period and cows detected in estrus were inseminated within 1 h of detected estrus. Cows not detected in estrus by d 7 (P7GPG) or d 14 (P7GGPG) received their assigned TAI treatment. Cows were examined by transrectal ultrasonography to determine pregnancy status 36 d after AI. Binary outcomes were analyzed by logistic regression. The rate at which cows were inseminated was analyzed using the Cox's proportional hazard model. No treatment difference was detected for the percentage of cows

inseminated based on detected estrus (72.2%). Presynch treatment ( $P < 0.01$ ), TAI ( $P < 0.01$ ), and the interaction between TAI and Presynch treatment ( $P = 0.02$ ) affected P/AI. Cows inseminated in estrus were more likely to become pregnant than TAI cows (30.5 vs. 23.1%). No difference was detected in P/AI for cows inseminated at estrus (30.5%). In contrast, for cows receiving a TAI, P/AI was ( $P < 0.01$ ) greater for P7GGPG than P7GPG cows (28.9 vs. 17.6%). Hazard of insemination was affected ( $P < 0.01$ ) by treatment because P7GPG cows were inseminated sooner than P7GGPG cows (AHR = 1.70 [95% CI = 1.53 to 1.89]). Presynchronizing cows with GnRH delays AI but improves P/AI of cows submitted to a TAI protocol.

**Key Words:** estrus, presynchronization, resynchronization

### **M236 Progesterone concentration at each treatment during an Ovsynch protocol affects fertility to timed AI in lactating**

**Holstein cows.** P. D. Carvalho\*, A. H. Souza, M. C. Wiltbank, and P. M. Fricke, *Department of Dairy Science, University of Wisconsin-Madison, Madison, WI.*

Our objective was to assess the association between progesterone (P4) concentration at each treatment of an Ovsynch protocol and pregnancies per AI (P/AI) to timed AI (TAI) in lactating Holstein cows. Data from 7,792 cows from 14 experiments in which P4 was measured during an Ovsynch protocol [GnRH (G1); 7 d PGF<sub>2α</sub> (PGF); 56 h, GnRH (G2); 16 h, TAI] were analyzed. Cows receiving exogenous P4 or that received 2 injections of PGF or an increased dose of PGF were excluded from the analysis. The association between P4 during the Ovsynch protocol and P/AI to TAI was analyzed independently because P4 was not measured for all cows at every time point in all experiments. Data were analyzed by logistic regression using the GLIMMIX procedure of SAS. At G1, cows ( $n = 6,144$ ) were stratified into 9 P4 categories from 0 to  $\geq 7$  ng/mL using 0.5 ng/mL increments. Overall, P/AI differed ( $P < 0.01$ ) among P4 categories at G1 with fewer P/AI for cows with  $P4 < 0.5$  ng/mL (30%; 375/1,259) or  $P4 > 7.0$  ng/mL (30%; 115/381) compared with cows with intermediate P4 (41%; 1,839/4,504; calculated relative decrease in P/AI of 27%;  $41 - 30 = 11/41 = 27\%$ ). At PGF, cows ( $n = 3,383$ ) were stratified into 9 P4 categories from 0 to  $\geq 8$  ng/mL using 1.0 ng/mL increments. Overall, P/AI differed ( $P < 0.01$ ) among P4 categories at PGF with a 51% relative decrease in P/AI for cows with  $P4 < 1.0$  ng/mL (21%; 140/681) compared with cows with  $P4 > 1.0$  ng/mL (43%; 1,151/2,702). At G2, cows ( $n = 3,148$ ) were stratified into 8 P4 categories from 0 to  $\geq 0.7$  ng/mL using 0.1 ng/mL increments. Overall, P/AI differed ( $P < 0.01$ ) among P4 categories at G2 with a 66% relative decrease in P/AI for cows with  $P4 > 0.4$  ng/mL (14%; 61/435) compared with cows with  $P4 < 0.4$  ng/mL (41%; 1,125/2,713). Based on this large data set, suboptimal P4 concentrations could be identified at G1 in 26% of cows (26% lower P/AI), at PGF in 21% of cows (51% lower P/AI), and at G2 in 14% of cows (66% lower P/AI). We conclude that achieving optimal P4 during an Ovsynch protocol may allow for a dramatic increase in fertility in lactating dairy cows. Supported by USDA NIFA Hatch project 23144.

**Key Words:** progesterone, fertility, Ovsynch

### **M237 Timed AI improves reproductive performance and reduces cost per pregnancy in dairy heifers.**

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Objectives were to evaluate the effects of implementing timed AI for first insemination on reproduction and cost per pregnancy in dairy heifers.

Non-pregnant Holstein heifers ( $n = 611$ ) at 400 d of age from 3 farms were allocated randomly to AI after detected estrus (Control,  $n = 306$ ) or to timed AI for first AI (TAI,  $n = 305$ ) followed by detection of estrus. Heifers in TAI were enrolled in the 5-d timed AI protocol on study d -6 [d -6, GnRH and an intravaginal progesterone insert; d -1, PGF<sub>2α</sub> and insert removal; d 0, PGF<sub>2α</sub>; d 2, GnRH + AI]. Starting on study d 0, heifers had tailheads painted daily with chalk for detection of estrus. Control heifers received PGF<sub>2α</sub> if not inseminated within 7 d of moving to the breeding pen. The study lasted 84 d to allow 4 21-d estrous cycle periods of breeding. A herd budget was created to determine cost per pregnancy. Data were analyzed by ANOVA, logistic regression, and the Cox's proportional hazard model using the MIXED, GLIMMIX, and PHREG procedures of SAS and models included the effects of treatment, farm, and treatment by farm interaction. The interval to first AI was 8 d shorter ( $P < 0.01$ ; 10 vs. 2 d) and the 21-d cycle insemination rate was greater ( $P < 0.01$ ) for TAI than Control heifers (82.4 vs. 91.4%). Pregnancy at first AI (PAI; Control = 58.3% vs. TAI = 62.8%) and all AI (Control = 56.0 vs. TAI = 57.4%) did not differ ( $P > 0.15$ ) between treatments. Implementing timed AI increased ( $P < 0.01$ ) the 21-d cycle pregnancy rate from 47.9% in Controls to 57.2% in TAI heifers and reduced ( $P < 0.01$ ) median d to pregnancy (Control = 14.0 vs. TAI = 2.0). In fact, the hazard of pregnancy improved ( $P < 0.01$ ) 60% for TAI than Control heifers (adjusted HR = 1.60; 95% CI = 1.35–1.89), resulting in increased ( $P < 0.01$ ) proportion of pregnant heifers by 84 d of breeding (Control = 85.2 vs. TAI = 91.5%). The cost per pregnancy for was \$17.16 less expensive ( $P < 0.01$ ) for TAI than Control heifers. Implementing timed AI for first service followed by detection of estrus improved reproductive performance and reduced the cost per pregnancy when compared with heifers managed under detected estrus only.

**Key Words:** dairy heifer, timed AI, reproduction

### **M238 Addition of a second dose of prostaglandin F<sub>2α</sub> to a fixed-time AI protocol improves fertility of anestrous dairy cows.**

Francisco R. Lopes Jr.\*, Marcos H. C. Pereira, Anderson K. Munhoz, and José L. M. Vasconcelos, *Sao Paulo State University, Botucatu, SP, Brazil.*

This study was conducted to evaluate if addition of a second prostaglandin (PG) F<sub>2α</sub> dose improves fertility of anestrous lactating Holstein cows submitted to an estradiol/progesterone (P4) based timed AI (TAI) protocol. Ovaries were scanned by ultrasound to determine if a corpus luteum (CL) was present at the protocol initiation (d-11) and on the day of PGF<sub>2α</sub> (d -4). Cows without CL on d -11 and d -4 were classified as anestrous ( $n = 436$ ). Anestrous cows were submitted to the TAI protocol: d -11: 2 intravaginal P4 devices (1.9g P4, CIDR, Zoetis) and 2.0 mg of estradiol benzoate (EB) i.m. (Estrogin, Farmavet); d -4, 25mg of PGF<sub>2α</sub> i.m. (Lutalyse, Zoetis) and withdrawal of one CIDR; d -2 withdrawal of the second CIDR and 1.0 mg i.m. of estradiol cypionate (ECP, Zoetis); on d 0 TAI. On d-4, cows were randomly assigned to 1 of 2 treatments: one dose of PGF<sub>2α</sub> on d -4 (1PGF) or 2 doses of PGF<sub>2α</sub> (2PGF), the first on d -4 and the second on d -2. Rectal temperature was measured on the day of TAI and 7 d later. The average rectal temperature was calculated and the cows were divided into 2 groups: without hyperthermia ( $< 39.1^\circ\text{C}$ ) or with hyperthermia ( $\geq 39.1^\circ\text{C}$ ). Pregnancy was diagnosed 60 d after AI. Binomial variables were analyzed using the GLIMMIX and continuous using the MIXED procedures of SAS. The 2PGF treatment tended to improve the synchronization rate (80.2% [ $n = 218$ ] vs. 72.4% [ $n = 218$ ]  $P = 0.07$ ). An interaction ( $P = 0.05$ ) between treatment and hyperthermia was observed for pregnancy per AI (P/AI) on d 60. For hyperthermic cows, P/AI were 9.1% ( $n = 123$ ) and 9.4% for 1PGF and 2PGF, respectively. For cows without hyperthermia, P/AI were 16.3%

(n = 95) and 30% (n = 88) in 1PGF and 2PGF, respectively. When only ovulated cows were included in the analysis, then an interaction ( $P = 0.05$ ) existed between treatment and hyperthermia on P/AI at 60 d. For cows with hyperthermia, P/AI were 13.8% (n = 80) and 12.1% (n = 100) in 1PGF and 2PGF, respectively. For cows not suffering of hyperthermia, P/AI were 19.4% (n = 78) and 34.8% (n = 75) in 1PGF and 2PGF, respectively. The addition of a second PGF dose during the TAI protocol increased fertility in anestrous dairy cows without hyperthermia on the first 7 d after AI.

**Key Words:** anestrous, PGF<sub>2α</sub>, TAI

### **M239 Effect of a second treatment with prostaglandin F<sub>2α</sub> during the Ovsynch protocol on pregnancy to the timed AI.**

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Lack of complete regression of the corpus luteum (CL) after prostaglandin F<sub>2α</sub> (PGF) treatment can reduce fertility during timed AI (TAI) protocols. The main objective of this study was to evaluate the effect of a second treatment with PGF during the Ovsynch protocol on fertility to the timed AI (TAI). A total of 2,148 lactating dairy cows were used on 11 dairy farms in 4 different regions of the USA, 3 in WI, 3 in CA, 2 in NY, one in PA, one in NM, and one in TX. Cows were synchronized with Ovsynch (GnRH [Gonadorelin acetate; 100 µg/mL; GONAbreed, Parnell]-7d-PGF[Cloprostenol sodium; 250 µg/mL; estroPLAN; Parnell]-56h-GnRH-16h-TAI) for TAI at 50 ± 3 DIM. The cows were randomized to receive: (1) No additional treatments with PGF = Ovsynch, or (2) A second PGF treatment at 24 h after the first PGF treatment = PROCEPT. Statistical analyses were performed using PROC Logistic in SAS, with treatment and parity as factors. In the overall logistic regression model there was no significant effect of Farm ( $P = 0.712$ ); although, treatment effects on individual farms ranged from positive ( $P = 0.01$ ) to a tendency for a negative effect ( $P = 0.12$ ). There was also no interaction of parity and treatment in the overall model ( $P = 0.741$ ). Overall, there was a tendency ( $P = 0.068$ ) for an effect of treatment on P/AI with an increase from 33.3% in Ovsynch compared with 36.1% in PROCEPT (2.8% absolute increase; 8.5% relative increase [2.8/33.3]). There was no effect of treatment in primiparous cows (37.1% [99/267] vs. 38.2% [99/259]; Ovsynch vs. PROCEPT;  $P = 0.393$ ). However, there was a tendency for an effect of treatment in multiparous cows (32.0% [259/809] vs. 35.4% [288/813]; 10.7% relative increase;  $P = 0.073$ ). When multiparous cows were evaluated by parity, it was found that there was an effect of treatment in cows of second and third lactation ( $P = 0.047$ ) but no effect of treatment in older cows ( $P = 0.517$ ). Thus, treatment with a second PGF during the Ovsynch protocol tends to increase pregnancies per TAI producing about 10% more pregnancies per TAI in multiparous cows, with a significant increase in second and third lactation cows.

**Key Words:** Ovsynch, prostaglandin F<sub>2α</sub>, fertility

### **M240 Efficacy of different fixed-time AI protocols using GnRH, estradiol, and progesterone in lactating dairy cows.**

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This study compared the reproductive efficiency of dairy cows submitted to fixed-time AI (FTAI) protocols using GnRH and/or different esters of estradiol. A total of 1,029 lactating Holstein cows (358 primiparous and 671 multiparous; BCS 2.99 ± 0.01, 159.8 ± 3.4 DIM; 36.4 ± 0.29 kg/d of milk; LS means ± SEM) were housed in freestall barns and randomly assigned to 1 of 4 treatment groups using a completely randomized design with a 2 × 2 factorial arrangements of treatments. Cows were assigned to 1 of 2 treatments at the beginning of the FTAI protocol (GnRH [G] or estradiol benzoate (EB)) and 1 of 2 treatments at the end of the protocol (EB or estradiol cypionate [ECP]), resulting in: G-EB (n = 263), G-ECP (n = 277), EB-EB (n = 259) and EB-ECP (n = 230). All cows received an intravaginal progesterone (P4) implant at the start of the protocol (d 0) and it was removed on d 8. All cows also received PGF<sub>2α</sub> (500 µg) on d 7 and d 8. At the end of the protocol, treatment with ECP was on d 8 or treatment with EB was on d 9 with FTAI on d 10. Statistical analyses were performed using Proc GLIMMIX of SAS 9.3. There were no interactions between treatments on pregnancies per AI (P/AI) at 30 and 60 d pregnancy diagnoses ( $P > 0.05$ ). At 30 d, there was no effect of EB or GnRH at start of protocol (33.9 vs. 37.0%;  $P = 0.30$ ) and no effect of treatment at end of protocol (33.1 vs. 37.9%; EB vs. ECP;  $P = 0.11$ ). At 60 d, there were no treatment effects at start (29.8 vs. 32.2%; EB vs. GnRH;  $P = 0.43$ ) or end (30.3 vs. 31.7%; EB vs. ECP;  $P = 0.63$ ) of the protocol. For pregnancy loss (d 30 to 60 of pregnancy), there were no interactions ( $P > 0.05$ ) and no effect of treatments at start ( $P = 0.72$ ), however, there was an effect of treatment at end (8.6 vs. 15.7%; EB vs. ECP;  $P = 0.04$ ) of the protocol. Independent of treatments, DIM tended to affect P/AI at 30 d ( $P = 0.08$ ) and at 60 d (34.8 vs. 27.4%, low vs. high DIM;  $P = 0.01$ ), but did not affect pregnancy loss ( $P = 0.05$ ). We concluded that in lactating Holstein cows, the use of EB or GnRH at the beginning or EB or ECP at the end of the protocol provided same P/AI at 30 and 60 d after FTAI, despite that ECP, at the end of protocol, provided greater embryonic/fetal loss. Also, cows with less DIM had greater P/AI at 60 d. Thanks to CNPq, FAPESP, CAPES and VALLÉE.

**Key Words:** fertility, synchronization.

### **M241 The use of a modified 5-day CIDR-Cosynch protocol for resynchronization in lactating dairy cows.** J. A. Spencer\*<sup>1</sup>, K. G. Carnahan<sup>1</sup>, B. Shafii<sup>1</sup>, C. Claypool<sup>1</sup>, J. C. Dalton<sup>2</sup>, and A. Ahmadzadeh<sup>1</sup>, <sup>1</sup>University of Idaho, Moscow, ID, <sup>2</sup>University of Idaho, Caldwell, ID.

Approximately 60% of lactating Holstein dairy cows fail to conceive to the first artificial insemination (AI) and are required reinsemination. Resynchronization facilitates timely reinsemination; however, pregnancy rates to the second AI (PR/AI) from resynchronization protocols have been inconsistent. The development of an effective resynchronization protocol is needed to improve PR/AI to second AI. The objective of this study was to determine the effect of the initial GnRH on PR/AI using a 5-d CIDR-Cosynch resynchronization protocol for second AI in lactating Holstein dairy cows. Approximately 37 d after first AI and upon non-pregnancy diagnosis, cows eligible for second AI (n = 429) were subjected into a modified 5-d CIDR-Cosynch protocol. On d 0, all cows received CIDR inserts and were assigned randomly to one of 2 treatments, initial GnRH (100µg; n = 226) or no GnRH (n = 203). On d 5, CIDR inserts were removed and all cows received one injection of PGF<sub>2α</sub> (500 µg). From d 5 to 7, cows were monitored daily for estrus. Animals detected in estrus on d 6 or 7 received AI and were not given GnRH. On d 8, all cows not identified in estrus on d 6 or 7 were timed AI (TAI) and received GnRH. Blood samples were collected from cows (n = 184) on d 0 and analyzed for progesterone (P<sub>4</sub>) concentration. Logistic



regression was used to examine the main effects of treatment, day of AI, sire, parity and all 2-way interactions with treatment on PR/AI. There were no effects of treatment (no GnRH, 27.1% vs. initial GnRH, 21.2%) on PR/AI ( $P > 0.10$ ). There were also no effects of method of AI (based on estrus vs. TAI), sire ( $n = 10$ ), or any 2-way interactions with treatment on overall PR/AI. There was a significant difference ( $P < 0.05$ ) between primiparous ( $n = 119$ ) and multiparous cows ( $n = 310$ ) in PR/AI (31% vs. 21%). Progesterone concentrations ( $n = 184$ ) were categorized into high ( $\geq 1$  ng/mL) and low ( $< 1$  ng/mL) groups. Overall, PR/AI tended ( $P = 0.09$ ) to be greater for high  $P_4$  ( $n = 136$ ) compared with low  $P_4$  ( $n = 48$ ) (26% vs. 16%). These results provide evidence that the initial GnRH injection in a 5-d CIDR-Cosynch resynchronization program may not be necessary to achieve a similar PR/AI following second AI.

**Key Words:** dairy cows, resynchronization, 5-d CIDR-Cosynch

**M242 Assessment of an application for mobile devices developed to evaluate the AI procedure.** Santiago Bas\* and Gustavo M. Schuenemann, *Department of Veterinary Preventive Medicine, College of Veterinary Medicine, The Ohio State University, Columbus, OH.*

Improving reproductive performance in dairy cattle is paramount to maximize dairy productivity and sustainability. Artificial insemination (AI) is the most common breeding practice for dairy cattle in the US and worldwide. It is common to observe large within- and among-herd variation in conception rate and performance of AI technicians. The objective was to assess a user-friendly application (APP) for mobile devices developed to allow the evaluation of the AI procedure under field conditions. The APP was designed to collect information during the AI procedure to allow the identification of breeding practices (e.g., semen handling, breeding procedures) that may affect reproductive outcomes. In addition, the APP generates a database for benchmarking metrics associated with performance of AI technicians. Novel components of the APP include: (1) login screen for individual users, (2) capture of farm information (e.g., type of facility, herd size), (3) sire identification, (4) semen used (sexed or conventional), (5) breeding strategies (e.g., estrous detection, synchronization programs), (6) animals inseminated (e.g., cows, replacement heifers), and (7) recording of the following metrics: number of semen straws simultaneously thawed, thawing time, AI catheter loading time, time from loading of AI catheter to the cow, time to AI, and time elapsed for each round of insemination. In addition, information is summarized to generate an electronic report, and all captured data are uploaded into a cloud database. The APP features a user-friendly interface for entering and managing data, and accurately (100%) captured, summarized and integrated multiple events associated with the AI procedure under field conditions.

**Key Words:** dairy, app, artificial insemination

**M243 Estradiol and GnRH on ovulation induction in crossbred cows estrus synchronized.** Fabrício Albani Oliveira, Jurandy Mauro Penitente Filho, Erly Luisana Triana Carrascal, Carlos Thiago Oliveira, Adriana Moreira Zolini, Italo Augusto da Costa Soares, and Ciro Alexandre Alves Torres\*, *Federal University of Viçosa, Viçosa, MG, Brazil.*

Three different ovulation inductors, estradiol benzoate (EB), estradiol cypionate (EC) and gonadotropin releasing hormone (GnRH) were evaluated in timed artificial insemination (TAI) of lactating crossbred cows. Two hundred and 16 (216) cows were used in 2 experiments. In experiment I ( $n = 45$ ), follicular dynamics and in the experiment II ( $n$

$= 171$ ) pregnancy per AI were evaluated. All cows were submitted to a synchronization protocol: d 0 (D0) insertion of a progesterone-release intravaginal device (PRID) plus IM injection of EB 2 mg; D8, PRID removal plus IM injection of PGF<sub>2a</sub> (0.150 mg) and eCG (400 IU). The cows were then randomly assigned into 1 of 3 groups: (1) EB9 ( $n = 61$ ): IM injection of EB (1 mg) on D9, and AI 52 h after PRID removal; (2) EC ( $n = 58$ ): IM injection of EC (1.0 mg) on D8 and AI 48 h after PRID removal and (3) GnRH ( $n = 52$ ): IM injection of GnRH (100  $\mu$ g) on D10 and AI 52 h after PRID removal. Ultrasonographic examinations of the ovaries were performed on d 0, 8, and on the day of TAI to determine follicular diameter and ovulation. Follicular data were analyzed using ANOVA and pregnancy per AI with Chi-squared test at 5% probability. No interaction was found between treatment and observation day on number of follicles or on the follicular classification ( $P > 0.05$ ). The intervals from TAI to ovulation were 12.4, 12.8 and 14.8 for EB, EC and GnRH, respectively ( $P > 0.05$ ). No interaction between treatment and day of observation was detected for diameter of the largest follicle (11.4, 11.0 and 10.7 mm for EB, EC and GnRH, respectively;  $P > 0.05$ ). The rate of follicle growth was slower in the EC cows than the other treatments (EB = 1.5, EC = 0.7 and GnRH = 1.9 mm;  $P < 0.05$ ). There were no effects ( $P > 0.05$ ) of treatment on ovulation rate (EB = 100, EC = 93.3, GnRH = 93.3%) and pregnancy per AI (EB = 62.3, EC = 37.9, GnRH = 51.9%). Sample size might have limited the ability to detect statistical differences among treatments. The use of EB, EC, and GnRH as ovulation inductors in TAI protocols in crossbred cows resulted in no differences in ovulatory responses or pregnancy per AI.

**Key Words:** ovulation, crossbred cows, timed AI

**M244 Effect of estrous synchronization program, season, body condition score and ovarian status on pregnancy rate to fixed-time AI in beef cows.** Federico Randi\*<sup>1,2</sup>, Mervyn Parr<sup>2</sup>, Peter Doolan<sup>2</sup>, Michael G. Diskin<sup>2</sup>, Alessio Valenza<sup>3</sup>, Pedro Rodriguez<sup>3</sup>, Pat Lonergan<sup>1</sup>, and David A. Kenny<sup>2</sup>, <sup>1</sup>University College Dublin, Dublin, Ireland, <sup>2</sup>AGRIC Teagasc Grange, Dunsany, Meath, Ireland, <sup>3</sup>CEVA Sante Animale, Libourne, France.

Reproductive efficiency in the Irish beef cow population is challenged by age at first calving, long calving intervals and limited AI usage. Low average herd size and often part-time nature of beef production increases the challenge of detecting cows in estrus. The objective was to establish an estrous synchronization regimen to facilitate fixed-time AI (TAI) in beef cows under Irish conditions. A total of 1410 suckled cows located on 61 farms were enrolled across 2 replicates (Spring,  $n = 703$  and Autumn,  $n = 707$ ). Presence of a corpus luteum (CL) and body condition score (BCS) were recorded at treatment initiation. Following a voluntary waiting period of 35 d postpartum, cows were assigned to (1) receive progesterone-releasing intravaginal device (PRID Delta) on d 0 and PRID removal on d 7 with an 25 mg injection of PF2 $\alpha$  (Enzaprost) (Group 1); (2) same as Group 1, but cows received a 100  $\mu$ g of GnRH analog (Ovarelin) on d 0 (Group 2); (3) same as Group 2, but cows received 400 IU equine chorionic gonadotropin (eCG) (Syncrestim) on d7; (Group 3). All cows received GnRH at TAI 72 h after PRID removal with pregnancy diagnosis by transrectal ultrasonography 35–40 d later. Data were analyzed using the GENMOD procedure of SAS. Presence of a CL (Spring 50.6%; Autumn 70.0%) increased pregnancy rate independent of treatment and season ( $P = 0.03$ ). Mean BCS in Spring and Autumn was not different but BCS positively affected pregnancy rate independent of treatment or season ( $P = 0.003$ ). There was a significant treatment  $\times$  season interaction for pregnancy rate ( $P = 0.0002$ ). In spring, overall pregnancy rate was 59.1% (416/703) and was affected by treatment (49.6 vs. 59.3 vs. 68.5%, for Groups 1, 2 and 3, respectively  $P <$

0.05). In contrast, in autumn, overall pregnancy rate (52.6%, 364/707) was unaffected by treatment (53.7 v 52.0 v 48.7%, respectively). In conclusion, GnRH at the initiation of a TAI synchronization protocol and eCG both increased pregnancy rate in spring-calving suckled beef cows. This effect was not evident in autumn-calving cows. Seasonal differences in outcome may reflect differences in management (grazing vs. confinement), days postpartum or breed type and remain to be elucidated.

**Key Words:** bovine, fertility, synchronization

**M245 Body weight loss of cows early postpartum is associated with negative effects on estrous expression.** Tracy A. Burnett\*, Muhammad A. Khan, Marina A. G. von Keyserlingk, and Ronaldo L. A. Cerri, *University of British Columbia, Vancouver, BC, Canada.*

The aim of this study was to determine the association of weight loss during the transition period with the expression of estrus. Sixty-seven Holstein cows were enrolled onto the study 2 wk before parturition. Body weight (BW) was determined 1 and 2 wk before expected calving date and again 5 wk postpartum. Beta-hydroxybutyrate (BHBA) and glucose concentrations were analyzed 2 wk postpartum. Cows were equipped with collar-mounted activity monitors (Heatime, SCR Engineering, Israel). Estrous expression was measured by an automated activity monitor (AAM) and quantified using 2 parameters: 1) peak activity and 2) duration of the estrus episode. Peak activity was defined as the maximum activity index during an estrus episode; the threshold activity was set at an index of 35 (an 80% increase relative to baseline). The duration of an estrus episode was calculated as the amount of consecutive time whereby the index level was greater than 35. Loss of BW from 2 wk prepartum until 5 wk postpartum was determined for all cows and used to retrospectively group them: (1) those that lost less than or equal to 100 kg of BW (Low; n = 46), and (2) BW loss more than 100 kg (High; n = 21). Data were analyzed by ANOVA using the MIXED procedure of SAS. Increased BW loss was associated with decreased estrous expression as measured with an AAM. Animals within the High group had decreased peak activity ( $63.6 \pm 3.0$  vs.  $74.0 \pm 3.9$  peak activity index;  $P = 0.04$ ) and decreased duration of estrus ( $8.4 \pm 0.7$  vs.  $11.5 \pm 0.9$  h;  $P < 0.01$ ). Ovulatory follicle diameter and estradiol concentration in plasma were unaffected. Additionally, cow within the High group had increased BHBA concentrations ( $0.57 \pm 0.12$  and  $0.90 \pm 0.10$  mmol/L;  $P = 0.03$ ) and reduced glucose concentrations ( $48.3 \pm 1.5$  vs.  $53.8 \pm 1.6$  mmol/L;  $P = 0.02$ ) at 2 wk postpartum. Also, the High group had increased days open in comparison to the Low group ( $163 \pm 13$  vs.  $113 \pm 17$  d open;  $P = 0.03$ ). In conclusion, excessive BW loss early postpartum has negative effects on estrous expression as measured by an AAM and is associated with increased days open.

**Key Words:** activity monitor, body weight loss, estrus

**M246 Automated detection of estrus using multiple commercial precision dairy farming technologies in synchronized dairy cows.** L. M. Mayo\*, W. J. Silvia, G. Heersche, I. C. Tsai, B. A. Wadsworth, A. E. Stone, and J. M. Bewley, *Department of Animal and Food Sciences, University of Kentucky, Lexington, KY.*

The objective of this study was to evaluate precision dairy farming technologies (PDFT) for estrous detection. Estrus was synchronized in 24 lactating Holstein dairy cows using a modified G7G-Ovsynch protocol (last GnRH injection withheld to permit expression of estrus) beginning 45–85 DIM. Resumption of ovarian cyclicity at enrollment, presence of a corpus luteum (CL) on the day of the final injection (designated experimental d 0), regression of the CL by d 5, and presence of

a new CL on d 11 were verified by transrectal ultrasonography. Cows were observed for estrous behaviors for 30 min, 4× per day, on d 2 to 5. Blood samples were collected on d -2, -1, 0, 1, 2, 5, 7, 9, and 11 to quantify progesterone to verify luteal regression and ovulation. Potential periods of estrus (gold standard) were defined by the temporal pattern of progesterone ( $>1.0$  ng/mL on days -2, -1 and 0,  $<1.0$  ng/mL on d 2 and  $>1.0$  ng/mL on d 9 and 11). Eighteen cows followed this pattern. Cows that failed to follow the pattern served as negative controls (n = 6). Detection of estrus by PDFTs, an estrous behavioral scoring system, and by visual observation of standing estrus were compared with the gold standard (Table 1). Sensitivity and specificity for detection of estrus was similar among all PDFT. Only 56% of cows that ovulated were observed standing by visual estrous detection. All systems tested are capable of detecting estrus at least as effectively as visual observation.

**Table 1 (Abstr. M246).** Detection of estrus using alerts generated by PDFT<sup>1</sup> and visual observation (n = 24)

Detection method	TP	FP	TN	FN	Sensitivity (%)	Specificity (%)
Afimilk AfAct Pedometer Plus	16	0	6	2	89	100
GEA CowScout (leg version)	15	0	6	3	83	100
ENGSTrack A Cow	14	0	6	4	78	100
Agis SensoOr	14	0	6	4	78	100
Estrus behavioral score <sup>2</sup>	12	1	5	6	67	83
Standing behavior	10	0	6	8	56	100

<sup>1</sup>Sensitivity = TP/(TP + FN) and specificity = TN/(TN + FP); where TP = true positive, TN = true negative, FP = false positive, and FN = false negative.

<sup>2</sup>Scoring system as defined by Van Eerdenburg et al. (1996).

**Key Words:** automated estrus detection, precision dairy technology, estrous behavior

**M247 Using the Draminski Estrous Detector in the dry cow to measure electrical resistance of vaginal mucus as it relates to animal factors pre- and postcalving.** Caitlin L. Widener\*, William M. Graves, and Jillian F. Bohlen, *University of Georgia, Athens, GA.*

Poor uterine health can be detrimental to a fresh cow's ability to efficiently re-breed. This 2-phase study aimed to establish changes in vaginal mucus resistance as it relates to calving date and uterine health post-calving. In phase one, nulliparous heifers (n = 11) and multiparous cows (n = 21) were examined approximately 2 weeks before and the week of calving with a device designed to measure changes in electrical resistance in ohms ( $\Omega$ ) of cervical/cranial vaginal mucus (Draminski Estrous Detector, Poland). Three consecutive readings were taken at each time point with the average being utilized for analysis. At time of Draminski readings, urine pH, rectal temperatures, body condition scores and udder edema levels were recorded. Increasing blood-flow and uterine contractions associated before calving as the result of estrogen may change the deposition of circulating minerals in mucus, as indicated by electrical resistance, which may have an impact on the uterine environment post-calving. In phase 2, 10 heifers and 14 cows from the original groups were assessed for incidence of metritis on d 7 and 14 ( $\pm 3$  d) post-calving. Vaginal mucus was visually examined using the Metricheck (Simcro, New Zealand) and subjectively scored by one individual (0-clear, 1-up to 25% white or off white pus, 2-25–75% pus, 3- > 75% pus). Draminski readings averaged  $183 \pm 28.8\Omega$  and  $184 \pm 42.8\Omega$  for the first and second readings, respectively. The Draminski readings did not consistently increase (n = 12) or decrease (n = 15) relative to calving date. Precalving data were analyzed using the PROC GLM procedure of SAS for their relationship to postcalving

mucous scores. The first model included overall increase or decrease in Draminski readings relative to calving, rectal temperature and body condition score with group (heifer or cow) held constant. The second model included the first or second Draminski reading with group held constant. There was no association between actual Draminski readings ( $P > 0.05$ ) or changes in Draminski readings over time ( $P > 0.05$ ) and the Metriceck scores postcalving. The lack of statistical associations was likely the result of high variability in Draminski readings.

**Key Words:** electrical resistance, metritis, periparturient

#### **M248 Estrus lying behavior of Holstein cows: Risk factors for estrus expression, ovulation risk and pregnancy per AI.**

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Objectives were (1) to quantify lying behavior (LB) changes around estrus, and (2) to assess risk factors for reduced changes in LB during estrus, ovulation risk and pregnancy per AI (P/AI). Holstein cows ( $n = 1,039$ ;  $45.6 \pm 10.7$  kg milk/d) were fitted with pedometers; 1,179 estrus episodes were evaluated. LB measurements were bout frequency (BOUT), total lying time/d (TOTAL), average lying bout duration (AVG), and ratio of lying/standing time (RATIO). Relative changes from d -7 to d -1 (d of estrus) and from d -7 to d +1 were calculated. Thresholds for high intensity estrus were set as a decrease in BOUT  $>25\%$  and in TOTAL and RATIO  $>22\%$ . AVG did not change with estrus. Descriptive statistics and logistic regressions were used to analyze the data. At d -1 BOUT, TOTAL, and RATIO were, respectively,  $19 \pm 34\%$  ( $-3 \pm 5$  bouts/d),  $20 \pm 30\%$  ( $-145 \pm 190$  min/d), and  $21 \pm 26\%$  ( $-11 \pm 21\%$ ) lower than at d -7. Major risk factors associated with reduced estrus expression were d 0 progesterone (P4) concentration, season and parity. Cows with  $P4 < 0.2$  ng/mL were twice as likely to have high intensity estrus (large decrease in LB from d -7 to d -1). High intensity estrus increased ovulation rate (OR = 2.7 [BOUT], 4.6 [TOTAL], 4.8 [RATIO]). Cold season also increased ovulation rate (OR = 1.8 [BOUT] and 1.9 [TOTAL and RATIO]). BOUT, TOTAL, and RATIO tended to affect P/AI at 32 and 60 d, but parity, BCS, and season were more important predictors. Interestingly, TOTAL, RATIO, and AVG were greater at d +1 than at d -7 by  $8 \pm 30\%$  (TOTAL),  $10 \pm 21\%$  (RATIO), and  $13 \pm 36\%$  (AVG). When d +1 TOTAL and RATIO were  $\geq 10\%$  d -7 values, there was greater likelihood of ovulation (OR = 2.6 [TOTAL] and 2.4 [RATIO]; 92% vs. 81%). P/AI at 32 d was 1.5 and 1.6 times greater for episodes followed by high d +1 TOTAL and RATIO (38% vs. 27%). BOUT, TOTAL and RATIO could improve the use of information from activity monitors (e.g.: increased estrus detection, fertility prediction). Relationships between P4 at AI, estrus behavior, ovulation and conception should be further investigated.

**Key Words:** estrus, lying behavior, progesterone

#### **M249 Characterization of ovarian function in nonpregnant previously inseminated lactating dairy cows.**

Robert Wijma\*, Matias L. Stangaferro, and Julio O. Giordano, Department of Animal Science, Cornell University, Ithaca, NY.

Our objective was to characterize ovarian dynamics in lactating dairy cows diagnosed nonpregnant after AI focusing on time points when resynchronization programs are initiated or PGF2 $\alpha$  can be used to induce estrus expression. After synchronization with the Presynch-Ovsynch

(PGF-14d-PGF-12d-GnRH-7d-PGF56h-GnRH-16h-TAI) protocol cows ( $n = 64$ ) received timed-AI. Blood was collected every 48 h from 14 to 42 d after AI to determine concentrations of progesterone (P4) and estradiol (E2). Daily, ovarian ultrasound (US) was used to determine size and location of all ovarian structures. Retrospectively, cows were classified as pregnant ( $n = 25$ ; PG) when a viable embryo was observed (excluded from analysis) or nonpregnant ( $n = 39$ ; NP) when no viable embryo was observed. A cow was considered to have an active CL (ACL) when  $P4 > 1$  ng/mL and  $\geq 1$  CL was observed. Also, a CL was considered mature (MCL) starting 7 d after ovulation based on the timing when the CL becomes sensitive to PGF2 $\alpha$ . Follicles were considered active (AFOL) when reached 10 mm until 2 consecutive days of reduction in diameter. Periods evaluated were: 24(D24), 32(D32), and 38(D38) d after AI. Parameters evaluated were cows with ACL, MCL, and an AFOL by day, E2 concentrations and E2 to P4 ratio. Quantitative data were analyzed by ANOVA using PROC MIXED whereas binary data were analyzed by logistical regression using PROC GLIMMIX of SAS. More ( $P < 0.01$ ) cows had an ACL on D32 (82.1%) and D38 (84.2%) than on D24 (35.9%). The greatest ( $P < 0.01$ ) proportion of cows with a MCL was for D38 (84.6%) followed by D32 (56.4%) and least for D24 (28.2%). The proportion of cows with an AFOL tended ( $P = 0.06$ ) to be affected by day with 79.5, 92.3, and 97.4% of cows with an AFOL for D24, D32, and D38, respectively. Concentrations of E2 were greater ( $P = 0.05$ ) for cows with an AFOL ( $1.1 \pm 0.3$  pg/mL) vs cows without AFOL ( $0.5 \pm 0.1$  pg/mL) across all days. Concentrations of E2 were affected by day because on D24 ( $1.3 \pm 0.2$  pg/mL) E2 was greater ( $P < 0.05$ ) than for D32 ( $0.7 \pm 0.2$  pg/mL) whereas on D38 ( $1.0 \pm 0.2$  pg/mL) E2 was intermediate. We conclude that there is more variation in ovarian function than expected based on the number of days after AI in nonpregnant previously inseminated cows. Supported by Hatch project NYC-127813.

**Key Words:** corpus luteum, follicle, dairy cow

#### **M250 The effects of ovulatory status of the dominant follicle and spatial relationship of the corpus luteum on diameter and average growth rate of that dominant follicle.**

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The aim of this study was to investigate the effects of ovulatory status of the dominant follicle (whether the follicle eventually ovulated or not) and spatial relationship of the corpus luteum (CL; contralateral or ipsilateral to the dominant follicle) on the diameter and average growth rate of that dominant follicle. Sixteen non-pregnant, multiparous beef cows were observed daily for estrus. Cows were randomly assigned to 1 of 2 treatments, daily ultrasound of: (1) the dominant anovulatory (AN) follicle (1st follicular wave), or (2) the ovulatory (OV) follicle (2nd or 3rd follicular wave). Ultrasonography exams were initiated on d 1 (AN treatment group) or on d 7 (OV treatment group) of the estrous cycle. Exams ended after 5 d of follicular dominance. Follicle diameter and whether the CL was ipsilateral or contralateral to the dominant follicle were recorded. The MIXED procedure of SAS was used for data analysis, with d of dominance (0 to 4) as a repeated measure; LSMeans and pooled SEM are reported. There was a treatment  $\times$  spatial relationship interaction ( $P = 0.056$ ) when evaluating average growth rate of the dominant follicle. Anovulatory follicles contralateral to the CL grew faster ( $1.137 \pm 0.045$  mm/d) than both ovulatory follicles contralateral to the CL ( $0.954 \pm 0.036$  mm/d) or ovulatory follicles ipsilateral to the CL ( $0.996 \pm 0.029$  mm/d). However, average growth rate of the dominant follicle was less ( $P = 0.041$ ) in cows in the OV group compared with cows in the AN group ( $0.975 \pm 0.023$  and  $1.07 \pm 0.040$  mm/d, respec-



tively). When evaluating diameter of the dominant follicle over the 5 d of dominance, treatment, spatial relationship of the CL, d of dominance, and treatment  $\times$  spatial relationship all affected diameter of the dominant follicle. However, when only evaluating the last d of dominance, these parameters did not significantly affect diameter of the dominant

follicle. In conclusion, additional research is necessary to understand the interactions between future ovulatory status of a dominant follicle and spatial relationship of the CL.

**Key Words:** corpus luteum, follicle diameter, follicle growth rate

# Production, Management and the Environment I

**M251 Canadian dairy producers' transition to automatic milking systems: preliminary results of a national survey.** Christina Tse\*<sup>1</sup>, Trevor DeVries<sup>2</sup>, Elsa Vasseur<sup>3</sup>, Herman Barkema<sup>1</sup>, and Ed Pajor<sup>1</sup>, <sup>1</sup>Department of Production Animal Health, University of Calgary, Calgary, AB, Canada, <sup>2</sup>Department of Animal and Poultry Science, University of Guelph, Kemptville, ON, Canada, <sup>3</sup>Organic Dairy Research Centre, University of Guelph, Alfred, ON, Canada.

Usage of automatic milking systems (AMS) is becoming increasingly common in Canada. The purpose of this study was to describe producers' transition to AMS and document their satisfaction. A total of 149 AMS producers were surveyed in 9 provinces by telephone and email. The average number ( $\pm$ SD) of robotic units/farm was  $2 \pm 1$ . The median AMS age was 32 mo, with a range of 4 to 170 mo. Of the respondents, 88% of farms used a free-flow traffic system, while 12% used a directed traffic system. Results show that 56% of producers built a new barn and 50% changed housing systems for the transition. Concerning animal training programs, 21% of producers trained cows before first milking with the robot and 38% used a training program for heifers. It took, on average, one week to train cows ( $7.4 \pm 6.0$  d) and heifers ( $7.7 \pm 5.9$  d), but  $2.7 \pm 0.4$  mo for the entire herd to adapt to the robot. Sixty percent of farms had cows that were not able to adapt to the robot. The median proportion of the herd culled for not adapting was 1% (with a range of 0 to 40%). The average number of lactating cows increased from  $92 \pm 56$  to  $102 \pm 68$ . The total hours devoted to milking/day, which included moving/fetching cows, milking time, preparation and cleaning of milking system, decreased by 40%. Milking frequency averaged  $3.1 \pm 0.4$  milkings/day. The majority (84%) of surveyed farms experienced an increase in milk yield from their previous milking system. The median bulk tank somatic cell count (SCC) was 180,000 cells/mL, with a range of 43,000 to 375,000 cells/mL. Changes in bulk tank SCC were variable with producers reporting increases, decreases or no change. Producers also reported an increase in time flexibility, life quality and a willingness to recommend AMS to other dairy producers. Transitioning to an AMS requires effort in training cows and learning a new management system. Producers reported that AMS have met their economic and lifestyle expectations, while improving animal welfare.

**Key Words:** automatic milking, dairy producer, transition

**M252 A stochastic dairy farm model to evaluate the effect of changes in technical or economic factors on performance and benefits.** Sergio Calsamiglia\*<sup>1</sup>, Susana Astiz<sup>2</sup>, Lorena Castillejos<sup>1</sup>, Carlos Lopez de Toro<sup>1</sup>, and Joaquim Baucells<sup>3</sup>, <sup>1</sup>Universitat Autònoma de Barcelona, Bellaterra, Spain, <sup>2</sup>INIA, Madrid, Spain, <sup>3</sup>Centre Veterinari Tona, Tona, Spain.

Dairy farms are constantly challenged with decisions. The evaluation of the outcome of such decisions requires the integration of technical and economic effects over time. The complexity of the multiple interactions can be resolved with the use of mathematical models that integrate biological and economical processes. The objective of the study was to develop a stochastic model to simulate technical and economic consequences of changes implemented in dairy farms. Individual cows of a dairy farm are uploaded. Data on reproduction, diseases, economics, labor, finances, management decisions, facilities and environment are introduced (default values are provided). Farm performance is modeled over a period and technical and economic performance is provided in the output. An 810 dairy cow farm (10,800 L/cow/year), 205 DIM, 15%

pregnancy rate with average incidences of diseases and prices was used as a model. Three scenarios were tested: (1) CON = No change in the current conditions; (2) CULL = Increase in voluntary culling rate by 5 percentage units (31 vs. 36%); and (3) SEMEN = Change from the use of low cost semen vs. high cost semen (€10 vs. €20) assuming that the use of the more expensive semen results in an increased genetic potential of 2% in the daughters. A simulation of 10 years was repeated 40 times with stable technical and economic conditions. At the end of the 10-year simulation, CULL increased benefits by 14%. Treatment SEMEN resulted in reduced income for the first 5 years, probably due to the increased cost of semen and the time required for the expression of the genetic improvement. After the 10-year simulation, SEMEN accumulated a 20% loss, although in the year 10 of simulation SEMEN had a 16% higher benefit. It took 13 years to reach the breakeven point for SEMEN. Although genetic improvement cannot be ignored in dairy farms, the benefits are long-term. In contrast, CULL resulted in short and medium term benefits.

**Key Words:** model, dairy farm, economic performance

**M253 Changes in temperature-humidity index and number of hot days related to heat stress of dairy cattle in Thailand.** Thirarat Sae-tiao<sup>1</sup>, Skorn Koonawootrittriron\*<sup>1</sup>, Thanathip Suwanasopee<sup>1</sup>, and Mauricio A. Elzo<sup>2</sup>, <sup>1</sup>Kasetsart University, Bangkok, Thailand, <sup>2</sup>University of Florida, Gainesville, FL.

Global climate is changing and becoming warmer. Higher ambient temperatures and humidity increase stress and lower dairy cattle production, particularly in tropical countries. Temperature-humidity index (THI) has been used to assess the risk of heat stress in dairy cattle. When THI exceeds 72, cows are likely to begin experiencing heat stress. The objective of this research was to characterize yearly changes in THI and number of days that THI exceeded 72 (NHD) in Thailand from 2002 to 2014. Average daily temperatures and relative humidity were measured in 17 meteorological stations located in 5 regions across Thailand ( $n = 78,713$ ). Regions were Northern, Northeastern, Central, Eastern, and Southern. The overall yearly means and SD were 81.9 and 4.2 for THI and 361.5 d and 13.8 d for NHD, and the corresponding ranges were 58.7 to 93.2 for THI and 166 to 366 d for NHD. The THI and NHD were analyzed using a model that included year, region and interaction between year and region as fixed effects, and residual as a random effect. All fixed effects influenced THI and NHD ( $P < 0.01$ ). Yearly LSM ranged from 79.3 (Northeastern-2011) to 84.1 (Central-2010) for THI, and from 315 (Central-2002) to 366 d (Southern-2012) for NHD. Trends across years were non-significant for THI ( $b = -0.003$  units/yr;  $P = 0.93$ ) and for NHD ( $b = 0.39$  d/yr;  $P = 0.34$ ). However, larger fluctuations in THI existed from 2009 to 2014 than from 2002 to 2008 (81.3 to 83.3 vs. 81.5 to 82.4). Conversely, NHD showed similar ranges in both periods (349 to 364 d vs. 341 to 357 d). The LSM for THI and NHD were higher in the Central (83.0 and 354 d), Eastern (83.0 and 363 d), and Southern (82.8 and 361 d) regions than in the Northeastern (80.6 and 344 d) and Northern (80.6 and 346 d) regions. Although increasing trends in THI and NHD did not occur during the years of the study, variability in THI increased since 2009. Regional effects indicated that more comprehensive strategies for reduction of heat stress in dairy cattle might be needed in the Central, Eastern, and Southern Thailand than in the Northern and Northeastern parts of the country.

**Key Words:** environment, dairy, tropic

**M254 Checks and balances: Evaluating reliability of dairy nutrient management data to better protect groundwater resources.** Christine Miller\* and Deanne Meyer, *University of California, Davis, Davis, CA.*

To protect groundwater from further nitrate contamination, California regulations prohibit dairy producers from applying more than 140% of the nitrogen (N) that their crops remove. The regulations require copious annual reporting of crop field management, farm infrastructure, and animal population. The data collected in these annual reports could be integral to evaluating and improving both farm practices and the regulations themselves. Data reliability and accuracy must be assessed to use the information responsibly. Annual Reports from 18 dairies were obtained to assess reliability. Mass balance calculations were performed to check the self-consistency of data within a facility. The results of mass balance calculations show that the data do not account for a remarkably large percentage of the nutrients being produced on the farms. Literature suggests that over 60% of N and 90% of P should be recovered; however, a median of only 25% of both N and P in cattle manure was recovered based on annual report data. This could be due to many different causes including inaccurate nitrogen sampling and analysis techniques, systematic reporting errors, or fraudulent reporting. Given that the accuracy of the majority of the recommended sampling and analysis protocols has not been assessed, it is likely that these methods are a significant source of error. Projects that should improve data collection protocols in both the short and long term are in progress. Online decision trees are being developed to help farmers self-assess their current data collection practices, and provide personalized suggestions for improvement. Additionally, I will use a statistical modeling approach paired with field experiments to examine the uncertainty in these recommended protocols (and thereby the overall uncertainty in regulations). By separating the various sources of measurement error, the model will identify the best ways to improve data collection and regulation efficacy. Results of this and future studies will influence future nutrient management regulations in California and other states with active livestock industries.

**Key Words:** dairy waste management, nitrate leaching

**M255 Reproductive performance of North American dairies by geographic region.** Cristian F. Vergara\*<sup>1,2</sup>, Flavio Bitencourt<sup>1</sup>, Lydia M. Johnson<sup>1</sup>, Diego Vallejo<sup>1</sup>, and Hernando Lopez<sup>1</sup>, <sup>1</sup>ABS Global Inc., DeForest, WI, <sup>2</sup>ABS Chile Ltda., Santiago, Chile.

Our objectives were to compare reproductive indicators from Holstein, Jersey and Crossbred dairies in 4 regions of United States of America and establish benchmarks for the 20% most efficient herds. Data from 290,351 cows and their records in DC305 were collected from February to November 2014 from a sample of 162 ABS customers. Regions (herds) represented were: Eastern (E; n = 28), Midwestern (MW; n = 58), Northwestern (NW; n = 10), and Southwestern (SW; n = 66). Univariate analyses were conducted with the MIXED and FREQ procedures of SAS using herd as the experimental unit. The Regions differed ( $P < 0.05$ ) by mean (SE) herd size (E = 1,022 ± 580; MW = 690 ± 537; NW = 4,919 ± 496; SW = 2,760 ± 533) and kg of daily milk production (E = 37.4 ± 1.7; MW = 36.1 ± 1.6; NW = 32.3 ± 1.5; SW = 36.5 ± 1.6). Crossbred were not represented in the South Regions and Jerseys not represented in MW, making this effect significant ( $P < 0.05$ ). The West Regions had a lower use of timed AI ( $P < 0.05$ ) than E and MW (E = 46.1%; MW = 49.1%; NW = 17.6%; SW = 15.4%); but a lower conception rate (CR) ( $P < 0.05$ ) than E and MW Regions (E = 36.6%; MW = 36.9%; NW = 31.5%; SW = 32.7%). Regarding insemination

risk, only MW did lower ( $P < 0.05$ ) than the other 3 Regions (E = 63.8%; MW = 52.8%; NW = 62.3%; SW = 61.8%). As a result, the E Region had the best pregnancy risk (PR) and MW the lowest (E = 22.9%; MW = 18.2%; NW = 20.0%; SW = 20.3%). The West Regions reported a higher ( $P < 0.05$ ) pregnancy loss (E = 11.4%; MW = 10.3%; NW = 16.0%; SW = 13.3%). The 20% most efficient herds based on PR ranking (n=71) had a mean PR of 24%. Breeds were represented in both strata and was not a significant effect ( $P > 0.05$ ). The top 20% had a 10 points higher insemination risk (mean 64%) and a 6 points higher CR (mean 38%) than bottom 80% ( $P < 0.05$ ). Additionally, they breed open cows at pregnancy diagnosis 3 days earlier (mean 10 ds), but did not differ in timed AI usage ( $P > 0.05$ ). The top 20% herds were not different in their daily milk production, voluntary waiting period, and pregnancy loss in comparison with the bottom 80% ( $P > 0.05$ ). Despite regional differences, the best combination of factors led the East Region to achieve the highest PR. The top 20% herds in PR had higher overall reproduction but did not differ in herd management performance with the bottom 80%. Further analysis is needed.

**Key Words:** dairy, reproduction

**M256 Lactation stage alters the rumen protozoal communities in three breeds of primiparous dairy cattle.** L. M. Cersosimo\*<sup>1</sup>, M. Bainbridge<sup>1</sup>, A.-D. G. Wright<sup>2</sup>, and J. Kraft<sup>1</sup>, <sup>1</sup>University of Vermont, Burlington, VT, <sup>2</sup>University of Arizona, Tucson, AZ.

Rumen protozoa are anaerobic ciliates that degrade fiber, starch, and proteins consumed by the host. The objective of our study was to determine if breed (B) or lactation stage (LS) affect the rumen protozoal community. Whole rumen digesta samples were collected at 3 and 93 d in milk (DIM) from 7 Holstein (H), 8 Jersey (J), and 7 Holstein-Jersey crossbreds (X) co-housed in free stalls. To characterize the rumen protozoa, the V3-V4 region of the 18S rRNA gene was amplified with the primers 316F and 758R. Sequence reads were produced by Illumina MiSeq (v.3) and the program Mothur was used for all bioinformatics analyses. The protozoal densities (cells/mL digesta) were determined by real-time PCR amplification of the 18S rRNA gene. The effects of B, LS, or B x LS on rumen protozoa were evaluated with the PROC MIXED model in SAS. At 3 DIM, 307,739 sequence reads clustered at a 4% cutoff into 3–21 operational taxonomic units (OTU) per animal with means of 5, 6, and 7 for H, J, and X, respectively. At 93 DIM, 536,846 sequence reads clustered into 1–15 OTU per animal with an average of 3 OTU for each breed. For both stages, greater than 98% of the sequences clustered with OTU 1, belonging to the family, *Ophryoscolecidae*. The OTU-based diversity measures, Shannon index ( $P < 0.01$ ), Inverse Simpson index ( $P < 0.05$ ), and Chao I estimator ( $P < 0.001$ ) showed greater diversities at 3 DIM. The abundances of protozoal genera *Epidinium* (0.4–9.3% abundance), *Ostracodinium* (3.5–21.6%), *Ophryoscolex* (0.2–3.0%), and *Polyplastron* (2.4–7.1%) were affected by LS ( $P < 0.05$ ). However, no differences in abundance were observed in the genera, *Entodinium* (19.8–36.3%), *Metadinium* (0.8–21.2%), *Isotricha* (0.3–1.7%), or *Diploplastron* (0.7–2.7%). *Eudiplodinium* spp were less abundant in H than in X ( $P < 0.05$ ), respectively, at 3 DIM (H: 2.0, X: 12.2%) and at 93 DIM (H: 2.6, X: 20.3%). Protozoal densities at 3 DIM ( $10^4$ – $10^6$  cells/mL) and 93 ( $10^3$ – $10^6$  cells/mL) DIM did not differ. The present study suggests that early and peak LS, but not B, affects the rumen protozoal community in primiparous dairy cattle.

**Key Words:** ciliates, 18S, diversity



**M257 Quality of hatching eggs from different broiler breeder ages.** Vanessa Michalsky Barbosa\*<sup>1</sup>, Bruno Delphino Medrado<sup>1,2</sup>, Isabela Cláudia Barbosa Santos<sup>1</sup>, Larissa Kiana Santos Azevedo Martins<sup>1</sup>, Tais Pinheiro Borges Silva<sup>1</sup>, Tayana Nery Franca<sup>1</sup>, Sylvania Belo Dourado<sup>1</sup>, Juliana Cantos Faveri<sup>1</sup>, and Lia Muniz Barretto Fernandes<sup>3</sup>, <sup>1</sup>Department of Animal Science, Faculty of Veterinary Medicine and Animal Science, Federal University of Bahia, Salvador, Bahia, Brazil, <sup>2</sup>Federal Institute of Education, Science and Technology–Santa Inês Campus, Santa Inês, Bahia, Brazil, <sup>3</sup>Department of Animal Production, Faculty of Veterinary Medicine and Animal Science, Federal University of Bahia, Salvador, Bahia, Brazil.

Several requirements before and during the artificial incubation process interfere with the success of the day-old chicks production, and the quality of the eggs to be incubated is an important factor to be considered. The aim of this study was to investigate the characteristics of young and old broiler breeder eggs. We studied 200 incubating eggs produced by Cobb broiler breeders aged 29 wk (n = 100) and 62 wk (n = 100). It was used a completely randomized design. The treatments were the age and each egg was considered replication. The eggs were weighed (EW), and also its components: eggshell (ES%), yolk (Y%) and albumen (A%). Specific weight analysis (SW) was determined by the solutions of different densities method. The eggshell resistance (ER) was analyzed by compression strength test. The means of eggshell thickness (ET) and number of pores (PO) were obtained in 3 different regions. Additionally we evaluated albumen pH (pHA), yolk pH (pHY), albumen height (AH), yolk height (YH) and Haugh Units (HU). Data were analyzed using PROC GLM of SAS 9.2 and the means were compared by F test. EW, Y%, pHA were higher ( $P < 0.05$ ) from the old flock as compared with the young flock (72.48 vs 57.91 g; 31.19 vs 26.30%; 8.84 vs 8.64; respectively). ES%, A%, SW, ER, ET, PO, AH, HU were higher ( $P < 0.05$ ) from the young flock as compared with the old flock (8.99 vs 8.71%; 64.70 vs 60.10%; 1.082 vs 1.076 g/mL H<sub>2</sub>O; 2320 vs 2158 g; 0,438 vs 0,428 mm; 122 vs 105 pores/cm<sup>2</sup>; 8,12 vs 7,19 mm; 82,22 vs 71,61 HU; respectively). There was no effect between the treatments for pH<sub>Y</sub> and YH. The hatching eggs quality from 62-wk-old broiler breeder is less than that of 29-wk-old broiler breeder. Due to the influence of the structural characteristics of eggs in embryo development, it is necessary to monitor the quality parameters and performing further studies that aim adapt the conditions of the incubation process according to the egg to be incubated.

**Key Words:** artificial incubation, eggshell, poultry production

**M258 Effects of turning duration during incubation and broiler breeder age on hatchability, chick organs, and poultry performance.** Vanessa Michalsky Barbosa\*<sup>1</sup>, Bruno Delphino Medrado<sup>1,2</sup>, Mariana André Pompeu<sup>3</sup>, Júlia Sampaio Rodrigues Rocha<sup>3</sup>, Edgard Onoda Luiz Caldas<sup>3</sup>, Genilson Bezerra Carvalho<sup>4</sup>, Leonardo José Camargos Lara<sup>3</sup>, and Nelson Carneiro Baião<sup>3</sup>, <sup>1</sup>Department of Animal Science, Faculty of Veterinary Medicine and Animal Science, Federal University of Bahia, Salvador, Bahia, Brazil, <sup>2</sup>Federal Institute of Education, Science and Technology–Santa Inês Campus, Santa Inês, Bahia, Brazil, <sup>3</sup>Department of Animal Science, Veterinary School, Federal University of Minas Gerais, Belo Horizonte, Minas Gerais, Brazil, <sup>4</sup>Department of Animal Science, Veterinary Medicine and Animal Science School, Federal University of Goiás, Goiânia, Goiás, Brazil.

We evaluated in an industrial scale, the effects of turning duration (15, 17 and 19 d) in the incubation phase and Cobb broiler breeder age (33 and 63 weeks) on the hatchability of fertile eggs (HF), the weights of the heart (HW) and yolk sac (YSW) and their relation to the weight

of chicks: (HW/CW and YSW/CW, respectively). The poultry performance was analyzed by chick weights during the housing (BW1), broiler weights at 42 d old (BW42), weight gain from 1 to 42d (WG), feed intake from 1 to 42d (FI), feed conversion from 1 to 42d (FC) and viability from 1 to 42d (VIAB). It was used a completely randomized design. Eighteen trays (replication) with 96 eggs each were used for the evaluation of hatchability. For the organs it were used 36 chicks per treatment, and the chick was considered the replication. To evaluate the poultry performance, we used 6 pens with 25 birds per treatment, with pen considered the repetition. Eggs were set in an incubator Casp CMg 125E multistage. On the 15th, 17th or 19th days of incubation, the cessation of egg turning was performed and the eggs were transferred to the hatcher. The unhatched eggs were analyzed to determine infertility. The male chicks were housed in an experimental stable and the performance parameters were measured weekly. Data were analyzed using PROC GLM of SAS 9.2 and the means were compared by F test. In the incubation phase, we used a split plot design and in the performance phase it was used a factorial design. Compared with old flocks, chicks and broilers from young breeder flocks had lower HW (0.36 vs 0.40 g), YSW (5.28 vs 6.47 g), YSW/CW (12.5 vs 13.5%), BW1 (40.0 vs 44.7 g), BW42 (3047.6 vs 3131.4 g), WG (3007.6 vs 3086.8 g) and FI (4860.8 vs 4989.3), independent of the turning duration, but there were no effects of breeder age on HF, HW/CW, FC, VIAB. There were no effects of turning duration, regardless the breeder age. The turning can be discontinued after 15 d of incubation without negative effects on hatchability, heart weight, yolk sac weight, and poultry performance.

**Key Words:** animal production, day-old chick, embryo development

**M259 Health, milk yield and quality for Parmigiano Reggiano cheese evaluated in cows fed OmniGen-AF from dry-off to 150 days in milk.** Ludovica M. E. Mammi\*, Mattia Fustini, Elena Bonfante, Alberto Palmonari, Giorgia Canestrari, and Andrea Formigoni, Department of Veterinary Medical Sciences, University of Bologna, Ozzano Emilia, Italy.

One hundred ninety-one Holstein and x-bred cows were used to study the effect of feeding OmniGen-AF (OG, Phibro Animal Health, Quincy, IL) from dry-off to 150 d in milk (DIM) on health, milk yield (MY) and milk components for making Parmigiano Reggiano cheese. Cows were assigned randomly to either a control (C, n = 95) or a group fed 55g/h/d of OG (n = 96). Groups were balanced on parity, breed, expected calving date, BCS and previous MY. Cows were housed in comparable pens during the dry and lactation phases with diets fed as TM rations. Cows were milked 2× and MY recorded daily using Afimilk. Individual cow milk was sampled at dry-off, once between DIM 4 and 10 and then every 4 wk to 150 DIM and analyzed for fat, casein, protein, lactose, somatic cell count (SCC), acidity and lactodynamographic analysis (LDG). Health events were recorded. Treated or culled cows (n = 43) were excluded from the statistical analysis for milk production and quality. Milk was analyzed by ANOVA-Repeated Measures, SCC by Kruskal-Wallis test and all data processed using Statsoft Statistica v.10. Significance declared at  $P \leq 0.05$ . MYs were not different, although OG cows tended ( $P = 0.25$ ) to produce more milk (+1.39kg/d) with a lower SCC linear score (−0.06,  $P < 0.10$ ) than C cows. A higher % of milk samples (23%) in C group had SCC > 300,000 cells/mL ( $P < 0.10$ ) compared with OG cows (18.2%). The same trend was observed in cows with 2 or more samples > 300,000 cells/mL (C = 32.4%, OG = 22.9%,  $P = 0.27$ ). Milk composition did not differ between C and OG cows, except milk from OG cows had a tendency ( $P = 0.12$ ) for a better LDG score (OG = 18.24, C = 19.24). Culling rates were higher in the C cows group (12.6%) compared with OG (7.4%) and culling tended

to occur sooner (DIM) for C cows than OG cows (57.3 vs. 102.6,  $P < 0.10$ ). Incidence rates (%) of mastitis and metritis were higher in the C (18.9, 17.9) versus the OG cows (10.4, 15.6). In this study, cows fed OG from dry-off to 150 DIM tended to have fewer diseases and culls, plus a tendency for improved MY and properties associated with quality Parmigiano Reggiano cheese.

**Key Words:** OmniGen-AF, Parmigiano Reggiano, health

**M260 Body weight gain of dairy heifers fed diets with *Origanum vulgare* or *Camellia sinensis* extracts.** Vivian Fischer<sup>\*1</sup>, Giovanni Jacob Kolling<sup>1</sup>, Débora Strider<sup>1</sup>, Dejana Maira Panazzolo<sup>1</sup>, Carolina da Silva dos Santos<sup>1</sup>, Alexandre Mossate Gabbi<sup>1</sup>, Mateus Wanderer<sup>1</sup>, Andress Sopelsa<sup>1</sup>, and Renata Wolf Sune Martins da Silva<sup>2</sup>, <sup>1</sup>Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil, <sup>2</sup>Empresa Brasileira de Pesquisa Agropecuária, Bagé, RS, Brazil.

Biological effects of plant extracts containing essential oils and polyphenols have been extensively studied for humans and nonruminant production animals, but in vivo studies with ruminants in field conditions are relatively scarce. This work aimed to investigate the effect of addition of *Origanum vulgare* or *Camellia sinensis* extracts upon average daily weight gain (ADG) of dairy heifers. Two trials were conducted for 70 d to test the levels of *Origanum vulgare* (0, 2.5, 5, 7.5 g/heifer/day) and *Camellia sinensis* (0, 1, 2, 3 g/heifer/day) extracts. Trial 1 used 32 Holstein heifers with initial overall BW 442.1 ± 96 kg and trial 2 used 34 heifers: 11 Jersey and 23 Holstein, with initial overall BW 305 ± 42 kg. Heifers grazed range pasture (trial 1) and Tifton (*Cynodon dactylon*; trial 2) and were individually supplemented with 2.5 and 1.5 kg of concentrate once a day in the morning, in trials 1 and 2, respectively. Heifers were weighed on day -14 (used as covariate), and afterward, on d 1, 28 and 56 after the inclusion of the extracts into the concentrate. Data of each trial was submitted to variance analysis, using the Mixed procedure of SAS according to a completely randomized design, considering the effect of diet (n = 4), period, interaction diet x period, breed and interaction diet x breed (just for trial 2) and the initial body weight as a covariate. Heifers supplemented with *Origanum vulgare* extract did not change daily weight gain and overall ADG was 0.56 kg. Heifers supplemented with 1 g of *Camellia sinensis* extract had greater ADG ( $P < 0.05$ ) than those fed with zero and 3 g of *Camellia sinensis* extract. Inclusion of 1 g of *Camellia sinensis* extract increased ADG 11%. In this study feeding *Camellia sinensis* at 1g/heifer daily improved ADG of dairy heifers.

**Table 1 (Abstr. M260).**

Item	Trial 1 – <i>Origanum vulgare</i> (g/heifer/day)				P > F	Trial 2 – <i>Camellia sinensis</i> (g/heifer/day)				P > F
	0	2.5	5.0	7.5		0	1	2	3	
ADG (kg/d)	0.62 <sup>a</sup>	0.68 <sup>a</sup>	0.66 <sup>a</sup>	0.73 <sup>a</sup>	NS	0.55 <sup>a</sup>	0.61 <sup>b</sup>	0.57 <sup>ab</sup>	0.52 <sup>a</sup>	*

\* $P < 0.05$ .

**Key Words:** *Camellia sinensis*, *Origanum vulgare*, weight gain

**M261 Effect of cow BCS during gestation on performance variables of the offspring.** Rodrigo Marques\*, Reinaldo Cooke, Murilo Rodrigues, Bruno Cappellozza, and David Bohnert, Oregon State University-EOARC, Burns, OR.

The objective of this experiment was to evaluate the effects of cow BCS during gestation on productive parameters of the offspring. Ninety-

seven Angus × Hereford cows pregnant to AI and to the same sire were assigned to the study based on their BCS during gestation: (1) ADQ = cows with adequate BCS throughout gestation (n = 20), (2) INDQ = cows with inadequate BCS throughout gestation (n = 20), (3) EARLY = cows that gained BCS during the first trimester of gestation and maintained BCS until calving (n = 19), (4) MID = cows that gained BCS during the second trimester of gestation and maintained BCS until calving (n = 19), and (5) LATE = cows that gained BCS during the third trimester of gestation and maintained BCS until calving (n = 19). Upon calving, calf BW was recorded. Approximately 60 d after calving, cow milk production was assessed using the weigh-suckle-weigh technique (12 h calf removal following by 30 min of nursing). Calves were weaned at 7 mo of age. All data were analyzed with the MIXED procedure of SAS. Cow BCS did not change ( $P \geq 0.30$ ) for ADQ and INDQ cows during gestation (average 4.6 and 6.0, respectively) and increased ( $P \leq 0.01$ ; based on BCS collected at AI, beginning of the second and third trimesters of gestation, and calving) for EARLY (4.4, 5.4, 6.0, and 6.1; SEM = 0.2), MID (4.4, 4.5, 6.1, and 6.1; SEM = 0.2), and LATE (4.5, 4.6, 4.7, and 5.9; SEM = 0.2). No treatment effects were detected ( $P \geq 0.42$ ) for calving rate (93, 100, 93, 100, and 100%, respectively; SEM = 5), calf BW at birth (45.2, 42.8, 43.2, 44.3, and 42.2 kg, respectively; SEM = 2.1), and estimated cow milk production (14.8, 13.7, 13.4, 14.2, and 15.7 kg/d, respectively; SEM = 1.6). However, calves from cows that gained BCS during gestation (EARLY, MID, and LATE) were heavier at weaning compared with calves from cows that maintained BCS during gestation (ADQ and INDQ; 253, 251, 260, 268, and 264 kg, respectively; SEM = 3.5). Therefore, these results indicate that increasing BCS during gestation in beef cows benefits weaning BW of the offspring.

**Key Words:** cow BCS, gestation, offspring performance

**M262 A comparison of two evaporative cooling systems on a commercial dairy farm in Saudi Arabia.** Xavier A. Ortiz<sup>\*1</sup>, John F. Smith<sup>1</sup>, Fernando Villar<sup>1</sup>, Laun W. Hall<sup>1</sup>, Jamison D. Allen<sup>2</sup>, Andrew Odde<sup>3</sup>, Adnan al-Haddad<sup>3</sup>, Peter Lyle<sup>4</sup>, and Robert Collier<sup>1</sup>, <sup>1</sup>The University of Arizona, Tucson, AZ, <sup>2</sup>Northwest Missouri State University, Maryville, MO, <sup>3</sup>Al Safi Dairy Company, Al-Kharj, Kingdom of Saudi Arabia, <sup>4</sup>Schaefer Ventilation Equipment, Sauk Rapids, MN.

Efficacy of 2 cooling systems, (Korral Kool, KK; Korral Kool Inc., Mesa, AZ, FlipFan dairy system, FF; Schaefer Ventilation Equipment LLC, Sauk Rapids, MN) was estimated utilizing 4 hundred multiparous Holstein dairy cows randomly assigned to one of 4 cooled California style shade pens (2 shade pens per cooling system). Each shaded pen contained 100 cows (DIM = 58 ± 39 d, milk production = 56 ± 18 kg/day and lactation = 3 ± 1). Production data (milk yield and reproductive performance) were collected during 4 mo (May–August, 2013) and physiological responses (core body temperature, respiration rates, skin temperatures and resting time) were measured, in June and July to estimate responses of cows to the 2 different cooling systems. Water and electricity consumption were recorded for each system. Cows in the KK system displayed slightly lower respiration rates in the month of June ( $P < 0.05$ ) and lower skin temperatures in June and July ( $P < 0.05$ ). However, no differences were observed in the core body temperature of cows, resting time, DMI, milk yield, services/cow and conception rate between systems. The FF system utilized less water and electricity during this study ( $P < 0.05$ ). In conclusion, both cooling systems (KK and FF) were effective in mitigating the negative effects of heat stress on cows housed in arid environments while the FF system consumed less water and electricity and did not require use of curtains on the shade structure.

**Key Words:** heat stress, dairy cow, evaporative cooling

**M263 Association between changes in body condition score and back fat thickness during the transition period with fertility and health events in Holstein cows.** P. D. Carvalho\* and P. M. Fricke, *Department of Dairy Science, University of Wisconsin-Madison, Madison, WI.*

Our objective was to assess the association between body condition score (BCS) change and back fat thickness (BFT) during the transition period with pregnancies per AI (P/AI) and health events in Holstein cows in a prospective cohort study. Measurement of BCS and BFT in Holstein cows ( $n = 645$ ) began ~3 wk before calving, and at  $4 \pm 3$ ,  $25 \pm 3$ ,  $52 \pm 3$ ,  $80 \pm 3$ , and  $113 \pm 3$  d after calving. All cows received a timed artificial insemination (TAI) at  $80 \pm 3$  DIM after a Double Ovsynch protocol. Health events included milk fever, ketosis, retained placenta, DA, mastitis, and respiratory disorders. Data were analyzed using logistic regression with GLIMMIX and ANOVA with MIXED procedures of SAS. Changes in BCS before but not after calving were associated with P/AI at first TAI. Cows were grouped based on BCS change (5 point scale; 0.25 point increments) before calving as follows: 1) cows that lost  $\geq 0.5$ ; 2) cows that lost 0.25; and 3) cows that maintained or gained BCS. Change in BCS before calving differed ( $P < 0.01$ ) among groups and was  $-0.57$ ,  $-0.25$ ; and  $0.03$  for cows in groups 1, 2, and 3, respectively. Change in BFT before calving differed ( $P < 0.01$ ) among groups and was  $-2.49$ ,  $-1.32$ , and  $-0.66$  mm for cows in groups 1, 2, and 3, respectively. The proportion of cows with  $\geq 1$  health event during the first 120 DIM differed ( $P = 0.01$ ) among groups and was 77% (54/70), 60% (105/174), and 62% (116/186) for cows in groups 1, 2, and 3, respectively. At 32 d after TAI, P/AI differed ( $P = 0.04$ ) among groups and was 37% (23/62), 48% (73/153), and 55% (92/168) for cows in groups 1, 2 and 3, respectively. Cows with  $\geq 1$  health event had fewer P/AI compared with healthy cows [47% (180/379) vs. 64% (127/199), respectively]. The proportion of cows culled by 300 DIM did not differ ( $P = 0.74$ ) among groups and was 25% (14/56), 27% (36/135) and 23% (33/145) for cows in groups 1, 2, and 3, respectively. We conclude that cows that lost BCS before calving lost more back fat, had fewer P/AI, and had more health events during the first 120 d of lactation. Supported by USDA NIFA Hatch project 231440.

**Key Words:** fertility, body condition score, back fat thickness

**M264 Effect of an automated teat preparation system on teat skin bacterial counts.** Cecilia Baumberger\* and Pamela L. Ruegg, *University of Wisconsin, Madison, WI.*

The aim of this study was to compare reduction in bacterial counts of teats cleaned using a commercial teat scrubber system (TS; Future-COW, Longwood, FL) or using conventional premilking preparation (CONV). Cows ( $n = 394$ ) from 10 WI dairy farms that were currently using TS were assigned to either CONV ( $n = 198$ ; 20/farm) or TS ( $n = 196$ ; 20/farm) premilking routines. CONV preparation consisted of forestripping, predipping using 0.5% iodine and drying with a cloth towel. Preparation with TS was performed using chlorine dioxide and varied according to each farm routine; most included forestripping and some included a dry prewipe. Teat skin swabs were collected before and after sanitation and analyzed for total bacteria count (TBC), *Strep. spp.*, *Staph. spp.* and gram-negative bacteria (GNB). Reduction (RED) in each bacterial count was the difference between the  $\log_{10}$  values before and after sanitation. RED was assessed using models that included treatment (CONV and TS), farm and treatment by farm interactions. For preparation using TS, the influence of the concentration of chlorine dioxide on RED was assessed using PROC GLM (SAS Institute, 2008). For teats prepared by CONV, RED was 2.26, 2.34, 2.23 and 2.04  $\log_{10}$  cfu/mL for TBC, *Strep. spp.*, *Staph. spp.* and GNB, respectively. For TS,

RED was 1.97, 2.14, 1.97 and 2.79  $\log_{10}$  cfu/mL for TBC, *Strep. spp.*, *Staph. spp.* and GNB, respectively. However, significant interactions of treatment by farm were identified for RED in all bacterial counts. Differences in RED based on preparation were not different for TBC (7 farms), *Strep. spp.* (8 farms), *Staph. spp.* (8 farms) or GNB (7 farms). Teats receiving CONV prep had greater RED of TBC on 3 farms, of *Strep. spp.* on 2 farms and of *Staph. spp.* on 2 farms. In contrast, RED in GNB was greater for teats prepped using TS for cows on 3 farms. For all bacterial counts, the concentration of chlorine dioxide was positively associated with increased RED. Results of this study suggest that TS can achieve similar RED in bacterial counts on teat skin but effectiveness of premilking teat preparation is influenced by management practices that differ among farms.

**Key Words:** teat sanitation, teat skin bacterial count

**M265 Effect of timing of insemination of lactating Jersey cows with sex-sorted semen on probability of pregnancy.** Gabriel D. Bombardelli<sup>1</sup>, Henrique F. Soares<sup>1</sup>, and Ricardo C. Chebel<sup>1,2</sup>, <sup>1</sup>University of Minnesota, St Paul, MN, <sup>2</sup>University of Florida, Gainesville, FL.

The objectives of the current experiment were to determine the effect of interval from reaching activity threshold to artificial insemination with sex-sorted semen on probability of pregnancy. Jersey cows ( $n = 500$ ) from a commercial dairy herd were fitted with a rumination/activity SCR collar at approximately at  $42 \pm 7$  d postpartum. Cows were presynchronized with 3 injections of prostaglandin-F<sub>2</sub> $\alpha$  given 14 d apart starting on d  $39 \pm 3$  postpartum. Cows in estrus, based on tail paint removal, after  $50 \pm 3$  d postpartum were inseminated by 1 of 3 technicians. Only cows inseminated in estrus were used in the experiment. Cows were examined for pregnancy at  $31 \pm 3$  and  $66 \pm 3$  d after insemination. A sub-sample of cows ( $n = 35$ ) were examined by ultrasound every 8 h after onset of estrus to determine time of ovulation, which was determined by the disappearance of a growing follicle  $\geq 10$  mm in diameter. Ovulation was estimated to have occurred between the exam in which it was last recorded and the exam in which it had disappeared. Dichotomous data were analyzed by logistic regression with a model that included interval from onset of estrus to insemination (linear, quadratic, and cubic), parity, yield of energy corrected milk, and insemination technician. Interval from onset of estrus to ovulation was analyzed by ANOVA with a model that included size of the follicle at the first ultrasound exam, number of follicles  $> 10$  mm in diameter in the first exam, and yield of energy corrected milk. The mean ( $\pm$ SEM) and median intervals from reaching activity threshold to ovulation were  $25.7 \pm 1.2$  h and 27 h (95% CI = 25, 28.2 h), respectively. Interval from onset of estrus to insemination tended to affect probability of pregnancy at  $31 \pm 3$  and  $66 \pm 3$  d after AI in a quadratic manner. Probability of pregnancy  $66 \pm 3$  d after AI increased from 23% to 44%, when AI occurred between 4 and 29 h, respectively, after cows reached activity threshold. Insemination of lactating dairy cows with sex-sorted semen closer to expected ovulation yields the highest probability of pregnancy.

**Key Words:** timing of insemination, lactating Jersey cow, sex-sorted semen



**M266 Determining the effect of transporting dairy cattle after calving on production and reproduction performance.** Danilo Domingues Millen<sup>\*1</sup>, Joaquim Baucells<sup>2</sup>, and Sergio Calsamiglia<sup>3</sup>, <sup>1</sup>Sao Paulo State University, Dracena, Sao Paulo, Brazil, <sup>2</sup>Centre Veterinari Tona, Tona, Spain, <sup>3</sup>Animal Nutrition and Welfare Service, Universidad Autonoma de Barcelona, Bellaterra, Spain.

Recently, it has been suggested that concentration of calvings in a centralized facility (FAC) may allow specialization of the drying, transition and freshening period. However, that requires the transportation of fresh cows to the host farm after calving. The objective of this study was to evaluate if transportation after calving affects milk production and reproductive performance of lactating dairy cows. A data set of 762 dairy cows from a commercial farm in Spain was used. After drying, 519 cows were transported from host farm to FAC (~80 km). The other 243 cows were at FAC, and they were not transported. Dry and production diets were identical in both farms. Milk production of previous (L-1; with no transportation) and current (L0; after transportation) lactation was recorded. Days open and current milk production were the dependent variables in the analysis, and previous milk production was used as co-variable. Transportation of cows after calving, from FAC back to the host farm, had greater 305-mature milk production when compared with cows that remained at FAC all the time (11,925 kg vs. 10,708 kg;  $P < 0.001$ ). Transportation had no effect ( $P > 0.20$ ) on days open (121 vs. 132). In a second analysis, the effect of DIM at time of transportation from FAC to the host farm on productive and reproductive performance was evaluated. The 519 transported cows were divided into 2 groups: 1) cows transported from FAC back to host farm at less than 8 DIM ( $n = 149$ ); and 2) cows transported from FAC back to host farm at more than 7 DIM ( $n = 370$ ). Cows transported earlier had greater 305-mature milk production (12,400 vs. 11,442 kg;  $P < 0.01$ ) but had more days open (163 vs. 112 d;  $P < 0.01$ ). Transporting dairy cows after calving does not negatively affect productive and reproductive performance. Transporting dairy cows at less than 8 DIM had positive effects on production but increased days open.

**Key Words:** centralized calving facility, postpartum transport, performance

**M267 Antibiotic resistance of *Escherichia coli* isolated from gastrointestinal tracts of dairy calves fed waste milk or milk replacer.** Georgina Maynou<sup>1</sup>, Anna Arís<sup>1</sup>, Alex Bach<sup>\*1,2</sup>, Francesc Fàbregas<sup>1</sup>, Alba Ferré<sup>1</sup>, and Marta Terré<sup>1</sup>, <sup>1</sup>Institut de Recerca i Tecnologia Agroalimentàries, Caldes de Montbui, Spain, <sup>2</sup>Institució Catalana de Recerca i Estudis Avançats, Barcelona, Spain.

The use of waste milk (WM) to feed calves is a common practice in dairy farms to avoid economical losses of milk disposal. The aim of this study was to determine whether there is an increase of antibiotic resistance patterns in *E. coli* isolated from feces of calves fed either WM or milk replacer (MR). Fecal swabs were collected from 20 ± 5 calves at 6 wk of age from 8 dairy farms. In 4 farms, calves were fed MR and in the other 4 they received WM. Fecal swabs were cultured to obtain 5 *E. coli* isolates per sample. Each colony was tested for its susceptibility to 9 antibiotics by disk diffusion. Binomial logistic regressions were used to analyze the resistance of *E. coli* isolates for each calf and for each antimicrobial agent tested. Feeding practice (MR or WM) and the use in the farm of the antibiotic tested (as a disease treatment) were the fixed effects of the model, and farm the random effect. Multidrug resistance profiles among *E. coli* isolates were described by cluster analyses using the Ward's minimum variance method. Then, a Chi-squared analysis was performed to assess the distribution of *E. coli* isolates in each of the 6 clusters defined within the 2 milk-feeding practices. The probability

of streptomycin (STR) resistant *E. coli* isolates was greater ( $P < 0.05$ ) in calves fed WM than in those fed MR, but the use of STR in the farm did not affect the incidence of *E. coli* resistance. The number of *E. coli* resistant to enrofloxacin (ENR) isolated from calves fed WM tended ( $P = 0.08$ ) to be greater than that from calves fed MR, and the use of ENR in the farm did not affect the incidence of *E. coli* resistance either. Most colonies isolated (93.9%) were included in 2 of the 6 clusters defined: 1 cluster included resistant *E. coli* isolates to 1 antibiotic and the other, resistant *E. coli* isolates to 3 antibiotics. The percentage of *E. coli* isolates within the most resistance cluster was greater ( $P < 0.001$ ) for WM treatment than for MR. It is concluded that feeding calves WM increases the presence of antimicrobial resistance in fecal *E. coli*.

**Key Words:** antibiotic resistance, dairy calf, waste milk

**M268 Effect of feeding calves waste milk on antibiotic resistance patterns of fecal *Escherichia coli*.** Georgina Maynou<sup>1</sup>, David Ziegler<sup>\*3</sup>, Hugh Chester-Jones<sup>3</sup>, Alex Bach<sup>2,1</sup>, and Marta Terré<sup>1</sup>, <sup>1</sup>Institut de Recerca i Tecnologia Agroalimentàries, Caldes de Montbui, Spain, <sup>2</sup>Southern Research and Outreach Center, Waseca, MN, United States, <sup>3</sup>Institució Catalana de Recerca i Estudis Avançats, Barcelona, Spain.

The use of waste milk in a calf feeding program is suspected to contribute to the development of bacterial antimicrobial resistance. Fifty-two calves allocated in individual pens were randomly assigned to milk replacer (MR) or pasteurized waste milk (pWM) fed twice daily for 42 d and once daily from d 43 to weaning (49 d). Waste milk was picked up from a local dairy farm twice weekly during the study and tested for detection of  $\beta$ -lactam antibiotic class from each load of milk. Fecal swabs were taken from each calf at 5 and 8 wk of age and cultured to obtain 3 *E. coli* isolates per sample. Each colony was tested for its susceptibility to 12 antibiotics by disk diffusion. Feeding practice (MR or pWM) and age (5 or 8 wk) effect on *E. coli* antimicrobial resistance were analyzed using binary logistic regressions for each antibiotic tested. Feeding practice, calf age and interaction between treatment and age were the fixed effects of the model, calf the random effect and calf age the repeated.  $\beta$ -lactam antibiotic residues were detected from each pWM load. Calves fed pWM had greater probability ( $P < 0.05$ ) to have ampicillin (AMP) and cephalothin (KF) ( $\beta$ -lactam antibiotics) resistant *E. coli* than calves fed MR, whereas the number of *E. coli* resistant to florfenicol tended ( $P = 0.07$ ) to be greater in calves fed pWM than in those fed MR. However, the probability of isolating *E. coli* resistant to AMP and KF tended ( $P = 0.11$ ;  $P = 0.06$ ) to decrease in pWM from 5 to 8 wk of age, in contrast to MR calves that had similar probability of isolating *E. coli* resistant to AMP and KF from 5 to 8 wk of age. It is concluded that treating cows with antibiotics generates antimicrobial residues in milk and feeding calves pWM triggers the presence of resistant *E. coli* in the gut of dairy calves.

**Key Words:** antibiotic resistance, *E. coli*, pasteurized waste milk

**M269 Fatty acid profile of bulk milk samples in commercial dairy herds in Quebec, Canada.** Melissa Duplessis<sup>\*</sup>, Debora E. Santschi, Rene Lacroix, and Daniel M. Lefebvre, Valacta, Ste-Anne-de-Bellevue, QC, Canada.

Consumer awareness of the potential health issues related to the consumption of saturated fats has increased over the past years. As milk fat contains more saturated fats than most plant oils, the effect of milk and dairy product consumption on human health has been questioned. The objective of this trial was to provide an assessment of the current

fatty acid (FA) profile, especially saturated FA, of bulk milk from 3 breeds [Holstein (HO), Jersey (JE), and Brown Swiss (BS)] in Quebec, Canada. Every wk from August 2014 to February 2015, milk samples (n = 97,999) from the bulk tank of 4,026 dairy herds were collected and sent to the Valacta laboratory (Ste-Anne-de-Bellevue, QC, Canada) for analysis. Milk fat and FA contents were determined using Fourier transform infrared spectroscopy. Data were analyzed using the MIXED and SQL procedures of SAS. As expected, bulk milk from JE had the highest milk fat content (5.24%) followed by bulk milk from BS (4.42%) and HO (4.15%;  $P < 0.0001$ ). Table 1 shows that bulk milk FA profile differs between the 3 studied breeds; milk from JE herds had the highest content of saturated FA and the lowest content of polyunsaturated FA. Saturated FA content in bulk milk increased from August to February, varying from 64.66 in August to 67.11 g/100 g of total FA in February ( $P < 0.0001$ ). Significant variation among herds was also observed ( $P < 0.0001$ ), with saturated FA ranging from 51.18 to 76.28 g/100 of total FA. These results suggest that bulk milk FA profile varies according to breed, sampling month, and herd.

**Table 1 (Abstr. M269).** Milk fatty acid (FA) profile in bulk milk of 3 breeds (g/100 g of total FA; least squares means  $\pm$  SEM)

Item	HO	JE	BS	P-value
C16:0	34.50 $\pm$ 0.01	35.12 $\pm$ 0.06	32.93 $\pm$ 0.10	<0.0001
C18:0	13.01 $\pm$ 0.00	13.11 $\pm$ 0.02	13.88 $\pm$ 0.04	<0.0001
C18:1	31.07 $\pm$ 0.01	28.00 $\pm$ 0.06	30.00 $\pm$ 0.10	<0.0001
Short-chain FA	9.62 $\pm$ 0.00	11.12 $\pm$ 0.02	10.74 $\pm$ 0.03	<0.0001
Medium-chain FA	43.75 $\pm$ 0.01	48.91 $\pm$ 0.11	46.00 $\pm$ 0.18	<0.0001
Long-chain FA	39.97 $\pm$ 0.01	36.90 $\pm$ 0.07	40.26 $\pm$ 0.12	<0.0001
Saturated FA	66.05 $\pm$ 0.01	69.36 $\pm$ 0.07	65.64 $\pm$ 0.12	<0.0001
Unsaturated FA	29.76 $\pm$ 0.01	27.77 $\pm$ 0.06	30.73 $\pm$ 0.11	<0.0001
Polyunsaturated FA	8.38 $\pm$ 0.00	7.43 $\pm$ 0.02	8.40 $\pm$ 0.03	<0.0001

**Key Words:** fatty acid profile, milk, cow

**M270 Determination of  $\beta$ -hydroxybutyrate concentration in hand-stripped milk samples taken at different times relative to milking.** Melissa Duplessis\*, Debora E. Santschi, Jean Durocher, and Daniel M. Lefebvre, *Valacta, Ste-Anne-de-Bellevue, QC, Canada.*

Analyzing  $\beta$ -hydroxybutyrate (BHBA) concentration in Dairy Herd Improvement (DHI) milk samples is a practical tool to determine incidence of hyperketonemia (HKET) in dairy herds. The objective of this trial was to determine BHBA concentration from hand-stripped milk samples outside DHI milking tests to assess HKET. A total of 31 cows with 90 d in milk or less in 3 herds was involved. Milk samples were taken just before, during and right after the morning milking and at 2, 4, 6, and 9 h after milking. Samples during milking were collected using approved metering devices used for DHI tests. Other milk samples were obtained by hand from one or several quarters. All samples were sent to the Valacta laboratory (Ste-Anne-de-Bellevue, QC, Canada). Two analytical methods were used to analyze BHBA concentration in milk, i.e., the Fourier transform infrared (FTIR) spectroscopy and the continuous flow analyzer (Skalar; which is the reference method used to calibrate FTIR). Threshold used to determine HKET was  $\geq 0.20$  mmol BHBA/L. Data were analyzed with the Glimmix and Corr procedures

of SAS. A significant analytical method  $\times$  time relative to milking interaction was noted on the incidence of HKET ( $P = 0.0004$ ). Incidence of HKET differed in the samples taken right after milking ( $P = 0.006$ ) and 2 h after milking ( $P = 0.10$ ) between the analytical methods whereas no difference was noted for other times relative to milking ( $P \geq 0.13$ ). Indeed, incidence of HKET was  $25.8 \pm 7.8\%$  and  $29.0 \pm 8.0\%$  for Skalar and  $61.3 \pm 8.7\%$  and  $70.0 \pm 5.0\%$  for FTIR right after and 2 h after the milking, respectively. Pearson correlation coefficient between Skalar and FTIR BHBA results was 0.95 for the samples collected during milking and was lower for other times relative to milking (ranging from 0.6 to 0.9). In conclusion, BHBA concentration in milk is a practical tool to assess HKET, especially for milk samples taken during DHI tests. Hand-stripped milk samples collected right after or 2 h after milking require further calibration of FTIR before recommending BHBA concentration analysis.

**Key Words:** hyperketonemia,  $\beta$ -hydroxybutyrate, dairy cow

**M271 Prevalence of *Salmonella* and *Campylobacter* from composite fecal samples on US dairy operations.** Charles P. Fossler<sup>1</sup>, Jason E. Lombard\*<sup>1</sup>, Paula J. Fedorka-Cray<sup>2,3</sup>, Jodie R. Plumblee<sup>2</sup>, Christine A. Kopral<sup>1</sup>, and R. Camilla Kristensen<sup>1</sup>, <sup>1</sup>USDA:APHIS:VS: Center for Epidemiology and Animal Health, Fort Collins, CO, <sup>2</sup>USDA:ARS:Bacterial Epidemiology and Antimicrobial Resistance Research Unit, Athens, GA, <sup>3</sup>North Carolina State University, Raleigh, NC.

During the National Animal Health Monitoring System's Dairy 2014 study, 6 composite environmental fecal samples were collected from 234 operations for culture of *Salmonella*. On 232 of the 234 operations, these samples were also tested for *Campylobacter*. One objective of the study was to estimate the herd-level prevalence of *Salmonella* and *Campylobacter* on US dairy operations. Samples were collected from March through July 2014. Estimates incorporated weighting procedures and were adjusted for study design. *Salmonella* was identified via culture from at least one sample on 39.2% of operations. Operations with 500 or more cows were more likely to be *Salmonella*-positive compared with operations with 30–99 cows (62.0% vs. 26.0%). No regional differences were observed in the percentage of *Salmonella*-positive operations. The largest percentages of *Salmonella*-positive samples were from manure pits (39.9%) and common alleyways (38.2%), while the lowest percentage positive were from gutter cleaners (23.9%); however, these percentages were not statistically different. Statistically, the percentage of *Salmonella*-positive operations from the Dairy 2014 study (39.2%) was not different from the percentage of *Salmonella*-positive operations from the NAHMS Dairy 2007 study (35.8%). *Campylobacter* was identified in at least one sample from 88.5% of operations. There were no differences by herd size or by region in the percentage of operations testing *Campylobacter*-positive. Overall, 54.8% of samples from holding pen floors were *Campylobacter*-positive, followed by 54.3% of samples from common alleyways, 51.1% from parlor exits, 45.5% from gutter cleaners, and 42.6% from common pens, and these locations were more likely to be positive than samples from manure spreaders (19.1%) and flush water (14.0%). A high percentage of US dairy operations can be expected to have *Campylobacter* on the operation. *Salmonella* is not as common, with less than half of operations expected to be positive at a single sampling. The percentage of operations positive for *Salmonella* did not change between the Dairy 2007 and Dairy 2014 studies.

**Key Words:** *Salmonella*, *Campylobacter*, prevalence

### **M272 Off-site dairy heifer rearing in the United States.**

Jason E. Lombard\*<sup>1</sup>, Ashley E. Adams<sup>1,2</sup>, Charles P. Fossler<sup>1</sup>, Natalie J. Urie<sup>1,2</sup>, Chelsey B. Shivley<sup>1,2</sup>, and Christine A. Kopral<sup>1</sup>, <sup>1</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO, <sup>2</sup>Colorado State University, Fort Collins, CO.

The National Animal Health Monitoring System's Dairy 2014 study collected information regarding off-site rearing of dairy heifer calves. Data for the study were collected via face-to-face interviews on operations in the top 17 dairy states. An objective of the study was to describe the use of off-site heifer-rearing facilities by US dairy operations. Size categories were created based on adult cow populations: small (30–99 cows), medium (100–499 cows), and large (500 or more cows). The percentage of operations that had their heifers raised off-site increased as herd size increased: 5.5% of small, 11.9% of medium, and 44.2% of large operations. Overall 11.7% of operations raised heifers off-site. The majority of small and medium operations (84.9% and 59.0% percent, respectively) sent weaned and non-pregnant heifers to off-site rearing facilities at an average age of 255.4 and 164.7 d, respectively. The majority of large operations (62.8%) sent heifers to off-site facilities as preweaned heifers at an average age of 6.8 d. About a quarter of operations that sent the majority of calves off-site as preweaned heifers (26.6%), provided waste milk to the off-site rearing operation. Additionally, 2-thirds of the rearing facilities (66.8%) fed pasteurized waste milk. More than 4-fifths of operations (84.6%) used off-site rearing facilities within 50 miles of the dairy operation. Approximately 50% of operations in each size category and 53.8% of all operations sent heifers to a single facility where heifers had contact with cattle from other operations. Additionally, 10.6% of operations sent heifers to multiple facilities where they had contact with cattle from other operations. Most operations (69.6%) brought back pregnant heifers at an average age of 21.5 mo. Off-site heifer rearing is an important component of the US dairy industry; however, off-site rearing increases the potential for disease exposure and spread. The development of biosecurity/biocontainment plans based on specific risks associated with stress from shipping and exposure of dairy heifers to cattle from other operations at off-site facilities should reduce disease transmission.

**Key Words:** off-site heifer rearing, biosecurity, dairy heifer

**M273 Management practices that may affect dairy heifer welfare on US dairy operations.** Ashley E. Adams\*<sup>1,2</sup>, Jason E. Lombard<sup>2</sup>, Chelsey S. Shivley<sup>1,2</sup>, Natalie J. Urie<sup>1,2</sup>, Ivette N. Roman-Muniz<sup>1</sup>, Charles P. Fossler<sup>2</sup>, and Christine A. Kopral<sup>2</sup>, <sup>1</sup>Colorado State University, Fort Collins, CO, <sup>2</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO.

The objectives of this study were to describe housing and management practices that may affect dairy heifer welfare on US dairy operations, and to compare dairy heifer housing and management practices on organic and non-organic dairy operations. This study was conducted as part of the National Animal Health Monitoring System's Dairy 2014 study, which included dairy operations in 17 states. Data on housing and management practices were collected on 1,261 dairy operations via in-person interviews. A total of 7.4% of operations were certified organic. Organic and non-organic operations differed only in the time at which calves were removed from the dam. On 50.1% of organic operations calves remained with their dam for 12 or more hours, compared with 26.9% of non-organic operations that kept calves with their dams for that long. Individual hutch/pen was the primary housing type for preweaned heifers on 69.6% of operations in 2013 compared with 74.9% of operations in 2006. Most dairy operations (94.3%) disbudded/dehorned calves. A hot iron was the most prevalent method used to disbud/dehorn calves

(69.9%); 30.0% of operations that disbudded/dehorned calves gave the calves analgesics and/or anesthetics. Tail-docking was performed on 31.7% of operations, of which 1.1% used analgesia/anesthesia. A band was the most common method of tail-docking (97.0% of operations that tail-docked); 31.9% of operations that tail-docked, docked tails when heifers were less than 2 mo of age, while 44.9% tail-docked when heifers were over 2 years of age. In total, 6.4% of preweaned dairy heifers and 1.9% of weaned dairy heifers died during 2013, compared with 7.8% of preweaned heifers and 1.8% of weaned heifers in 2006. Of preweaned heifers and weaned heifers that died in 2013, 3.5% and 4.2%, respectively, were euthanized. The percentage of operations using an analgesia/anesthesia during disbudding/dehorning increased from 13.8% in 2006 to 30.0% in 2013; however, 70% of dairy operations do not use analgesia/anesthesia when disbudding/dehorning cows, which highlights the need for education regarding pain management during surgical procedures.

**Key Words:** dairy heifer, welfare, organic

**M274 Management practices that may affect dairy cow welfare on US dairy operations.** Ashley E. Adams\*<sup>1,2</sup>, Jason E. Lombard<sup>2</sup>, Ivette N. Roman-Muniz<sup>1</sup>, Charles P. Fossler<sup>2</sup>, and Christine A. Kopral<sup>2</sup>, <sup>1</sup>Colorado State University, Fort Collins, CO, <sup>2</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO.

The objectives of this study were to describe housing and management practices that may affect dairy cow welfare on US dairy operations, and to compare dairy cow housing and management practices on organic (ORG) and non-organic (NORG) dairy operations. This study was conducted as part of the National Animal Health Monitoring System's Dairy 2014 study, which included data from dairy operations in 17 states. Data on housing and management practices were collected on 1,261 dairy operations via in-person interviews. A total of 7.4% of operations were certified organic. Freestall barns were the primary housing type for lactating cows on 39.0% of operations, while tie-stall or stanchion barns were used on 38.2%. The primary bedding material used in operations with freestall barns were sand (44.0%), sawdust (19.0%), and straw (14.2%). Operations with tie-stall or stanchion barns primarily used straw for bedding (46.4%) or saw dust (25.3%). For organic operations, the predominant flooring types for cows not being milked were concrete (45.0%), pasture (31.1%), and rubber mats over concrete (14.9%). In comparison, the predominant flooring types used on NORG operations were concrete (62.2%) and rubber mats over concrete (21.8%). Fans for cooling dairy cows were used on 59.6% of ORG operations and 77.0% of NORG operations. Sprinklers/misters were used on 12.1% of ORG and 26.3% of NORG operations. A total of 59.8% of operations provided personnel with training in milking procedures, 56.0% in animal handling, 33.4% in management of nonambulatory cattle, 29.8% in surgical procedures, and 20% in euthanasia. In total, 5.6% of dairy cows died during 2013, of which 40.6% were euthanized. While a smaller percentage of ORG operations used concrete as the primary flooring for dairy cows, more NORG operations provided heat abatement, suggesting that there is room to improve cow welfare on both types of operations. This finding is also evidenced by the percentage of dairies that do not train personnel in animal handling, surgical procedures, management of nonambulatory cattle, and euthanasia techniques, all of which may affect dairy cow welfare.

**Key Words:** dairy cow, welfare, organic



**M275 Evaluation of colostrum management, feeding management, and weaning practices on US dairy operations.** Chelsey B. Shivley<sup>\*1,2</sup>, Jason E. Lombard<sup>2</sup>, Ashley E. Adams<sup>1,2</sup>, Natalie J. Urie<sup>1,2</sup>, Charles P. Fossler<sup>2</sup>, and Christine A. Kopral<sup>2</sup>, <sup>1</sup>Colorado State University, Fort Collins, CO, <sup>2</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO.

The National Animal Health Monitoring System's Dairy 2014 study evaluated colostrum management, feeding management, and weaning practices on 1,261 operations from 17 of the nation's top dairy states. Results from the study indicate that on average heifer calves were given their first feeding of colostrum 3.6 h following birth. The highest percentage of operations (53.2%) administered colostrum by hand, followed by both hand-feeding and suckling the cow (40.5%), and only suckling the cow (6.3%). For the 93.7% of operations that hand-fed colostrum, 87.4% fed colostrum with a bottle, 8.1% with an esophageal feeder, and 4.5% with a bucket. Most dairies (53.0%) fed 2 L of colostrum at the first feeding, and 62.1% of operations fed another 2 L within the first 24 h following birth. About half of all operations (49.3%) stored colostrum, with freezing being the most common method of storage (73.0% of operations). About half of dairies that hand-fed colostrum (53.3%) estimated colostrum quality, with visual appearance being the most common method used (45.1% of operations). Only 6.2% of operations monitored serum proteins to test passive-transfer status. Almost half of dairies (49.9%) fed milk replacer (MR), and 63.2% fed waste milk. Of operations that fed MR, percent protein was 20–24% on 89.2% of operations. About half of operations (56.4%) fed less than 4.7 L of waste milk or MR per day, with most operations (94.6%) feeding twice per day. The highest percentage of operations used bottles and buckets to feed waste milk or MR (77.2% and 72.3%, respectively). On average, heifer calves were first offered water at 17.3 d of age, starter grain at 10.8 d of age, and hay at 36.0 d of age. The average age at weaning was 8.8 weeks, and age was the most commonly used determinant for weaning on 50.2% of operations. These results show that less than 2 thirds of producers are feeding the recommended 3–4 L of colostrum at the first feeding on US dairies, indicating that producer education on colostrum management is warranted.

**Key Words:** dairy heifer calves, colostrum management, feeding management

**M276 Locomotion scoring dairy cows: A comparison among three different locomotion scoring on intra- and interrater reliability.** Ashley E. Adams<sup>\*1,2</sup>, Jason E. Lombard<sup>2</sup>, and Ivette N. Roman-Muniz<sup>1</sup>, <sup>1</sup>Colorado State University, Fort Collins, CO, <sup>2</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO.

Lameness in dairy cattle is usually identified by judging the locomotion of dairy cows. The objective of this study was to determine differences in intra- and interrater reliability among 3 different locomotion scoring systems for dairy cows. The 3-, 4-, and 5-point locomotion scoring systems were used by 42 observers to score the gait of cows in a set of 45 video clips. Each observer scored the set of clips 4 times, once using each of the 3 scoring systems, then again repeating one of the 3 systems. The order of presentation of the video clips was randomized between each viewing, and participants were randomly assigned to repeat one of the 3 scoring systems. Intra- and interrater reliability were calculated as the weighted kappa coefficient ( $\kappa_w$ ). The mean intrarater reliability for the 3-point system was  $\kappa_w = 0.57$  (95% CI: 0.43–0.71), for the 4-point system  $\kappa_w = 0.60$  (95% CI: 0.53–0.68), and  $\kappa_w = 0.64$  (95% CI: 0.55–0.72) for the 5-point system. The CI for intrarater reliability suggests that no difference exists in  $\kappa_w$  among the 3 systems. The mean

interrater reliability for the 3-point system was  $\kappa_w = 0.42$  (95% CI: 0.41–0.43), for the 4-point system  $\kappa_w = 0.43$  (95% CI: 0.42–0.44), and  $\kappa_w = 0.47$  (95% CI: 0.46–0.48) for the 5-point system. These intervals indicate that interrater reliability is greater for the 5-point system when compared with the 3- and 4-point systems. Using a level of acceptable reliability of  $\kappa_w > 0.60$ , none of the scoring systems achieved good reliability for interrater comparisons, while both the 4- and the 5-point scoring systems achieved good intrarater reliability. This result suggest that within observer reliability is adequate for the 4- and 5-point scoring systems, but not for the 3-point system. In contrast, between observer reliability is lacking for the 3 locomotion scoring systems examined in this study. Further analysis is needed to determine if cows are consistently identified as lame and severely lame using the 3 scoring systems.

**Key Words:** dairy cow, locomotion scoring, reliability

**M277 Association between changes in body condition score, NEFA, and BHBA concentrations during the transition period on fertility of Holstein cows.** R. V. Barletta<sup>\*1,2</sup>, P. D. Carvalho<sup>1</sup>, M. Madureira<sup>3</sup>, T. A. Del Vale<sup>2</sup>, E. R. Madureira<sup>3</sup>, A. S. Netto<sup>2</sup>, G. M. Baez<sup>1</sup>, P. M. Fricke<sup>1</sup>, and M. C. Wiltbank<sup>1</sup>, <sup>1</sup>Department of Dairy Science, University of Wisconsin-Madison, Madison, WI, <sup>2</sup>Department of Nutrition and Animal Production, School of Veterinary Medicine and Animal Sciences, University of São Paulo, São Paulo, Brazil, <sup>3</sup>Department of Animal Reproduction, School of Veterinary Medicine and Animal Sciences, University of São Paulo, São Paulo, Brazil.

Our objective was to assess the association between body condition score (BCS) change during the transition period with pregnancies per AI (P/AI) and NEFA and BHBA concentrations in Holstein cows in a retrospective cohort study. Holstein cows ( $n = 232$ ) were assessed for BCS (5 point scale; 0.25 point increments) at 21 d before, on the day of, and 21 d after calving. Blood samples were collected 21 and 7 d before, on the day of, and 7 and 21 d after calving and were assayed for NEFA and BHBA concentrations. All cows received a timed artificial insemination (TAI) after a Presynch-Ovsynch protocol at  $75 \pm 3$  DIM. Data were analyzed by logistic regression with GLIMMIX and ANOVA with repeated measures using the MIXED procedures of SAS. Both NEFA and BHBA concentrations after calving differed ( $P < 0.01$ ) for cows that lost, maintained, or gained BCS from 21 d before to 21 d after calving (NEFA:  $0.49 \pm 0.01$ ;  $0.40 \pm 0.01$ ;  $0.43 \pm 0.01$  mmol/L; BHBA:  $0.72 \pm 0.02$ ;  $0.70 \pm 0.02$ ;  $0.67 \pm 0.02$  mmol/L; respectively). Day of sampling relative to calving also affected ( $P < 0.01$ ) NEFA and BHBA concentrations, with the greatest increase occurring 7 d after calving. Change in BCS after ( $P < 0.01$ ) but not before calving affected P/AI. Cows were grouped based on BCS change after calving as to whether they: 1) lost, 2) maintained, or 3) gained BCS. By design, BCS change after calving differed ( $P < 0.01$ ) among groups and was  $-0.38$ ;  $0.00$ ; and  $0.35$  for cows in groups 1, 2 and 3, respectively. On the day of calving, BCS differed ( $P < 0.01$ ) among groups and was  $2.89$ ;  $2.69$ ; and  $2.49$  for cows in groups 1, 2 and 3, respectively. At 21 d after calving, BCS differed ( $P < 0.01$ ) among groups and was  $2.51$ ;  $2.69$ ; and  $2.85$  for cows in groups 1, 2 and 3, respectively. At 32 d after TAI, P/AI differed ( $P < 0.01$ ) for cows that lost [13% (11/84)], maintained [33% (26/80)], or gained [47% (32/68)] BCS. In conclusion, changes in BCS during the transition period affected NEFA and BHBA concentrations during the transition period; however, P/AI was only affected by changes in BCS after calving.

**Key Words:** body condition score, fertility, transition period

**M278 Compost bedded pack on bacterial counts and milk composition in lactating dairy cows.** Lorena Castillejos, María Rodríguez, Adriana Siurana, and Sergio Calsamiglia\*, *Animal Nutrition and Welfare Service, Department of Animal and Food Science, Universitat Autònoma de Barcelona, Bellaterra, Spain.*

The compost bedded pack dairy barn is an alternative loose housing system that has been implemented in the recent years. The objective of this study was to evaluate the effect of compost bedded pack on bacterial counts in bedding and milk in lactating dairy cows. A field study was conducted in Girona (Spain) between May 2013 and July 2014 in 2 commercial dairy farms (816 and 394 lactating cows). Cows were distributed in equal barns ( $90 \pm 14$  and  $66 \pm 7$  cows) with different loose housing system: 2 compost-bedded-pack (compost) barns were compared with 2 conventional-bedded-pack (control) barns, 4 barns per farm. Compost barns were aerated twice daily, but less sawdust was added as bedding material. Farms were visited 6 times (spring, summer and autumn of 2013 and winter, spring and summer of 2014). During each visit, bedding material samples throughout each barn and milk samples of the total production of each barn of the morning and afternoon milking were collected. Data on milk composition (lactose, protein, fat and fat free total solids) and somatic cells count (SCC) were analyzed. Bacterial counts of bedding material were determined for mesophilic aerobic bacteria (total bacteria counts, TBC), other gram-negative, total coliforms, *E. coli*, *Streptococcus* spp., *Strep. agalactiae*, and *Klebsiella* spp. Bacterial counts of milk were determined for TBC, other gram-negative, lactose (-), total coliforms, *E. coli*, coagulase-negative staphylococci, *Staph. aureus*, *Streptococcus* spp., *Strep. agalactiae*, and *Klebsiella* spp. No significant effects in bacterial counts were found in bedding material (TBC  $1.1 \times 10^{10} \pm 3.3 \times 10^9$  cfu/g) or milk (TBC  $5.1 \times 10^4 \pm 9.3 \times 10^3$  cfu/mL). Milk quality (SCC 240,000  $\pm$  173,000 cells/mL) and composition did not change, except for lactose that was higher in compost cows than in control cows (4.91 vs  $4.89 \pm 0.008\%$ ,  $P < 0.05$ ). These results suggest that udder health and milk quality were not compromised when housing lactating dairy cows were in compost barns. Saving of sawdust and manure storage costs without negative effect in milk quality, encourage the implementation of compost barns in dairy farms.

**Key Words:** compost bedded pack barn, bacterial count, milk composition

**M279 Economic impact of nutritional grouping in dairy herds.** Afshin S. Kalantari\*, Louis E. Armentano, Randy D. Shaver, and Victor E. Cabrera, *University of Wisconsin-Madison, Madison, WI.*

Study's objective was to evaluate the economic impact of nutritional grouping in commercial dairy herds. A dynamic, stochastic Monte Carlo simulation model was developed to represent individual cow DMI,  $NE_L$ , and MP requirements and the resulting changes in body energy throughout a year. The amount of energy consumed directly affected the BW and BCS changes in the model. Moreover, to control the range of observed BCS in the model constraints on lower and upper bounds of BCS were set. Cows that would drop to  $BCS < 2.0$  were assumed to stay at  $BCS = 2.0$ , but at a decreased milk production {New daily milk (kg/d) = daily milk (kg/d) - [deficient  $NE_L$  intake (Mcal/d)/ $NE_L$  in milk (Mcal/kg)]}, and cows that would reach  $BCS > 4.5$  were assumed to stay at  $BCS = 4.5$ , but at decreased DMI {New DMI (kg/d) = DMI (kg/d) - [extra  $NE_L$  intake (Mcal/d)/dietary  $NE_L$  (Mcal/kg)]}. The model was validated by comparing its projections against actual data regarding involuntary culling, BW, BCS,  $NE_L$  and MP concentrations of the offered diet. The model was initialized by separate data sets obtained from 5 dairy herds with different herd size and structure. Status of each cow was updated on a daily basis together with the required diet nutrient

concentrations. Each month, lactating cows were regrouped according to the studied grouping strategies and their nutrient concentration requirements. The calculated income over feed costs (IOFC, \$/cow per yr) of having more than 1 nutritional group among the herds ranged from \$51 to \$70, with an average of \$58 for 2 groups and \$70 for 3 groups, when group was fed at average  $NE_L$  and average+1xSD MP concentration. The improved IOFC was explained by increased milk sales and lower feed costs. Higher milk sales were a result of fewer cows having a milk loss associated with low BCS in multi-group scenarios. Lower feed costs in multi-group scenarios were mainly due to less RUP consumption. The percentage of total  $NE_L$  consumed captured in milk for greater than 1 nutritional group did not show a consistent increase and the values were rather similar among different group numbers. The percentage of N fed that was captured in milk increased with more than 1 group, and was the most important factor for improved economic efficiency of the grouping strategies.

**Key Words:** stochastic, simulation, nutrition

**M280 Influence of calving weight on milk yield of dairy Gyr cows.** Manuela Pires Monteiro Gama\*<sup>1</sup>, Gabriela Geraldi Mendonça<sup>2</sup>, Anibal Eugênio Vercesi Filho<sup>3</sup>, André Rabelo<sup>4</sup>, Lenira El Faro Zadra<sup>3</sup>, and Cláudia Cristina Paro Paz<sup>1,3</sup>, <sup>1</sup>*Departamento de Genética, FMRP-USP, Ribeirão Preto, SP, Brazil*, <sup>2</sup>*Instituto de Zootecnia (IZ/APTA/SAA), Nova Odessa, SP, Brazil*, <sup>3</sup>*Centro APTA Bovinos de Corte, Instituto de Zootecnia (IZ), Sertãozinho, SP, Brazil*, <sup>4</sup>*ABCGIL – Associação Brasileira de Criadores de Gir Leiteiro, Uberaba, MG, Brazil.*

The weight of cows at calving is not a trait commonly measured by producers, but may influence milk yield and reproduction because of the negative energy balance. The effect of calving weight on milk yield of dairy Gyr cows was analyzed using data from 2 herds comprising 570 records of total yield during lactation of 363 cows that had calved between 2000 and 2014. The contemporary groups containing at least 5 observations were defined as herd and year and season of calving. The latter was divided into dry season (April to September) and rainy season (October to March). The analyses were performed using linear models containing the fixed effects of contemporary group and lactation duration (linear coefficient), and by regression of milk yield on calving weight nested within classes of age at calving (1 to 8), with class 1 corresponding to animals aged 2 to 3 years and so on until animals aged 10 years at calving. The PROC GLM module of the SAS program was used for analysis. The mean calving weight was 434.156 kg and this trait was influenced by age of cow at calving (linear and quadratic effects). The trend of regression indicated an increase in calving weight up to 90 mo, declining thereafter. The effect of calving weight for each class of age at calving presented linear regression coefficients of 5.19, 7.34, 9.33, 8.97, 2.43, -0.93, -3.70 and -4.97, respectively. These results demonstrate an increase in calving weight and total yield until physiological maturity, which occurs in this breed at about 93 mo when production starts to decrease slowly with increasing age of the animal. The influence of calving weight on milk yield is more important for younger cows. This importance decreases for older cows as expected, since they have stopped growing and have reached their maximum genetic production potential. Furthermore, older cows produce less milk compared with younger animals whose genetic potential for milk production and lactation persistence is higher due to the genetic improvement performed.

**Key Words:** body weight, Zebu, age at calving

**M281 Effects of dietary forage and protein levels on the concentration and total load of *Escherichia coli* and *Listeria monocytogenes* in feces of dairy cows.**

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*Escherichia coli* and *Listeria monocytogenes* are animal waste borne pathogens. The study aimed to determine if the basic dietary nutrient composition affected total *E. coli* and *Listeria* load and concentration in dairy cow feces. Twelve Holstein cows were assigned to a 2 × 2 factorial arrangement of 2 forage levels [37 (LF) vs. 53% (HF)] and 2 CP levels [15.2 (LP) vs. 18.5% (HP)] in a 4 × 4 Latin square design with 4 periods. After a 14 d adaptation period, total feces mass (kg/cow/d) was determined using total collection approach for 3 d. Six fecal samples were collected periodically to represent the course of a day. Dry matter intake (kg/cow/d), and *E. coli* and *Listeria* concentrations in all feed ingredients were measured to determine pathogen intake via feed. Pathogen concentrations in solid samples were determined as colony forming units (cfu) per gram according to the Bacteriological Analytical Manual recommended by US FDA. Total pathogen load was calculated based on the pathogen concentration and feces mass. Treatment effects were analyzed by a mixed model with the random effect of cow. Concentration of *E. coli* and *Listeria* were measured for all feed ingredients. Estimated average *E. coli* and *Listeria* concentrations in LF and HF diets were  $1.4 \times 10^6$  and  $1.9 \times 10^6$ , and  $3.8 \times 10^4$  and  $5.4 \times 10^4$  cfu/kg of DM, respectively. Interaction effects between dietary fiber and CP contents were found for both *E. coli* concentration ( $P < 0.05$ ) and load ( $P < 0.01$ ) in feces. Fecal *E. coli* concentration and load were greater ( $P < 0.05$ ) in HFHP ( $5.4 \times 10^6$  cfu/g and  $6.4 \times 10^{10}$  cfu, respectively) than HFLP ( $3.1 \times 10^6$  cfu/g and  $3.64 \times 10^{10}$  cfu, respectively). Greater dietary CP content significantly decreased fecal concentration of *Listeria* regardless of dietary fiber content ( $P < 0.01$ ). Concentration of *Listeria* was higher in LP (110.5 cfu/g) than HP (68.2 cfu/g). There was no association between DM, CP, NDF, or starch of feces and *E. coli* or *Listeria*. Dietary forage and CP interactively affect the concentration and total load of *E. coli*. Only dietary CP level affects concentration of *Listeria* in the feces of dairy cows.

**Key Words:** dairy cow, *E. coli*, *Listeria*

**M282 Urinary disposition kinetics and fecal excretion of two intramammary antibiotic preparations in dairy cows.**

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Appropriate assessment of the contribution of antibiotic use in animal agriculture to development of antibiotic resistance in bacterial pathogens and development of efficient mitigation strategies are limited due to a lack of data on the elimination of antibiotics from animals. The objective was to investigate urinary disposition kinetics and fecal excretion of pirlimycin and cephalixin, 2 antibiotics commonly used on dairy farms as intramammary infusion. Three lactating cows were administered 2 doses of pirlimycin (50 mg/cow; 24 h apart), and 3 end of lactation dairy cows were administered cephalixin (1200 mg/cow) intramammary. Feces and urine were collected just before antibiotic infusion and at 2, 4, 8, 12, 16, and 24 h after 1st dose of pirlimycin, at 2, 4, 8, 12, 16, 24, 36, 48, and 72 h after 2nd dose of pirlimycin, and at 2, 4, 8, 12, 16, 24, 36, and 72 h post-cephalixin infusion. Antibiotics were quantified using UPLC-MS/MS. Urinary disposition kinetics parameters were derived by fitting urine concentration-time data into noncompartmental model.

Elimination rate constants in urine were estimated using regression analyses of antibiotic concentrations after peak time. The elimination half-life, mean residence time, and elimination rate constant for 1st and 2nd dose of pirlimycin and cephalixin were  $11.6 \pm 3.6$ ,  $20.2 \pm 8.8$ , and  $14.9 \pm 1.9$  h,  $22.1 \pm 9.3$ ,  $33.1 \pm 14.5$ , and  $25.2 \pm 4.9$  h, and  $0.039 \pm 0.034$ ,  $0.038 \pm 0.016$ , and  $0.048 \pm 0.006$  h<sup>-1</sup>, respectively. Fecal concentration of pirlimycin peaked ( $252 \pm 162$  ng/g) at  $34.9 \pm 9.2$  h after the 2nd dose. Despite intramammary administration, antibiotics reached blood and the digestive tract, were excreted in urine and feces, and thus may influence microbial antibiotic resistance in the gut and following excretion. Slow elimination and long residence time of antibiotics indicate that manure may need to be treated to reduce antibiotic loading to the environment. The data obtained in this study will allow development of efficient manure management strategies to reduce the contribution of dairy farms to the challenge of antibiotic resistance.

**Key Words:** dairy cow, disposition kinetics, intramammary antibiotic preparation

**M283 Performance and health of Holstein calves fed different amounts of milk supply with or without a symbiotic complex.**

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The objective of this study was to evaluate performance and health of Holstein calves fed low or high milk supply (MSP) and with an inclusion or not of symbiotic complex (SYM). Thirty-two Holstein calves ( $34 \pm 7$  kg BW) with an age of 6 d were distributed in a randomized block design in a 2 × 2 factorial arrangement. The MSP consisted in: low MSP - 10% BW for the 1st through 8th weeks after birth; and high MSP - 20% BW for the 1st and 2<sup>nd</sup> weeks after birth, 15% BW for the 3rd and 4th weeks after birth and 10% BW for the 5th and 8th weeks after birth. The SYM tested was Bioformula Leite (prebiotics, probiotics, and fibrolytic enzymes). Solid ration was supplied in addition to milk. Intake, ADG, diet digestibility, and fecal consistency index were evaluated. MSP influenced calf growth ( $P < 0.10$ ), with different final weight (77.8 and 85.0 kg), post-weaning average weight gain (548 and 788 g/d), and final average weight gain (549 and 646 g/d) in calves fed 10% and 20% BW, respectively. MSP affected intake variables; calves fed 10% and 20% BW had pre-weaning dry ration intakes of 291 and 237 g/d, pre-weaning total DM intake of 715 and 1,672 g/d, and total DM intakes over the experimental period of 867 and 1,016 g/d. The digestibility of DM, CP, NDF, and non-fiber carbohydrates were not different for both variables ( $P > 0.10$ ): 0.73, 0.69, 0.77, and 0.92 g/kg, respectively. There was an interaction between MSP and SYM on the digestibility's of DM and NDF ( $P < 0.10$ ). The digestibility of DM (0.728 g/kg) and NDF (0.771 g/kg) of the calves fed 10% BW were not affected by inclusion of SYM ( $P > 0.10$ ). On the other hand, for calves fed 20% BW, inclusion of SYM increased digestibility of DM (0.720 to 0.736 g/kg) and NDF (0.758 to 0.783 g/kg). The use of SYM improved fecal score of the animals ( $P < 0.10$ ; 0.31 and 0.42, respectively). The milk-feeding level is an important factor in animal performance. In addition, the present study demonstrated positive results for the DM and NDF digestibilities, and animal's health; Thereby SYM might be use as an option to control diarrhea.

**Key Words:** fecal score, milk supply, weaning



**M284 Impact of feeding betaine-containing molasses to transition dairy cows during late summer.** Ana Paula A. Monteiro\*<sup>1</sup>, John K. Bernard<sup>1</sup>, Stephen Emanuele<sup>2</sup>, Randy Davis<sup>2</sup>, Charles R. Staples<sup>3</sup>, Jundi Liu<sup>1</sup>, Geoffrey E. Dahl<sup>3</sup>, and Sha Tao<sup>1</sup>, <sup>1</sup>University of Georgia, Tifton, GA, <sup>2</sup>Quality Liquid Feeds, Dodgeville, WI, <sup>3</sup>University of Florida, Gainesville, FL.

Betaine (tri-methyl glycine) is a natural compound found in sugar beets and serves as a methyl donor when fed to animals. Under heat stress it can act as an osmolyte, reducing dehydration. The objective was to evaluate the effect of feeding betaine-containing molasses on performance of transition dairy cows during late summer. In early September, cows were randomly assigned to betaine (B) or control (C) groups either at dry off (B: n = 10; C: n = 10) or 24 d before calving (B: n = 8; C: n = 8) based on their previous mature equivalent milk yield. Cows were fed a common diet supplemented either with a 28% CP molasses-based liquid supplement made from sugar cane or a 28% CP liquid supplement made of molasses from sugar cane (67%) and condensed beet solubles containing ~30% betaine (33%) for C and B cows, respectively, until 60 d in milk. The liquid supplement was fed at a rate of 1.1 and 1.4 kg DM/d for pre and postpartum cows, respectively. Before calving, all cows were housed in the same free-stall barn without supplemental cooling. After calving, all cows were cooled by misters and fans and were milked thrice daily. Feed intake was recorded daily and BW and BCS were assessed every 2 wk. Milk yield was recorded at each milking and composition was analyzed weekly. Blood samples were collected weekly from a subset of cows to assess hematocrit and concentrations of cortisol, betaine, and metabolites. There were no treatment effects on intake of DM and BW in the prepartum and postpartum periods, but cows fed B had lower ( $P = 0.05$ ) BCS at 4 and 6 wk postpartum compared with C. For those cows enrolled at dry off, B tended to have lower ( $P < 0.15$ ) hematocrit at 8 and 5 wk before calving compared with C, but no difference was detected postpartum. All cows fed B tended ( $P < 0.10$ ) to produce more milk (44.2 vs. 41.5 kg/d) and 3.5% FCM (50.0 vs. 47.0 kg/d) compared with C, but no differences in milk composition were observed. In conclusion, supplementing transition cows with betaine-containing liquid supplement during late summer may prevent dehydration of dry cows based on a trend for lower hematocrit and increase 3.5% FCM yield in early lactation.

**Key Words:** dairy cow, heat stress, betaine

**M285 Growth and behavior of group-fed dairy calves fed once or twice daily in an organic production system.** Myrrh-Anna J. Kienitz\*<sup>1</sup>, Bradley J. Heins<sup>1</sup>, and Hugh Chester-Jones<sup>2</sup>, <sup>1</sup>University of Minnesota, West Central Research and Outreach Center, Morris, MN, <sup>2</sup>University of Minnesota, Southern Research and Outreach Center, Waseca, MN.

Heifer calves (n = 102) were used to evaluate the effect of once or twice daily feeding on growth and behavior of calves in an organic group management system. Calves were assigned to replicate feeding groups of 10 in super hutches by birth order, during 2 seasons from September to December 2013 and March to May 2014 at the University of Minnesota West Central Research and Outreach Center, Morris. Calves in groups were the experimental unit. Breed groups of calves were: Holsteins (HO; n = 26), crossbreds (n = 45) including combinations HO, Montbéliarde, and Viking Red selected for high production, and crossbreds (n = 31) including combinations of HO, Jersey, Normande, and Viking Red selected for robustness. Treatment groups were 1) once daily feeding (1×) or 2) twice daily feeding (2×). Calf groups were fed 6 L per calf/daily (2×, 3 L/feeding) of 13% total solids organic milk then weaned at 60 d when the group consumption averaged 0.91 kg starter/

calf daily. Body weight and hip height were recorded at birth, once/wk, at weaning, and at 90 d of age. Hobo Pendant G loggers were applied to the right rear leg of calves to measure total lying and standing time. Data were analyzed using PROC MIXED of SAS. Independent variables for analyses were the fixed effects of birth weight (co-variable), season of birth, treatment group, along with replicate as a random effect. Weaning group performance was gain per day, 1× (0.79 kg) and 2× (0.81 kg;  $P = 0.33$ ); weaning weight, 1× (92.7 kg) and 2× (93.3 kg;  $P = 0.80$ ); and weaning hip height, 1× (95.2 cm), and 2× (95.3 cm;  $P = 0.94$ ). Daily gain to 90 d were 0.85 vs. 0.85 kg, and daily gain to 120 d were 0.85 vs. 0.83 kg for 1× and 2× calves, respectively ( $P = 0.62$ ). For lying time, 1× (988 min/d) and 2× (995 min/d) were not different ( $P = 0.57$ ) from each other. During the evening hours, the 2× calves had lower ( $P < 0.01$ ) lying times (34 min/h for 1×; 28 min/h for 2×) per h because they were fed at 6pm every evening. In summary, group-fed calves fed once per day in an organic production had similar average daily gains and body dimensions compared with calves fed twice per day.

**Key Words:** organic, calf growth, crossbreeding

**M286 The sex ratio of female to male calves may be affected by number of services to achieve conception and lactation number of the lactating dairy cow.** Andy Mendes\*<sup>1</sup>, Michael R. Murphy<sup>2</sup>, Peter S. Erickson<sup>3</sup>, and David P. Casper<sup>1</sup>, <sup>1</sup>South Dakota State University, Brookings, SD, <sup>2</sup>University of Illinois, Champaign Urbana, IL, <sup>3</sup>University of New Hampshire, Durham, NH.

Commercial dairy producers get frustrated by the low ratio of female to male calves born because female calves are more valuable than bull calves. The objective of this data analysis was to determine if any parameters could be identified that may be influencing the ratio of female to male calves born on the dairy operation. Data from the University of Illinois and the University of New Hampshire Dairy herds were collected and summarized for calf sex, number of services to achieve conception, and lactation number of the lactating dairy cow when she conceived. Logistical regression procedures were used to analyze the data set via version 9.4 of SAS. The data set was edited to delete those cow observations when the number of services to achieve conception (n = 6) or lactation number (n = 2) were greater than 9. These cows would typically be classified as “do not breed” and sold for harvest. The final data set contained 2,987 calvings, which consisted of 1,406 females and 1,581 males (47.1 and 52.9% for females and males, respectively). The frequency distribution of number of services to achieve conception was highest for first service and progressively declined with increasing services (52.1, 21.7, 10.8, 6.7, 4.2 and 4.7% for 1 to 6 services, respectively). The frequency distribution of calvings by lactation number was highest for 1st lactation cows becoming pregnant with their 2nd calf and declined with increasing lactation number (35.5, 28.2, 17.0, 9.6, 5.0, 2.5, 1.1, 0.1 and 0.3% for lactation number 1 to 9, respectively). Logistic stepwise regression indicated that number of services to achieve conception was significant ( $P < 0.02$ ) in predicting the ratio of female to male calves. Calculation of odds ratios indicated that as the lactation number increased the likelihood of getting a bull calf decreased. These data demonstrate that increasing number of services to achieve conception and age of the cow increase the probability of getting a heifer calf. The earlier in DIM or age a cow conceives will increase the odds of the dairy producer getting a male calf.

**Key Words:** female, male, sex ratio

**M287 Margin over concentrates as a performance indicator for the dairy farms.** Aadi Remmik\*, *Estonian University of Life Sciences, Tartu, Estonia.*

Margin over concentrates (MoC) was identified as a practical indicator of management performance at dairy farms. It was found to be a more objective metric than income over feed costs (IOFC), yet it has been scarcely used in dairy farm management literature. Estonian University of Life Sciences and partner farmers conducted a study recording data on production processes in 10 midsize (190–750 milking cows) Estonian dairy farms on a monthly basis in 2012–2014. The study included detailed information on feeding and milk output. One of the key objectives of the study was to find reliable performance indicators that would assist dairy farmers in diagnosing and measuring potential weaknesses in farms' production processes. In the study, MoC was calculated as milk sales turnover less cost of concentrate feeds for the milking herd (including dry cows, but excluding youngstock). Concentrate feeds included all feeds except grass, silages (grass and corn), hay and straw. Concentrate feeds were determined the biggest single cost component at the farms (29% of all dairy-related costs on average) and its efficient use has a significant effect on total farm profitability. MoC was found to effectively summarize the financial result of several other metrics, such as milk yield, milk price, percentage of discarded milk, and so on. MoC excludes cost of forage feeds that often lack reliable market price and can have very different physical and nutritional characteristics based on preparation timing, plant material and technology. Dairy farmers could benefit from using MoC as a broad measurement tool for the milk herd performance.

**Key Words:** dairy, performance, measurement

**M288 Factors affecting the success of an embryo transfer program in dairy cattle.** Priscila Ferraz\*<sup>3</sup>, Clay Burnley<sup>4</sup>, John Karanja<sup>5</sup>, Achilles Vieira-Neto<sup>1</sup>, Jose Eduardo P. Santos<sup>1</sup>, and Klíbs N. Galvão<sup>2</sup>, <sup>1</sup>*Department of Animal Sciences, University of Florida, Gainesville, FL*, <sup>2</sup>*Department of Large Animal Clinical Sciences, University of Florida, Gainesville, FL*, <sup>3</sup>*Escola de Medicina Veterinária, Universidade Federal da Bahia, Salvador, Bahia, Brazil*, <sup>4</sup>*Southern Embryo, Athens, GA*, <sup>5</sup>*North Florida Holsteins, Bell, FL.*

Objective of the study was to evaluate factors affecting pregnancy per embryo transfer (PET) in dairy cattle. A total of 10,634 transfers in 6,734 Holstein dairy cows (primiparous = 2,287; multiparous = 1,907) and heifers (n = 6,440) from a 4,500-cow commercial herd in Northern Florida, were used. Data on collection year (2011–2014), transfer season (fall, winter, spring, summer), semen type [conventional (C), sexed (S)], embryo type (Fresh, Frozen, IVF, IVF-Frozen), embryo developmental stage (4 - morula, 5 - early blastocyst, 6 - blastocyst, 7 - expanded blastocyst, 8 - hatched blastocyst), embryo quality grade (1 - excellent/good, 2 - fair, 3 - poor), recipient parity (N - nulliparous, P - primiparous, M - multiparous), recipient estrous cycle day (5, 6, 7, 8, 9), technician (1, 2, 3, 4), DIM, and milk yield at the time of transfer were collected. The embryos were produced in vivo (superstimulation and uterine flush 7 d after AI) or in vitro (ovum pick-up, fertilization, in vitro culture for 7 d). Pregnancy was diagnosed at 41 ± 3 d of gestation. Data were evaluated by mixed logistic regression using the GLIMMIX procedure of SAS using all data listed as fixed effects and recipient ID as random. PET was affected by embryo type (Fresh = 48.1%, Frozen = 36.6%, IVF = 38.9%, IVF-Frozen = 32.6%;  $P < 0.001$ ), embryo stage (4 = 36.4%, 5 = 34.1%, 6 = 38.2%, 7 = 45.2%, 8 = 37.1%;  $P < 0.001$ ), embryo quality (1 = 42.1%, 2 = 32.6%, 3 = 23.5%;  $P < 0.001$ ), recipient parity (n = 41.9%,  $P = 37.7%$ , M = 31.6%;  $P < 0.001$ ), recipient estrous cycle day (5 = 32.6%, 6 = 36.4%, 7 = 40.8%, 8 = 40.4%, 9 = 36.6%;

$P < 0.001$ ), technician (1 = 33.9%, 2 = 33.5%, 3 = 38.0%, 4 = 43.8%;  $P < 0.001$ ), semen type (C = 39.9%, S = 37.7%,  $P = 0.03$ ), transfer year (2011 = 44.2%, 2012 = 41.5%, 2013 = 38.2%, 2014 = 36.5%,  $P < 0.001$ ). In conclusion, PET was affected by year, semen type, embryo type, embryo stage and quality, recipient parity, recipient estrous cycle day, and technician, but not by transfer season, DIM or milk yield at embryo transfer.

**Key Words:** embryo transfer, pregnancy, dairy cattle

**M289 High cow ration recipe: Preparation and feeding times.** Yolanda Trillo\*<sup>1</sup>, Sonia Rodriguez<sup>1</sup>, Alfonso Lago<sup>2</sup>, and Noelia Silvaldel-Rio<sup>1</sup>, <sup>1</sup>*UC Davis School of Veterinary Medicine, VMTRC, Tulare, CA*, <sup>2</sup>*DairyExperts, Tulare, CA.*

The objective of this study was to describe the high cow ration (HCR) recipe preparation and feeding times on 26 California dairies ranging in size from 1,100 to 6,900 cows. Consecutive records from a 12-month period were extracted from the feeding management software FeedWatch 7.0. The variables included were data, recipe, recipe number, ingredient, loading sequence, start loading time and end loading time. A total of 33 ingredient types were used across dairies. Data set included information from 50,909 recipe loads (487,218 ingredient loads). Descriptive statistics were conducted with SAS 9.3. Across dairies, the median recipe preparation time was [median (range)] 15 min 0 s (9 min 18 s to 27 min 0 s). The interquartile range (IQR =  $Q_3 - Q_1$ ) per dairy was <13 min 30 s ( $Q_1$ ) and >16 min 58 s ( $Q_3$ ). At least 20% (up to 68%) of the time, recipe preparation was < 10 min in 5 dairies, and at least 10% (up to 70%) of the time it was >25 min in 4 dairies. Across dairies, the median elapsed time from recipe preparation to start feeding was [median (range)] 3 min 48 s (1 min 54 s to 9 min 0 s). The IQR per dairy was <2 min 30 s ( $Q_1$ ) and >5 min 10 s ( $Q_3$ ). On 6 dairies, at least 70% (up to 80%) of the time, recipe preparation to start feeding took <3 min in 6 dairies. On 6 dairies, at least 20% (up to 30%) of the time it took >10 min. Across dairies, the median feeding time was [median (range)] 4 min 12 s (1 min 30 s to 10 min 48 s). The IQR per dairy was <3 min 20 s ( $Q_1$ ) and >6 min 30 s ( $Q_3$ ). At least 20% (up to 75%) of the time, feeding took <2 min in 4 dairies; and between 20 to 55% of the time, it was >10 min in 4 dairies. Overall, from recipe preparation to feeding, 25% of the times ( $Q_1$ ) it took <15 min (n = 10), 15 to 20 min (n = 12) and >20 min (n = 4); and 25% of the time ( $Q_3$ ) it took <25 min (n = 5), 25 to 35 min (n = 16) and >35 min (n = 5). There were important differences within and across dairies on time elapsed from recipe preparation to feeding. This may have implication on mixing and processing of the HCR recipe.

**Key Words:** dairy cattle, high cow ration, feeding management software

**M290 Using first test milk yield and previous lactation data to assess herd transition cow management in Brazilian dairy farms.**

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The objective of this study was to develop an index to assess the management applied to dairy cows during transition period, known as Transition Cow Index (TCI), using information from official DHIA Paraná herds, Southern Brazil. Data set was assigned by Holstein Cattle Breeders Association from Paraná State (APCBRH) and it had originally 310,007 first monthly test-days collected between 5 and 40 d after calving from 2000 to 2013. To generate this index several information from

previous lactation were used. First-lactation data were excluded and animals were categorized in 2nd, 3rd, 4th, and 5th lactation or more. After editing, only Holstein data were kept in the final data set, which had 94,508 first test-days from 445 herds. Pearson correlations among independent variables and milk yield on first test-day were estimated by CORR procedure from SAS. Previous cumulative milk yield was the variable with the highest correlation with the first test-day milk yield ( $r = 0.42$ ). Previous cumulative milk yield, DIM in prior lactation (linear and quadratic effects), linear somatic cell score at the last test-day of previous lactation, days dry (linear and quadratic effects), lactation number, month of calving, milking frequency and DIM at first test were the independent variables used in GLM procedure from SAS to generate an expected milk yield in the first test-day after calving. The expected daily production was compared with the actual production collected, to generate TCI in kg/d. During the whole period investigated, 50.9% of the analyzed data had positive TCI (above expectations) and 49.1% had negative TCI (below expectations) and the overall TCI average was  $-0.283$  kg/d. The 5 best herds with 100 or more test-days had TCI average of  $3.908$  kg/d and the 5 worst herds with 100 or more test-days had TCI average of  $-5.589$  kg/d. Transition Cow Index proved to be an efficient tool for evaluation of transition period management of Paraná herds. It is recommended to not overlook individual cows that fail and to use TCI only at herd level.

**Key Words:** early lactation, fresh cow performance, transition cow index

**M291 Differences in rumination time, lying time, and rectal temperature between cows with and without metritis, ketosis, and subclinical hypocalcemia.** I. C. Tsai\*, L. M. Mayo, A. E. Stone, B. A. Wadsworth, and J. M. Bewley, *University of Kentucky, Lexington, KY.*

The objective of this study, conducted at the University of Kentucky Coldstream Dairy from June 2014 to January 2015, was to quantify differences in rumination time, lying time, and rectal temperature between cows with and without metritis, ketosis, and subclinical hypocalcemia. Precision dairy farming technologies used for these measurements included: SensOor (Agis Automatisering, Harmelen, the Netherlands) which measured ear movement, feeding time, rumination time, and ear skin temperature; Track-a-Cow (ENGS, Hampshire, UK) which measured lying time and time at the feed bunk; and Precision Xtra (Abbott Laboratories, Abbott Park, Illinois) which measured blood  $\beta$ -hydroxybutyrate (BHBA). Ten days before due date, technologies were assigned to cows. Blood serum samples were collected on 3, 7, 14, and 21 DIM and sent to the University of Kentucky Veterinary Diagnostic Laboratory for calcium analysis. The GLM procedure of SAS (Version 9.3, Cary, NC) was used to evaluate factors affecting metritis, ketosis, and subclinical hypocalcemia. Mean rumination time, lying time, and rumen temperature were calculated for the first 21 DIM. Average lying time percent for cows with subclinical hypocalcemia ( $63\% \pm 8\%$ ) was greater than for cows without subclinical hypocalcemia ( $57\% \pm 9\%$ ;  $P < 0.002$ ). Average rectal temperature of metritis cows ( $39.13 \pm 0.22$ ) was higher than for cows without metritis ( $37.75 \pm 0.29$ ;  $P \leq 0.05$ ). No differences in rumination time were observed between cows with or without metritis, ketosis, and subclinical hypocalcemia. Precision dairy farming technologies could be useful for detecting metritis, ketosis, and subclinical hypocalcemia.

**Key Words:** transition period, metabolic disease, precision dairy farming technology

**M292 Bacterial and dry matter content of bedding substrates utilized on Canadian dairy farms.** Ivelisse Robles\*<sup>1</sup>, David F. Kelton<sup>2</sup>, Herman W. Barkema<sup>3</sup>, Greg P. Keefe<sup>4</sup>, Jean-Philippe Roy<sup>5</sup>, Marina A. G. von Keyserlingk<sup>6</sup>, and Trevor J. DeVries<sup>1</sup>, <sup>1</sup>*Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada*, <sup>2</sup>*Department of Population Medicine, University of Guelph, Guelph, ON, Canada*, <sup>3</sup>*Faculty of Veterinary Medicine, University of Calgary, Calgary, AB, Canada*, <sup>4</sup>*Atlantic Veterinary College, University of Prince Edward Island, Charlottetown, PEI, Canada*, <sup>5</sup>*Faculté de Médecine Vétérinaire, Université de Montréal, Montréal, QC, Canada*, <sup>6</sup>*Animal Welfare Program, University of British Columbia, Vancouver, BC, Canada.*

The objective of this study was to determine the bacteria and DM content of bedding substrates utilized on dairy farms. Seventy-two dairy farms in Ontario, Canada, were visited 3 times, 7 d apart. At each visit, composite samples of the unused and used bedding material were collected for DM determination and bacterial culturing of major pathogen groups (gram-positive, gram-negative, and *Klebsiella* spp.). Bedding substrates were classified by farm as following: sand ( $n = 12$ ), straw and other dry forage ( $n = 34$ ), wood products (shavings, sawdust;  $n = 17$ ), and other (recycled manure solids, compost, digestate, peat moss;  $n = 10$ ). Tests of bacteria counts and DM content were made between bedding types, as well as between unused and used samples, in a general linear mixed model. DM content of unused bedding varied with type ( $P < 0.01$ ): sand was driest ( $92 \pm 1.4\%$ ), followed by straw ( $88 \pm 0.9\%$ ) and wood ( $85 \pm 1.2\%$ ), while other types were wettest ( $39 \pm 1.6\%$ ). DM content of used bedding was reduced ( $P < 0.01$ ) for wood ( $75 \pm 1.7\%$ ) and straw ( $77 \pm 1.2\%$ ), increased for other types ( $48 \pm 2.3\%$ ;  $P < 0.01$ ), and did not change for sand ( $95 \pm 2.1\%$ ;  $P = 0.17$ ). Gram-positive bacteria counts did not vary with bedding type ( $P > 0.25$ ) in unused ( $2.4 \pm 1.2$  ln cfu/mL) and used ( $16.1 \pm 0.9$  ln cfu/mL) samples. Gram-negative bacteria counts in unused bedding varied with type ( $P < 0.01$ ): straw was greatest ( $12.6 \pm 0.6$  ln cfu/mL), followed by other types ( $8.1 \pm 1.3$  ln cfu/mL), then wood ( $4.7 \pm 0.8$  ln cfu/mL), while sand was least ( $2.4 \pm 1.0$  ln cfu/mL). Gram-negative counts in used samples were greater ( $P < 0.01$ ) for other types ( $16.8 \pm 1.1$  ln cfu/mL), sand ( $14.2 \pm 0.8$  ln cfu/mL), and wood ( $10.0 \pm 0.7$  ln cfu/mL), while straw did not change ( $13.4 \pm 0.5$  ln cfu/mL;  $P = 0.17$ ). *Klebsiella* spp. counts in unused bedding varied with type ( $P < 0.01$ ): straw was greatest ( $11.4 \pm 0.6$  ln cfu/mL), while wood ( $4.2 \pm 0.9$  ln cfu/mL), other types ( $3.7 \pm 1.4$  ln cfu/mL), and sand ( $2.5 \pm 1.0$  ln cfu/mL) were similarly ( $P > 0.6$ ) lower. *Klebsiella* spp. counts for used bedding were greater ( $P < 0.01$ ) for other types ( $12.3 \pm 1.3$  ln cfu/mL) and sand ( $11.2 \pm 1.0$  ln cfu/mL), while wood tended ( $P = 0.1$ ) to be greater ( $5.4 \pm 0.9$  ln cfu/mL), and straw did not change ( $11.4 \pm 0.6$  ln cfu/mL;  $P = 0.99$ ). Overall, bedding types varied in their DM, particularly between unused and used samples.

**Key Words:** bedding, dry matter, bacteria count

**M293 Risk factors affecting expression of estrus measured by activity monitors in lactating dairy cows.** Augusto M. L. Madureira\*<sup>1,2</sup>, Bruna F. Silper<sup>2</sup>, Liam B. Polsky<sup>2</sup>, Eraldo L. Drago Filho<sup>1</sup>, Sergio Soriano<sup>3</sup>, Alex F. Sica<sup>3</sup>, José L. M. Vasconcelos<sup>1</sup>, and Ronaldo L. A. Cerri<sup>2</sup>, <sup>1</sup>*Sao Paulo State University, Botucatu, SP, Brazil*, <sup>2</sup>*University of British Columbia, Vancouver, BC, Canada*, <sup>3</sup>*Colorado Dairies, Araras, SP, Brazil.*

The objective of this study was to determine risk factors related to increase in physical activity during estrus. Estrus events ( $n = 2,510$ ) from 1,358 lactating Holstein cows from 2 commercial dairies were recorded. Cows were monitored continuously by 3 automated activity monitors (AAM). A collar-mounted accelerometer (Collar; Heatime,



SCR Engineers) and a leg-mounted pedometer (Leg1; Boumatic Heat-seeker-TX) were used in farm 1. A second leg-mounted pedometer (Leg2; AfiMilk Pedometer Plus) was used in farm 2. For cows with collar and Leg1, only spontaneous estrus (SE) events were registered (1,099 events; 318 cows). Cows with Leg2-tags were induced to ovulate (IO; 1,411 events; 1,040 cows) through an E2/P4-based TAI protocol. Body condition score (BCS; 1 to 5 scale) was measured at moment of AI. SE cows' also had secondary signs of estrus (clear vaginal mucus, uterine tonus, signs of mounting) recorded. Milk production was recorded at each milking. Ovarian ultrasonography was performed at estrus. Data were analyzed by Pearson correlation and ANOVA. Collar, Leg1 and Leg2 systems' peak of activity (PA) and Collar's duration (DU) were affected by parity as multiparous expressed lower PA ( $69.3 \pm 0.8$  vs.  $75.9 \pm 1.1$  index [Collar];  $323.9 \pm 6.0$  vs.  $354.8 \pm 8.5\%$  [Leg1];  $323.04 \pm 4.6$  vs.  $336.3 \pm 5.3\%$  [Leg2]) and shorter duration ( $10.7 \pm 0.2$  h vs.  $12.0 \pm 0.3$  h) than primiparous. Cows with  $BCS \leq 2.5$  showed less intense estrus measurements (PA and DU). Lactation stage did not influence PA or DU of estrus. Cows that displayed secondary signs of estrus had greater PA and DU measured by Collar and Leg1 systems. Intensity progressively increased as more secondary signs were displayed. Milk production was negatively correlated with PA ( $r = -0.20$ ), but no difference was observed in PA and DU when milk production was categorized into quartiles. Quantitative data from activity monitors can be used to survey estrus behavior in dairy cows, but must be calibrated based on risk factors to ensure accurate behavior interpretation.

**Key Words:** pedometer, estrus, body condition score

#### **M294 Description of close-up cow recipes in California dairies.**

Sonia Rodriguez<sup>\*1</sup>, Yolanda Trillo<sup>1</sup>, Alfonso Lago<sup>2</sup>, and Noelia Silvaldel-Rio<sup>1</sup>, <sup>1</sup>UC Davis School of Veterinary Medicine, VMTRC, Tulare, CA, <sup>2</sup>DairyExperts, Tulare, CA.

The objective of this study was to describe close-up cow (CU) recipes prepared on 25 California dairies ranging in size from 1,100 to 6,900 cows. Records from a consecutive 12-mo period were extracted from the feeding management software FeedWatch 7.0. The variables included were date, recipe, recipe number, ingredient, loading sequence, target weight, actual weight and tolerance level. Descriptive statistics were conducted with SAS 9.3. Dairies prepared a median of one ( $n = 24$ ) or two ( $n = 1$ ) CU recipes per day. The median number of ingredients included in the CU recipe ranged from 3 to 5 ( $n = 18$ ) and 6 to 9 ( $n = 7$ ); and varied over time within dairy, by one ( $n = 7$ ), 2 to 3 ( $n = 2$ ), or zero ( $n = 16$ ) ingredients. The most commonly used ingredients in CU recipes were rolled corn ( $n = 19$ ), premix ( $n = 17$ ); 5 prepared it on farm), liquids ( $n = 8$ ), mineral-vitamins ( $n = 7$ ), anionic salts ( $n = 6$ ) and canola ( $n = 5$ ). The most common forages included were corn silage ( $n = 24$ ) and alfalfa hay ( $n = 20$ ), and some dairies also used straw hay ( $n = 5$ ) and oat hay ( $n = 7$ ). The ingredients most frequently added first were alfalfa hay ( $n = 11$ ), straw hay ( $n = 5$ ) oat hay ( $n = 5$ ), premix ( $n = 2$ ), yeast ( $n = 1$ ) and wheat ( $n = 1$ ). The ingredients most frequently added last were corn silage ( $n = 11$ ), liquids ( $n = 7$ ), premix ( $n = 2$ ), mineral-vitamins ( $n = 2$ ), canola ( $n = 1$ ), earlage ( $n = 1$ ) or rolled corn ( $n = 1$ ). The tolerance level (TL) (kg) of the various ingredients for all dairies enrolled was: 0 (11.2%), 0.5–20 (31.7%), 25–40 (33.5%), 50–75 (18.0%) and 100–150 (5.6%) kg. The TL allowed a deviation from the median formulated target for the various ingredients across dairies of: 0% (11.2%), > 0–2% (14.9%), > 2–5% (25.5%), > 5–10% (18.0%) and > 10% (30.4%). Deviation from target > 10% was allowed on 20 ingredient types on 20 dairies [alfalfa hay ( $n = 11$ ), rolled corn ( $n = 8$ ), mineral-vitamins ( $n = 4$ ), corn silage ( $n = 3$ ), canola ( $n = 3$ ) and others ( $n = 2$ )]. Corn silage, alfalfa hay, rolled corn and premix were ingredients

commonly found on CU recipes. In many dairies, the TL allowed to CU ingredients should be re-evaluated.

**Key Words:** dairy cattle, close-up cow ration, feeding management software

#### **M295 Agritourism: Demographics and views of customers—In-depth look at Fair Oaks Farms.** Ann Cummins\*, Nicole Widmar, Candace Croney, and Joan Fulton, *Purdue University, W. Lafayette, IN.*

This research will use data from 3 mutually exclusive data sets, where all are representative of households in a given sample for age, sex, income, and region of residence. The 3 samples only differ in geographical location (National Survey  $n = 1004$ , 5-State Survey  $n = 1029$ , Indiana Survey  $n = 797$ ). Through this presentation, we will highlight the rates of attendance at a variety of agritourism locations (i.e., national parks, museums, amusement parks, livestock farms, pick-your-own farms) and look at the demographics of those who attend these attractions, including an in-depth look at Fair Oaks Farms (FOF). In this survey we have questions which gather respondents self-reported perceptions of how well educated they are about agriculture and livestock production and questions which test their general knowledge level. We will look at consumers who attend livestock agritourism locations to see if they care more about animal welfare but are misinformed about production practices or if they care more about animal welfare and are more informed. The methods used for this research are the survey data collected and analysis via z-scores and cross tabulations. A brief highlight on the national sample includes that 15% of the sample had heard of FOF and 7% had attended FOF, we see that those who had been to FOF tended to be male, from the younger age categories, they tended to have pets, and were more highly concerned about animal welfare. They are less supportive than those who had not been to FOF for the growth of livestock agriculture in their county, but tended to more strongly believe in the importance of agriculture in the state which they reside as well as tended to be high dollar food purchasers in the supermarket.

**Key Words:** agritourism, consumer demographics, perceptions

#### **M296 Management practices on Virginia dairy farms utilizing automated calf feeders.** Alyssa M. Dietrich\* and Robert E. James, *Virginia Polytechnic Institute and State University, Blacksburg, VA.*

The objective of this study was to evaluate management practices and calf performance on 6 Virginia dairy farms using automated calf feeders (Förster Technik, Engen, Germany) in their preweaning programs. Following an initial management survey, farms were visited biweekly for approximately 28 wk. At each visit, facilities were scored on cleanliness of feeders and quality of bedding, calf starter, and drinking water. Weights (estimated by heart girth) and hip heights were measured on calves within approximately 5 d of entering the feeder system or 5 d of being weaned and averaged by farm for each age group to estimate average daily gain (ADG) and average daily height gain (ADH) over the preweaning period. One farm was an exception to this procedure, where farm staff regularly measured hip heights and calf weights with a digital scale and ADG and ADH were calculated for individual calves. Blood was collected from calves 1–7 d of age by jugular venipuncture, transported on ice, and centrifuged to estimate total serum proteins by refractometry. Calf treatment and mortality records were collected at each farm visit. Median lactating herd size was 260, ranging from 110 to 850 cows. Each farm sorted calves by age into 2 pens per feeder, with a mean number of calves  $\pm$  SD per pen of  $10 \pm 3$ . Mean ADG and

ADH  $\pm$  SD of calves were  $0.73 \pm 0.11$  kg and  $0.21 \pm 0.05$  cm. Mean  $\pm$  SD serum total protein levels were  $5.80 \pm 1.04$  g/dl. According to treatment and mortality records, mean  $\pm$  SD morbidity and mortality rates were  $61.05 \pm 18.48$  and  $3.14 \pm 2.67\%$ . All farms fed milk replacer and utilized a feeding plan with a mean starting allotment of 5.4 L/d, ranging from 4.0 to 6.0 L/d, and a mean peak allotment of 9.0 L/d, ranging from 7.0 to 16.0 L/d. Mean milk replacer concentration was 150 g powder/L water, with a range of 140 to 160 g powder/L water. Mean length of enrollment on the feeder system was 54.3 d, ranging from 48 to 63 d. The information from this study may provide insight to producers using or considering using automated feeders in their calf programs.

**Key Words:** dairy calves, management, automated calf feeder

**M297 Calculation method alters the ratio of milk true protein production to milk urea nitrogen production in late-lactation cows fed four levels of dietary crude protein.** Margaret A. Quaas-sdorff\* and Michel A. Wattiaux, *University of Wisconsin-Madison, Madison, WI.*

Our preliminary data suggested that, using DHIA data only, the ratio of milk protein production (MPP, g/d) to milk urea nitrogen production (MUNP, g/d) might be a reliable and inexpensive indicator of nitrogen (N) use efficiency of dairy cows on commercial dairy farms. Although milk urea N (MUN, mg/dL) is an indicator of N intake and urinary urea-N excretion, it does not reflect milk protein synthesis. Our main objective was to compare 5 methods of calculation of MPP:MUNP ratio with the hypotheses that the ratio would be altered neither by calculation methods that use fewer DHIA variables nor by dietary CP level. Data were from samplings collected from 2 consecutive milkings (pm and am) in wk 12 of a study with 122 Holstein cows (mean  $\pm$  SD: 303  $\pm$  55 DIM; 761  $\pm$  77 kg BW) fed a 16.2, 14.4, 13.1 or 11.8% CP (DM basis) TMR for 12 weeks. Methods of calculation were as follows: A) daily MPP divided by daily MUNP, where daily values were calculated using 6 DHIA values (milk production, milk protein %, and MUN for am and pm sampling, respectively); B) milk protein % divided by MUN (averages of am and pm for both); C) method B using am values only; D) method B using pm values only; and E) average of methods C and D. Ratios were analyzed in SAS 9.3 with PROC MIXED and single df orthogonal contrasts to compare methods B, C, D and E to method A. Overall, there was no difference between methods A and B, or A and E, but method C overestimated, and method D underestimated the MPP:MUNP ratio relative to method A (see Table 1.). These differences, however, were associated with high MPP:MUNP ratios observed when dietary CP were 13.1 and 11.8%. In this study, the MPP:MUNP ratios remained similar when calculated with average concentrations alone or average concentrations weighted for am and pm milk production.

*Contd.*

**Table 1 (Abstr. M297).** Comparison of 5 methods of calculation of MPP:MUNP ratio

Dietary CP	Method					Contrast <i>P</i> -value			
	A	B	C	D	E	A vs. B	A vs. C	A vs. D	A vs. E
16.2% of DM	291	289	309	274	291	0.906	0.222	0.239	0.982
14.4% of DM	333	329	364	304	334	0.838	0.119	0.150	0.946
13.1% of DM	494	480	627	397	512	0.760	0.009	0.043	0.686
11.8% of DM	763	744	1042	647	844	0.884	0.048	0.383	0.536
Overall	470	461	585	405	495	0.596	<0.001	<0.001	0.164

**Key Words:** nitrogen use efficiency, MUN, DHIA

**M298 Seasonality distributions of number of breedings and conception rate of Florida dairy farms.** Fernanda Ferreira\*<sup>1,2</sup> and Albert De Vries<sup>1</sup>, <sup>1</sup>*University of Florida, Gainesville, FL,* <sup>2</sup>*Embrapa Gado de Leite, Juiz de Fora, MG, Brazil.*

Florida dairy farms are seasonal in their reproductive performance due to the hot summers. The amount of seasonality across farms has not been quantified, however. The objective was to describe the distribution of seasonality in the number of breedings (NB) and the average conception rate (CR) of dairy farms in Florida. We used reproductive data from USDA-AGIL, collected through DHIA, from the year 2010. Farms with annual NB < 100 and annual average CR > 0.75 were not used. We calculated the ratio of breedings (BR) as the NB in a calendar month divided by the average monthly NB. We also calculated the CR per calendar month for each farm. A sigmoidal function was fitted per farm to measure seasonality as is common in economic studies. The final data set had 36 (BR) and 33 (CR) farms. Goodness of fit of the sigmoidal function was measured by the mean square of errors (MSE). Measures of seasonality were the range (max – min) and minmax ratio (min/max) from the sigmoidal functions. Means and 5th, 25th, 50th, 75th and 95th percentiles are reported as well as Pearson correlations. For the BR, the mean  $\pm$  SD of MSE were  $0.31 \pm 0.73$ . In the fitted model, the mean  $\pm$  SD of BR were  $1.21 \pm 1.08$ . The 5 percentiles were 0.94, 0.98, 0.99, 1.04 and 1.46, respectively. The mean BR range was 1.261 and the 5 percentiles were 0.10, 0.35, 0.695, 1.32 and 2.46, respectively. The mean minmax ratio of BR was 0.45 and 5 percentiles were –0.03, 0.27, 0.49, 0.70 and 0.90. For CR, the mean  $\pm$  SD of MSE were  $0.01 \pm 0.01$ . The mean  $\pm$  SD of CR were  $0.40 \pm 0.13$ . The 5 percentiles were 0.26, 0.30, 0.38, 0.43 and 0.63, respectively. The mean CR range was 0.30 and 5 percentiles were 0.09, 0.21, 0.27, 0.36 and 0.71 respectively. The mean minmax ratio of CR was 0.46 and the corresponding 5 percentiles were 0.05, 0.33, 0.46, 0.58 and 0.80, respectively. Correlations between the ranges and minmax ratios were –0.96 for BR and –0.72 for CR. Maximum values were typically observed around March and minimum values around August for both BR and CR. The sigmoidal function did not necessarily describe the seasonal pattern well. In conclusion, many farms were very seasonal but different measures of seasonality may be needed.

**Key Words:** seasonality, reproduction, dairy farm

**M299 An evaluation of automated milking systems in the Midwest United States.** Marcia I. Endres<sup>1</sup> and Jim A. Salfer\*<sup>2</sup>, <sup>1</sup>University of Minnesota, St. Paul, MN, <sup>2</sup>University of Minnesota Extension, St. Cloud, MN.

Automatic or robotic milking systems (AMS) are being adopted by dairy producers in the Upper Midwest United States at a relatively fast rate. There is limited research available on AMS in US dairies. This ongoing study is summarizing data collected from 52 AMS dairy farms in Minnesota and Wisconsin to describe characteristics of AMS systems and investigate factors that may influence AMS efficiency in US farms. Farms were visited once to collect housing and management information and then daily data from the AMS were remotely collected for approximately 18 mo. These data showed that on average cows were milked  $2.61 \pm 0.30$  (mean  $\pm$  SD) times per day, produced  $32.25 \pm 6.81$  kg of milk per day, and consumed  $5.03 \pm 0.95$  kg of concentrate in the AMS per day. The number of cows per robot box was  $70.25 \pm 14.06$  and it was greater for guided flow compared with free flow AMS farms ( $75.0$  vs.  $65.5$  cows per robot). Forty farms had exclusively free flow cow traffic. Milking speed was  $2.24 \pm 0.40$  L/min and total milk yield per robot unit was  $1843.5 \pm 490.9$  L/day. Total daily milk yield per robot has been suggested to be an important characteristic to assess the efficiency of AMS. Proc CORR in SAS was used to conduct a preliminary analysis of factors associated with yield per robot and milk yield per day. Factors most strongly associated ( $P < 0.001$ ) with yield per robot were milk per cow per day ( $r = 0.81$ ) and average milking speed ( $r = 0.76$ ). Other factors moderately associated ( $P < 0.001$ ) with yield per robot were average concentrate consumed per cow per day ( $r = 0.31$ ) and exit length from the AMS ( $r = 0.32$ ). Protected AMS exit lane was  $3.1 \pm 2.4$  m long and ranged from 0.3 to 8.5 m. Factors associated ( $P < 0.001$ ) with average daily milk yield per cow were milking speed ( $r = 0.79$ ), exit length ( $r = 0.51$ ) and average concentrate consumed per cow per day ( $r = 0.41$ ). Further multilevel regression analysis will provide a clearer picture of factors influencing efficiency of AMS in the United States.

**Key Words:** automated milking, robotic milking

**M300 Factors affecting vaginal temperature in high-producing lactating Holstein cows.** Eraldo L. Drago Filho\*<sup>1</sup>, Augusto M. L. Madureira<sup>1</sup>, Liam B. Polsky<sup>2</sup>, Sergio Soriano<sup>3</sup>, Alex F. Sica<sup>3</sup>, Jose L. M. Vasconcelos<sup>1</sup>, and Ronaldo L. A. Cerri<sup>2</sup>, <sup>1</sup>Sao Paulo State University, Botucatu, SP, Brazil, <sup>2</sup>University of British Columbia, Vancouver, BC, Canada, <sup>3</sup>Colorado Dairies, Araras, SP, Brazil.

The aim of this experiment was to evaluate the continuous collection of vaginal temperature for 72 h (every 10 min) in lactating Holstein cows ( $n = 480$ ; 591 data collections) allocated in one single cross-ventilation barn. Cows were fitted with a thermometer attached to a progesterone intravaginal device before AI. Milk production, DIM, skin thickness, coat color, body condition score (BCS), and pregnancy per AI (P/AI) data were collected and recorded for further analyses. Ambient temperature and humidity was measured for 72 h using a data logger for calculation of the temperature and humidity index (THI). Data were analyzed using ANOVA and Pearson correlations using proc GLM, Corr and Logistic of SAS. Maximum THI (MAX) and percentage of time above a vaginal temperature of  $39^\circ\text{C}$  (PCT) were used as data logger responses, class variables were created for MAX and PCT (High vs Low) using the median threshold. There was a low correlation between THI and PCT ( $r = 0.01$ ). Skin thickness was also poorly correlated with PCT ( $r < 0.01$ ). Cows with black color coat spent less time with high vaginal temperatures ( $P = 0.05$ ). Primiparous ( $P = 0.04$ ) and cows with low BCS ( $P < 0.01$ ) had greater PCT. Milk production was affected by parity ( $P < 0.01$ ) and PCT ( $P = 0.02$ ; High =  $43.5$  vs Low =  $41.2$  kg/d). There was a milk

production by MAX interaction for PCT ( $P < 0.01$ ), whereas only cows in the highest milk production quartile ( $>50.7$  kg/d) spent more time with high vaginal temperatures. Among the independent variables included in the model, parity ( $P < 0.01$ ), PCT ( $P = 0.03$ ;  $26.1$  vs  $17.4\%$  for Low and High, respectively) and a PCT by milk production interaction ( $P = 0.05$ ) affected P/AI. The decrease in P/AI in cows with High PCT only occurred in cows with the highest production. In summary, there is a large variability on how individual cows respond to heat stress. Parity, BCS, coat color and milk production affect PCT, particularly under high ambient temperatures. Selection of animals with efficient control of body temperature in spite of high milk production should be further approached as a strategy to maintain adequate fertility.

**Key Words:** heat stress, milk production, pregnancy per AI

**M301 Prepartum activity is associated with increased plasma NEFA and body weight loss in postpartum dairy cows.** Yu Zang\*, Ida Holásková, and Joseph W. McFadden, West Virginia University, Morgantown, WV.

Monitoring physical activity of cattle is an estrus detection tool utilized on dairy farms. Lower than normal dairy cow activity can enhance postpartum metabolic disease risk (e.g., ketosis). Considering that NEFA mobilization and BHBA synthesis can promote metabolic disease, our objective was to investigate the relationship between activity and metabolic health markers in peripartur dairy cows. Seventy-one multiparous Holstein cows ( $713 \pm 65$  kg BW) housed in a commercial herd setting were continuously enrolled. Collar mounted activity meters (DeLaval) were worn by cows  $-45$  d before expected calving until 10 DIM. Data were recorded at 1 h intervals. Blood was collected regularly. Plasma variables were measured and insulin sensitivity was estimated using RQUICKI. Cows were retrospectively categorized according to pre- and postpartum activity into those with the lowest (LOW; 25%), modest (MOD; 50%), and highest (HIGH; 25%) activity. Data were analyzed using a mixed model with repeated measures (fixed effects of activity and time). Nonparametric correlations were analyzed. For activity categorized from d  $-10$  to  $-4$ , LOW had higher plasma NEFA on d  $-15$  and 1, relative to HIGH (e.g.,  $744$  vs.  $367$   $\mu\text{M}$  on d 1;  $P < 0.01$ ). LOW had decreased RQUICKI values and glucose levels on d  $-15$  and 1, respectively, relative to HIGH ( $P < 0.01$ ). For activity categorized from d 1 to 7, LOW had higher plasma NEFA on d 1, relative to HIGH ( $P < 0.01$ ). Plasma BHBA and insulin were not modified by activity. Plasma NEFA levels were negatively correlated with previous 24-h activity on d 1 and 4 ( $r = -0.33$  and  $-0.26$ , respectively;  $P < 0.05$ ). RQUICKI values were positively correlated with previous 24-h activity on d 1 ( $r = 0.25$ ;  $P < 0.05$ ). Similar results were observed on d 1 and 4 when evaluating daytime activity. Prepartum activity was negatively correlated with postpartum BCS loss ( $P < 0.01$ ). Lower prepartum activity of dairy cows is associated with reduced insulin sensitivity, and accelerated NEFA mobilization and BCS loss postpartum. Activity monitoring within large herds has the potential to serve as an inclusive alternative to laboratory NEFA testing.

**Key Words:** activity, dairy cow, metabolic health

**M302 Adding antioxidants to ram sperm improves sperm binding capability after cryopreservation.** Jonathan M. S. Costa<sup>1</sup>, Wildelfrancys L. Souza<sup>1</sup>, Elenice A. Moraes\*<sup>1</sup>, and James K. Graham<sup>2</sup>, <sup>1</sup>Federal University of San Francisco Valley, Petrolina, PE, Brazil, <sup>2</sup>Colorado State University, Fort Collins, CO.



Cryopreserved mammalian sperm generally exhibit lower fertility than fresh sperm. An increasing number of antioxidants have been tested in attempts to improve semen quality, but improvements have not often been consistent or repeatable. The objective of this study was to determine if adding several different antioxidant combinations to ram sperm before cryopreservation could improve sperm binding to perivitelline membrane (PM) after thawing. Thirty ejaculates from 3 rams were split and diluted to  $200 \times 10^6$  sperm/ml in an egg yolk tris diluent containing different antioxidants: control, with no antioxidant; 100  $\mu$ M Melatonin plus 0,05% ascorbic acid (MEL+AA); 100  $\mu$ M MEL plus 90 $\mu$ L Trolox C (MEL+TRO); 90 $\mu$ L TRO plus 0,05% AA (TRO+AA); and 100  $\mu$ M MEL plus 0,05% AA plus 90  $\mu$ L TRO (MEL+AA+TRO). The samples were then cooled to 5°C, over 2h, and upon reaching 5°C were packaged into 0.5mL straws and frozen in static liquid nitrogen vapor for 15min before being plunged into liquid nitrogen. Straws were thawed for 30s in 37°C water and the motility and zona binding ability were determined using a CASA and fluorescence microscopy, respectively. Prior to binding analysis, sperm were stained with 35  $\mu$ g/mL Hoechst 33342 for 15 min at 37°C, and then centrifuged at 400g for 5min and the sperm suspended in 1mL TALP and 5- $\mu$ L aliquots (10,000 sperm prepared) were added to each PM. The membranes and sperm incubated together at 37°C for 2 h in an atmosphere of 5% of CO<sub>2</sub> in air, after which the PM were washed 5 times, placed onto a glass slide and examined using fluorescence microscopy at 400 $\times$ . The number of sperm bound to the membrane in 6 predetermined fields of each membrane piece was counted. Data were analyzed by ANOVA using Tukey test. All antioxidants combinations maintained higher percentages of total and progressive motile cells after thawing, than control sperm ( $P < 0.05$ ), with MEL+AA+TRO maintaining the highest percentage of motile sperm (49%). Sperm treated with MEL+AA+TRO exhibited the highest the number of sperm binding to the PM after thawing (177.63 cells;  $P < 0.05$ ) compared with other treatments. Adding antioxidants to ram sperm before freezing increased the number of sperm surviving cryopreservation. Supported by FACEPE, CAPES and UNIVASF (Brazil).

**Key Words:** ascorbic acid, melatonin, vitamin C

**M303 Distribution of detected estrus following 14-day CIDR and prostaglandin F<sub>2 $\alpha$</sub>  treatment as a pre-synchronization strategy in dairy heifers.** Courtney K. Claypool\*<sup>1</sup>, Jennifer A. Spencer<sup>1</sup>, Saulo Menegatti Zoca<sup>3</sup>, Bahman Shafii<sup>1</sup>, William J. Price<sup>1</sup>, Amin Ahmadzadeh<sup>1</sup>, Neil R. Rimbey<sup>2</sup>, and Joseph C. Dalton<sup>2</sup>, <sup>1</sup>University of Idaho, Moscow, ID, <sup>2</sup>University of Idaho, Caldwell, ID, <sup>3</sup>UNESP, Botucatu, Sao Paulo, Brazil.

Pre-synchronization is an effective management strategy in lactating dairy cows to increase the number of animals in estrus and facilitate AI during a desired time interval. However, it is not a strategy often used in nulliparous dairy heifers. The objective was to characterize the distribution of detected estrus following a 14-d CIDR and prostaglandin F<sub>2 $\alpha$</sub>  (PG) treatment as a pre-synchronization strategy in cyclic dairy heifers. Holstein heifers (n = 119) received a CIDR on d -30, and had the CIDR removed 14 d later on d -16. All heifers then received an Estroject patch and were observed daily to facilitate detection of estrus. Following removal of the CIDR, 115/119 (96.7%) heifers were detected in estrus within 5 d. The proportion of heifers detected in estrus  $\pm$  SE for each d after CIDR removal was: d 1: 12.6%  $\pm$  3% (15/119); d 2: 55.5%  $\pm$  5% (66/119); d 3: 18.5%  $\pm$  4% (22/119); d 4: 8.4%  $\pm$  3% (10/119); d 5: 1.7%  $\pm$  1% (2/119). Four heifers were not detected in estrus. Sixteen d after CIDR removal, all heifers received an injection of PG (25 mg i.m.) upon entry to the AI pen (study d 0). All heifers received tail paint and were observed for behavioral estrus daily. Following PG treatment, 114/119

(95.8%) heifers were detected in estrus within 5 d. The proportion of heifers detected in estrus  $\pm$  SE for each d after PG treatment was: d 1: 3.3%  $\pm$  2% (4/119); d 2: 16.0%  $\pm$  3% (19/119); d 3: 59.7%  $\pm$  5% (71/119); d 4: 16.8%  $\pm$  3% (20/119); d 5: 0% (0). Five heifers were not detected in estrus. For the 2 estrus events observed, peak activity was detected 2 d after CIDR removal and 3 d after PG treatment. Pre-synchronization with a 14 d CIDR and PG protocol appears to be an effective strategy to facilitate detection of a high percentage of dairy heifers in estrus.

**Key Words:** dairy heifer, pre-synchronization, CIDR

**M304 Feeding time variation on California dairies.** Yolanda Trillo\*<sup>1</sup>, Sonia Rodriguez<sup>1</sup>, Alfonso Lago<sup>2</sup>, and Noelia Silva-del-Rio<sup>1</sup>, <sup>1</sup>UC Davis School of Veterinary Medicine, VMTRC, Tulare, CA, <sup>2</sup>DairyExperts, Tulare, CA.

The objective of this study was to describe the variation of high cow ration (HCR) recipe feeding on 26 California dairies ranging in size from 1,100 to 6,900 cows. Records from a consecutive 12-mo period were extracted from the feeding management software FeedWatch 7.0. The variables included were date, recipe, recipe number, feeding sequence, start feeding time, end feeding time, target weight, actual weight and pen number. Data set included information from 51,195 recipes. Descriptive statistics were conducted with SAS 9.3. The median day-to-day variation on start feeding time [median (range)] was |12| min (|6| to |60| min). The interquartile range (IQR = Q<sub>3</sub> - Q<sub>1</sub>) per dairy was |<15| min (Q<sub>1</sub>) to |>33| min (Q<sub>3</sub>). On 23 dairies, at least 70% (up to 93%) of the feedings were done within 15 min from the previous day feeding time. Across dairies, at least 2% of the time feedings were delayed by 60 to 90 min (n = 23; up to 17% of the time), by 90 to 120 min (n = 23; up to 5%), and by >120 min (n = 26; up to 9.6%). On 9 dairies, at least 5% (up to 25%) of the total feedings were delayed at least by 2 h relative to the previous day. On those dairies, delays affected 75 to 100% of the pens, at least once. Feeding delays of 2 h or more were observed in all dairies, ranging from 0.05 to 2% (n = 18), 2 to 5% (n = 7) and 9.6% (n = 1). The median time elapsed between the last feeding to the first feeding of the next day was 23 h 50 min, 14 h 20 min to 23 h 30 min and 15 h 50 min to 19 h 00 min for dairies feeding once a day (n = 2), twice a day (n = 18), and thrice a day (n = 6). The IQR for the time elapsed between the last feeding to the first feeding was <1 h 20 min (Q<sub>1</sub>) to >4 h 25 min (Q<sub>3</sub>). The median time to complete the first feeding to all HCR pens [median (range)] was 1 h 18 min (10 min 58 s to 4 h 24 min). The IQR was <18 min (Q<sub>1</sub>) to >45 min (Q<sub>3</sub>). At first feeding, the amount of HCR recipe delivered [median (range)] was 49.7% (33.1 to 100%) of the total amount fed per day. The IQR was <1.8% (Q<sub>1</sub>) and >19.4% (Q<sub>3</sub>). Based on our data, some dairies could improve their time consistency at first feeding.

**Key Words:** dairy cattle, high cow ration, feeding time consistency

**M305 Association among body condition score change, milk yield, and reproductive performance of Holstein cows.** Ricardo C. Chebel\*<sup>1</sup>, Pablo J. Pinedo<sup>2</sup>, Jose E. P. Santos<sup>1</sup>, Gustavo Schuene-mann<sup>3</sup>, Guilherme J. M. Rosa<sup>4</sup>, Robert Gilbert<sup>5</sup>, Rodrigo C. Bicalho<sup>5</sup>, Klubs N. Galvao<sup>1</sup>, Christopher Seabury<sup>6</sup>, John Fetrow<sup>7</sup>, William W. Thatcher<sup>1</sup>, and Sandra L. Rodriguez-Zas<sup>8</sup>, <sup>1</sup>University of Florida, Gainesville, FL, <sup>2</sup>Texas A&M AgriLife Research, Amarillo, TX, <sup>3</sup>Ohio State University, Columbus, OH, <sup>4</sup>University of Wisconsin, Madison, WI, <sup>5</sup>Cornell University, Ithaca, NY, <sup>6</sup>Texas A&M University, College Station, TX, <sup>7</sup>University of Minnesota, St Paul, MN, <sup>8</sup>University of Illinois at Urbana-Champaign, Urbana, IL.

Objectives were to determine the association among body condition score change from calving to 35 DIM, resumption of cyclicity by 50 DIM and both productive and reproductive performance. Holstein cows ( $n = 5,175$ ) from 9 dairy herds from the Southwest, Southeast and Midwest of the USA were used in this experiment. Cows were BCS at  $3 \pm 3$  and  $35 \pm 3$  DIM. Cows were classified according to BCS change [Gained(G), No change(NC), Moderate loss(ML) =  $-0.25$  to  $-0.75$ , and Extreme loss(EL)  $< -0.75$ ]. Study personnel examined cows for retained placenta and metritis. Blood sampled at 7 DIM was used to diagnose ketosis (BHB  $> 1.0$  mmol/L). Calf gender, occurrence of calving problems (twins, stillbirth, and dystocia), and occurrence of mastitis, displacement of abomasum, and respiratory illness within the first 60 DIM were recorded. Cows were milked thrice daily and average milk yield in the first 90 DIM recorded. Continuous data were analyzed by ANOVA, dichotomous data were analyzed by logistic regression, and pregnancy hazard ratio was analyzed by Cox Proportional Hazard Ratio. The interaction between BCS at 3 DIM and BCS change was associated with milk yield ( $P < 0.01$ ). Among cows calving with BCS  $< 3.25$ , milk yield was lowest for cows gaining BCS and cows with extreme BCS loss from 3 to 35 DIM (G =  $34.4 \pm 0.3$ , NC =  $37.1 \pm 0.3$ , ML =  $38.2 \pm 0.3$ , XL =  $34.4 \pm 4.4$  kg/d). Among cows calving with BCS = 3.25 to 3.5 (G =  $35.4 \pm 0.5$ , NC =  $36.3 \pm 0.3$ , ML =  $37.9 \pm 0.2$ , XL =  $39.1 \pm 1.0$  kg/d) and cows calving with BCS  $> 3.5$  (G =  $26.0 \pm 2.3$ , NC =  $35.0 \pm 1.0$ , ML =  $37.5 \pm 0.4$ , XL =  $38.5 \pm 0.9$  kg/d) milk yield was greatest when cows had extreme BCS loss from 3 to 35 DIM. Change in BCS from 3 to 35 DIM was associated ( $P < 0.01$ ) with likelihood of cows resuming cyclicity by 50 DIM (G = 78.8%, NC = 76.4%, ML = 74.8%, XL = 75.9%). Although BCS change was not associated ( $P < 0.02$ ) with the probability of pregnancy after first postpartum AI, pregnancy hazard ratio was associated with BCS change from 3 to 35 DIM [G = 1.2 (0.99, 1.56), NC = 1.21 (0.98, 1.51), ML = 1.10 (0.90, 1.36), EL = referent]. In conclusion, excessive loss of BCS from 3 to 35 DIM was associated with greater milk yield and reduced cyclicity and reproductive performance.

**Key Words:** Holstein cow, body condition score, reproductive performance

**M306 Animal and dietary factors associated with daily rumination time during the periparturient period.** Daniela N. Liboreiro\*<sup>1</sup>, Luiz Ferraretto<sup>2</sup>, Randy Shaver<sup>2</sup>, and Ricardo C. Chebel<sup>1,3</sup>, <sup>1</sup>University of Minnesota, St Paul, MN, <sup>2</sup>University of Wisconsin, Madison, WI, <sup>3</sup>University of Florida, Gainesville, FL.

The objectives of the current experiment were to determine the association among animal and feed factors associated with daily rumination of periparturient dairy cows. Holstein animals ( $n = 297$ ) were fitted with collars that record minutes of rumination every 2 h and activity every 2 h (SCR) from  $-21$  to 21 d relative to calving. Rumination data were analyzed in 24 h intervals. Animal data collected were parity, twinning, calf sex, animal body condition score at  $-21$  d relative to calving, and days relative to calving. Total mixed ration samples were collected twice weekly. Samples from the same week were combined and analyzed for crude protein, neutral detergent fiber, ether extract, ash, and non-fiber carbohydrate. Furthermore, weekly feed samples were evaluated for particle size using a 4-panel particle separator. Physically effective fiber was calculated. Each pen in which study cows were housed had a temperature humidity HOBO and temperature humidity index were calculated. Daily rumination data were analyzed, separately for the prepartum and postpartum periods, by ANOVA using the MIXED procedure of SAS. A stepwise backward elimination procedure was conducted and only independent variables with  $P < 0.10$  were kept in the final model. During the prepartum period, non-fiber carbohydrate ( $P < 0.01$ ) was

positively associated with daily rumination time and parity ( $P = 0.04$ ), days relative to calving ( $P = 0.01$ ), BCS at  $-21$  d relative to calving ( $P = 0.05$ ), twinning ( $P = 0.08$ ), daily average THI ( $P < 0.01$ ), crude protein ( $P < 0.01$ ), neutral detergent fiber ( $P < 0.01$ ), ash ( $P < 0.01$ ), and physically effective NDF ( $P = 0.01$ ) were negatively associated with daily rumination time. During the postpartum period, days relative to calving ( $P < 0.01$ ), crude protein ( $P < 0.01$ ), ether extract ( $P = 0.01$ ), non-fiber carbohydrate ( $P < 0.01$ ) were positively associated with daily rumination time and parity ( $P < 0.01$ ), daily average THI ( $P < 0.01$ ), neutral detergent fiber ( $P < 0.01$ ), ash ( $P < 0.01$ ), were negatively associated with daily rumination time. The findings of the current experiment demonstrate that variability in daily rumination time is dependent on several animal, dietary, and environmental factors.

**Key Words:** Holstein cow, rumination

**M307 Deviation from the formulated target weight for ingredients loaded into the high cow ration on California dairies.**

Yolanda Trillo\*<sup>1</sup>, Alfonso Lago<sup>2</sup>, and Noelia Silva-del-Rio<sup>1</sup>, <sup>1</sup>UC Davis School of Veterinary Medicine, VMTRC, Tulare, CA, <sup>2</sup>DairyExperts, Tulare, CA.

The objective of this study was to describe the deviation from target weight of the various ingredients added into the high cow ration (HCR) on 26 California dairies ranging in size from 1,100 to 6,900 cows. Records from a consecutive 12-mo period were extracted from the feeding management software FeedWatch 7.0. The variables included were date, recipe, recipe number, ingredient, loading sequence, target weight, actual weight and tolerance level (TL, deviation allowed per ingredient during loading). Descriptive statistics were conducted with SAS 9.3. The median TL assigned to ingredients across dairies ranged from 9 to 90 kg. The TL allowed a deviation from the median formulated target for the various ingredients across dairies of 0% (8.7%),  $> 0$ –2% (45.2%),  $> 2$ –5% (25.5%),  $> 5$ –10% (11.6%) and  $> 10$ % (9.0%). A total of 2.5% of the observations did not reach the target weight set by TL, and represented from 0.1 to 21.1% of all loads within a given dairy. Across dairies the median deviation from the target weight for the HCR recipe varied from  $< -2$  ( $n = 2$ ),  $-2$  to  $< -1$  ( $n = 2$ ),  $-1$  to 0 ( $n = 6$ ),  $> 0$  to 1 ( $n = 11$ ),  $> 1$  to 2% ( $n = 3$ ),  $> 2$ % ( $n = 2$ ). Seven dairies had 20 to 47% of their loads with a deviation from target  $> 40$  kg. On 2 dairies 59% and 27% of the observations had a deviation from target of  $< -40$  kg. Rolled corn and almond hulls were loaded with an interquartile range (IQR =  $Q_3 - Q_1$ )  $< 20$  kg on 68% of the dairies. The deviation from target ( $Q_1$ ) was  $< 10$  kg on 77% of the dairies. Overall, alfalfa hay, corn silage and canola had a IQR  $> 20$  kg on 60 to 62% of the dairies, and a deviation from target ( $Q_3$ )  $> 40$  kg on 35 to 45% of the dairies. This represents a deviation (as percentage) of 2.1 to 12.9% (alfalfa hay), 2.2 to 5.5% (corn silage), and 2.3 to 7.3% (canola). Some dairies were able to load ingredients very close to the target weight, however, dairy producers should evaluate if readjusting the TL settings of some ingredients might reduce the deviation from target weight.

**Key Words:** dairy cattle, loading deviations from target, feeding management software

**M308 Feeding frequency of the high cow ration recipe on California dairies.** Yolanda Trillo\*<sup>1</sup>, Alfonso Lago<sup>2</sup>, and Noelia Silva-del-Rio<sup>1</sup>, <sup>1</sup>UC Davis School of Veterinary Medicine, VMTRC, Tulare, CA, <sup>2</sup>DairyExperts, Tulare, CA.

The objective of this study was to describe the feeding frequency of high cow ration (HCR) recipe on 26 California dairies ranging in size

from 1,100 to 6,900 cows. Records from a consecutive 12-mo period were extracted from the feeding management software FeedWatch 7.0. The variables included were date, recipe, recipe number, feeding sequence, start time, end time and pen number. Descriptive statistics were conducted with SAS 9.3. The median frequency of feedings per pen within a dairy varied from 1 (n = 2), 2 (n = 10), 1 to 2 (n = 6), 1 to 3 (n = 4) and 2 to 4 (n = 1) times per day. Sixteen dairies had at least 90% of the pens fed the same number of times consistently (80% of the times or more) and 10 dairies had at least 30% of the pens fed the same number of times inconsistently (60% of the times or less). For dairies with a median of 2 feedings per day (n = 18), the time elapsed between the first and second feedings was <3 h (n = 8), from 3 to 6 h (n = 8), and from 6 to 9 h (n = 8). For dairies with 3 feedings per day (n = 6), the time elapsed between the first and the second feeding ranged from 2 to 4 h, and between the second and the third feeding was between 2 to 5 h. Dairies with 1, 2, or 3 feedings per day started feeding at 0300 or 0530 a.m. (n = 2), 0215 to 1030 a.m. (n = 18), and 0200 to 0815 a.m. (n = 6) respectively. The last feeding was done from 0530 a.m. to 1530 p.m. for dairies feeding 2 times (n = 18) and from 0800 a.m. to 1330 p.m. (n = 6) for dairies feeding 3 times. Some dairies were consistent (at least 85% of the time), feeding each recipe load to a single pen (n = 4), 2 pens (n = 2) or 3 pens (n = 2). However, most dairies (n = 18) split a recipe load among a combination of 1, 2, 3, 4, or more than 4 pens. At least 50% of the times, a recipe load was split among 4 or more pens in 3 dairies. One dairy split 37% of the times a recipe load among 8 pens. There was an important variation on the number of feedings per pen within and across dairies. Many dairies were inconsistent on how they split a recipe load among pens.

**Key Words:** dairy cattle, high cow ration, feeding management software

**M309 The effects of milk production class on serum haptoglobin concentrations in mid-lactation Holstein cows with and without clinical mastitis.** G. Taasoli\*<sup>1</sup>, F. Kafilzadeh<sup>1</sup>, D. Ghadimi<sup>2</sup>, and M. A. Ballou<sup>3</sup>, <sup>1</sup>*Department of Animal Science, Razi University, Kermanshah, Iran*, <sup>2</sup>*Institute of Physiology and Biochemistry, MRI,*

*Karlsruhe, Germany*, <sup>3</sup>*Department of Animal and Food Sciences, Texas Tech University, Lubbock, TX.*

This experiment investigated the link between production class and inflammation in mid-lactation Holstein cows with or without clinical mastitis. Forty cows were sampled, 20 positive for clinical mastitis and 20 healthy control cows. Within the mastitis and healthy groups, cows were further classified by level of milk production (Low = 23 to 25 kg/d vs. High = 28 to 35 kg/d). Therefore the experimental design was a 2 × 2 factorial with health status and level of milk production as the main effects (n = 10 cows per health status × level of milk production). All cows were sampled from a single commercial dairy farm with 1200 lactating Holstein cows with an average milk yield of 28 kg/d. Sampled cows were 80 to 120 DIM in their 2nd or 3rd parity. A positive mastitis case was defined as clinical signs and a positive California mastitis test. Cows had ad libitum access to feed and water. A peripheral blood sample was collected from each cow and analyzed for plasma haptoglobin concentrations. Among the healthy cows, plasma haptoglobin concentrations were greater for the high production group compared with the low production group (132 ± 12.9 vs. 93 ± 23.0 µg/mL; *P* ≤ 0.05). Although cows with mastitis had greater plasma haptoglobin concentrations compared with the healthy cows, there was no difference (*P* = 0.71) between the low and high milk production groups for cows with clinical mastitis (1,193 ± 238 vs. 1,145 ± 211 µg/mL, high and low, respectively). These data indicate that increased milk production among healthy Holstein cows may cause an increase in low level inflammation when compared with cows that produced less milk. The mild increase in plasma haptoglobin concentrations may be associated with a greater intake of certain nutrients and/or overall metabolic activity. Clinical mastitis caused a large increase in plasma haptoglobin concentrations that was independent of milk production class.

**Key Words:** inflammation, mastitis, production



# Ruminant Nutrition: Beef I

**M310 Effect of narasin in mineral mix to Nellore heifers fed with high forage.** Renan G. Silva\*<sup>1</sup>, Marcos V. C. Ferraz Junior<sup>1</sup>, Vinicius N. Gouvea<sup>1</sup>, Daniel M. Polizel<sup>1</sup>, Marcelo H. Santos<sup>1</sup>, Alexandre A. Miszura<sup>1</sup>, Thiago S. Andrade<sup>1</sup>, Mariana F. Westphalen<sup>2</sup>, Marcos V. Biehl<sup>2</sup>, and Alexandre V. Pires<sup>2,1</sup>, <sup>1</sup>University of São Paulo, Pirassununga, São Paulo, Brazil, <sup>2</sup>University of São Paulo, Piracicaba, São Paulo, Brazil.

The use of feed additives for ruminants is a further tool that can improve feed conversion in animal protein. The objective of this study was to determine the effect of narasin in mineral mix (MM) of heifers fed high-forage diet. Thirty Nellore heifers (12 mo old) were penned (3 heifers / pen) by BW ( $222 \pm 3.0$ ). Heifers were fed daily with Tifton 85 haylage ad libitum. Experimental treatments were 0 (control); 650 and 1300 mg of narasin / kg MM (0N, 6.5N and 13N, respectively). The variables analyzed were dry matter intake (DMI), and mineral mix intake (MMI) daily, average daily gain (ADG) and feed:gain ratio. The experimental design was a change-over (all pens underwent all treatments) with 3 experimental periods of 28 d. Between periods there was a 7 d wash-out. Heifers were weighed at the beginning and end of each experimental period to determine ADG. The variables ADG and feed:gain were analyzed by PROC MIXED (SAS 9.3), MMI and DMI measures were analyzed as repeated measures over time. Treatments, periods and pens were placed in the model. The addition of narasin in MM did not effected MMI (0N -  $60.70 \pm 3.05$ ; 6.5N -  $55.62 \pm 3.18$ ; 13N -  $57.44 \pm 2.50$  g / heifer / d,  $P = 0.7497$ ), corresponding to 0; 7.30 and 15.62 ppm of narasin intake. There was no change on DMI (0N -  $4.85 \pm 0.09$ ; 6.5N -  $4.95 \pm 0.11$ ; 13N -  $4.78 \pm 0.09$  kg / heifer / d,  $P = 0.6944$ ). However, adding 1300 mg of narasin / kg MM increased ADG (0N -  $0.486 \pm 0.02$ ; 6.5N -  $0.512 \pm 0.02$ ; 13N -  $0.582 \pm 0.02$  kg / d,  $P = 0.0181$ ) and improved feed:gain ratio (0N -  $12.71 \pm 0.85$ ; 6.5N -  $11.70 \pm 0.85$ ; 13N -  $9.50 \pm 0.85$ ,  $P = 0.0488$ ) compared with the control group. In conclusion, adding 1300 mg of narasin / kg MM increased ADG and improved the feed:gain ratio.

**Key Words:** narasin, Nellore, high-forage

**M311 Ruminant microbiology of Nellore steers fed different sources of forage in diets with crude glycerin in feedlot.** Telma T. Berchielli\*, Andressa F. Ribeiro, Yury G. Salcedo, Antonio Jose Neto, Luis G. Rossi, Monaliza O. Santana, Ana Laura E. G. F. Carvalho, Erick E. Dallantonia, and Juliana D. Messana, Sao Paulo State University, Jaboticabal, Sao Paulo, Brazil.

The aim of this study was to evaluate the effect of alternative forages for corn silage such as sugar cane and sugar cane bagasse included in a similar forage NDF level (fNDF) in diets with crude glycerin (80.64% of glycerol) on ruminal microbiology. Nine ruminally cannulated Nellore steers ( $300.0 \pm 30$  kg of BW and  $18 \pm 2$  mo of age) were used in a 3x3 Latin Square experimental design. Experimental periods were 15 d (14 d for adaptation and 1 d for ruminal sampling). Ruminal samples were taken 3 h after feeding. Protozoa population were preserved in formalin and quantified in Sedgewick-Rafter chamber. The technique to quantify total bacteria was qPCR. Data were analyzed using R software (version 3.1.1) as a triple Latin Square design with 3 treatments and 3 animals in 3 simultaneous replicates. The fixed effects were treatments and Latin Square, and random effects were time, animal and error. The statistical test used was Tukey, and the significance was  $P < 0.05$ . The protozoa population was not influenced by the different sources of forage with

crude glycerin, except *Dasytricha* and *Isotricha*. The concentration of *Dasytricha* population was increased in animals fed with sugar cane which not differed from animals fed with corn silage diet ( $P > 0.05$ ). Additionally, *Isotricha* population concentration increased ( $P < 0.05$ ) in animals fed with sugar cane. The population of fibrolytic bacteria (*Ruminococcus flavefaciens*, *Ruminococcus albus* and *Fibrobacter succinogenes*) were similar ( $P > 0.05$ ) among diets. On the other hand, the population of *Selenomonas ruminantium* increased ( $P < 0.01$ ) in animals fed with corn silage. Corn silage and sugar cane included in 15% of fNDF in diets with crude glycerin (10% DM) altered ruminal microorganisms populations.

**Key Words:** bacteria, corn silage, protozoa

**M312 Performance of young Nellore bulls during the dry season with two supplements.** Marcella de Toledo Piza Roth\*<sup>1</sup>, Matheus Henrique Moretti<sup>2</sup>, Flavio Dutra de Resende<sup>3,4</sup>, Rodolfo Maciel Fernandes<sup>1</sup>, Ana Paula de Toledo Piza Roth<sup>2</sup>, and Gustavo Mazende Siqueira<sup>3,4</sup>, <sup>1</sup>UNIFEB, Barretos, São Paulo, Brazil, <sup>2</sup>Agrocercos Multimix, Rio Claro, São Paulo, Brazil, <sup>3</sup>Agência Paulista de Tecnologia dos Agronegócios, Colina, São Paulo, Brazil, <sup>4</sup>UNESP, Jaboticabal, São Paulo, Brazil.

The effects of 2 supplements on the performance of Nellore cattle in their post-weaning phase on a marandu-grass pasture during the dry season were evaluated. It was expected that differences would occur in the average daily gain (ADG), thereby modifying the carcass gain (CG) and carcass gain percent (CGP). The experiment was divided into 3 experimental periods (EP) with 35 d each (105 d), beginning in 08/2009. A total of 129 animals aged  $8 \pm 2$  mo, with an initial BW of  $204 \pm 5.2$  kg, were used in the experiment. The following supplements were tested: protein supplement (PS) ( $1 \text{ g} \cdot \text{kg}^{-1}$  BW, low-moderate ADG rate) and protein energy supplement (PES) ( $5 \text{ g} \cdot \text{kg}^{-1}$  BW, moderate-high ADG rate). Animals were weighed at the beginning and end of the EP (after 16 h deprivation) to determine the ADG. For the calculation of CG, the initial carcass weight was estimated through the equations obtained with 3 animals slaughtered at the onset. The CGP was calculated by dividing the carcass gain by the BW gain. The animals were allocated to a 48.2-ha pasture area (deferred in April 2009) divided into 18 paddocks (9 per treatment). The experimental design was completely randomized, with the paddock considered the experimental unit. To evaluate performance, 9 paddocks were considered per treatment (63 animals), while for the evaluations of carcass and body components, 3 animals were considered per treatment (6 animals); these results were subjected to ANOVA at 10% probability. Treatment PES provided a 44% higher ADG ( $P < 0.01$ ) as compared with PS. The animals ADG was 0.72 kg (PES) and 0.50 kg (PS), resulting in a different BW at the end of the dry season ( $P < 0.01$ ), which was 26 kg greater in the animals fed PES (285 kg) as compared with PS (259 kg). CG was not affected ( $P = 0.35$ ) by the type of supplementation; however, the final carcass weight in the animals fed PES was 14.7% higher than in those which received PS ( $P = 0.05$ ). The CGP (average  $547.3 \text{ g carcass kg}^{-1}$  BW) was not affected ( $P = 0.94$ ) by the supplements. Supplementation with PES increases the ADG and provides heavier carcasses.

**Key Words:** average daily gain, protein supplement, protein-energy supplement

**M313 Transcriptional profiles of adipose tissue from finishing beef steers fed diets supplemented with *Aspergillus oryzae*.** Kristen M. Brennan\*, Daniel E. Graugnard, Allison C. Smith, Tatijana M. Fisher, and Leya F. Spangler, *Alltech Inc., Center for Animal Nutri-genomics and Applied Animal Nutrition, Nicholasville, KY.*

The objective of this study was to compare the transcriptional profiles in adipose tissue of steers fed diets supplemented with *Aspergillus oryzae* (Amaize, Alltech Inc.). Angus-cross steers were randomly assigned to one of 3 treatments (n = 9/treatment): basal diet (CON, control), basal diet plus 10g/hd/d of *A. oryzae* introduced gradually during the transition phase (GRAD, gradual), or basal diet plus 10g/hd/d of *A. oryzae* introduced at full level at the end of the transition phase (FULL). For the GRAD treatment, *A. oryzae* was added gradually to the ration by increasing the dose 2.5 g every 3 d until steers were receiving 10g/d at d 19. From receiving until d 21, starter and step-up diets were fed to transition steers to a low-forage finishing ration. On d 140, adipose tissue was biopsied. Samples were analyzed for gene expression using the Affymetrix Bovine Genome Array. Performance did not differ between treatment groups, however GRAD and FULL steers had numerically greater hot carcass weights ( $P = 0.54$ ) and rib eye areas ( $P = 0.15$ ) than CON steers. Relative to CON, there were 930 (396 upregulated, 534 downregulated) differentially expressed genes (DEG) in FULL steers and 425 (196 upregulated, 229 downregulated) DEG in GRAD steers. In FULL steers, DEG categorized to both the oxidative phosphorylation and mitochondrial dysfunction pathways were downregulated, suggesting a decrease in cellular energy production. Key DEG included components of complex I, III, and IV and the ATP synthase subunits. Surprisingly, these pathways were unaffected in GRAD steers. Although performance or carcass characteristics did not differ ( $P < 0.05$ ) between treatment groups, the DEG suggest that, at least in FULL steers, energy production may be decreased. A future study is warranted to examine response to Amaize in terms of steer carcass composition and transcriptional patterns in skeletal muscle.

**Key Words:** enzyme, gene expression, beef

**M314 Earlage: Yield, harvest timing, composition, and in situ digestibility.** L. Brown, S. Soderlund, and F. N. Owens\*, *DuPont Pioneer, Johnston, IA.*

To increase return per hectare and profit, production of earlage (ensiled high-moisture harvested chopped corn ears) has increased. Compared with high moisture shelled corn grain (HMSC), yield of DM as earlage is 15 to 19% greater; field grain loss is less, harvest is simpler (grain need not be shelled from the cob), and "built-in-roughage" (cob and husk) reduces the need for added forage with no additional cost of production. Effects of harvest timing on yield, composition, and in situ digestibility were examined. In trial 1, ears from 6 hybrids grown at 2 locations harvested at 3 grain maturities (56 to 87% grain DM) were separated into grain, cob, and husk plus shank portions. Within the optimum harvest moisture for HMSC (minimum of 65% for maximum grain yield and maximum of 74% for adequate fermentation), earlage contained 4.0 to 5.6% less DM. Thereby; the optimum DM range for earlage harvest to parallel HMSC would be 60 to 69% DM. For simplicity, the optimum stage for harvest for earlage can be appraised by DM content of samples of shelled grain (optimum being 65 to 74% DM). Across hybrids and growing locations, ranges in percentage of earlage DM derived from grain, cob, and husk were 81 to 85%, 7.6 to 10.8%, and 5.7 to 8.9%, respectively. Heavier ears had less cob due to incomplete grain fill of cob tips, so high plant density or other factors that decrease ear size increase the percentage of cob in earlage. Irrigation timing and other traits (e.g., stay-green) may alter moisture distribution within ears. In trial 2, ears

from 4 hybrids, one being a BMR, were harvested on 6 dates (60.6 to 80.2% DM) and ensiled for 0, 30, 60, 120, or 240 d. NDF content of earlage differed with hybrid and maturity due to changes in the break point between the cob and the shank. In situ NDFD was greater ( $P < 0.01$ ) for the BMR hybrid (67 vs 58 to 62%). Delaying harvest decreased NDF content from 26% to 20% and in situ NDFD at 48 h from 71 to 57% (due to cob maturation) without changing DM disappearance. With longer durations of storage, curvilinear decreases in NDF content (22 vs 24% of DM) and in situ NDFD (60 vs 65% of NDF) were detected, but ranking of hybrids was not altered by fermentation time.

**Key Words:** earlage, harvest, NDF digestibility

**M315 Effect of prenatal trace mineral source on neonatal and growing calf mineral status.** Deborah M. Price\*<sup>1</sup>, Alex F. Swain<sup>1</sup>, Joseph M. Guevera<sup>2</sup>, Carley R. Trcalek<sup>2</sup>, Meaghan M. O'Neil<sup>1</sup>, Max Irsik<sup>2</sup>, Owen Rae<sup>2</sup>, Matthew J. Hersom<sup>1</sup>, and Joel V. Yelich<sup>1</sup>, <sup>1</sup>*Department of Animal Sciences, University of Florida, Gainesville, FL,* <sup>2</sup>*College of Veterinary Medicine, University of Florida, Gainesville, FL.*

The effect of cow prenatal trace mineral (TM) supplement source on calf TM status from birth through 30 d of age was examined. Factorial treatment (TRT) arrangements (Angus = AN, n = 95 and Brangus = BN, n = 96 cows; Inorganic = ING, n = 98, and Organic = ORG yeast, n = 93) utilized calves born to cows supplemented (3d/wk at a rate of 0.4 kg<sup>-1</sup>454 kg BW<sup>-1</sup>d in a pellet) with TM  $\geq 90$  d before expected parturition. Calf BW were collected at birth (n = 191). A subset of calves (n = 43) had BW, serum and plasma collected by jugular vein puncture at birth (0 h, before colostrum intake), 12 h, 24 h and 30 d of age. Processed samples were frozen at -20°C and plasma was refrigerated at 4°C until analyzed for TM (serum: Co, Cu, Fe, Mn, Mo, Se, Zn; plasma: Se), by ICP-MS. Data were analyzed in SAS using Proc Mixed and repeated measures, with TRT, breed and time as fixed effects. Breed, TRT, nor interactions affected ( $P > 0.34$ ) BW at birth (36  $\pm$  0.4 kg) and 30 d (69  $\pm$  1.1 kg). With the exception of plasma Se, there were no ( $P \geq 0.39$ ) 3-way interactions. Time affected ( $P < 0.02$ ) all 0–24 h serum and plasma TM except for Co ( $P = 0.60$ ). Calf Mn concentrations were undetectable at 0–24 h, but were 1.9  $\pm$  0.2 ng/mL at 30 d sample. Both serum Se (ING = 57.5  $\pm$  1.7 and 44.7  $\pm$  1.1  $\mu$ g/mL vs. ORG = 46.5  $\pm$  1.7 and 33.4  $\pm$  1.2  $\mu$ g/mL) and plasma Se (ING = 149.4  $\pm$  3.2, 129.9  $\pm$  3.2 vs. ORG = 118.7  $\pm$  3.3, 95.4  $\pm$  3.3 ng/mL) were affected ( $P < 0.001$ ) by TM source at 0–24 h and 30 d, respectively. At 30 d, Co was greater ( $P = 0.002$ ) for ING (2.0  $\pm$  0.3 ng/mL) than ORG (0.6  $\pm$  0.3 ng/mL). At 0–24 h, Fe and Se were greater ( $P \leq 0.05$ ) in AN (87.1  $\pm$  5.9  $\mu$ g/dL and 54.7  $\pm$  1.7 ng/mL) than BN (70.1  $\pm$  5.8  $\mu$ g/dL and 49.2  $\pm$  1.7 ng/mL). At d 30, AN (40.7  $\pm$  1.2 ng/mL) serum Se was greater ( $P = 0.048$ ) than BN (37.4  $\pm$  1.1 ng/mL). In contrast, BN had greater ( $P < 0.01$ ) Cu (0.3 vs. 0.2  $\pm$  0.01  $\mu$ g/mL) than AN at 0–24 h and Mo (52.9  $\pm$  8.5 vs. 10.0  $\pm$  1.7 ng/mL) at 30 d, respectively. These data demonstrate calf TM status varies by time, prenatal TM source and breed. Pre-calving nutritional strategies need to consider TM source and calf breed.

**Key Words:** trace mineral, prenatal nutrition, calves

**M316 Effects of feeding a complete pelleted feed consisting of treated corn stover and distillers byproducts on total tract digestion and performance of growing cattle.** Jana L. Harding\*, Curt J. Bittner, Dirk B. Burken, Galen E. Erickson, and James C. MacDon-ald, *University of Nebraska Lincoln, Lincoln, NE.*

Two studies evaluated the effect of replacing traditional growing diets with a complete pelleted feed. In Exp. 1, 6 steers were utilized in a 4 × 6 Youden square to evaluate the effects of a complete pelleted feed on total-tract digestion. Four treatments consisted of 2 control diets containing 18% modified distillers grains (MDGS), 18% solubles, and either 60% untreated corn stover (-CON) or 60% alkaline treated corn stover (+CON). The remaining 2 diets were 1 of 2 complete pelleted feeds (Pellet A or Pellet B). Pellets contained alkaline treated corn stover, dry distillers grains, solubles, and supplement. Corn stover used in Pellet A and B were harvested using different methods causing Pellet B to have less stem than Pellet A. In Exp. 2, a 92-d growing study utilized 360 (initial BW = 316 ± 22 kg) yearling steers to evaluate replacing a diet consisting of 60% corn stover, 18% solubles, and 18% MDGS with a complete pelleted feed on growing cattle performance. The pelleted feed was the Pellet A treatment in Exp. 1. Pellets were either pair-fed (Pel-PF) to the control (CON) treatment (CON fed ad libitum) or fed ad libitum (Pel-AL). In Exp.1, differences ( $P < 0.01$ ) in total-tract digestibility of OM (OMD) and NDF (NDFD) between the 4 treatments were observed. Pellet B had the greatest OMD and NDFD (75.55% and 63.93%, respectively) and the -CON had the lowest (67.58% and 54.36%, respectively). The +CON and Pellet A were intermediate and statistically similar to one another ( $P \geq 0.22$ ). In Exp. 2, there were no differences ( $P > 0.50$ ) between the CON and Pel-PF in ending BW, DMI (by design), or ADG (1.29 ± 0.17 and 1.27 ± 0.22 kg/d, respectively). Greater DMI (12.18 ± 0.71 kg/d) and ADG (1.63 ± 0.20 kg/d) were observed for the Pel-AL compared with the CON and Pel-PF treatments ( $P < 0.01$ ). The Pel-AL treatment (0.134) had lower G:F ( $P = 0.05$ ) compared with the CON and Pel-PF treatments (0.143 and 0.140, respectively). Replacing a traditional growing diet with a complete pelleted feed resulted in similar or improved diet digestibility, depending on stover harvesting method. Pelleting increased DMI and ADG if cattle were fed ad libitum but decreased G:F.

**Key Words:** byproducts, pellet, treated corn stover

**M317 Additives in supplements to fed Nelore young bulls in pasture.** Erick Escobar Dallantonia<sup>1</sup>, Josiane Fonseca Lage<sup>\*2</sup>, Elias San Vito<sup>1</sup>, Gabriela Moraes Delamagna<sup>1</sup>, Patricia Baldi Garrido<sup>1</sup>, Lutti Maneck Dellevatti<sup>1</sup>, Ricardo de Andrade Reis<sup>1</sup>, Marco Antonio Alvares Balsalobre<sup>2</sup>, and Telma Teresinha Berchielli<sup>1</sup>, <sup>1</sup>*Universidade Estadual Paulista Julio de Mesquita Filho, Jaboticabal, São Paulo, Brazil*, <sup>2</sup>*Trouw Nutrition Brazil, Mirassol, São Paulo, Brazil.*

This trial aimed to evaluate the blood parameters and growth of Nelore young bulls finished in pasture in the rainy season supplemented with monensin (MON), virginiamycin (VM) or both associated. Ninety animals were used with the initial BW of 360 kg ± 24.98 kg, receiving daily supplementation in 0.3% of BW, distributed in 12 paddocks of *Brachiaria brizantha* 'Xaraés', with about 2 ha each (3 paddocks/treatment) and fed daily with supplement without additives (SUP), with MON (SUPM - 80 mg/kg), with VM (SUPV - 150 mg/kg) and supplement with MON and VM (SUPMV - 80 and 150 mg/kg, respectively). Supplements had 25% of CP. After 14 d of adaptation, 8 animals were slaughtered as a reference group to obtain the initial dressing percentage (DP) to calculate the carcass gain (CG) at the end of the experiment. The remaining animals (n = 82) were allotted in paddocks (7 animals/paddock). In one paddock were allotted 5 animals, due to the smaller size

of the area. Blood was collected after 14 h withdrawal period from feed and water at the 0, 63 and 118 d of feeding. To obtain the ADG, animals were weighted in shrunk. Animals were slaughtered after 140 d of trial. Data were analyzed using the Proc Mixed of SAS with a randomly design (5% of probability). The inclusion of additives in supplements does not affect the insulin ( $P = 0.81$ ) and glucose ( $P = 0.29$ ) concentrations. The inclusion of the additives in supplements does not affect the final BW ( $P = 0.11$ ), hot carcass weight ( $P = 0.45$ ), DP ( $P = 0.14$ ) and CG in relation the ADG ( $P = 0.91$ ). However, the ADG was affected ( $P = 0.03$ ) by inclusion of additives in supplements. Young bulls fed SUPM had similar ADG (0.867 kg/d) in relation young bulls fed SUPV (0.896 kg/d), but lower ADG than animals fed SUP (0.949 kg/d) and SUPMV (0.969 kg/d). Animals fed SUPV had similar ADG than animals fed SUP, but lower than animals fed SUPMV. The inclusion of MON, VM or both associated in supplements to fed bulls in pasture at the rainy season not promotes additional weight in carcass or BW.

**Key Words:** beef cattle, monensin, virginiamycin

**M318 Relationship between infrared thermography and heat production in young bulls.** Mario L. Chizzotti<sup>2</sup>, Rafael A. Gomes<sup>\*1,3</sup>, Karina C. Busato<sup>1</sup>, Marcio M. Ladeira<sup>1</sup>, Matheus C. Galvão<sup>1</sup>, Priscilla D. Teixeira<sup>1</sup>, Aline C. Rodrigues<sup>1</sup>, Antonio Henrique Ribeiro Junior<sup>1</sup>, and Maria Helena Oliveira<sup>1</sup>, <sup>1</sup>*Universidade Federal de Lavras, Lavras, MG, Brazil*, <sup>2</sup>*Universidade Federal de Viçosa, Viçosa, MG, Brazil*, <sup>3</sup>*Washington State University, Pullman, WA.*

Infrared thermography (IRT) was tested for predicting heat production in Nelore and Angus young bulls. A total of 32 animals with initial body weight (BW) of 380.6 ± 6.1 kg were housed in individual stalls. Eight animals of each breed were fed ad libitum with a whole shell corn (WSC) diet (85% whole shell corn and 15% of a pellet based on soybean meal) or a silage/concentrate (SC) diet (30% silage and 70% concentrate based on corn and soybean meal). After 50 experimental days, skin and eyeball temperatures were obtained by infrared thermal images (Fluke Ti 55ft, Fluke Corporation) and the pictures were analyzed using the software SmartView 3.0. The daily intake was measured and a metabolic trial was conducted with total collection of feces and urine to estimate the metabolizable energy intake (MEI). With 90 d of growth, the cattle were slaughtered. The retained energy (RE) was calculated using comparative slaughter technique. Heat production (HP) was calculated as the difference between MEI and RE. The data were analyzed using the GLM and REG procedures of SAS adopting significance level of 0.05. Angus cattle show greater daily MEI, daily HP and body temperatures than Nelore (Table 1). SC diets show greater MEI and HP, but similar eye and skin temperatures. We found a moderate correlation ( $P < 0.05$ ) between daily HP and maximum ( $r = 0.46$ ) and average skin ( $r = 0.47$ ) and maximum ( $r = 0.44$ ) and average eyeball ( $r = 0.40$ ) temperatures. We concluded that IRT technique has a potential to be used to evaluate HP in cattle.

*Contd.*



**Table 1 (Abstr. M318).** Metabolizable energy intake (Mcal/d), heat production (Mcal/d), and maximum and average temperatures of skin and eyeball (°C) of Nellore and Angus bulls fed whole shell corn (WSC) or a silage/concentrate (SC) diets

Item	Nellore		Angus		SEM	P-value		
	SC	WSC	SC	WSC		Breed	Diet	Breed × Diet
ME intake	26.3 <sup>c</sup>	21.1 <sup>d</sup>	38.1 <sup>a</sup>	32.7 <sup>b</sup>	1.8	<0.0001	0.006	NS
Heat production	21.6 <sup>bc</sup>	17.2 <sup>c</sup>	31.2 <sup>a</sup>	26.0 <sup>b</sup>	1.6	<0.0001	0.006	NS
Temperature								
Maximum skin	35.9 <sup>b</sup>	36.4 <sup>b</sup>	36.8 <sup>a</sup>	36.6 <sup>a</sup>	0.2	0.0118	NS	NS
Average skin	34.6 <sup>b</sup>	35.1 <sup>b</sup>	35.6 <sup>a</sup>	35.7 <sup>a</sup>	0.2	0.0015	NS	NS
Maximum eye	36.6 <sup>b</sup>	36.8 <sup>b</sup>	37.2 <sup>a</sup>	37.1 <sup>a</sup>	0.2	0.0184	NS	NS
Average eye	34.8 <sup>b</sup>	35.3 <sup>b</sup>	35.5 <sup>a</sup>	35.5 <sup>a</sup>	0.2	0.0328	NS	NS

**Key Words:** Nellore, Angus, metabolizable energy intake

**M319 Intake, digestibility and rumen fermentation in beef cattle fed warm-season legume (*Stylosanthes* ‘Campo Grande’) silage with two dietary protein levels.** Leandro D. Silva, Odilon G. Pereira\*, Thiago C. Silva, Sebastião C. Valadares Filho, and Karina G. Ribeiro, *Universidade Federal de Viçosa, Viçosa, MG, Brazil.*

The *Stylosanthes* ‘Campo Grande’ (*Stylosanthes capitata* + *S. macrocephala*) is adapted to tropical conditions and exhibit a nutritional value comparable to other high efficiency feeds being used in the tropics. The objective of this study was to evaluate the nutrient intake, total apparent digestibility, ruminal pH and ammonia concentration and the microbial efficiency of beef cattle fed *Stylosanthes* ‘Campo Grande’ (StS) or corn silages (CS) with 2 dietary protein levels (PL). Four Nellore rumen cannulated, average (BW) of 449.8 ± 9.47 kg were assigned to a 4 × 4 Latin square with a 2 × 2 factorial design of treatments with 4 16-d periods. The treatments were StS and CS with 2 PL (11% and 13% CP on DM basis). Data were analyzed including in the model the fixed effects of silage (S), PL and the interaction S × PL, and the random effects of animal and period. The intake and digestibility were not affected ( $P > 0.01$ ) by the interaction S × PL. However, the diets containing StS showed lower ( $P < 0.01$ ) TDN intake than CS (4.52 vs. 6.01 kg/d). Similar pattern was observed for DM, OM, CP and NDF digestibilities. The ruminal pH and ammonia concentration were affected ( $P < 0.01$ ) by S and PL, respectively. The microbial efficiency estimated from urinary purine derivatives was not affected ( $P > 0.01$ ) by treatments (average of 75.77 g/kg TDN). The StS resulted in lower TDN intake and lower total apparent digestibility of DM, OM, CP and NDF without affecting the microbial efficiency. Supported by Fapemig, CNPq and INCT-CA.

**Key Words:** stylo silage, microbial efficiency, ruminal pH

**M320 Performance of Nellore bulls supplemented with energy sources associated crude glycerin on finishing phase.** Antonio Jose Neto\*, Luis Gustavo Rossi, Erick Escobar Dallantonia, Monaliza de Oliveira Santana, and Telma Teresinha Berchielli, *Department of Animal Science, São Paulo State University, UNESP, Jaboticabal, SP, Brazil.*

The objective of this study was to evaluate the effects of crude glycerin (Gly) combined with energy sources on performance and carcass characteristics of Nellore bulls fed tropical pasture during the finish phase. Sixty Nellore bulls (424 ± 34 kg; 20 ± 2 mo) were distributed in a completely randomized design (5 animals per paddock and 3 paddocks per treatment) with 4 experimental treatments in a 2 × 2 factorial arrangement (high or low starch, with or without a source of oil). The

experimental period lasted 133 d, divided in adaptation (21 d) and 4 periods of 28 d each. The animals were allocated into 12 paddocks of 1.8 ha, consisting of *Brachiaria brizantha* ‘Xaraés’ in the dry season, and supplemented at the rate of 1000 g · 100 kg<sup>-1</sup> of BW, daily, at 1000 h. The supplements were: T1 - Gly with high starch (corn), T2 - Gly with low starch (soybean hulls), T3 - Gly with high starch associated with a source of oil (corn and soybeans), and T4 - Gly with low starch associated with a source of oil (soybean hulls and soybeans). Crude glycerin (83.9% of glycerol), a byproduct from the biodiesel agroindustry, was used in all supplements to replace (28% of DM) corn or soybean hulls. Average daily gain (ADG) was measured by weight changes during successive weightings of the animals in each experimental period. After 133 d of feeding, all the animals were slaughtered with 546 ± 43 kg of shrunk BW and was determined the hot carcass weight (HCW), dressing percent (DP), and daily carcass gain (CrG). Data were analyzed using the PROC MIXED of the SAS with  $\alpha = 0.05$  and F-test, animal was the experimental unit. There was no interaction between starch × oil for final BW, ADG, HCW, DP, and CrG ( $P = 0.11$ ). However, animals supplemented with oil increased final BW ( $P = 0.01$ ), ADG ( $P = 0.02$ ), HCW ( $P < 0.01$ ), and CrG ( $P = 0.01$ ). Independently of starch level utilized, there was an increase of 7.4% (ADG), and 2.5% (HCW) for animals fed with oil compared with without oil. Oil supplementation associated with Gly may be effective to improve performance and final carcass characteristics of Nellore bulls finished on pasture.

**Key Words:** beef cattle, glycerol, oil

**M321 Ruminant health of feedlot beef cattle fed with crude glycerin during adaptation period or finishing time.** Josimari Regina Paschoaloto\*, Jane Maria Bertocco Ezequiel, Marco Tulio Costa Almeida, Ivaldo Monsignati, Vanessa Barbosa Carvalho, Henrique Leal Perez, Antonio Carlos Homem Junior, Henrique Boselli Bussioli, Rhaony Gonçalves Leite, Anibal Garcia Camargo Junior, and Gustavo Leite Vieira, *São Paulo State University, UNESP/FCAV, Jaboticabal, São Paulo, Brazil.*

This trial evaluated the inclusion of glycerin in high concentrate diets as an ingredient is able to of reducing ruminal metabolic problems. Sixty Nellore cattle (initial BW of 300 ± 50kg) were used in a completely randomized block. Diets consisted of corn silage, corn, crude glycerin (83% glycerol), urea, sunflower meal, soybean hulls and mineral. Finishing diets presented 15% of roughage and 85% of concentrate. The crude glycerin derived from vegetable used is soybean oil and 30% was included in the diets of DM. Four treatments were used: glycerin during adaptation step up and finishing period (1) or glycerin just during adaptation period (2); absence glycerin during adaptation and finishing period (3) or absence of glycerin during adaptation and finishing period (4). The animals were housed in individual pens with free access to water. The diets were offered twice daily at 0800 and 1600h. Hepatic abscesses incidence (HAI) was determined according the presence and severity. At harvest rumenitis incidence (RI) was determined, on the entire washed rumen, using a scale of 0 (no lesions noted) to 10 (severe ulcerative). A fragment of 1 cm<sup>2</sup> of each rumen was collected from ventral sac. Manually, the number of papillae per cm<sup>2</sup> of rumen wall (NOP) was determined and 10 papillae were randomly collected from each fragment; scanned, and mean papillae area (MPA) in cm<sup>2</sup> was measured by software for image analysis. RASA in cm<sup>2</sup> was calculated as follows: 1 + (NOP × MPA) – (NOP × 0.002). The data were analyzed using GLM procedure of SAS statistical software 9.2 and were compared the means by Tukey’s test (5%). No significant ( $P > 0.05$ ) effect was observed for RI, HAI, MPA and NOP between diets, however there are difference for ASA and PSA ( $P < 0.05$ ). Diets without

glycerin during adaptation showed reduction on absorptive surface area per cm<sup>2</sup> of rumen wall and lower participation of rumen papillae in the total area of absorptive surface (%). Use of glycerin during adaptation led to greater development of ruminal epithelium, which may represent a new technology to control rumen acidification.

**Key Words:** Nellore, papillae, rumenitis

### **M322 Feeding behavior of feedlot beef cattle fed with high level of crude glycerin with crude glycerin during adaptation period or finishing time.**

Josimari Regina Paschoaloto\*, Jane Maria Bertocco Ezequiel, Marco Tulio Costa Almeida, Bruno Henrique Ferreira Araujo, Vanessa Barbosa Carvalho, Henrique Leal Perez, Ivaldo Monsignati, Henrique Boselli Bussioli, Rhaony Gonçalves Leite, Anibal Garcia Camargo Junior, and Gustavo Leite Vieira, *São Paulo State University-UNESP/FCAV, Jaboticabal, São Paulo, Brazil.*

This trial evaluated the inclusion of glycerin in high concentrate diets as an ingredient is able to of reducing ruminal metabolic problems, for this was evaluated the behavior feeding of 60 Nellore cattle (initial BW of 300 ± 50 kg) assigned in a completely randomized block. Diets consisted of corn silage, corn, crude glycerin (83% glycerol), urea, sunflower meal, soybean hulls and mineral. Finishing diets presented 15% of roughage and 85% of concentrate. The crude glycerin derived from vegetable used is soybean oil and 30% was included in the diets of DM. Four treatments were used: glycerin during adaptation step up and finishing period (1) or glycerin just during adaptation period (2); absence glycerin during adaptation and finishing period (3) or absence of glycerin during adaptation and finishing period (4). The animals were housed in individual pens with free access to water. The diets were offered twice daily at 0800 and 1600h with 50% of total in each meal. The data were analyzed using GLM procedure of SAS statistical software 9.2 and were compared the means by Tukey's test considering 5% of significance level. There were no difference treatments among the variables: ruminating time, total idle, ruminating efficiency, bolus number and mastication per day ( $P > 0.05$ ). The exceptions were the variables: dry matter intake, feeding time, feeding dry matter efficiency ( $P < 0.05$ ). Diets with glycerin finishing promoted greater feeding time and lower dry matter intake, probably this effect is due to energy contribution of glycerin, which modulated the animal intake. Glycerin increases the frequency of meals and reduces meal size which is beneficial for ruminal fluid pH. It is concluded, that the use of glycerin results in better synchronization in time between feed intake (acid production) and rumination (saliva production).

**Key Words:** feed intake, Nellore, ruminating time

### **M323 Supplementary levels of *Macleaya cordata* plant extract Sangrovit-RS on feedlot performance and carcass traits of finishing bullocks.**

Rubén Barajas\*<sup>1</sup>, Billy J. Cervantes<sup>2</sup>, Ingo Rogge<sup>3</sup>, Alejandro Plascencia-Jorquera<sup>4</sup>, Alejandro Camacho<sup>1</sup>, and Marco A. Osuna<sup>1</sup>, <sup>1</sup>FMVZ-Universidad Autónoma de Sinaloa, Culiacán, Sinaloa, México, <sup>2</sup>Ganadera Los Migueles, S.A. de C.V., Culiacán, Sinaloa, México, <sup>3</sup>Phytobiotics Futtermittelzusatzstoffe GmbH, Eltville, Germany, <sup>4</sup>FMVZ-Universidad de Baja California, Mexicali, Baja California, México.

*Macleaya cordata*, is a plant of the *Papaveraceae* family that contains quaternary benzophenanthridine and protopine alkaloids, showing systemic anti-inflammatory effects. Despite *Macleaya cordata* plant extract being used in Europe as feed additive for livestock (Sangrovit RS; Phytobiotics, Eltville, Germany), its effects on feedlot cattle per-

formance are not well documented. Hence, 60 bullocks (368 ± SD 44.7 kg) were used to determine the effect levels of *M. cordata* plant extract Sangrovit-RS (S-RS) supplementation level on feedlot performance and carcass traits of finishing bullocks. Animals were crowded by breed in groups of 5 placed in 12 dirt-floor pen and randomly assigned to receive treatments as follows: 1) A 90% concentrate finishing diet (Control); 2) Control plus daily supplementation of 3 g of Sangrovit-RS (SRS)/bullock (SRS3); and 3) Control plus daily supplementation of 8 g of SRS/bullock (SRS8). Results were analyzed as a completely randomized design. Pen was the experimental unit. Body weight at d -21 and hot carcass weight were used as the associated co-variables, for performance and carcass characteristics, respectively. The influence of SRS level was analyzed using unequal spaced polynomials. Feed efficiency (Gain/feed ratio) responded quadratically to inclusion level of SRS ( $P = 0.04$ ), with means of 0.17, 0.19 and 0.15 kg/kg for Control, SRS3, and SRS8, respectively; bullocks in SRS3 shown the best response, with 12% improved feed efficiency compared with Control (0.19 vs. 0.17 kg gain/kg DM). Dry matter intake tended ( $P = 0.08$ ) to respond quadratically to SRS supplementation level, with the lowest value for SRS3. Hot carcass dressing was augmented linearly ( $P = 0.03$ ) by SRS level. KPH-fat tended to decrease linearly as SRS intake was increased ( $P = 0.11$ ). Results of this experiment suggest that feeding Sangrovit-RS in doses close to 3 g/day improves feed efficiency of finishing bullocks.

**Key Words:** feedlot performance, *Macleaya cordata*, sanguinarine

### **M324 Energy requirements of Nellore and Angus young bulls.**

Rafael A. Gomes\*<sup>1,3</sup>, Mario L. Chizzotti<sup>2</sup>, Karina C. Busato<sup>1</sup>, Jose Rodolfo R. Carvalho<sup>1</sup>, Marcio M. Ladeira<sup>1</sup>, Matheus C. Galvão<sup>1</sup>, Maria Helena Oliveira<sup>1</sup>, Aline C. Rodrigues<sup>1</sup>, and Priscilla D. Teixeira<sup>1</sup>, <sup>1</sup>Universidade Federal de Lavras, Lavras, MG, Brazil, <sup>2</sup>Universidade Federal de Viçosa, Viçosa, MG, Brazil, <sup>3</sup>Washington State University, Pullman, WA.

Fourty-8 animals with initial body weight (BW) of 380.2 ± 4.73 kg were used to determine energy requirements of Nellore and Angus young bulls using comparative slaughter technique. Four animals of each breed were slaughtered at beginning of the experiment (baseline animals). The remainder were housed in individual stalls. Eight animals of each breed were fed ad libitum with a whole shell corn (WSC) diet (85% WCS and 15% of a pellet based on soybean meal) or a silage/concentrate (SC) diet (30% silage and 70% concentrate based on corn and soybean meal). Other 4 animals of each breed were fed with the SC diet at 50% of feed restriction, based on dry matter intake adjusted for metabolic BW of animals that received the SC diet ad libitum. We measured the daily intake and a metabolic trial was conducted with total collection of feces and urine to estimate the metabolizable energy intake (MEI). At 90 d of growth, the bulls were slaughtered. Heat production (HP) was calculated as the difference between MEI and retained energy (RE). The net energy requirements for maintenance (NE<sub>m</sub>) was estimated considering the intercept of the equation  $HP = a \times e^{(b \times MEI)}$ . The metabolizable energy requirements for maintenance (ME<sub>m</sub>) was calculated by iteration, assuming the value which HP and MEI are the same. The efficiency of ME utilization for maintenance (k<sub>m</sub>) was obtained as the NE<sub>m</sub> divided by the ME<sub>m</sub>. The slope of the regression of RE on MEI was assumed to be the efficiency of energy utilization for growth (k<sub>g</sub>). The data were analyzed using the GLM and NLIN procedures of SAS adopting significance level of 0.05. NE<sub>m</sub> was different between Angus and Nellore (99.8 vs 75.8 kcal/kg EBW<sup>0.75</sup>/d). The ME<sub>m</sub> was 150 and 122 kcal/kg EBW<sup>0.75</sup> and the k<sub>m</sub> was 67 and 62% for Angus and Nel-

lore cattle, respectively. We did not find difference between breeds ( $P > 0.05$ ) in  $k_g$  (20%).

**Key Words:** maintenance, growth

**M325 Influence of particle size of sugar cane and concentrate level on intake and digestibility of nutrients, and rumen fermentation in crossbred steers.** T. C. da Silva\*, O. G. Pereira, D. R. da Costa, R. M. Martins, M. C. N. Agarussi, L. D. da Silva, K. G. Ribeiro, and S. C. Valadares Filho, *Universidade Federal de Vicosa, Vicosa, Minas Gerais, Brazil.*

The objective of this study was to evaluate the nutrient intake, ruminal and intestinal digestibility, and the ruminal fermentation of beef cattle fed fresh sugarcane (SC) chopped in 2 particle sizes (PS) and 2 levels of concentrate (40 and 60%, DM basis). The treatments were: 1) Fine-chopped sugarcane and 40% C (FSC-40); 2) Fine-chopped sugarcane and 60% C (FSC-60); 3) Coarse-chopped sugarcane and 40% C (CSC-40); and 4) Coarse-chopped sugarcane and 60% C (CSC-60). Four Holstein  $\times$  Zebu steers (average BW of  $510 \pm 11$  kg) were used, distributed in a  $4 \times 4$  Latin square design, with a  $2 \times 2$  factorial. The experiment lasted 64 d and was divided in 4 periods of 16 d each (10 d for adaptation and 6 d for samples and data collection). Data from intake and digestibility were analyzed including in the model the fixed effects of PS, C and the interaction PS  $\times$  C, and the random effects of animal and period. For the ruminal fermentation data, a scheme of time-repeated measurements was used considering the sampling time (before feeding, 2, 4 and 6 h after feeding). The geometric means ( $X_{gm}$ ) were 8.30 and 15.92 mm for the FSC and CSC, respectively. The intake of DM, OM and NDF were affected by the interaction PS  $\times$  C ( $P < 0.05$ ). The diet containing FSC showed higher intake of these nutrients than CSC-diet when 60% C was added. The amounts of OM and NDF digested in the rumen were affected by the interaction PS  $\times$  C ( $P < 0.05$ ). The diet FSC-60 had higher amounts of OM and NDF digested in the rumen than the diet CSC-60. The amounts of OM and NDF digested in the intestine were higher in the diets containing 60% than the diets with 40% C ( $P < 0.05$ ). The diet FSC-60 had the lowest ( $P < 0.05$ ) pH (5.75). The ruminal ammonia-nitrogen was affected by sampling time ( $P < 0.05$ ) and had a quadratic behavior. We concluded that fine-chopped sugarcane ( $X_{gm} = 8.30$  mm) in diets with 60% of concentrate increases the intake and digestibility of nutrients without compromise the ruminal fermentation of crossbred steers.

**Key Words:** effective fiber, ruminal digestibility, ruminal pH

**M326 Supplementation with a DHA-rich microalgae alters muscle gene expression profiles in finishing beef heifers.** Kristen M. Brennan\*, Daniel E. Graugnard<sup>1</sup>, Allison C. Smith<sup>1</sup>, James S. Drouillard<sup>2</sup>, and Kate A. Jacques<sup>1</sup>, <sup>1</sup>Alltech Inc., Center for Animal Nutrigenomics and Applied Animal Nutrition., Nicholasville, KY, <sup>2</sup>Kansas State University, Manhattan, KS.

The objective of this study was to determine gene expression patterns in the muscle tissue of finishing heifers fed diets supplemented with DHA-rich algae. Angus-cross heifers ( $n = 285$ ;  $453 \pm 24$  kg initial BW) were randomly assigned to one of 4 treatments: control (CON) or 50, 100, or 150 g/d of DHA-rich microalgae (ALG, *Schizochytrium limacinum* CCAP 4087/2, Alltech Inc., Nicholasville, KY). Muscle samples ( $n = 9$  per treatment) were obtained via biopsy from the *Longissimus dorsi* and gene expression was analyzed using the Affymetrix Bovine Gene 1.0 ST Array. Relative to the CON, a total of 2047 differentially expressed genes (DEG, 919 upregulated and 1128 downregulated), 711 genes (343

upregulated and 368 downregulated), 956 genes (440 upregulated and 516 downregulated), were differentially expressed in the 50, 100 and 150 g/d treatment groups, respectively. In the 50-g treatment group, DEG encoding enzymes involved in the synthesis of n-3 PUFA included ELOVL2 (-1.15 fold), ACOX1 (1.19 fold) and SCP2 (1.22 fold). These genes were unchanged in the 100-g and 150-g treatment groups. Biological functions commonly enriched in all 3 treatment groups included cellular development and lipid metabolism. DEG involved in free radical scavenging, including catalase (1.16 fold), superoxide dismutase (1.18 fold) and peroxiredoxin (1.14 fold) were increased in the 150-g treatment group, supporting previous findings that markers of lipid oxidation in muscle were increased in this group. Overall, these results suggest that supplementing finishing heifer diets with ALG alters the expression of genes involved in lipid metabolism and lipid accumulation in muscle tissue, and that these alterations are dose dependent.

**Key Words:** microalgae, gene expression, omega-3 fatty acid

**M327 Influence of a tannins extract preparation supplementation level on the feedlot performance of feedlot bullocks.** Rubén Barajas\*, Billy J. Cervantes<sup>2</sup>, Juan A. Vázquez<sup>2</sup>, Alejandro Camacho<sup>1</sup>, and Javier A. Romo<sup>1</sup>, <sup>1</sup>FMVZ-Universidad Autónoma de Sinaloa, Culiacán, Sinaloa, México, <sup>2</sup>Ganadera Los Migueles, S.A. de C.V., Culiacán, Sinaloa, México.

Eighty Brahman cross bullocks ( $399.3 \pm$  SD 19.41 kg) were utilized in a 56-d experiment to evaluate the influence of a tannins extract preparation supplementation level on the feedlot performance of feedlot bullocks. Bullocks were individually weighed and blocked by weight. Groups of 5 bullocks were placed in 16 dirt-floor pens ( $6 \times 12$  m). Pens within a block were randomly assigned to treatments follows: 1) Finishing diet (13.6% CP, 2.11 Mcal  $NE_m$ /kg) formulated with ground corn (Control, CTL); 2) Control added with 2 g of tannins extract preparation/ kg of DM (TE2); 3) Control supplemented with 4 g of tannins extract preparation/ kg of DM (TE4); and 4) Control added with 6 g of tannins extract preparation/ kg of DM (TE6). The tannins extract preparation (TE) was provided as TMP Protein Enhancer + ByPro (Tecnica Mineral Pecuaria, Mexico, that contains 50% of condensed and hydrolysable tannin blend. TE treatments were top dress on the diet in the feed bunk. Results were analyzed by ANOVA for a randomized complete block design. Three orthogonal contrasts were performed: I = 0 vs. 2 + 4 + 6 g of TE/kg DM; II = 0 vs. Two g of TE/kg DM; and III = 2 vs. Four + 6 g of TE/kg DM. and the linear, quadratic or cubic trend were tested by polynomial contrasts. The final weight of TE2 bullocks was higher than CTL ( $P = 0.05$ ), and a tendency for a quadratic response was observed ( $P = 0.10$ ). TE2 increased 13.8% ( $P = 0.05$ ) the ADG respect to CTL (1.53 vs. 1.35 kg/d). ADG show a quadratic tendency ( $P = 0.10$ ). Dry matter intake was not affected by treatments ( $P > 0.20$ ). Feed efficiency tended ( $P = 0.06$ ) to be better in TE2 treatment than CTL (0.174 vs. 0.156 kg gain/kg food). Gain/feed ratio responded in a quadratic manner ( $P = 0.05$ ) as TE supplementation was increased, the best level was calculate by polynomial regression as 3 g of TE by kg of diet DM ( $R^2 = 0.3$ ;  $P = 0.05$ ). It is concluded that performance of feedlot bullocks respond in a quadratic manner to tannins extract supplementary level, and the best response could be expected with addition of 3 g of tannins extract by kg of diet DM.

**Key Words:** bovine, feedlot performance, tannin



### M328 Effect of monensin withdrawal on intake and digestion in *Bos indicus* and *Bos taurus* steers consuming bermudagrass hay.

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Effects of monensin withdrawal and subspecies of cattle on the utilization of bermudagrass hay (13.7% CP) were evaluated using ruminally cannulated steers (5 *Bos indicus*, BI and 5 *Bos taurus*, BT; 398 kg BW). Subspecies were concurrently subjected to a 2 period, 2 treatment crossover design. Each period consisted of a 42 d adaptation phase with treatments including 0 (CON) or 200 (MON) mg·hd<sup>-1</sup> monensin, fed in 0.91 kg DDGS daily. Withdrawal of monensin was then evaluated for a 28 d phase following adaptation. Hay, ort, and fecal grab samples were collected d 23–27 after the cessation of MON feeding for determination of intake and digestion. Steers were group housed throughout the trial, but moved to individual covered pens to facilitate sampling. Data were analyzed using the MIXED procedure of SAS 9.3 (SAS Inst. Inc., Cary, NC) with terms in the model including treatment, subspecies, subspecies × treatment and period, with animal as a random effect. No subspecies × treatment interactions were observed ( $P \geq 0.15$ ). A tendency for greater ( $P = 0.09$ ) forage OM intake (FOMI; 21.2 vs 19.2 g/kg BW) and greater ( $P < 0.01$ ) OM digestibility (OMD; 72.4 vs 63.0%) resulted in greater ( $P < 0.01$ ) total digestible OM intake (TDOMI; 16.8 vs 13.2 g/kg BW) in BT vs BI steers. Previous monensin feeding had no effect ( $P \geq 0.45$ ) on FOMI, OMD, or TDOMI. Greater ( $P = 0.04$ ) forage NDF intake (FNDFI; 16.68 vs 14.94 g/kg BW) and greater ( $P < 0.01$ ) NDF digestibility (NDFD; 73.8 vs 65.6%) led to greater ( $P < 0.01$ ) total digestible NDF intake (TDNDFI; 12.9 vs 10.3 g/kg BW) in BT vs BI steers. Previous monensin feeding had no effect on FNDFI, NDFD, or TDNDFI. Increased FOMI, FNDFI, OMD, NDFD, TDOMI, and TDNDFI by BT steers suggest they are better able to utilize the bermudagrass hay offered them than BI steers. Results also indicate that upon treatment withdrawal, no intake or digestibility differences are observed between steers previously fed monensin and control.

**Key Words:** cattle subspecies, ionophore, digestibility

### M329 The influence of pellet size and a pellet binder on rumen fermentation and total tract digestibility in beef heifer.

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Using by-product feeds can be a means of reducing feed costs, however, challenges with the pelleting process may affect pellet quality. The effect of pellet size and durability of a by-product based pellet was evaluated in 8 crossbred ruminally cannulated heifers. The experiment was conducted as a replicated 4 × 4 Latin square design with a 2 × 2 factorial treatment arrangement. Pellets were prepared using a 3.96 mm (SP) or 12 mm (LP) die and contained Nutraflex Plus (3 kg/T) and Ameri-Bond 2X (5 kg/T) as binding agents (B) or no binding agent (NB). Heifers were fed a partial mixed ration (81.8% grass hay and 18.2% oat hull, DM basis) ad libitum and pellet was offered separately at a rate of 0.09% of BW. Heifers were provided 15 d for adaptation followed by 3 d for total fecal and urine collection, rumen fluid sampling and continuous rumen pH measurement using indwelling pH probes. Statistics were conducted using PROC MIXED in SAS with the fixed effect of

pellet size, binder inclusion, and the interaction. Tukey's adjusted post hoc mean separation test was used to compare treatment means where significance was declared at  $P \leq 0.05$ . Heifers fed pellet with binder had increased ( $P \leq 0.05$ ) ADG and G:F, did not differ ( $P \geq 0.33$ ) in DM intake. The molar proportion of acetate was greater (interaction,  $P = 0.03$ ) in heifers fed large pellet with binder (66.0%) compared with small pellet with binder (63.9%) whereas the large and small pellets without binder were intermediate (64.6 and 63.9%, respectively). Propionate was greater (interaction,  $P = 0.05$ ) in heifers fed large pellet (17.7%) with no binder than with binder (15.6%) and was intermediate for heifers fed the small pellet with and without binder (17.6 and 17.2%, respectively). Heifers fed binder had butyrate concentrations that were 0.68% greater ( $P = 0.05$ ) than without binder. Total-tract digestibility was not affected. The results of this experiment indicate that overall digestibility of a by-product based range pellet was not influenced by size or binder; however, pellet size and use of a binder may influence rumen fermentation, ADG, and feed conversion.

**Key Words:** pellet size, binder, beef

### M330 Effect of monensin concentration on rumen pH, short-chain fatty acid absorption, total-tract digestibility, and barrier function in beef heifers.

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The objective of this study was to evaluate whether the monensin concentration affects DMI, rumen fermentation, short-chain fatty acid (SCFA) absorption across the rumen and total-tract barrier function. Four ruminally cannulated Hereford × Angus heifers were used in a 4 × 4 Latin square design with 24-d periods. Heifers were fed a barley-based finishing diet (76% rolled barley grain, 12% barley silage, 8% mineral and vitamin supplement, and 4% canola meal) containing either 0, 22, 33 or 48 ppm monensin. Urinary recovery of Cr-EDTA was used as an indicator of total-tract barrier function (d 18 to 20). Days 20 to 23 were used to evaluate rumen fermentation and total-tract digestibility measurements, and SCFA absorption was measured on d 24. Data were analyzed using PROC MIXED of SAS with linear and quadratic contrasts. Increasing monensin inclusion decreased DMI linearly (10.0, 9.9, 9.3, and 9.1, kg/d DM respectively,  $P = 0.01$ ), but as the dose of monensin increased, BW, ADG and G:F ratio were not affected ( $P \geq 0.11$ ). Total Cr-EDTA recovery was not ( $P \geq 0.61$ ) affected by increasing dose of monensin, nor was rumen pH (mean, minimum, maximum, duration <5.5, area under curve;  $P \geq 0.21$ ). The acetate-to-propionate ratio decreased linearly (1.9, 1.8, 1.4, 1.3, respectively,  $P = 0.03$ ) as dose of monensin increased. A quadratic response (35.7, 47.5, 41.8, 35.4%, respectively,  $P = 0.02$ ) for the absorption of acetate was detected with a maximal value at 22 ppm. Ethanol soluble carbohydrate digestibility increased linearly (77.2, 84.7, 88.0, 94.0%, respectively,  $P = 0.003$ ) with increasing doses of monensin. Starch digestibility showed a quadratic response (93.8, 93.9, 88.0, 94.0%, respectively,  $P < 0.001$ ), where 33 ppm inclusion had a minimal value. The results from this study indicate that increased monensin inclusion decreases DMI without affecting SCFA concentrations, SCFA absorption, and total-tract barrier function.

**Key Words:** monensin, digestibility, barrier function

**M331 Effects of essential oils and exogenous enzymes on in vitro rumen fermentation kinetics.** Camila Delveaux Araujo Batalha<sup>1</sup>, Lucas Jado Chagas<sup>1</sup>, João Ricardo Rebouças Dórea<sup>\*2</sup>, Tiago Sabella Acedo<sup>2</sup>, Luis Fernando Tamassia<sup>2</sup>, Cristina Simões Cortinhas<sup>2</sup>, and Flávio Augusto Portela Santos<sup>1</sup>, <sup>1</sup>University of São Paulo, Piracicaba, SP, Brazil, <sup>2</sup>DSM Produtos Nutricionais Brasil SA, São Paulo, SP, Brazil.

The objective with this trial was to evaluate the combination of essential oils and exogenous enzymes on ruminal DM degradation kinetics in feedlot diets. The treatments were MON (Monensin, Tortuga – 26 mg/kg DM), CRINA (Essential Oils: Crina Ruminants, DSM – 90 mg/kg DM), CRINA+MON (90 and 26 mg/kg DM, respectively), CRINA+RUM (CRINA +  $\alpha$ -amylase: Ronozyme RumiStar, DSM – 90 and 560 mg/kg DM, respectively) and CRINA+RUM+P (CRINA+RUM+Protease: Ronozyme Proact, DSM – 90; 560 and 840 mg/kg DM, respectively). The incubated diets were composed by corn (82.5%), sugarcane bagasse (8.5%), soybean meal (5%), mineral (3%) and urea (1%). The experimental design was completely randomized, with 5 treatments and 3 replicates per incubation time. The estimated parameters of rumen degradation kinetics were soluble fraction (A), potentially degradable fraction (B), degradation rate (kd), lag time (Lag), potential degradability after 120 h of incubation time (PD = A+B). The substrate (1 g) was incubated with rumen inoculum (10 mL) and bath culture (90 mL). For each incubation time (0, 2, 4, 8, 16, 24, 48, 72, 96 and 120 h) the flask contents were filtered, dried and weighted to determine de DM disappearance. The rumen degradation parameters were analyzed using SAS system. The A and B fractions and kd were not affected by treatments ( $P > 0.05$ ). The MON diet had higher Lag ( $P < 0.05$ ) in comparison with other treatments. The reduction on lag is related to the increases on dry matter PD for CRINA+MON, CRINA+RUM and CRINA+RUM+P diets, probably because the rumen microorganisms starts the DM degradation earlier, reaching more extension of degradation compared with MON. In conclusion, the use of enzymes (amylase and protease) improves rumen DM degradation.

**Table 1 (Abstr. M331).** Combination of essential oils and exogenous enzymes on in vitro rumen fermentation kinetics

Parameter	MON	CRINA	CRINA +MON	CRINA +RUM	CRINA +RUM+P	P-value	SEM
A	20.16	20.97	19.48	21.79	19.83	0.6986	1.24
B	56.56	57.55	58.22	58.36	58.98	0.7486	1.32
kd	0.0822	0.0687	0.0739	0.0698	0.0723	0.7317	0.0064
Lag	5.34 <sup>a</sup>	3.74 <sup>b</sup>	3.07 <sup>bc</sup>	2.78 <sup>bc</sup>	2.16 <sup>c</sup>	0.0025	0.40
PD	76.71 <sup>c</sup>	78.51 <sup>abc</sup>	77.70 <sup>bc</sup>	80.15 <sup>a</sup>	78.80 <sup>ab</sup>	0.0700	0.73

**M332 Effects of essential oils and exogenous enzymes for finishing Nellore cattle in feedlot during the adaptation period.** Murillo Alves Porto Meschiatti<sup>1</sup>, Diandra Leziér<sup>1</sup>, João Ricardo Rebouças Dórea<sup>2</sup>, Tiago Sabella Acedo<sup>\*2</sup>, Luis Fernando Tamassia<sup>2</sup>, Cristina Simões Cortinhas<sup>2</sup>, and Flávio Augusto Portela Santos<sup>1</sup>, <sup>1</sup>University of São Paulo, Piracicaba, SP, Brazil, <sup>2</sup>DSM Produtos Nutricionais Brasil SA, São Paulo, SP, Brazil.

The objective with this trial was to evaluate the combination of essential oils and exogenous enzymes during the adaptation period on performance of Nellore bulls finished in feedlot. Three hundred Nellore bulls (initial BW = 330 ± 33 kg) were fed diets containing 82.5% corn, 8.5% sugarcane bagasse, 5% soybean meal, 3% mineral, 1% urea and were randomly allocated to 50 pens. Animals were blocked based on initial BW. The treatments were MON (Monensin, Tortuga – 26 mg/kg DM), CRINA (Essential Oils: Crina Ruminants, DSM – 90 mg/kg DM),

CRINA+MON (90 and 26 mg/kg DM, respectively), CRINA+RUM (CRINA +  $\alpha$ -amylase: Ronozyme RumiStar, DSM – 90 and 560 mg/kg DM, respectively) and CRINA+RUM+P (CRINA+RUM+Protease: Ronozyme Proact, DSM – 90; 560 and 840 mg/kg DM, respectively). Response variables included: dry matter intake (DMI), average daily gain (ADG), feed efficiency (FE, G:F) after 28 d on feed. The data were analyzed using PROC MIXED of SAS and means were compared by Tukey test considering the block as random effect and treatments as fixed effects. Cattle fed CRINA and CRINA+RUM diets presented greater ( $P < 0.05$ ) DMI in comparison with the other treatments. The improvements on DMI were 7.8% and 9.1% for CRINA and CRINA+RUM, respectively, compared with MON. For ADG the improvements were 14.5% and 23.3% for CRINA and CRINA+RUM, respectively, compared with MON. Feed efficiency was greater ( $P < 0.05$ ) for cattle fed CRINA+RUM diets. In conclusion, the use of essential oils and its combination with amylase improved DMI, ADG during the adaptation period. Nellore bulls were 12.4% more efficient when diets containing Amylase and essential oils were fed compared with monensin diets.

**Table 1 (Abstr. 332).** Combination of essential oils and exogenous enzymes on feedlot performance of finishing Nellore bulls during the adaptation period

Parameter	MON	CRINA	CRINA +MON	CRINA +RUM	CRINA+ RUM+P	P-value	SEM
Initial BW, kg	330.8	330.8	330.9	330.6	330.7	0.5422	10.9
Final BW, kg	382.5 <sup>b</sup>	388.2 <sup>a</sup>	382.8 <sup>b</sup>	391.2 <sup>a</sup>	379.6 <sup>b</sup>	0.0050	11.0
DMI, kg/d	7.73 <sup>b</sup>	8.26 <sup>a</sup>	7.73 <sup>b</sup>	8.41 <sup>a</sup>	7.63 <sup>b</sup>	0.0160	0.31
ADG, kg/d	1.352 <sup>b</sup>	1.549 <sup>a</sup>	1.354 <sup>b</sup>	1.667 <sup>a</sup>	1.251 <sup>b</sup>	0.0050	0.080
FE, G:F	0.177 <sup>bc</sup>	0.190 <sup>ab</sup>	0.177 <sup>bc</sup>	0.199 <sup>a</sup>	0.166 <sup>c</sup>	0.0440	0.010

**M333 Shifts in rumen microbiota in response to inoculation with *Propionibacterium acidipropionici* strain P169.** Elnaz Azad<sup>\*1</sup>, Nelmy Narvaez<sup>2</sup>, Hooman Derakhshani<sup>1</sup>, Awfa Y. Alazze<sup>2,3</sup>, Yuxi Wang<sup>2</sup>, Tim A. McAllister<sup>2,1</sup>, and Ehsan Khafipour<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Manitoba, Winnipeg, MB, Canada, <sup>2</sup>Agriculture and Agri-Food Canada, Lethbridge Research Centre, Lethbridge, AB, Canada, <sup>3</sup>Department of Clinical Nutrition, College of Applied Medical Sciences, University of Hail, Hail, Saudi Arabia.

Inoculation of cattle with *Propionibacterium acidipropionici* strain P169, a commensal rumen bacterium known for its lactate and glucose utilizing properties, has been reported to modify the profile of ruminal VFAs in favor of increased propionate production. The objective of this study was to document shifts in rumen microbiota as a result of rumen inoculation with P169. Eight ruminally cannulated steers (434 ± 33 kg) were either not inoculated (control) or inoculated with P169 ( $1 \times 10^{11}$  cfu/head/day) by including it in the diet for 101 d. Rumen samples from liquid and solid fractions were collected every 3 wks. Genomic DNA was extracted and subjected to qPCR for quantification of P169 and Illumina sequencing of the V4 region of the 16S rRNA gene for community profiling. On average, 36,250 high quality sequences were generated per sample, resulting in identification of 16 and 15 bacterial phyla in liquid and solid fractions, respectively. No significant difference was detected in  $\alpha$ -diversity indices between treatments. While PERMANOVA analysis of the UniFrac distances of microbial communities revealed distinct clustering of rumen liquid from solids ( $P = 0.03$ ),  $\beta$ -diversity did not differ between treatments. Quantitative PCR identified a trend for an increase ( $P = 0.06$ ) in the abundance of P169 in inoculated steers. Partial Least square discriminant analysis of sequence data found that the proportion of members of the Firmicutes (Clostridiaceae, Lachnospiraceae, and Ruminococcaceae, and *Phascolarctobacterium*), Bacteroidetes (Bacteroidales, families

RFP12 and BS11), and Synergistetes (Pyramids) were increased ( $P < 0.05$ ) in response to P169 ( $R^2 = 0.86$ ,  $Q^2 = 0.57$ ). Data suggest that while inoculation with P169 altered the proportions of bacteria within the rumen, it did not change the  $\alpha$  or  $\beta$  diversities of bacteria in either liquid or solid bacterial communities.

**Key Words:** *Propionibacterium acidipropionici* (P169), rumen microbiota, 16S rRNA sequencing

**M334 A comparative assessment of dried distillers grain, ionophore, bambermycin, saponin, and condensed tannin for methane emission abatement in beef cattle.** M. A. Fonseca\*<sup>1</sup>, L. O. Tedeschi<sup>1</sup>, T. R. Callaway<sup>2</sup>, and W. L. Crossland<sup>1</sup>, <sup>1</sup>Texas A & M University, College Station, TX, <sup>2</sup>USDA-ARS, College Station, TX.

The objectives of this study was to assess in vitro (IV) potential abatement of CH<sub>4</sub> production rate of 4 commercially available products [ionophore (I), bambermycin (B), saponin (S), and condensed tannin (CT)] when fed in combination with dried distillers grain (DDG) on growing feedlot type diets. The IV gas production technique was used to determine the fermentability of 3 base diets with 3 levels of DDG (0, 20, and 40% DM). Diets contained alfalfa hay, ground corn, and Bermudagrass hay. The 15 diets (DDG0, DDG20, DDG40, I0, I20, I40, B0, B20, B40, S0, S20, S40, CT0, CT20, and CT40) were incubated in duplicates, 6 times each (15 × 2 × 6 × 2; n = 360 bottles), and the alfalfa hay was incubated alone in each run as standard. Blanks (n = 2 bottles/incubation time), containing rumen fluid and medium only, were used to adjust the CH<sub>4</sub> production. Methane was computed as adjusted CH<sub>4</sub> concentration divided by the amounts of NDF (CH<sub>4</sub>NDF, mM/g NDF), NFC (CH<sub>4</sub>NFC, mM/g NFC), fermentable carbohydrate (FCHO = sugar + starch + available NDF) (CH<sub>4</sub>FCHO, mM/g FCHO), OM (OM = 100 – Ash) (CH<sub>4</sub>OM, mM/g OM), and fermentable OM (FOM = OM – Ash) (CH<sub>4</sub>FOM, mM/g FOM). The PROC MIXED of SAS (SAS Inst., Cary, NC) was used to analyze the CH<sub>4</sub> data assuming a completed randomized design. Diets, products, and products within levels were assumed to be fixed factors and replicate within diet to be random. The convergence method was REML. Products affected CH<sub>4</sub> production for total concentration ( $P = 0.0003$ ), Adj CH<sub>4</sub> ( $P < 0.0001$ ), CH<sub>4</sub>NDF ( $P < 0.0001$ ), CH<sub>4</sub>NFC ( $P < 0.0001$ ), CH<sub>4</sub>FCHO ( $P < 0.0001$ ), CH<sub>4</sub>FOM ( $P < 0.0001$ ), and CH<sub>4</sub>OM ( $P < 0.0001$ ). Ionophores had the greatest effect ( $P > 0.05$ ) among all tested products to further decrease CH<sub>4</sub> production, being the only significantly different from the DDG control diets. Levels (0, 20 and 40% DM) of DDG were not significant ( $P > 0.2076$ ) in decreasing CH<sub>4</sub> production. This is probably because the fat content in the diets were not high enough. Previous IV data from our laboratory had shown that responses in CH<sub>4</sub> mitigation due to fat content in the diet was only significant when DDG levels were over 50% or 7.3% EE in the diet. These IV results suggested that ionophores have the potential to further decrease methane production from ruminants fed DDG based diet.

**Key Words:** greenhouse gas emissions, dried distillers grain (DDG), methane mitigation

**M335 Effect of dietary supplementation of  $\beta$ -mannanase in Hanwoo growing heifers.** Jakyoom Seo<sup>1</sup>, Jeongsoo Park\*<sup>1</sup>, Junsung Lee<sup>1</sup>, Jae-Hwan Lee<sup>2</sup>, Jung-Jin Lee<sup>2</sup>, Dong Keun Kam<sup>3</sup>, and Seongwon Seo<sup>1</sup>, <sup>1</sup>Department of Animal Biosystem Sciences, Chungnam National University, Daejeon, Republic of Korea, <sup>2</sup>CTC Bio Inc., Seoul, Republic of Korea, <sup>3</sup>Cargill Agri Purina Inc., Seongnam, Republic of Korea.

Supplementation of  $\beta$ -mannanase, an enzyme that breaks down mannan, in a diet composed of high mannan contained feedstuffs (e.g., palm kernel meal, copra meal, soy hull; HMCF) improves utilization of feeds not only in monogastric animals, but also in goats and Holstein calves. No study, however, has been conducted to evaluate the effect of  $\beta$ -mannanase supplementation in Hanwoo (*Bos taurus coreanae*) growing heifers. Moreover, little is known whether  $\beta$ -mannanase supplementation also has beneficial effects in conventional corn-soy based diets (CS). The objective of this study was thus to investigate the effect of supplementation of  $\beta$ -mannanase in Hanwoo growing heifers fed a diet composed of either a CS or HMCF based concentrate mix. For this purpose, a feeding trial was conducted for 12 weeks using a total of 40 Hanwoo growing heifers (BW: 236.2 kg  $\pm$  1.1 kg, 11 mo of age). Total mixed rations (250 g/kg of ryegrass and 750 g/kg of concentrate mix) were offered ad libitum. The experimental design was 2 × 2 factorial approach with 2 different concentrate mixes (CS or HMCF based) and with or without a commercial feed  $\beta$ -mannanase (800,000 U/kg DM) product (CTCZYME, CTC Bio Inc., Seoul, Korea) at a level of 1 g/kg in concentrate mixes. In both diets, supplementation of  $\beta$ -mannanase significantly increased ADG of Hanwoo heifers by 95 g/d on average (111 g/d or 90 g/d for CS and HMCF, respectively;  $P < 0.05$ ). No significant difference was observed in DMI among treatments ( $P > 0.10$ ), and thus,  $\beta$ -mannanase supplementation tended to decrease feed conversion ratio (DMI/ADG) by 1.3 on average ( $P = 0.06$ ). The ADG of CS (943 g/d) was significantly greater than that of HMCF (854 g/d), as expected ( $P < 0.05$ ). These results indicated that supplementation of  $\beta$ -mannanase enhanced growth rate and feed efficiency of Hanwoo heifers regardless of the basal diets. Therefore, we conclude that supplementation of  $\beta$ -mannanase can be beneficial to improve the feed utilization in Hanwoo growing heifers.

**Key Words:**  $\beta$ -mannanase, growing heifer, growth rate

**M337 Calculating residual feed intake using high-frequency partial body weights.** Ann Kenny\*<sup>1</sup>, David Benfield<sup>2</sup>, Camiel Huisma<sup>2</sup>, and Kevin Garossino<sup>2</sup>, <sup>1</sup>University of Missouri, Columbia, MO, <sup>2</sup>GrowSafe Systems Ltd., Airdrie, AB, Canada.

Residual feed intake (RFI) calculations were compared using either conventional chute weighing (CW) or partial body weights (PBW) collected with specialized data acquisition systems (DAS). Per-second PBW, CW and feed intake (FI) data of beef cattle (n = 3743) from 49 contemporary groups located throughout Canada, Australia and USA was compiled. Data for PBW and FI was collected using automated DAS which non-invasively collected per-second information on individual animals while eating or drinking (GrowSafe Systems Ltd., Airdrie, AB, Canada). Chute weights were taken at varying time points while cattle were on feed. Total BW was converted from PBW using a constant conversion factor. Correlations of RFI calculation, between FI and metabolic weight ( $W^{0.75}$ ) and between FI and ADG using CW, PBW or a combination of CW and PBW (COMB) were examined. Paired T-Tests were performed to compare calculated R<sup>2</sup> values. Linear regression on CW and PBW demonstrated a high degree of correlation ( $R^2 = 0.962$ ), which remained similar when data were sorted by sex (Bull n = 5077  $R^2 = 0.962$ ; Heifer n = 4942  $R^2 = 0.953$ ; Steer n = 4331  $R^2 = 0.972$ ). The R<sup>2</sup> values of the calculated RFI were greater (0.614 vs 0.590;  $P = 0.0016$ ) for CW compared with PBW. The correlation between FI and ADG also had greater R<sup>2</sup> values for CW vs COMB ( $P = 0.0224$ ). The correlation between FI and  $W^{0.75}$  tended to have lower R<sup>2</sup> values for PBW compared with CW ( $P = 0.0551$ ) and COMB ( $P = 0.0551$ ). Comparison of individual animal RFI calculations using PBW or CW identified differences in RFI values of greater than one standard



deviation (SD) in 2.7% of animals and greater than 2 SD in 0.2% of animals. These differences were associated with chute weight errors and nonlinear growth. RFI rankings within contemporary groups changed < 23% in 90% of animals evaluated when weighing techniques were compared. The use of DAS for collecting PBW and FI measurements was determined to be a viable method for RFI ranking of animals within contemporary groups. High-frequency PBW information also provided the ability to calculate RFI with better rejection of errors caused by incorrect chute weights and issues caused by nonlinearity in growth.

**Key Words:** residual feed intake, body weight

**M338 Effect of different supplementation strategies on grazing and ingestive behavior in cattle finished on pasture.** Guilherme Felipe Berti<sup>1</sup>, Rodolfo Maciel Fernandes<sup>2</sup>, Matheus Henrique Moretti<sup>2</sup>, Mauricia Brandao Silva<sup>3</sup>, Paloma Helena Gonçalves<sup>1</sup>, Michele Aparecida Prado Alves<sup>1</sup>, Flávio Dutra Resende<sup>4,2</sup>, and Gustavo Rezende Siqueira<sup>4,2</sup>, <sup>1</sup>Centro Universitario de Barretos, Barretos, Sao Paulo, Brazil, <sup>2</sup>Universidade Estadual Paulista, Jaboticabal, Sao Paulo, Brazil, <sup>3</sup>Universidade Estadual Paulista, Botucatu, Sao Paulo, Brazil, <sup>4</sup>Agencia Paulista de Tecnologia dos Agronegocios, Colina, Sao Paulo, Brazil.

The objective of this observational study was to describe grazing and ingestive behavior of Nelore cattle under field conditions fed 2 grain supplementation strategies during the dry season. Forty-eight finish Nelore bulls ( $\pm 340$  kg of initial body weight and  $\pm 20$  mo of age)

were blocked by body weight and assigned to 6 paddocks of 2.4 ha each of *Brachiaria brizantha* 'Marandu' pastures. The experiment was conducted as a randomized complete block experiment with 2 treatment factors for 140 d from May to September 2012. Treatment factors consisted of 2 levels of grains supplementation - 1.5% BW/daily (DMI) (SUPL1.5) and 2.0% BW/daily (DMI) (SUPL2.0) offered once a day (at 8 a.m.). Were evaluated the animals daily grazing time and time spent near the trough (min/day) during continuous 72h after supplementation, with intervals of 5 min. The observation occurred in 2 different period – Day (8 a.m. until 7:59 p.m.) and Night (8 p.m. until 7:55 a.m.). Data were analyzed using the MIXED procedure of SAS with sampling time serving as a repeated measure. Paddocks served as the experimental unit. The SUPL1.5 animals had greater ( $P < 0.01$ ) grazing activity than SUPL2.0 (298.5 min and 244.2 min, respectively). Although, SUPL2.0 animals spend 20% more time near to the trough compared with SUPL1.5 animals ( $P < 0.01$ ). Period significantly affected grazing time, with animals spend more time in grazing activity during the day (166.6 min) compared with the night (104.7 min;  $P < 0.01$ ). The time spend near to the trough was greater during the day period ( $P < 0.01$ ), increased rapidly in the firsts 3 h after the grain supplementation. This study indicates that the high supplementation (2.0% of the body weight) decrease the grazing time and increase the time spent near to the trough. Supported by CNPq/BELLMAN.

**Key Words:** beef cattle, grazing behavior, finishing pasture supplementation

# Ruminant Nutrition: Dairy I

**M339 The effect of decreasing dietary cation-anion difference in the prepartum diet on urine pH and plasma minerals in multiparous Holstein cows.** B. M. Sweeney<sup>\*1</sup>, C. M. Ryan<sup>1</sup>, T. Stokol<sup>2</sup>, K. Zanzalari<sup>3</sup>, D. Kirk<sup>3</sup>, and T. R. Overton<sup>1</sup>, <sup>1</sup>Department of Animal Science, Cornell University, Ithaca, NY, <sup>2</sup>Department of Population Medicine and Diagnostic Sciences, Cornell University, Ithaca, NY, <sup>3</sup>Prince Agri Products Inc., Quincy, IL.

The objective of this study was to determine the effect of decreasing dietary cation-anion difference (DCAD) in the prepartum period on prepartum urine pH and peripartum plasma mineral concentrations. Multiparous Holstein cows ( $n = 89$ ) were allocated randomly to one of 3 prepartum diets formulated with decreasing DCAD: CON (+17.5 mEq/100 g DM), MED (+3.6 mEq/100 g DM), or LOW (-10.9 mEq/100 g DM), beginning 24 d before expected parturition. Analyzed DCADs were +18.3, +5.9, and -7.4 mEq/100 g DM. Cows were fed a common postpartum diet from parturition until 63 d in milk (DIM). Blood was collected 1 × /wk prepartum, 2 × in 24 h postpartum, 1 × /d until 5 DIM, and 3 × /wk until 14 DIM. Repeated measures analyses were conducted using the MIXED procedure of SAS with linear and quadratic effects of decreasing prepartum DCAD as contrasts. There was a quadratic effect on urine pH (CON = 8.20, MED = 7.84, LOW = 5.98;  $P < 0.01$ ). There tended to be an interaction between DCAD and parity (2nd lactation vs. 3rd and greater) for prepartum Mg ( $P = 0.08$ ) such that Mg decreased with decreasing DCAD for older cows ( $P < 0.05$ ) but not for 2nd lactation cows. Postpartum Ca increased linearly with decreasing DCAD (CON = 8.84, MED = 8.89, LOW = 9.19;  $P < 0.01$ ) with greater increases through 5 DIM (DCAD × Day;  $P = 0.06$ ). There tended to be an interaction between DCAD and parity ( $P = 0.06$ ) such that Ca was increased more for older cows fed lower DCAD levels (CON = 8.68, MED = 8.63, LOW = 9.16;  $P < 0.01$ ) than for 2nd lactation cows. Postpartum P tended to decrease linearly ( $P = 0.08$ ). For 2 d postpartum cows fed LOW had lower Mg (DCAD × Day;  $P < 0.01$ ). Incidence of hypocalcemia (HC, plasma Ca < 8.5 mg/dL) was significantly different at 1 DIM ( $P = 0.02$ ). When parity groups were analyzed separately, incidence of HC in older cows was decreased by decreasing prepartum DCAD at 0.6 ( $P = 0.07$ ), 1 and 2 DIM ( $P < 0.01$ ) but did not differ for 2nd lactation cows. Decreasing prepartum DCAD improved Ca status, decreased incidence of HC, and had varied effects on other minerals over the periparturient period.

**Key Words:** hypocalcemia, dietary cation anion difference, transition cow

**M340 Canola meal in dairy cow diets with varying concentration of starch sources.** Nadeesha K. Jayasinghe<sup>1</sup>, Kenneth F. Kalscheur<sup>\*2</sup>, Jill L. Anderson<sup>1</sup>, and David P. Casper<sup>1</sup>, <sup>1</sup>Dairy Science Department, South Dakota State University, Brookings, SD, <sup>2</sup>US Dairy Forage Research Center, USDA-ARS, Madison, WI.

Synchronization of the degradability of non-structural carbohydrate and rumen degradable protein has been identified as an effective method of increasing intestinal AA flow through increased microbial protein synthesis and more efficient ruminal fermentation, thereby increasing performance of dairy cows. Therefore, the objective was to determine the performance of lactating cows fed either corn and barley starches at varying proportions in diets containing canola meal as the major source of supplemental protein. Twelve multiparous and 4 primiparous Holstein cows ( $94 \pm 25$  DIM) were used in a  $4 \times 4$  Latin square design

with 28-d periods. The ratio of starch from ground corn and rolled barley within each treatment was 100:0, 67:33, 33:67, and 0:100. Diets contained 36% corn silage, 20% alfalfa haylage, and 44% concentrate (DM basis). Varying proportions of corn and barley had no effect ( $P > 0.10$ ) on dry matter intake (26.5 kg/d) or milk production (41.2 kg/d). Milk fat percentage (3.52%) and yield (1.42 kg/d) and milk protein percentage (2.95%) and yield (1.21 kg/d) were not affected by starch. Lactose percentage (4.86, 4.83, 4.90, and 4.88%, for 100:0, 67:33, 33:67, and 0:100, respectively) and MUN (14.8, 14.5, 15.4, and 15.1 mg/dL) responded cubically ( $P < 0.05$ ) to the changes in dietary starch proportions. Treatments did not affect energy-corrected milk (40.6 kg/d) nor feed efficiency (1.53). Increasing the proportion of barley to corn had no effect on the molar proportion of ruminal acetate and butyrate, however, propionate increased quadratically as barley increased in the diets ( $P < 0.01$ ). Ruminal ammonia concentration averaged 11.5 mg/dl and was not affected by starch source. Apparent total-tract digestibilities of DM, OM, and NDF decreased linearly ( $P < 0.05$ ) and, CP and ADF tended to decrease linearly ( $P < 0.10$ ) when the proportion of barley starch increased in the diet. Total-tract digestibility of starch was not affected by starch source and averaged 95.5%. Overall, lactation performance was not affected by feeding varying proportions of corn and barley when the diets were formulated with canola meal as the primary protein supplement.

**Key Words:** canola meal, starch source, lactating dairy cow

**M341 Dietary grape marc supplementation alters the milk protein and fatty acid profile produced by pasture-based dairy cattle.** Reuben Harland<sup>1</sup>, Aysha Morrow<sup>1</sup>, Roland Harrison<sup>1</sup>, Jana Kraft<sup>2</sup>, and Sabrina L. Greenwood<sup>\*1,2</sup>, <sup>1</sup>Lincoln University, Lincoln, New Zealand, <sup>2</sup>The University of Vermont, Burlington, Vermont.

Grape marc (GM) is a byproduct of the wine-making industry and is a rich source of polyphenols. This experiment evaluated the efficacy of feeding supplementary GM to late-lactation dairy cows fed pasture-based diets as a means to manipulate the milk fatty acid (FA) and protein content produced. The effects of condensed tannins (CT) within GM were determined through the feeding of polyethylene glycol (PEG), a CT inhibitor. Forty late-lactation Friesian x Jersey cows were used in a  $4 \times 2$  factorial design, receiving either (1) pasture only (control), (2) pasture + 350 g PEG/cow/d, (3) pasture + 2 kg DM GM/cow/d, (4) pasture + 2 kg DM GM/cow/d + 350 g PEG/cow/d, (5) pasture + 4 kg DM GM/cow/d, (6) pasture + 4 kg DM GM/cow/d + 350 g PEG/cow/d, (7) pasture + 6 kg DM GM/cow/d, or (8) pasture + 6 kg DM GM/cow/d + 350 g PEG/cow/d. Cows were offered increasing amounts of GM for 10 d and maintained on the full amount of supplement for a further 6 d. Milk yield was determined at each milking. Milk samples were collected at a.m. and p.m. milking on d 0 and 16 and analyzed for milk components (milk fat, protein, and lactose), protein profile ( $\alpha$ -,  $\kappa$ - and  $\beta$ -casein,  $\alpha$ -lactalbumin, and  $\beta$ -lactoglobulin variants A and B) and FA profile by ANOVA using GenStat. Total milk and component yields were not affected by GM or PEG ( $P > 0.05$ ). Of the short-chain FA analyzed ( $n = 13$ ; C4 to C15), 63% were negatively affected ( $P < 0.05$ ) by GM inclusion and 15% increased with PEG intake. PEG positively affected 25% of long-chain FA ( $n = 24$ ; C17 to C26), while 29% were negatively affected by GM and 8% were positively affected by GM. Inclusion of GM did not affect the content (mg/mL) of any milk protein analyzed. Cows that received dietary PEG had lower  $\alpha$ - ( $P = 0.06$ ) and  $\beta$ - ( $P = 0.02$ ) casein content than those not receiving PEG. In conclusion, GM

negatively affected the profile of some short-chain (de novo synthesized) and long-chain (diet-derived) FA, the effects of which could not be fully eradicated by dietary PEG inclusion (CT inhibition). Dietary PEG inclusion decreased the content of  $\alpha$ - and  $\beta$ - casein, suggesting that diet CT can increase the output of these proteins in milk.

**Key Words:** casein, fatty acid, byproduct

**M342 Integrating nutrient and hormonal effects on mTOR phosphorylation in the mammary cell.** Juan J. Castro Marquez\* and Mark D. Hanigan, *Virginia Tech, Blacksburg, VA.*

The objective of this work was to integrate experimental data on the effect of insulin, EAA and acetate into a model of transcription control in the mammary cell. Current representations of milk protein synthesis consider energy and dietary protein effects separately with no regulation other than by limiting production when supply of either is short. In reality, protein synthesis is simultaneously affected by EAA, energy substrates and hormones, by upregulating the initiation and elongation phases of mRNA translation. A central element to this process is the mammalian target of rapamycin (mTOR) which transfers nutritional and hormonal signals onto translation initiation and elongation proteins that, through phosphorylation, modulate protein synthesis. Phosphorylation data for protein kinase B (Akt), AMP activated protein kinase (AMPK), mTOR, eukaryotic elongation factor binding protein (4EBP1) and eukaryotic elongation factor 2 (eEF2) from a series of experiments in MACT cells and lactogenic mammary tissue slices conducted over the past 5 years was used to build a dynamic model representing each one of the above signaling proteins in their phosphorylated and dephosphorylated states. Akt and mTOR phosphorylation reactions were defined as Michaelis-Menten, whereas AMPK, 4EBP1 and eEF2 representations used mass action kinetics. Inference was based on resampling with replacement and resulting nonparametric confidence intervals (CI). The affinity constants for insulin effects on Akt (CI = 6.6, 32.8  $\mu$ g) and isoleucine on mTOR (CI = 90, 443  $\mu$ M) were within physiological concentration ranges, but the rate constant for acetate effects on AMPK was null (CI =  $2.71 \times 10^{-9}$ ,  $9.8 \times 10^{-8}$ ). Sensitivity analysis coefficients indicate isoleucine was the strongest driver (0.71) of mTOR phosphorylation compared with insulin (0.01) and acetate (0.01). Phosphorylated Akt, AMPK, mTOR, 4EBP1 and eEF2 were predicted without bias and had errors of 16, 33, 29, 28 and 33%, respectively. Insulin and isoleucine effects seem to be well represented but more work is needed to improve our understanding of the role of acetate and other AA on mTOR regulation.

**Key Words:** mTOR, casein, modeling

**M343 Prediction of daily energy status in early and mid lactation using milk and body traits.** Päivi Mäntysaari\*<sup>1</sup>, Tuomo Kokkonen<sup>2</sup>, Martin Lidauer<sup>1</sup>, and Esa A. Mäntysaari<sup>1</sup>, <sup>1</sup>*Natural Resources Institute Finland, Green technology, Jokioinen, Finland,* <sup>2</sup>*Department of Agricultural Sciences, University of Helsinki, Helsinki, Finland.*

Monitoring cow's energy status at the individual level in early lactation is important for management, but also for breeding purposes. Energy status of a cow can be estimated by calculating the energy balance (EB) from cow's energy intake and output. Alternatively, indicator traits such as body weight (BW) and body condition score (BCS) changes and milk fat-protein ratio (FP) have been proposed. However, precision of these predictions has been low. This may be related to the lack of precision in estimated EB itself, because standard estimates for energy requirements are used in its calculation. We used the plasma nonesterified fatty acids (NEFA) concentration as a biomarker of energy mobilization and

energy status, and addressed associations between NEFA concentration and energy status indicators. The data included 10032 daily BW, intake and milk, 279 BCS, and 261 NEFA measurements of 56 Nordic Red primiparous dairy cows. Plasma samples for NEFA were collected twice on lactation wk 2 and 3 and once on wk 20. The milk samples were taken on the same days as NEFA samples and on monthly test days. Daily BWs were smoothed by a regression model with fixed effect of days in milk and random animal part. The NEFA concentration on wk 20 was considered as base level and deviations from base level (dNEFA) were used in calculations. The mean ( $\pm$ sd) ECM (kg/d), milk fat and protein (%), BW (kg) and BCS (1–5) were 30.3 ( $\pm$ 4.60), 4.43 ( $\pm$ 0.51), 3.59 ( $\pm$ 0.32), 574 ( $\pm$ 53), and 3.19 ( $\pm$ 0.38), respectively. On lactation wk 2 and 3 the average NEFA concentrations were 0.704 ( $\pm$ 0.363) and 0.526 ( $\pm$ 0.275), and 0.123 ( $\pm$ 0.035) mmol/l on lactation wk 20. From all indicators daily BW change had the highest correlation (0.57) with NEFA, followed by daily FP (0.53) and BCS change (–0.20). To predict dNEFA by available indicators, a multiple linear regression model was developed. The best fit was achieved with a model including BW change, FP, BCS and its change, BCS\*BCS change, and days in milk. The correlation between predicted and observed dNEFA was 0.77, which was higher than the correlation (0.69) between dNEFA and EB.

**Key Words:** dairy cow, energy balance

**M344 Performance of dairy calves receiving probiotic containing *Bacillus subtilis* and *Bacillus licheniformis*.** Thais M. Torrezan<sup>1</sup>, Jackeline T. Silva<sup>1</sup>, Nathalia B. Rocha<sup>1</sup>, Evangelina Miqueo<sup>1</sup>, Fernanda L. M. Silva<sup>1</sup>, Samyra Baldassin<sup>1</sup>, and Carla M. M. Bittar\*<sup>1,2</sup>, <sup>1</sup>*University of Sao Paulo, ESALQ, Piracicaba, Sao Paulo, Brazil,* <sup>2</sup>*CNPq, Brasilia, DF, Brazil.*

Methods to improve calf growth and health, with reduced antibiotics use are essential for a successful and profitable dairy. This study evaluated the performance and fecal pH of dairy calves receiving milk replacer supplemented or not with probiotic. Twenty-four newborn Holstein male calves were utilized in a randomized blocks experimental design, and distributed into 2 treatments: Control: no probiotic supplementation; Supplementation of 2g/d of a *Bacillus subtilis* ( $1.6 \times 10^9$  cfu) and *Bacillus licheniformis* ( $1.6 \times 10^9$  cfu) containing probiotic (Bioplus, Chr. Hansen), via milk replacer. Calves were individually housed, with free access to water and starter (20% CP and 80% TDN), and received 4L/d of milk replacer (FeedTech, DeLaval, 21.6:15.5, 12.5% solids) until the eighth week of life, when weaned. Feed intake was monitored daily; while body weight, height withers, heart girth, hip width and fecal pH were weekly measured. Data were analyzed as repeated measures, considering treatment, age, and their interaction effects. There were no probiotic supplementation effects on growth and starter intake ( $P > 0.05$ ). As animals were growing, all parameters were significantly affected by age ( $P > 0.05$ ); however, there were no treatment and age interaction effect ( $P > 0.05$ ). On the other hand, probiotic supplementation tended to reduce fecal pH, suggesting a change in intestine microorganisms and probably a reduction on pathogens growth.

*Contd.*



**Table 1 (Abstr. M344).** Performance of calves receiving milk replacer supplemented with probiotics

Item	Treatment			$P <^1$		
	Control	Probiotic	SEM	T	A	T × A
Initial BW, kg	36.54	35.47	0.82	0.48	—	—
Weaning BW, kg	57.22	56.64	1.99	0.89	—	—
ADG, kg	0.39	0.44	0.03	0.49	0.0001	0.81
Average starter intake, g/d	372.8	362.0	28.6	0.89	0.0001	0.72
Withers height, cm	78.8	79.2	0.2	0.68	0.0001	0.98
Heart girth, cm	81.1	80.7	0.4	0.71	0.0001	0.39
Hip width, cm	22.0	22.1	0.09	0.68	0.0001	0.86
Fecal pH	6.75	6.79	0.04	0.07	0.001	0.02

<sup>1</sup>T = treatment effect; A = age effect; T × A = treatment and age interaction effect.

**Key Words:** additive, health, supplement

**M345 Growth performance in Crossbred (Holstein x Gyr) calves differing in phenotypic residual feed intake on pre-weaned period.** Juliana Mergh Leão<sup>\*1</sup>, Fernanda Samarini Machado<sup>2</sup>, Mariana Magalhães Campos<sup>2</sup>, Juliana Campos Carneiro<sup>3</sup>, Paulo Campos Martins<sup>1</sup>, Isabela Carvalho Costa<sup>4</sup>, Paulo Sérgio Dornelas Silva<sup>4</sup>, Brenda Karoline Alcântara Faria<sup>3</sup>, Juliana Aparecida Mello Lima<sup>2</sup>, Rayanne Soalheiro de Souza<sup>1</sup>, and Sandra Gesteira Coelho<sup>1</sup>, <sup>1</sup>Universidade Federal de Minas Gerais-UFMG, Belo Horizonte, Minas Gerais, Brazil, <sup>2</sup>Embrapa Dairy Cattle, Juiz de Fora, Minas Gerais, Brazil, <sup>3</sup>Instituto de Ciências Agrárias da UFMG, Montes Claros, Minas Gerais, Brazil, <sup>4</sup>Instituto Federal de Educação, Ciência e Tecnologia do Sudeste de Minas Gerais-IFSEMG, Rio Pomba, Minas Gerais, Brazil.

The aims of this study were to quantify the variation in residual feed intake (RFI) of calves F<sub>1</sub> (Holstein × Gyr) until 60 d of age and evaluate their productive performance. Eighteen calves received colostrum after birth (10% of body weight) and were housed in individual sand bed stalls in the experimental farm of Embrapa Dairy Cattle (Coronel Pacheco, Brazil). All animals were subjected to the same nutrition management which consisted of 6 L of whole milk (TS; 11.75%) in equal amounts twice a day. Solid diet consisting of 95% of concentrated (88% DM; 20% CP and 3% Fat), 5% Tifton 85 hay (81% DM; 13.4% CP; 72.8% NDF; 32.3% ADF) and water were provided ad libitum from the first day of life. Feed solid diet, milk and water intakes were measured daily and body weight and morphometric measurements (withers height, hip height, chest circumference and rump width) were done at birth and at 60 d of age. RFI was calculated for each animal as the difference between actual DMI and expected DMI. Expected DMI was computed for each animal by regressing average daily DMI on mean BW<sup>0.75</sup> and ADG over a 60 d period. Twelve animals were ranked according to the RFI into 2 groups: low (efficient) and high (inefficient). The data were analyzed in a completely randomized design by ANOVA using GLM procedure of SAS. High RFI calves had DMI 12.39% higher than the low group ( $P < 0.05$ ). There was no difference in ADG, water intake and rump width between RFI groups ( $P > 0.05$ ). Withers height, hip height, chest circumference were higher ( $P < 0.05$ ) for the low RFI group.

Contd.

**Table 1 (Abstr. M345).** Main effect means and SE to intake and growth performance parameters in crossbred (Holstein x Gyr) calves differing in phenotypic residual feed intake (RFI)

Parameters	Low RFI	High RFI	SE	P-value
RFI, kg DM/d	-0.13	0.07	—	—
ADG, kg/d	0.730 <sup>a</sup>	0.746 <sup>a</sup>	0.02	>0.05
DMI, kg/d	0.941 <sup>b</sup>	1.074 <sup>a</sup>	0.03	<0.05
Water intake, L/d	1.00 <sup>a</sup>	1.38 <sup>a</sup>	0.20	>0.05
Milk intake, kg/d	0.803 <sup>a</sup>	0.795 <sup>a</sup>	0.04	>0.05
Hip height, cm	92.25 <sup>a</sup>	91.87 <sup>b</sup>	0.96	<0.05
Chest circumference, cm	94.00 <sup>a</sup>	93.50 <sup>b</sup>	1.33	<0.05
Rump width, cm	25.67 <sup>a</sup>	25.50 <sup>a</sup>	0.40	>0.05
Withers height, cm	88.20 <sup>a</sup>	87.25 <sup>b</sup>	0.89	<0.05

Means within rows followed by the same letter are not significantly different ( $P \geq 0.05$ ).

**Key Words:** measurement, body weight, efficiency

**M346 Dietary grape marc supplementation lowers urinary nitrogen excretion from pasture-based dairy cattle.** Aysha Morrow<sup>1</sup>, Reuben Harland<sup>1</sup>, Roland Harrison<sup>1</sup>, Jana Kraft<sup>2</sup>, and Sabrina L. Greenwood<sup>\*1,2</sup>, <sup>1</sup>Lincoln University, Lincoln, New Zealand, <sup>2</sup>The University of Vermont, Burlington, Vermont.

On-farm nitrogen (N) losses in pasture-based dairy systems are an environmental issue. Grape marc (GM) is a byproduct that contains condensed tannins (CT) and low crude protein concentrations, constituents that alter N partitioning in ruminants. The objective of this experiment was to determine if GM could decrease the urine N concentration and the urine N output (g N/d) from cows grazing a typical perennial ryegrass/white clover pasture. A second objective of the current experiment was to determine if the CT in GM are a component causing any changes in fecal and urine N concentrations through the use of polyethylene glycol (PEG), a CT inhibitor. The experiment was arranged in a 4 × 2 factorial design, including 4 levels of GM supplementation (0, 2, 4, and 6 kg DM/cow/d) and 2 levels of PEG (0, and 350 g/cow/d) in addition to their daily pasture allowance. Forty Friesian x Jersey crossbred multiparous cows in late lactation were evenly divided into one of 8 treatment groups. Cows were fed increasing amounts of GM for 10 d and fed the full treatment amounts of GM for a further 6 d. The pasture DMI were estimated for each treatment group daily, and intakes of GM and PEG were determined for each cow daily. Urine, feces, plasma and milk samples were collected on d 0, 10, and 16 of the trial. Fecal N % and DM % were analyzed, as well as urine N %, and concentrations of urine ammonia, urine urea-N, plasma urea-N, and milk urea-N. Estimated urine N excretion (g N/d) was calculated. Results were analyzed using GenStat. The dietary inclusion of PEG decreased fecal N % ( $P < 0.001$ ), while dietary supplementation with GM lowered urine N % ( $P = 0.005$ ) and urine urea ( $P = 0.003$ ) concentrations. Cows fed GM also had lower plasma urea-N ( $P = 0.024$ ) and milk urea-N ( $P = 0.026$ ) concentrations. There was a tendency toward lower ammonia (mmol/L;  $P = 0.09$ ) and N excretion in urine (g N/d;  $P = 0.08$ ) due to the supplementation of dietary GM. These results indicate that the dietary inclusion of grape marc is effective at increasing fecal N and reducing urine N concentrations and that the CT within GM aids in increasing fecal N loss.

**Key Words:** nitrogen loss, environment, byproduct

**M347 Energy expenditure and methane emission in dairy heifers using the face-mask method.** Carlos Alberto Alves Oliveira Filho<sup>1</sup>, Fernanda Samarini Machado<sup>2</sup>, Alexandre Lima Ferreira<sup>2</sup>, Luiz Gustavo Ribeiro Pereira<sup>\*2</sup>, Thierry Ribeiro Tomich<sup>2</sup>, Mariana Magalhães Campos<sup>2</sup>, José Augusto Gomes Azevêdo<sup>3</sup>, Rogério Martins Maurício<sup>4</sup>, Alexandre Vieira Chaves<sup>5</sup>, and Camilla Flávia Portela Gomes Silva<sup>6</sup>, <sup>1</sup>*Universidade Estadual do Sudoeste da Bahia, Itapetinga, Bahia, Brazil*, <sup>2</sup>*Embrapa Dairy Cattle, Juiz de Fora, Minas Gerais, Brazil*, <sup>3</sup>*Universidade Estadual de Santa Cruz, Ilhéus, Bahia, Brazil*, <sup>4</sup>*Universidade Federal de São João Del Rei, São João Del Rei, Minas Gerais, Brazil*, <sup>5</sup>*Faculty of Veterinary Science, Sydney, New South Wales, Australia*, <sup>6</sup>*Instituto Federal de Educação, Ciência e Tecnologia Baiano, Santa Inês, Bahia, Brazil*.

The aim of this study was to evaluate the effect of feeding levels (FL) and breed (B) on energy expenditure as heat production (HP) and methane emission in dairy heifers using the face-mask method. Twenty-four heifers, 8 Holstein, 8 Gyr and 8 Holstein-Gyr crossbreed (F<sub>1</sub>) with average live weight (LW) of 440 ± 88 kg and average age of 27.5 ± 0.8 mo, were housed in tie stall and randomly distributed to the treatments in a 2 × 3 factorial design (feeding level of 1.17% LW or 1.46% LW, on dry matter (DM) basis and breed). The diet was offered as a total mixed ration (700g/kg of corn silage and 300g/kg of concentrate, on DM basis). O<sub>2</sub> consumption, CO<sub>2</sub> production and CH<sub>4</sub> emission data were measured using Sable System (Sable Systems, Henderson, NV) coupled with a face-mask for 30 min per day, for 3 d. Heart rate (HR) (beat/min) was registered over 72 h using Polar Equine transmitter (model RS800CX G3, Polar Electro Inc.). Three measurements of oxygen pulses (O<sub>2</sub>P) (mL O<sub>2</sub>/beat) were registered. Total daily O<sub>2</sub> consumption (L/d) was calculated as O<sub>2</sub>P times daily mean HR. Daily HP was calculated as total daily O<sub>2</sub> consumption times the constant 20.47 KJ/L of O<sub>2</sub>. Data were subjected to ANOVA and means were compared by Student-Newman-Keuls test ( $P < 0.05$ ). No effect of the interaction between B and FL was observed for any of the variables analyzed. HR, O<sub>2</sub>P and HP (kcal/kg LW<sup>0.75</sup>) did not differ among B and FL. F<sub>1</sub> heifers presented higher DM intake (DMI) and higher daily mean gain (DMG) in comparison to Holstein and Gyr breeds (6.18, 5.36 and 4.14 kg DM/d; 674.3, 480.8 and 435.4 g/d, respectively). Animals fed at 1.46% LW level presented higher DMI, DMG, BW and methane emission (g/d). Methane emission was higher for F<sub>1</sub> animals (161.3 g/d), but did not differ from Holstein breed (141.1 g/d). Animals from Gyr breed fed at 1.17% LW presented lower DMI, DMG and BW as well as inferior CH<sub>4</sub> production (98.3 g/d). This research project was funded by FAPEMIG, CAPES, CNPq and Embrapa.

**Key Words:** greenhouse gas, heat production, zebu

**M348 Methane production in dairy cows consuming corn milling co-products.** K. G. Saathoff<sup>\*1</sup>, C. J. R. Jenkins<sup>1</sup>, S. C. Fernando<sup>1</sup>, D. Hostetler<sup>2</sup>, and P. J. Kononoff<sup>1</sup>, <sup>1</sup>*Department of Animal Science, University of Nebraska-Lincoln, Lincoln, NE*, <sup>2</sup>*The School of Veterinary Medicine and Biomedical Sciences, University of Nebraska-Lincoln, Lincoln, NE*.

A study using 4 multiparous Holstein dairy cows, which were 93.5 ± 22.2 DIM, was conducted to determine the effect of conventional and corn milling co-products, specifically dried distillers grains and solubles (DDGS), on milk production, composition, and methane production. A 4 × 4 Latin square was utilized and included 4 treatments, namely a zero control (C) and diets that contained 30% of the diet DM as either conventional DDGS (ConDG), or 15% reduced fat DDGS (RFDG) or a mixture (Mix) of 15% conventional DDGS and 15% reduced fat DDGS. In all 3 treatment diets, DDGS were included in replace of corn and

soybean meal. Cows were housed and fed in individual stalls and fed once per day and milked twice per day for 4–28 d periods. During the last 2 d methane production was measuring using indirect calorimeters. Cows consuming DDGS consumed more ( $P = 0.05$ ) feed (22.7, 24.8, 26.4 and 27.3 ± 1.57 for the C, RFDG, Mix and ConDG respectively). Likely in response feed intake, milk yield was also increased ( $P < 0.01$ ) by feeding DDGS (29.4, 39.4, 38.2, and 38.0 ± 3.22 for the C, RFDG, Mix and ConDG respectively). The concentration of fat in the milk was not affected ( $P = 0.47$ ) by treatment and averaged 3.41 ± 0.29%. In comparison, protein in milk was higher ( $P = 0.05$ ) in consuming DDGS (2.74, 2.85, 2.95 and 2.91 ± 0.12 for the C, RFDG, Mix and ConDG respectively). Although total methane was not different ( $P = 0.69$ ) across treatments averaging 443.8 ± 20.6 L/d, cows consuming DDGS produced less ( $P = 0.01$ ) methane per unit of milk produced (14.6, 11.3, 12.1 and 12.1 ± 0.84 kg milk/kg feed for the C, RFDG, Mix and ConDG respectively). Results of this study further support the notion that corn milling co-products may be used to replace both corn and soybean meal in dairy rations and also suggest that doing may also result in less methane per unit of milk produced.

**Key Words:** methane, dried distillers grains and solubles

**M349 Implementing multi-variate statistical process control to detect variability on a commercial dairy farm.** Robb W. Bender<sup>\*1,2</sup>, James A. Barmore<sup>2</sup>, David E. Cook<sup>1</sup>, and David K. Combs<sup>1</sup>, <sup>1</sup>*University of Wisconsin-Madison, Madison, WI*, <sup>2</sup>*GPS Dairy Consulting LLC, Calmar, IA*.

The objective of this study was to characterize variability in both individual animal and pen data in data streams on a well-managed commercial dairy farm. Additional objectives were to assess the effect of outside events on the variability of data streams and utilize multi-variate statistical process control (SPC) to improve detection time of out-of-control data streams. A 1,400-cow dairy in Eden, Wisconsin, was equipped with milk meters and rumination/activity collars to record individual cow milk production, rumination, physical activity, and pen-based feed intake. Data were collected over a 3 mo period. Milk production was analyzed for out-of-control data points via the Shewhart procedure, and rumination, physical activity and feed intake were analyzed multivariately via the MVP procedures of SAS. On this dairy, milk production averaged 45.1 kg, with a standard deviation of 1.3 kg among days within a pen, 11.5 kg among individual cows within a pen, and 20.1 kg among days within individual cows. Rumination (min per day) averaged 441.6 min, with a standard deviation of 14.0 among days within a pen and 120.7 min among individual cows within a pen. Physical activity (measured in arbitrary units) averaged 489.2, with a standard deviation of 17.1 among days within a pen, 103.4 among individual cows within a pen. Feed intake (DMI kg/cow/d) averaged 26.6 kg, with a standard deviation of 1.8 kg among days within pens. Multi-variate SPC increased sensitivity when compared with individual single-variate SPC analyses. Out-of-control milk production values were preceded by a deviation from normal variance in the multi-variate analysis of rumination, physical activity, and feed intake. Thus, multi-variate SPC could be used as an early determinant of extreme variability in data streams on a commercial dairy.

**Key Words:** statistical process control, variation, dairy

**M350 Evaluation of apparent starch digestibility in commercial dairy herds.** R. A. Silva<sup>1</sup>, J. H. Carneiro<sup>1</sup>, I. Q. Carvalho<sup>2</sup>, J. F. Santos<sup>3</sup>, R. B. Navarro<sup>4</sup>, P. F. Menegucci<sup>5</sup>, M. Caetano<sup>6</sup>, D. P. D. Lanna<sup>7</sup>, and R. Almeida\*<sup>1</sup>, <sup>1</sup>Universidade Federal do Paraná, Curitiba, PR, Brazil, <sup>2</sup>Fundação ABC, Castro, PR, Brazil, <sup>3</sup>Castrolanda Cooperativa Agroindustrial, Castro, PR, Brazil, <sup>4</sup>Capal Cooperativa Agroindustrial, Arapoti, PR, Brazil, <sup>5</sup>Chr. Hansen, Valinhos, SP, Brazil, <sup>6</sup>University of Adelaide, Roseworthy, SA, Australia, <sup>7</sup>ESALQ/USP, Piracicaba, SP, Brazil.

The objective of this study was estimate the apparent total-tract starch digestibility (ATTSD) in dairy herds, as well as its correlation with corn ensiling time. There were 20 commercial herds in the Paraná State, South Brazil, TMR-fed with predominantly Holstein cows. In all herds, only the high-producing groups were included in the study, with an average milk yield of  $41 \pm 6$  L/cow/d, representing  $33 \pm 13\%$  of lactating cows, and a total of 1655 cows. To check the influence of silage fermentation time on the starch digestibility, each farm was sampled in 2 distinct periods; fall season when most producers were using corn silage (CS) with short ensiling time ( $132 \pm 132$  d), and spring season with CS of longer ensiling time ( $260 \pm 46$  d). In each period 3 samples were collected per herd; TMR, CS and feces. Starch contents were determined in quadruplicate by enzymatic colorimetric method. To estimate ATTSD 2 equations were used: only with fecal starch as input (Eq1 =  $100 \times (1 - 0.0125 \times \% \text{fecal starch})$ ); and using lignin as marker (Eq2 =  $1 - ((\% \text{TMR lignin} / \% \text{fecal lignin}) \times (\% \text{fecal starch} / \% \text{TMR starch})) \times 100$ ). The average values of 40 samples of corn silage and TMR were  $31.15 \pm 3.44\%$  DM,  $42.36 \pm 3.86\%$  NDF,  $21.19 \pm 2.11\%$  ADF,  $2.49 \pm 0.68\%$  lignin,  $30.45 \pm 4.29\%$  starch, and  $45.38 \pm 3.75\%$  DM,  $34.99 \pm 3.21\%$  NDF,  $16.72 \pm 2.16\%$  ADF,  $1.92 \pm 0.66\%$  lignin,  $27.02 \pm 4.37\%$  starch, respectively. The average fecal starch content was  $3.40 \pm 3.1\%$  and  $3.93 \pm 2.50\%$  for spring and fall samples, respectively. Apparent TTSD estimates were  $95.42 \pm 2.38\%$  and  $95.43 \pm 5.15\%$  to Eq1 and Eq2, respectively, with a high correlation between them ( $r = 0.83$ ;  $P < 0.01$ ). Positive correlation between ensiling time and ATTSD calculated by Eq2 was also detected ( $r = 0.31$ ;  $P < 0.05$ ), showing that CS with longer fermentation contributes positively to the increased starch utilization. We also observed moderate negative correlation between CS dry matter and ATTSD ( $r = -0.28$ ;  $P < 0.05$ ), indicating that dried CS reduces TTSD. In general, dietary starch was efficiently utilized, but some factors such as ensiling time and plant maturity influence starch digestibility of the dairy cow diets.

**Key Words:** corn silage, fecal starch

**M351 Effects of partial replacement of corn and alfalfa silage with tall fescue hay on total-tract digestibility and lactation performance in lactating dairy cows.** Robb W. Bender\*, Fernanda Lopes, David E. Cook, and David K. Combs, *University of Wisconsin-Madison, Madison, WI.*

Our objective was to evaluate the effects of partial replacement of corn and alfalfa silage with tall fescue hay on total-tract NDF digestibility and lactation performance in dairy cows. Twenty-four primi- ( $75 \pm 35$  DIM) and 40 multi-parous ( $68 \pm 19$  DIM) Holstein cows were blocked by parity and randomly assigned to 1 of 4 treatment groups in a pen equipped with 32 feeding gates to record intake by cow. Each gate was randomly assigned to 1 treatment group, thus each cow had access to all 8 gates within the respective treatment and cow was the experimental unit. Treatments were formulated to partially replace corn silage (CS) and alfalfa silage (AS) with tall fescue hay (TF) as follows (DM basis): 67% CS and 33% AS (control), 60% TF and 40% AS (60TF40AS), 60% TF and 40% CS (60TF40CS), and 33% TF and 67% CS (33TF67CS).

The experiment was a 7-week continuous lactation trial with a 2-week covariate period. Data were analyzed using the MIXED procedure of SAS. The model included parity, treatment, week, and relevant interactions as fixed effects and cow within treatment as a random effect. Dry matter intake and milk production did not differ, and averaged 23.5 and 41.4 kg/d, respectively. Fat concentration, protein yield and concentration, and SCC did not differ and averaged 4.00%, 1.28 kg/d, 3.15%, and 92.3, respectively among all treatments. Fat yield was greater ( $P < 0.01$ ) for the control (1.65 kg/d), 60TF40AS (1.66 kg/d), and 33TF67CS (1.66 kg/d) treatments compared with the 60TF40CS treatment (1.50 kg/d). Total-tract dry matter digestibility and total-tract organic matter digestibility did not differ, and averaged 67.9% and 70.0%, respectively. Total-tract NDF digestibility was lowest ( $P < 0.01$ ) for control (40.8%), and greater ( $P < 0.01$ ) for 60TF40CS (51.2%) than 60TF40AS (45.9%). 33TF67CS (46.7%) had similar total-tract NDF digestibility to the latter 2 treatments. Inclusion of highly digestible tall fescue grass hay has the potential to replace corn silage and alfalfa silage without influencing DMI and production.

**Key Words:** NDF, fiber digestion, dairy cow

**M352 Methyl-donors choline and methionine differentially alter hepatic methyl carbon metabolism.** Tawny L. Chandler\*<sup>1</sup>, Courtney L. McCourt<sup>1</sup>, and Sandra J. Bertics<sup>1</sup>, Barbara A. Barton<sup>2</sup>, and Heather M. White<sup>1</sup>, <sup>1</sup>University of Wisconsin-Madison, Madison, WI, <sup>2</sup>Balchem Corporation, New Hampton, NY.

Overlap in hepatic methyl pathways highlights a role for potential competition and compensation of methyl donors. The objective of this experiment was to examine the regulation of genes controlling methyl group transfer in response to increasing concentrations of choline chloride (CC), DL-Met (dLM), and added fatty acids (FA). Primary hepatocytes isolated from 4 Holstein calves were maintained as monolayer cultures for 24 h before treatment with CC (33, 100, 200, 450  $\mu\text{M}$ ) and dLM (16, 30, 100, 300  $\mu\text{M}$ ), with or without a 1 mM FA cocktail in a factorial design. Concentrations mimicked expected physiological concentrations. After 24 h, media was collected for quantification of reactive oxygen species (ROS) by fluorometric assay and cells were collected for quantification of gene expression. Data were analyzed using PROC MIXED of SAS 9.4 with linear and quadratic contrasts in a model with fixed effect of treatment and random effects of calf. Interactions were not significant and therefore only main effects are discussed. Met can be generated from betaine via BHMT, or from homocysteine via MTR which also serves as the final step in regeneration of met after methyl-group donation. Increasing dLM concentration did not alter BHMT expression but did decrease ( $P = 0.003$ ) MTR expression. Increasing concentration of CC did not alter BHMT but did increase ( $P = 0.02$ ) MTR expression, suggesting that CC plays a key role in regeneration of met after methyl group donation. FA increased ( $P = 0.05$ ) BHMT expression but decreased ( $P = 0.0001$ ) MTR expression, which favors regeneration of met that is coupled with downstream glutathione production, which may aid the cell with oxidative stress associated with FA metabolism. Both CC and dLM decreased ( $P = 0.02$ ) expression of MAT1A, the enzyme that generates SAM from met. PEMT expression was not affected by CC or dLM suggesting that dLM was not used to generate phosphatidylcholine. ROS tended to decrease ( $P = 0.08$ ) with increasing CC treatment but was not changed with dLM treatment. These data suggest that CC may play a critical role in donating methyl groups and in decreasing ROS within the hepatocyte.

**Key Words:** choline, methionine, methyl-donor



**M353 The effect of nitrate or live yeast culture on methane mitigation in a continuous culture system.** Caitlyn M. Massie<sup>\*1</sup>, Benjamin A. Wenner<sup>1</sup>, Amanda M. Gehman<sup>2</sup>, Zhongtang Yu<sup>1</sup>, Kelly C. Wrighton<sup>1</sup>, and Jeffrey L. Firkins<sup>1</sup>, <sup>1</sup>The Ohio State University, Columbus, OH, <sup>2</sup>Alltech, Nicholasville, KY.

Nitrates have been successfully fed to dairy cows to decrease methane emissions in several experiments. In the nitrate assimilatory pathway, bacteria reduce nitrate to nitrite to ammonia. Because the second step can be rate-limiting, nitrite accumulation poses health risks such as methemoglobinemia, which would hinder adoption of nitrate feeding to compete with methanogens for H<sub>2</sub> while assimilating the N from nitrate into microbial protein. The yeast *Saccharomyces cerevisiae* has the potential to anaerobically respire nitrite through its cytochrome c oxidase; it also can stimulate populations of bacteria that express nitrate and nitrite reductases. For this project, 4 dual flow continuous culture fermenters were used in 5 periods with 7 d of adaptation and 3 d of sampling. Fermenters were fed 40 g DM (50:50 ratio of concentrate:alfalfa pellet). Treatments were arranged in a 2 × 2 factorial with NO<sub>3</sub> (1.5% of DM) or urea as an isonitrogenous control and without or with Yea-Sacc (Alltech Inc., Nicholasville, KY) fed at a recommended 0.010 g/d. Gas production was measured over 3 d by closed circuit respirometry; 1 fermenter's gas production was omitted for all periods (unrelated to treatment). Objectives were to test the hypothesis that the combination of live yeast culture and nitrate would mitigate methane production in continuous culture compared with the control (a statistical interaction). However, there were no interactions ( $P > 0.10$ ). The main effect of nitrate decreased ( $P < 0.05$ ) CH<sub>4</sub> emission compared with urea control (29.6 vs 21.0 mmol/d). There was no difference ( $P > 0.10$ ) for H<sub>2</sub> emission for nitrate or yeast (averaging 0.149 mmol/d; SEM = 0.051), but the main effect of nitrate was decreased ( $P < 0.01$ ) for aqueous H<sub>2</sub> concentration compared with urea (1.23 vs 1.88 μM). Total VFA production (averaging 148 mmol/d; SEM = 15) and acetate:propionate (averaging 3.37, SEM = 0.12) did not differ ( $P > 0.10$ ) among treatments. Nitrate decreased methanogenesis without affecting H<sub>2</sub> variables. No interactions were detected, but live yeast might offer a useful protection against incompletely adapted rumen microbial populations.

**Key Words:** dairy, methane, yeast

**M354 Performance, heat production and methane emission in dairy heifers under different nutritional plans.** Carlos Alberto Alves Oliveira Filho<sup>1</sup>, Fernanda Samarini Machado<sup>2</sup>, Alexandre Lima Ferreira<sup>2</sup>, Luiz Gustavo Ribeiro Pereira<sup>\*2</sup>, Thierry Ribeiro Tomich<sup>2</sup>, Mariana Magalhães Campos<sup>2</sup>, José Augusto Gomes Azevêdo<sup>3</sup>, Rogério Martins Maurício<sup>4</sup>, Alexandre Vieira Chaves<sup>5</sup>, and Camilla Flávia Portela Gomes Silva<sup>6</sup>, <sup>1</sup>Universidade Estadual do Sudoeste da Bahia, Itapetinga, Bahia, Brazil, <sup>2</sup>Embrapa Dairy Cattle, Juiz de Fora, Minas Gerais, Brazil, <sup>3</sup>Universidade Estadual de Santa Cruz, Ilhéus, Bahia, Brazil, <sup>4</sup>Universidade Federal de São João Del Rei, São João Del Rei, Minas Gerais, Brazil, <sup>5</sup>University of Sydney, Sydney, New South Wales, Australia, <sup>6</sup>Instituto Federal de Educação, Ciência e Tecnologia Baiano, Santa Inês, Bahia, Brazil.

This study aimed to evaluate the effect of feeding levels (FL) and breed (B) on performance, heat production (HP) and enteric CH<sub>4</sub> emission in dairy heifers. Thirty 6 heifers, 12 Holstein, 12 Gyr and 12 crossbreed Holstein-Gyr (F1) with average live weight (LW) of 445.8 ± 98 kg and average age of 27.5 ± 0.8 mo, were housed in tie stall and randomly distributed to the treatments in a 3x3 factorial design (feeding levels of 1.95% LW, 1.46% LW and 1.17% LW, on dry matter (DM) basis and breeds). The diet was offered as a total mixed ration (700g/kg of corn silage and 300g/kg of concentrate, on DM basis; 140g of crude protein

(CP) per kg of DM). Respiratory exchanges (oxygen consumption and CO<sub>2</sub> and CH<sub>4</sub> production) were measured over 2 periods of 24h using 4 open-circuit respiration chambers and a Sable System (Sable Systems, Henderson, NV) of Embrapa's Bioenergetic Laboratory (Coronel Pacheco, Minas Gerais, Brazil). The equation from Brouwer was used to estimate HP. Data were subjected to ANOVA and means were compared through the Student-Newman-Keuls test ( $P < 0.05$ ). Significant effects were found for the interaction between B and FL for DM, organic matter (OM) and neutral detergent fiber (NDF) intakes (kg/d, %LW and g/kg LW<sup>0.75</sup>) and enteric methane emissions (L/day, g/kg DMI, g/kg OMI and g/kg NDFI). Animals fed at 1.17% LW level presented lower HP (kcal/kg LW<sup>0.75</sup>), feeding efficiency (FE) (kg LW/kg DM<sub>ing</sub>) and CH<sub>4</sub> emission (L/kg LW<sup>0.75</sup>). Gyr breed presented lower heat production (132.5 kcal/kg LW<sup>0.75</sup>) and CH<sub>4</sub> emission (2.03 L/kg LW<sup>0.75</sup>). For Gyr heifers, FE was higher than Holstein heifers but did not differ ( $P > 0.05$ ) from F1 animals (0.11 and 0.10 Kg LW/kg DMI, respectively). F1 heifers presented higher ( $P < 0.05$ ) daily mean gain (DMG), but did not differ ( $P > 0.05$ ) from Holstein breed (0.81 and 0.67 kg/day, respectively). Animals fed at the level of 1.17% of LW presented lower DMG (0.40 kg/day). This research project was funded by Fapemig, CAPES, CNPq and Embrapa.

**Key Words:** greenhouse gas, respirometric chamber, zebu

**M355 Effects of rumen-protected choline and B vitamins during the transition period on serum metabolites and milk composition in periparturient dairy cattle.** C. M. Melo<sup>1</sup>, L. C. Copetti<sup>1</sup>, O. F. Stuani<sup>2</sup>, R. Locatelli-Dittrich<sup>1</sup>, and R. Almeida<sup>\*1</sup>, <sup>1</sup>Universidade Federal do Paraná, Curitiba, PR, Brazil, <sup>2</sup>Safeeds Nutrição Animal, Toledo, PR, Brazil.

The effects of rumen-protected choline and B vitamins (RPB) on β-hydroxybutyrate (BHBA), nonesterified fatty acids (NEFA), total cholesterol, triglycerides, HDL, LDL and VLDL and milk fat to protein ratio (FPR) were evaluated in periparturient dairy cows. In a commercial farm in Southern Brazil, 132 Holstein cows (104 multiparous and 28 primiparous) were blocked by parity, expected day of calving and body condition score (BCS). The supplementation period was 18 d pre and 21 d postpartum. Cows in the treatment group were individually top-dressed with 100 g/d Vicomb (Jefo, Quebec, Canada), to provide 20.9 g of rumen-protected choline plus protected riboflavin and folic acid, while the control cows (CON) were supplemented with corn meal. Five blood samples were collected from each animal (14 d and 7 d before calving, at calving, 7 d and 14 d after calving). Additionally 4 samples were collected (3, 5, 7 and 10 d after calving) for BHBA analysis using a Precision Xtra meter. Milk samples to estimate FPR were collected on d 7, 14 and 21 after parturition. Data were analyzed using MIXED procedure of SAS with a model containing the effects of block, treatment, time, and treatment × time interaction as fixed effects and cow within treatment as a random effect. The incidence of hyperketonemia (BHBA concentration ≥ 1.2 mmol/L) was 25.0% (16/64) for RPB and 30.0% (19/63) for CON. Body weight and BCS did not differ ( $P > 0.05$ ) between RPB and CON cows in all 3 observations (21 d before calving, at calving, and 21 d after calving). No differences on serum metabolites between RPB and CON cows were detected ( $P > 0.05$ ). NEFA concentrations peaked ( $P < 0.01$ ) at calving; 0.82 ± 0.04 mmol/L, whereas BHBA peaked ( $P < 0.01$ ) at 7 d after calving; 0.78 ± 0.05 mmol/L. The lowest cholesterol concentration was at calving; 73.78 ± 1.53 mg/dL. Finally no milk FPR differences ( $P > 0.05$ ) were observed between RPB and CON cows in the first 3 weeks after calving; 1.29 vs. 1.31. In the particular

conditions of this on-farm trial with low incidence of hyperketonemia no benefits on RPB supplementation were detected.

**Key Words:** cholesterol, ketosis, lipoproteins

**M356 Microbial protein synthesis of Jersey heifers supplemented with chitosan or omega-6 fatty acids source.** Murilo Vendramini<sup>1</sup>, Helder Amaral<sup>1</sup>, Hayne Araki<sup>1</sup>, Marcia Vaz<sup>2</sup>, Dargon Salvia<sup>1</sup>, Euclides Oliveira<sup>1</sup>, Rafael Goes<sup>1</sup>, Marcelo Barros<sup>2</sup>, Bruno Secundino<sup>1</sup>, and Jefferson Gandra<sup>\*1</sup>, <sup>1</sup>Faculdade de Ciências Agrárias, Universidade Federal da Grande Dourados, Dourados, MS, Brazil, <sup>2</sup>Faculdade de Ciências da Saúde, Universidade Federal da Grande Dourados, Dourados, MS, Brazil.

The objective of this study was to evaluate the microbial protein synthesis of Jersey heifers supplemented with chitosan or omega-6 fatty acids (FA) source. Eight animals (average body weight of 158.62 ± 1.75 kg, mean ± SD) were used in replicated 4 × 4 Latin square experimental design, balanced and contemporary, in 2 × 2 factorial arrangements. The experimental period consisted of 18 d (12 d for adaptation and 6 d for data collection) and 5 d were used for wash out. The experimental diets were (1) control (CO), without omega-6 FA and chitosan supplementation; (2) whole raw soybean (WRS, source of omega-6 FA) with 200 g/kg of DM of WRS; (3) chitosan (CHI), with 2 g/kg of DM of chitosan; (4) chitosan and WRS (CHWS), with chitosan and WRS in the same level used in previously cited treatments. The diets were formulated according to the NRC (2001) to achieve weight gain of 700 g/day. Spot urine samples were collected on d 17 of each period 4 h after feeding. The analyses were performed according to Chen and Gomes (1992). Data were analyzed using PROC MIXED of SAS 9.3. Interaction effect ( $P < 0.05$ ) was observed between CHI and WRS for uric acid (mmol/L), which CHI reduced (1.74 mmol/L) and CHIWS increased uric acid (3.34 mmol/L). Animals fed CHI presented decrease ( $P < 0.05$ ) in total purine and absorbable purine derivatives. The output of microbial nitrogen and protein (g/d) was reduced ( $P < 0.05$ ) when heifers were supplemented only with CHI. Animals fed CHI presented decrease in the output of microbial nitrogen and protein in 31.55, 35.53 and 15.07% when compared with CO, WRS and CHWS, respectively. The chitosan supplementation reduced the microbial protein synthesis of dairy Jersey heifers.

**Key Words:** nitrogen metabolism, purine derivative, rumen manipulation

**M357 Nutritional value of hemp byproducts as ruminant feeds.** George N. Gozho\* and Jan C. Plaizier, *University of Manitoba, Winnipeg, MB, Canada.*

Hemp byproducts have potential as ruminant feeds, especially now that very low tetrahydrocannabinol (THC) hemp varieties contents are available in Canada. Three hemp by-products, i.e., hemp meal, hemp bran, and hemp meal fines, were, therefore, analyzed for their value as ruminant feeds. These values were compared with ruminant feeds that these byproducts may replace, including canola meal, soybean meal, and grain screenings. Analyses included proximate analysis, in vitro dry matter digestibility (IVTDMD) analysis using the Dairy II system, and in situ rumen degradability using Dacron bags. The rumen in situ dry matter digestibilities were fitted to an exponential equation using the SAS nonlinear regression procedure. Calculated parameters included washing loss (A), potential digestibility after washing (B), potential total digestibility (A + B) and digestibility rate (c). Results showed that hemp meal contained less CP, and more fiber and fat, and had lower

in situ and in vitro dry matter digestibilities than soybean meal. Hemp meal had a similar CP content, contained more fiber and fat, and had lower in situ and in vitro dry matter digestibilities than canola meal. Hemp bran had a very high fiber content and was less digestible than grain screenings. Hemp seed fines had a similar chemical composition than hemp meal, but it was more digestible than hemp meal. Hemp meal could be an alternative for other high protein feeds, but its relatively low digestibility must be taken into account.

**Table 1 (Abstr. M357).** Evaluation of hemp byproducts for ruminant feeds

	Hemp meal	Hemp bran	Hemp seed fines	Canola meal	Soybean meal	Grain screenings
CP, % DM	36.2	15.9	29.1	37.1	52.9	10.3
NDF, % DM	56.5	76.2	52.1	30.1	10.5	49.7
ADF, % DM	34.0	56.0	29.6	18.0	5.5	27.5
Crude fat, %	9.5	12.1	ND	2.4	0.7	2.4
Ash, % DM	6.3	3.2	4.5	6.6	6.7	8.2
IVTDMD, %	49.4	46.7	70.8	67.2	86.5	54.1
Rumen digestibility						
A, %	37.3	23.0	61.0	47.4	37.3	35.3
B, %	23.3	6.33	16.6	40.5	63.2	48.0
A + B, %	61.6	31.4	76.6	87.5	99.9	83.3
c, %/h	0.08	0.01	0.25	0.06	0.08	0.12

**Key Words:** hemp byproduct, ruminant, feed

**M358 Effect of corn type, particle size, enzymes, and time ensiled on chemical composition of rehydrated corn silage.** Naina M. Lopes<sup>\*1,2</sup>, Marcos N. Pereira<sup>2</sup>, and Felipe C. Cardoso<sup>1</sup>, <sup>1</sup>University of Illinois, Urbana, IL, <sup>2</sup>Universidade Federal de Lavras, Lavras, MG, Brazil.

Utilization of enzymes have been reported to improve feedstuff quality for dairy cows. The objective of this study was to evaluate 2 corns, Flint (F) and Floury (D); 2 particle sizes, grinded Fine (Fi) or Coarse (Co); application of 3 enzyme combination, amylase (A), protease (P) and both (AP); during 3 times of ensiling (TP) at 1, 3, or 5 mo in a complete randomized design resulting in 36 treatments. Each of the 5 silos for each treatment (replicates) consisted of 1kg of corn at 35% moisture and were stored in vacuum sealed bags at environmental temperature. Enzymes used were 650 g/t of amylase and 200 g/t of protease or the combination of the 2. Corn weight, pH and temperature were measured at time of bag opening. Samples were analyzed for starch, crude protein (CP) and prolamin concentration. Statistical analysis was performed using the MIXED procedure in SAS. Corn F had higher ( $P < 0.001$ ) CP (9.52 vs. 7.61%DM) and prolamin (9.70 vs. 8.3 %DM) before ensiling than D. On TP5, F had increased prolamin concentration (7.95%DM,  $P = 0.003$ ) than D (4.91%DM). Corn F had lower weigh variation (11.34 vs. 12.12 g,  $P < 0.0001$ ), lower starch concentration (65.03 vs. 66.63%DM,  $P = 0.01$ ) and lower NFC ( $P < 0.0001$ ) than D. Particle size Co had lower DM (29.28 vs. 29.41%;  $P = 0.04$ ), higher weigh variation (12.12 vs. 11.34 g,  $P < 0.0001$ ) and higher pH (4.07 vs. 3.98,  $P < 0.0001$ ) than Fi. Enzymes P and AP had lower prolamin concentration for A (6.09%DM), P (4.62%DM) and AP (4.92%DM,  $P < 0.0001$ ); for starch, A had lower concentration for A (64.40%DM), P (65.30%DM) and AP (67.80%DM,  $P = 0.0002$ ). Enzyme A had lower NFC, for A (8.75%DM), P (8.86%DM) and AP (8.89%DM,  $P < 0.0001$ ). Corn weight increased with time 1.73g in TP1, 11.4g in TP3 and 22.02g in TP5 ( $P < 0.0001$ ). Corn CP decreased ( $P = 0.04$ ) with time ensiled from TP1 to TP3 by 0.35%DM. Prolamin concentration changed ( $P =$

0.01) during time of ensiling, 5.19, 4.0 and 6.43%DM for TP1, TP3 and TP5, respectively. In conclusion, P seemed to have reduced the matrix starch protein. Small particle size, enzyme and time ensiled, made F better feed but not as good as D.

**Key Words:** matrix starch protein, amylase, protease

**M359 Nitrogen balance of Jersey heifers supplemented with chitosan or omega-6 fatty acids source.** Murilo Vendramini<sup>1</sup>, Helder Amaral<sup>1</sup>, Maria Gabriela Lobo<sup>1</sup>, Marcia Vaz<sup>2</sup>, Natalia Silva<sup>1</sup>, Euclides Oliveira<sup>1</sup>, Rafael Goes<sup>1</sup>, Marcelo Barros<sup>2</sup>, Caio Takiya<sup>3</sup>, and Jefferson Gandra<sup>\*1</sup>, <sup>1</sup>Faculdade de Ciências Agrárias, Universidade Federal da Grande Dourados, Dourados, MS, Brazil, <sup>2</sup>Faculdade de Ciências da Saúde, Universidade Federal da Grande Dourados, Dourados, MS, Brazil, <sup>3</sup>Departamento de Nutrição e Produção Animal, Universidade de São Paulo, Pirassununga, Brazil.

The objective of this study was to evaluate the nitrogen balance of Jersey heifers supplemented with chitosan or omega-6 fatty acids (FA) source. Eight animals (average body weight of 158.62 ± 1.75 kg, mean ± SD) were used in replicated 4 × 4 Latin square experimental designs, balanced and contemporary, in 2 × 2 factorial arrangements. The experimental period consisted of 18 d (12 d for adaptation and 6 d for data collection) and 5 d were used for wash out. The experimental diets were (1) control (CO), without omega-6 FA and chitosan supplementation; (2) whole raw soybean (WRS, source of omega-6 FA) with 200 g/kg of DM of WRS; (3) chitosan (CHI), with 2g/kg of DM of chitosan; (4) chitosan and WRS (CHWS), with chitosan and WRS in the same level used in previously cited treatments. The diets were formulated according to the NRC (2001) to achieve weight gain of 700 g/day. Blood samples were collected on d 15 of each period before feeding, by puncture of coccygeal vein and immediately centrifuged at 2000 g × 15 min, supernatant serum was transferred to tubes and submitted to analyses. Spot urine samples were collected on d 17 of each period 4 h after feeding. For the evaluation of nitrogen compounds were dosed concentrations of urea and creatinine (mg/dL) in blood and urine by enzymatic colorimetric method. Data were analyzed using PROC MIXED of SAS 9.3. Animals fed CHI presented lower ( $P < 0.05$ ) concentration of urea and higher ( $P < 0.05$ ) concentrations of creatinine (0.80 mg/dL) in blood than other diets. Higher concentration of urea (43.12 mg/dL) was observed for heifers fed WRS and lower concentration of creatinine than others experimental diets. The renal clearance of urea (24 h period) presented highest value ( $P < 0.05$ ) for animals fed WRS and lowest ( $P < 0.05$ ) for animals fed CHI. Fractional urea excretion (%) was highest for animals fed CHI and lowest for animals fed WRS. Chitosan and omega-6 FA source supplementation altered the nitrogen balance of dairy Jersey heifers.

**Key Words:** rumen manipulation, renal clearance, soybean

**M360 Effects of amylase and protease on degradability and gas production of rehydrated corn grain silage.** Naina M. Lopes<sup>\*1,2</sup>, Marcos N. Pereira<sup>2</sup>, and Felipe C. Cardoso<sup>1</sup>, <sup>1</sup>University of Illinois, Urbana, IL, <sup>2</sup>Universidade Federal de Lavras, Lavras, MG, Brazil.

Protein starch matrix is a physical barrier to starch digestion in corn grain. The objective of this study was to evaluate 2 corns, Flint (F) and Floury (D); 2 particle sizes, grinded Fine (Fi) or Coarse (Co); application of 3 enzyme combinations, amylase (A), protease (P) and both (AP); during 3 times of ensiling (TP) at 1, 3, or 5 mo in a complete randomized design resulting in 36 treatments. Each of the 5 silos for each treatment (replicates) consisted of 1kg of corn at 35% moisture and were stored

in vacuum sealed bags at environmental temperature. Enzymes used were 650 g/t of amylase and 200 g/t of protease or the combination of the 2. Samples were analyzed for in vitro degradability for 18 h and fermentation of gas production for 24 h (Ankom RFS, Ankom Tech.). Statistical analysis was performed using the MIXED procedure in SAS. Corn F had lower soluble fraction (Fs) degradability (6% of incubated,  $P < 0.0001$ ) but higher degradable fraction (Fd,  $P = 0.02$ ) than D (42 and 43% of incubated, respectively). Corn Co had higher indigestible fraction (Fi, 53% of incubated,  $P = 0.002$ ) than Fi (51% of incubated). Enzyme P resulted in higher ( $P < 0.0001$ ) Fd (43% of incubated) and AP (44% of incubated) than A (39% of incubated). Addition of AP reduced Fi portion and increased the rate of degradation ( $P < 0.0001$ ) over time. Time points 3 and 5 had higher Fd (30% on TP1 vs. 50% on TP3 and 5;  $P = 0.002$ ) than TP1, thus, the rate of degradation increased in the last month of ensiling (from 0.02 to 0.04;  $P = 0.004$ ). Timepoint 3 had a higher ( $P < 0.0001$ ) absolute pressure 37.42 psi for TP1, 42.81 psi for TP3 and 38.08 psi for TP5 and cumulative pressure 9.77 psi for TP1, 16.5 psi for TP3 and 8.92 psi for TP5 ( $P < 0.0001$ ). Data from gas production were divided in 2 periods: 0–12 h and 12–24 h. Period 1 (6.17 psi) had lower ( $P < 0.001$ ) gas production than period 2 (17.30 psi). In conclusion, corn ensiled and treated with enzymes for 3 mo had improved in vitro degradability than non-ensiled corn. Treated corn has great potential to improve farmer's profitability when fed to cattle.

**Key Words:** enzyme, corn fermentation, gas production

**M361 Stereo microscopy and scanning electron microscopy of manure samples from late lactation dairy cows when fed cobalt-lactate in a high-forage total mixed ration.** Jon P. Pretz<sup>\*1</sup>, Jianping Wu<sup>2</sup>, Madam Jao<sup>2</sup>, Bill Holloway<sup>3</sup>, Del Davis<sup>3</sup>, and David P. Casper<sup>1</sup>, <sup>1</sup>South Dakota State University, Brookings, SD, <sup>2</sup>Gansau Agricultural University, Lanzhou, Gansu, China, <sup>3</sup>Ralco Inc., Marshall, MN.

Feeding higher Co amounts showed increased ruminal fiber digestion. Cobalt-lactate is a highly rumen soluble source of Co and has demonstrated increased ruminal molar acetate concentrations and decreased ruminal ammonia concentrations when lactating dairy cows were fed a high forage ration (Pretz et al., 2014). These results indicated increased fiber digestion and synthesis of microbial protein. Twenty-four late-lactation ( $\mu = 238$  DIM and 36.5 kg milk) Holstein dairy cows (10 primiparous and 14 multiparous), were block by milk yield, DIM, and parity and randomly assigned to 1 of 2 treatments being: 1) control (C): ration containing cobalt carbonate fed at 58 mg/cow/d and 2) (T): ration containing 5 g/cow/d of a 1% Co-lactate product (Co-Max). Total mixed rations (TMR) were 70% forage (60% alfalfa baleage and 40% corn silage) and 30% of the respective experimental grain mix on a DM basis. Lactational responses were previously reported. This study further evaluated the total-tract nutrient digestion by collecting TMR and manure samples for measuring nutrient digestibility via internal markers (acid insoluble ash and iron). In addition, manure samples were submitted for stereo microscopy and scanning electron microscopy evaluation. Manure samples were collected at 4 h intervals for 3 consecutive d from each cow during wk 3 and 4. Total-tract DM (53.7 and 56.8% for C and T, respectively), CP (63.8 and 64.0%), NDF (46.3 and 48.9%), and ADF (39.8 and 42.9%) digestibility coefficients were numerically greater, but not significantly ( $P > 0.10$ ) different for cows fed T compared with cows fed C. Stereo microscopy demonstrated that manure samples from cows fed T were more transparent than manure samples from cows fed C. Scanning electron microscopy indicated small visible improvements in digestibility of fiber components observed as appearance of hollow pits in the fiber particles. Feeding additional Co as cobalt-lactate visu-



ally appeared to increase digestibility of fiber particles when evaluated by stereo and scanning electron microscopy.

**Key Words:** scanning electron microscope, cobalt-lactate, stereo microscopy

**M362 Plasma metabolites of Jersey heifers supplemented with chitosan or omega-6 fatty acid source.** Helder Amaral<sup>1</sup>, Murilo Vendramini<sup>1</sup>, Leticia Parangaba<sup>1</sup>, Grazielle Rosa<sup>1</sup>, Caio Takiya<sup>2</sup>, Euclides Oliveira<sup>1</sup>, Rafael Goes<sup>1</sup>, Antonio Machado<sup>1</sup>, André Santos<sup>1</sup>, and Jefferson Gandra\*<sup>1</sup>, <sup>1</sup>*Faculdade de Ciências Agrárias, Universidade Federal da Grande Dourados, Dourados, MS, Brazil*, <sup>2</sup>*Departamento de Nutrição e Produção Animal, Universidade de São Paulo, Pirassununga, Brazil*.

The objective of the present study was to evaluate plasma metabolites of blood of Jersey heifers supplemented with chitosan or omega-6 fatty acids (FA). Eight Jersey heifers (average body weight of 158.62 ± 1.75 kg, mean ± SD) were used in replicated 4 × 4 Latin square experimental design, balanced and contemporary, in 2 × 2 factorial arrangement. The experimental period consisted of 18 d (12 d for adaptation and 6 d for data collection) and 5 d were used for wash out. The experimental diets were (1) control (CO), without omega-6 FA and chitosan supplementation; (2) whole raw soybean (WRS, source of omega-6 FA) with 200g/kg of DM of WRS); (3) chitosan (CHI), with 2 g/kg of DM of chitosan; (4) chitosan and WRS (CHWS), with chitosan and WRS in the same level used in previously cited diets. The diets were formulated according to the NRC (2001) to achieve weight gain of 700 g/day. Blood samples were collected on d 15 of each period before feeding, by puncture of coccygeal vein and immediately centrifuged at 2000 g × 15 min, supernatant serum was transferred to tubes and submitted to analyses of cholesterol (CHO), low density cholesterol (LDL), high density cholesterol (HDL) urea and blood urea nitrogen (UN) by enzymatic colorimetric method. Data were submitted to PROC MIXED of SAS 9.3. The CHI diet decreased ( $P < 0.05$ ) and WRS increased ( $P < 0.05$ ) blood concentrations of TC, LDL, urea and UN. Heifers presented blood concentration (mg/dL) of TC (78.12 and 131.25), LDL (44.15 and 92.35), urea (35.50, 43.12) and UN (16.54 and 20.09) for fed CHI and WRS, respectively. Interaction effect for HDL blood concentration was observed ( $P < 0.05$ ) between CHI and WRS. The animals fed CHWS presented higher concentration (46.37 mg/dL), than WS (32.87 mg/dL), CO diet (28.00 mg/dL), and CH (23.75 mg/dL). Chitosan and omega 6 fatty acid influenced plasma metabolites of Jersey dairy heifers.

**Key Words:** alternative rumen modulator, metabolic profile, rumen manipulation

**M363 Effect of top-dressing rumen-protected methionine in lactating Holstein cows: I. Profile of plasma amino acids, milk yield, and milk composition.** Mateus Z. Toledo\*<sup>1</sup>, Giovanni M. Baez<sup>1</sup>, Eduardo Trevisol<sup>1</sup>, Nelson E. Lobos<sup>1</sup>, Alvaro Garcia-Guerra<sup>1</sup>, Jerry N. Guenther<sup>1</sup>, Daniel Luchini<sup>2</sup>, Randy D. Shaver<sup>1</sup>, and Milo C. Wiltbank<sup>1</sup>, <sup>1</sup>*University of Wisconsin-Madison, Madison, WI*, <sup>2</sup>*Adisseo, Alpharetta, GA*.

Experimental objectives were to evaluate the effects of supplementation with rumen-protected methionine (RPM) from 31 ± 2 until 127 ± 2 DIM on circulating amino acid concentrations and lactation performance of dairy cows. Holstein cows (n = 309) were housed in a freestall barn, milked twice daily, fed a basal diet formulated to 16.7% CP to deliver 2521 g of metabolizable protein (MP) with 6.93 lysine as % of MP and randomly assigned to once daily top-dressing with either: 1) RPM, 21.2

g of Smartamine M mixed with 38.8 g of dry distillers grains (2.34 methionine as % of MP) or 2) Control (CON), 60 g of dry distillers grain (1.87 methionine as % of MP). Plasma was assayed for free amino acids by gas chromatography using a commercial kit (EZ:faast, Phenomenex). Amino acid data were analyzed using a linear mixed model with repeated measures. Milk yield and composition were determined monthly and analyzed using a linear mixed model with treatment as a fixed effect and enrollment week as a random effect. Cows treated with RPM had increased milk protein content at all 3 milk tests (mean 58.7, 86.0, 113.5 DIM) (2.96 vs. 3.03%,  $P = 0.003$ ; 3.02 vs. 3.07%,  $P = 0.02$ ; 3.05 vs. 3.12%,  $P = 0.02$ ) with no difference in milk yield. Blood samples were collected from a subset of cows (n = 8 CON; n = 12 RPM) at 0, 3, 6, 9, 12, 18, and 24 h after feeding RPM. Plasma methionine did not differ between treatments at 0 ( $P = 0.37$ ) and 3 h ( $P = 0.50$ ) but was greater in RPM cows at 6 ( $P = 0.03$ ) and 9 h ( $P < 0.0001$ ), peaking at 12 h (52.4 vs 26.0 nmol/mL,  $P < 0.0001$ ), decreasing by 18 h ( $P = 0.12$ ), and back to basal by 24 h ( $P = 0.44$ ). Plasma lysine ( $P = 0.47$ ) and histidine ( $P = 0.42$ ) were unaffected by treatment. Another subset of cows (n = 16 CON; n = 24 RPM) was evaluated at 12 h after top-dressing. Cows fed RPM had increased methionine in both primiparous (23.9 vs. 46.4;  $P = 0.007$ ) and multiparous (23.5 vs. 38.7;  $P = 0.02$ ) cows. Thus, top-dressing RPM resulted in a surprisingly large but acute change in circulating methionine and a small but consistent change in milk protein content.

**Key Words:** methionine, dairy cattle, milk protein

**M364 Milk protein and fat production are regulated by histidine and glucose supply in the lactating dairy cow.** John Doelman\*<sup>1,2</sup>, Michelle Carson<sup>1</sup>, John P. Cant<sup>2</sup>, and John A. Metcalf<sup>1</sup>, <sup>1</sup>*Nutreco Canada Agresearch, Guelph, ON, Canada*, <sup>2</sup>*Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada*.

The lactational response to essential amino acid (EAA), histidine and glucose supply was determined in 6 early lactation fistulated dairy cows (92 ± 17 DIM) fed a diet to provide an NE<sub>L</sub> of 6.9 MJ/kg DM and 11.7% crude protein. Treatments consisted of abomasal infusions of saline (Sal), 1 kg/d glucose (Glc), 563 g/d of an EAA mix (equivalent to EAA in 1kg casein) with (EAA + Glc) and without (EAA) glucose, or EAA less histidine with (-His + Glc) and without glucose (-His) in a 6 × 6 Latin square design. Data were analyzed using the MIXED procedure in SAS where period and treatment were fixed effects and cow a random effect. Milk yield was significantly increased in response to EAA + Glc compared with Sal ( $P < 0.001$ ), while the removal of histidine from EAA + Glc significantly decreased milk yield ( $P < 0.001$ ). Compared with Sal, milk protein yield increased 146 g/d ( $P < 0.001$ ) in response to EAA. The -His treatment decreased milk protein yield 266 g/d ( $P < 0.001$ ) compared with EAA, and EAA + Glc generated an increase of 346 g/d over -His + Glc ( $P < 0.001$ ). Milk protein concentration increased 0.21 and 0.23 percentage points during EAA and EAA + Glc infusion compared with Sal ( $P \leq 0.003$ ), while the imbalance created with the omission of histidine, with and without glucose, significantly reduced milk protein concentrations to below those observed for Sal ( $P \leq 0.002$ ). With the exception of the complete EAA treatment, milk fat yield was significantly greater for -His compared with all other treatments ( $P \leq 0.03$ ). Milk fat concentration was 0.99 and 1.08 percentage points greater for -His compared with Glc ( $P < 0.001$ ) and EAA + Glc ( $P < 0.001$ ). Supplemental glucose did not stimulate a milk protein response and amplified the negative effects of a histidine deficiency. Additionally, histidine deficiency generated a positive milk fat response while glucose

**Table 1 (Abstr. M366).**

Item	Strategy					
	L-H	S-H	H-H	L-S	S-S	H-S
Farms, no.	12	14	12	6	12	8
Cows sampled	178	192	187	90	194	131
% HYK	9.0	15.0	12.0	7.8	33.3	23.7
BHBA mmol/L	0.46 <sup>a</sup>	0.51 <sup>ac</sup>	0.53 <sup>ac</sup>	0.52 <sup>ac</sup>	0.69 <sup>b</sup>	0.58 <sup>bc</sup>
[95% CI]	[0.41–0.53]	[0.46–0.57]	[0.48–0.59]	[0.45–0.60]	[0.62–0.77]	[0.52–0.65]
Farms/risk group (%)						
Low	8 (66.7)	7 (50.0)	8 (66.7)	5 (83.3)	2 (16.7)	3 (37.5)
Moderate	4 (33.3)	6 (42.9)	4 (33.3)	1 (16.7)	4 (33.3)	3 (37.5)
High	0	1 (7.1)	0	0	6 (50.0)	2 (12.5)

<sup>a,b,c</sup>Row means with different superscript letters differ ( $P < 0.05$ ) Tukey's HSD.

supplementation inhibited milk fat production. These results demonstrate the effect of histidine and glucose supply on lactation performance.

**Key Words:** amino acid, dairy cow, milk protein

**M365 Development of an in vitro subacute ruminal acidosis (SARA) model.** Allan B. Chestnut<sup>\*1</sup>, Jim M. Aldrich<sup>1</sup>, Tammy K. Miller Webster<sup>2</sup>, Wenping Hu<sup>1</sup>, Wibe B. Fokkink<sup>1</sup>, and Howard G. Bateman<sup>1</sup>, <sup>1</sup>Provimi North America, Brookville, OH, <sup>2</sup>Rumen Fermentation Profiling Laboratory, West Virginia University, Morgantown, WV.

To test efficacy of additives to reduce SARA, a protocol was developed to model SARA conditions using an in vitro continuous culture fermentor system at the Rumen Fermentation Profiling Laboratory, West Virginia University. A BASE diet was formulated with a 60:40 forage:concentrate (F:C) ratio (DM basis). Inclusion of corn silage, alfalfa balage, grass hay, ground corn (GC), SBM, corn gluten meal, corn gluten feed, soybean hulls, dried molasses, sodium bicarbonate and a mineral-vitamin supplement were, respectively, 36.1, 12.1, 12.1, 11.00, 13.7, 1.0, 1.0, 8.3, 2.0, 2.0 and 1.8% of DM. Mean particle size of GC was 870  $\mu\text{m}$  (SD = 2.7). In Trial 1, diet SARA1 was formulated by reducing the BASE diet F:C ratio to 52:48. Corn was increased to 20.9% with 50% (DM basis) of the GC replaced with steam flaked corn (SFC). Treatments were: BASE diet fed as 25-g meals 4 times daily for 10 d (B) and BASE diet fed as 25-g meals 4 times daily for 7 d followed by the SARA1 diet fed as 50-g meals 2 times daily for 3 d (S1). Treatments were replicated in triplicate. Data collected the last 3 d included pH at 30-min intervals and NDF digestibility. Daily average pH were calculated for each fermentor. All data were analyzed using PROC MIXED of SAS. Average and minimum pH were analyzed with a repeated-measures model. Fermentor was treated as a random variable and AR(1) was selected as the appropriate covariance structure. Average pH of S1 (6.25) vs B (6.28) was similar ( $P = 0.15$ ). Minimum pH of S1 (6.06) vs B (6.15) tended to be less ( $P = 0.08$ ). NDF digestion was less ( $P < 0.01$ ) for S1 (41.5%) vs B (46.6%). In Trial 2, treatment S1 was replaced with S2 by altering the SARA1 diet to create SARA2 as follows: corn replaced sodium bicarbonate and the GC:SFC ratio changed to 1:2. Trial 2 protocol was similar to Trial 1. Average pH of S2 (6.17) was less ( $P < 0.05$ ) than B (6.30). Minimum pH of S2 (5.88) was less ( $P < 0.01$ ) than B (6.18). NDF digestion was less ( $P < 0.01$ ) for S2 (30.5%) vs B (40.4%). It was concluded that using the tested protocol with diets BASE and SARA2 provided an appropriate SARA model.

**Key Words:** subacute ruminal acidosis, in vitro, model

**M366 Association of periparturitional nutritional strategy with concentration of postpartum  $\beta$ -hydroxybutyrate in dairy cows.**

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The objective was to determine the effect of transition cow nutritional management on  $\beta$ -hydroxybutyrate (BHBA) concentration of postpartum dairy cows. Commercial Holstein herds ( $n = 64$ ) based in New York and Vermont were enrolled in a prospective cohort study in one of 6 herd nutritional strategy groups: 1) low energy dry cow (<16% starch), high energy lactating (>25% starch) (L-H); 2) Step-up dry (far-off < 16% starch, close-up > 16% starch), high energy lactating (S-H); 3) high energy dry (>16% starch), high energy lactating (H-H); 4) low energy dry, step-up fresh (fresh < 25% starch, high > 25% starch) (L-S); 5) step-up dry, step-up fresh (S-S); 6) high energy dry, step-up fresh (H-S). Blood samples were collected from 972 cows, 3–14 DIM and BHBA concentration measured using a Precision Xtra meter. Concentrations of BHBA were log-transformed and a multivariable linear model was used to assess the fixed effects nutritional strategy, season, and parity on BHBA concentration with farm as a random effect. The proportion of cows with hyperketonemia (HYK; BHBA  $\geq 1.2$  mmol/L) in each nutritional strategy as well as risk group (low: <15%, moderate:  $\geq 15\%$ , <40%, high:  $\geq 40\%$  of sampled cows HYK) was evaluated. Multiparous cows had higher BHBA concentrations than primiparous (0.61 [0.57–0.67 CI] vs. 0.48 [0.43–0.53 CI] mmol/L,  $P < 0.0001$ ) and concentrations were higher in summer compared with winter (0.61 [0.56–0.67 CI] vs. 0.48 [0.44–0.53 CI] mmol/L,  $P < 0.0001$ ; Table 1). Overall, a step-up approach to both dry and fresh diets led to the highest BHBA concentration among all strategies in this study.

**Key Words:** transition period,  $\beta$ -hydroxybutyrate, nutritional strategy

**M367 Evaluation of rumen outflow in dairy cows by use of reticular and omasal sampling as an alternative to sampling from abomasal cannula.** José Esler Freitas Jr.<sup>\*1</sup>, Tiago Dell Vale<sup>2</sup>, Vitor Pereira Bettero<sup>2</sup>, Marjorye Kametani<sup>2</sup>, Pablo Gomes Paiva<sup>2</sup>, Rodrigo Gardinal<sup>2</sup>, Caio Seiti Takiya<sup>2</sup>, Filipe Zanferari<sup>2</sup>, Thiago T. H. A. Vendramini<sup>2</sup>, Elmeron Ferreira Jesus<sup>2</sup>, Gustavo Delfino Calomeni<sup>2</sup>, and Francisco Palma Renno<sup>2</sup>, <sup>1</sup>Department of Animal Science, Federal University of Bahia, Salvador, Bahia, Brazil, <sup>2</sup>Department of Nutrition and Animal Production, Faculty of Veterinary Medicine, University of São Paulo, Pirassununga, São Paulo, Brazil.

The aim of this study was to evaluate the effects rumen outflow in dairy cows fed unsaturated fatty acid by use of reticular and omasal sampling as an alternative to sampling from the abomasal canal. Four Holstein dry cows cannulated in the rumen and abomasum (602 ± 21 kg of body weight; mean ± SD) were assigned randomly into a 4 × 4 Latin square design experiment, fed the following diets: (1) control (C), without unsaturated fatty acid supplementation; (2) soybean oil (SO), addition of 3% of refined soybean oil in DM basis of total diet; (3) whole raw soybean (WS), addition of 16% of whole raw soybeans in DM basis of total diet; and (4) calcium salts of unsaturated fatty acids (CSFA), addition of 3% of CSFA in DM basis of total diet. Nutrient flow was calculated using the reconstitution system based on 3 markers (cobalt-EDTA, ytterbium chloride and indigestible NDF). Large and small particles and the fluid phase were recovered from digesta collected of different sites. Samples were collected 8 times during 20 4 h and were combined into one pool sample for further reconstitution. Data were analyzed using PROC MIXED of SAS 9.1 using the Tukey test to test pair wise comparisons. The DM flow was higher ( $P < 0.01$ ) to abomasum compared with reticulum and omasum (4.0; 4.0 and 4.8 kg to sites reticulum, omasum and abomasum respectively). However, the NDF flow was higher ( $P < 0.01$ ) (1.5; 1.2 and 1.4 kg to reticulum, omaso and abomasums respectively), to reticulum compared with omaso and abomasums. There was no difference for the 18:3 flow. The 18:2 flow was higher (50.9; 10.3 and 24.3 g to sites reticulum, omasum and abomasum respectively), to reticulum compared with omaso and abomasums, and 18:1 flow was less ( $P < 0.01$ ) for to omasum compared with reticulum and abomasum. The reticular sampling technique provided reliable estimates for DM flow and fatty unsaturated fatty acids in dairy cows

**Key Words:** dairy cow, omasal sampling, nutrient flow

**M368 Feed milk value and protein supply to dairy cows of new co-products (carinata meal) from bio-fuel processing in comparison with canola meal.** Yajing Ban<sup>\*</sup>, David A. Christensen, John J. McKinnon, and Peiqiang Yu, Department of Animal and Poultry Science, College of Agricultural and Bioresources, University of Saskatchewan, Saskatoon, Saskatchewan, Canada.

*Brassica carinata* is a newly developed oilseed for bio-fuel production in Canada. The bio-fuel processing results in a large amount of co-products (carinata meal), which could potentially be utilized as a protein source for animal feed. However, there is little research on metabolizable protein characteristics of carinata meal for dairy cattle. The objectives of this study were to determine differences among carinata meal, extruded carinata meal and canola meal for dairy cattle in terms of (1) rumen protein degradation, (2) intestinal digestion of protein, (3) total truly digestible protein supply, and (4) feed milk value. The animal trial was carried out in University of Saskatchewan dairy research facility. Statistical analyses were performed using PROC MIXED procedure of SAS 9.3 with significance declared at  $P \leq 0.05$ . The results indicated that extruded carinata meal had higher rumen degraded protein than canola meal ( $P \leq 0.05$ ). Intestinal digestible protein was the highest

in canola meal but the lowest in extruded carinata meal ( $P < 0.0001$ ), while total digestible protein was the highest in extruded carinata meal but the lowest in canola meal ( $P < 0.0001$ ). Extruded carinata meal had the most truly absorbed rumen-synthesized microbial protein in the intestine and degraded protein balance, but had the least truly absorbed rumen undegraded feed protein and truly digested protein in the small intestine ( $P \leq 0.05$ ). The canola meal was the highest in truly absorbed rumen undegraded feed protein in the small intestine but the lowest in truly absorbed rumen-synthesized microbial protein in the intestine and degraded protein balance ( $P \leq 0.05$ ). Feed milk values had no significant difference in carinata meal and canola meal using the Dutch DVE/OEB model. In NRC model, canola meal was the highest in FMV. The results indicated that carinata meal could be used as a potential protein supplement. Extrusion processing seems to have a negative effect on protein utilization and production in dairy cattle.

**Key Words:** carinata meal, metabolizable protein characteristics, feed milk value

**M369 Dairy calves changes in serum total protein and albumin concentration according to time after colostrum intake.** Nathalia B. Rocha<sup>1</sup>, Fernanda L. M. Silva<sup>1</sup>, Jackeline T. Silva<sup>1</sup>, Carolina C. F. Monteiro<sup>2</sup>, Marília R. Paula<sup>1</sup>, and Carla M. M. Bittar<sup>\*1,3</sup>, <sup>1</sup>University of Sao Paulo, ESALQ, Piracicaba, SP, Brazil, <sup>2</sup>Universidade Federal de Pernambuco, Recife, PE, Brazil, <sup>3</sup>CNPq, Brasilia, DF, Brazil.

Determination of serum total protein (TP) is being used as a tool to evaluate failure of passive immune transfer in dairy calves. However, time of evaluation recommendation is not well established. The objective of this study was to evaluate these changes, aiming a better recommendation of the better time to assess TP in neonatal calves. Blood from 47 cows was drawn just after calving, as well as from the calves at 0 (before colostrum feeding), 1, 2, 4, 6, 12, 24, 48, 72, 96 and 120h after colostrum intake. Crossbred calves (Holstein × Gir and Jersey × Gir) were separated from their mothers and 3L of high quality colostrum were fed. Hematocrit was determined and samples were centrifuged for determination of TP using a refractometer and albumin and TP using a commercial enzymatic kit. Regression curves were constructed for each parameter according to time, using SAS PROC REG. Correlations between mother TP at calving and calf TP at different times were also calculated using SAS PROC CORR. Quadratic regressions showed higher  $R^2$  for all parameters. Total protein concentration estimated by the protein refractometer was higher than that determined by enzymatic method, as shown by the higher intercept in both linear and quadratic prediction equation (Table 1). Correlation between cow's TP at calving were positive and significant only at time 0 ( $r = 0.2989$ ;  $P < 0.05$ ) and 1h ( $r = 0.24591$ ;  $P < 0.1$ ). The quadratic behavior of the regression analysis of TP, by both methods, suggest that the maximum time after colostrum feeding for a better evaluation of passive immune transfer is around 12–24h after colostrum feeding.

*Contd.*



**Table 1 (Abstr. M369).**

Prediction equation <sup>1</sup>	R <sup>2</sup>	P <
Albumin = 2.32 - 0.00038 T	0.0009	0.497
Albumin = 2.39783 - 0.00951T + 0.00008507T <sup>2</sup>	0.0418	0.0001
TPe = 5.304999 + 0.01931 T	0.2876	0.0001
TPe = 4.79507 + 0.08244 T - 0.00058577 T <sup>2</sup>	0.5176	0.0001
TPr = 6.13537 + 0.02468 T	0.2486	0.0001
TPr = 5.53535 + 0.10027 T - 0.00070451 T <sup>2</sup>	0.4233	0.0001

<sup>1</sup>TPe = TP determined by enzymatic method; TPr = TP estimated by refractometer.

**Key Words:** refractometer, passive immune transfer

**M370 Relationship between rumen methanogens and methane production in crossbred Holstein-Gyr steers.** Shirley Motta de Souza<sup>1</sup>, Daniela Batista Oss<sup>2</sup>, Luiz Gustavo Ribeiro Pereira<sup>1</sup>, Cláudia Braga Pereira Bento<sup>2</sup>, Hilário Cuquetto Mantovani<sup>2</sup>, Marcos Inácio Marcondes<sup>2</sup>, Fernanda Samarini Machado<sup>1</sup>, Thierry Ribeiro Tomich<sup>1</sup>, Mariana Magalhães Campos<sup>1</sup>, Adriana Santana Carmo<sup>1</sup>, Ellen de Almeida Moreira<sup>1</sup>, Sávio Augusto Toledo Moreira<sup>1</sup>, and Pedro Braga Arcuri<sup>4</sup>, <sup>1</sup>Brazilian Agricultural Research Corporation-Embrapa (Dairy Cattle), Juiz de Fora, MG, Brazil, <sup>2</sup>Federal University of Viçosa, Viçosa, MG, Brazil, <sup>3</sup>Federal University of Minas Gerais, Belo Horizonte, MG, Brazil, <sup>4</sup>EMBRAPA Liaison Officer for Multilateral, Regional & National Entities in Europe, Rome, Italy.

The aim of this study was to evaluate the bacterial community composition by denaturing gradient gel electrophoresis (PCR-DGGE). Eighteen steers Holstein × Gyr with average body 8 155 ± 5 kg d<sup>-1</sup> were randomly distributed in a completely randomized design with 3 treatments and 6 repetitions. The diet was calculated for average daily gain of 1.2 kg/d and an average weight of 240 kg, using the requirements for crossbred steers estimated by BR-Corte. Forage:concentrate ratio (based on DM) used was 60:40 and the animals received dietary treatment 1.2% MS of BW; 1.9% MS of BW and ad libitum intake, as maintenance, intermediate and high gain treatment, respectively. The daily feed intake was recorded and animals were weighed weekly each 28 d. The production of methane enteric by the animals was measured by open-circuit respiration chambers for 2 consecutive 24-h days. To assess the genetic diversity of the ruminal microbial community, 50 mL of rumen fluid samples were collected at the slaughter. DNA was extracted and processed by phenol-chloroform and bead beating method. PCR reaction used universal primers to amplify the V3 region to amplify the 16S rRNA of archaea Nested-PCR was performed to amplify a shorter region of the archaea 16S rRNA, using the primers ARC344f-GC/517r for archaea. PCR-DGGE patterns were analyzed using BioNumerics software 5.1 with which hierarchical cluster comparisons were carried out to group similar profiles and to generate a binary matrix of band classes. All the images were normalized using the internal control samples and the comparison among whole profiles was performed using the Dice similarity coefficient. The total number of the detected bands represented the species richness. Shannon-Wiener index was calculated based on relative band intensity and the total of number bands of each DGGE profile. The statistical analyses were done using the software R. The methane emission was affected by the treatments ( $P < 0.05$ ) but there was no effect of the treatments on the richness index also by Shannon-Wiener ( $P > 0.05$ ). No differences in archaeal population were detected between treatments.

**Key Words:** archaea, greenhouse gas, global warning

**M371 Evaluation of different oral rehydration protocols for dairy calves affected by diarrhea.** Evangelina Miqueo<sup>1</sup>, Thais M. Torrezan<sup>1</sup>, Nathalia B. Rocha<sup>1</sup>, Jackeline T. Silva<sup>1</sup>, Marília R. Paula<sup>1</sup>, Samyra Baldassin<sup>1</sup>, and Carla M. M. Bittar<sup>1,2</sup>, <sup>1</sup>University of Sao Paulo, ESALQ, Piracicaba, SP, Brazil, <sup>2</sup>CNPq, Brasilia, DF, Brazil.

Different oral rehydration therapies are used to restore the electrolyte level in diarrheic dairy calves. Most of them demand great amount of labor and time, however the use of commercial supplements administered with the liquid diet may make it easier. The objective of this study was to evaluate 3 rehydration protocols to determine the most effective in restoring electrolytes and water while maintaining animal performance. Thirty male Holstein bull calves were blocked by birth weight and serum total protein 24 h after birth, and distributed to one of 3 protocols of oral rehydration when presenting fecal score higher than 3 in a scale of 1 to 5: (1) Common electrolytes oral solution: 25 g dextrose, 10 g of sodium bicarbonate and diluted 5 g of sodium chloride in 1 L of water supplied at 10:00 and at 14:00; (2) commercial electrolyte solution vial for calves added to the milk replacer; and (3) Common electrolytes oral solution containing 2.2 g of glutamine and glutamate diluted in 1 L of water supplied at 10:00 and 1L at 14:00 h. All animals received 4 L/d milk replacer divided into 2 meals (7 h and 17 h) and were weaned with 56 d. Starter concentrate and water intake were measured daily and calves were weighed weekly. Although there were no significant differences among treatments for consumption, weight gain and days in diarrhea, voluntary water intake was significantly higher for diarrheic calves that received Glutellac, which together with the simplicity of use, represent the major advantages of this method of rehydration.

**Table 1 (Abstr. M371).** Performance of dairy calves receiving different rehydration treatments when presenting diarrhea

Item	Dextrose solution	Commercial electrolytes	Dextrose	SEM	P <
			+ AA solution		
Total DMI, g/d	713	747	670	41.3	0.41
Starter intake, g DM/d	267	199	225	37.0	0.35
Milk replacer intake, g DM/d	447	448	445	2.2	0.6
Total water intake, L/d	3.50	2.96	3.31	0.20	0.201
Voluntary water intake, L/d	1.50 <sup>b</sup>	2.96 <sup>a</sup>	1.30 <sup>b</sup>	0.20	0.0001
Average weight, kg	40.7	40.6	39.1	2.5	0.63
ADG, g	309	340	292	32.4	0.53
Feed efficiency	0.36	0.34	0.37	0.03	0.35

**Key Words:** amino acid, dextrose solution, commercial electrolyte

**M372 Pretreatment with saturated and unsaturated fatty acids regulates [1-<sup>14</sup>C] C16:0 metabolism in Madin-Darby bovine kidney cells.** Katherine E. Boesche\*, Stephanie L. Koser, and Shawn S. Donkin, Purdue University, West Lafayette, IN.

Metabolic fates of fatty acids (FA) may be influenced by circulating FA concentration. Previous work in our lab demonstrated an ability of C18:3n-3 *cis* to ameliorate gene expression of pyruvate carboxylase (PC) after depression by either C16:0 or C18:0. PC catalyzes oxaloacetate (OAA) synthesis and ostensibly links gluconeogenesis and FA metabolism. Our objective was to determine effects of copresence of saturated and unsaturated FA pretreatments on cellular partitioning of [1-<sup>14</sup>C] C16:0 metabolism to CO<sub>2</sub> or acid-soluble products (ASP) in Madin-Darby bovine kidney (MDBK) cells. Cells at 80% confluence were exposed for 21h to either individual FA bound to BSA (C16:0,

C18:0, C18:1n-9 *cis* or C18:3n-3 *cis*) or FA cocktails in 10:90, 25:75, 50:50, 75:25 or 90:10 ratios for combinations of C16:0: C18:3n-3 *cis* or C18:0: C18:3n-3 *cis* or C18:1n-9 *cis*: C18:3n-3 *cis*. Total pretreatment FA concentration was 1.0 mM. Following pretreatment, cells were then incubated in the presence of 1.0 mM [ $^{14}\text{C}$ ] C16:0 for 3h. Pretreatments with either C16:0 alone or C18:0 alone significantly ( $P < 0.01$ ) depressed subsequent oxidation of [ $^{14}\text{C}$ ] C16:0 to ASP by 62.7% and 41.2%, respectively, compared with C18:3n-3 *cis* pretreatments. Pretreatments with C18:1n-9 *cis* either alone or in any combination with C18:3n-3 *cis* did not significantly ( $P > 0.10$ ) depress subsequent oxidation of [ $^{14}\text{C}$ ] C16:0 to ASP. Similar patterns were seen with [ $^{14}\text{C}$ ] C16:0 oxidation to  $\text{CO}_2$ . ASP production from [ $^{14}\text{C}$ ] C16:0 was positively correlated ( $r = 0.68$ ,  $P < 0.01$ ) with PC gene expression levels while  $\text{CO}_2$  production from [ $^{14}\text{C}$ ] C16:0 did not show a correlation ( $r = 0.30$ ,  $P > 0.10$ ) with PC expression. Activation of PC gene expression by unsaturated FA may play a critical role in setting the capacity for OAA synthesis and determining metabolic fates of FA. Results show the regulation of ketone production by bovine kidney cells in response to saturated and unsaturated FA pretreatments.

**Key Words:** fatty acid oxidation, ketogenesis, pyruvate carboxylase

**M373 Ca(OH) $_2$ -treated corn stover as an alternative for hay-crop forage or corn silage in diets for lactating dairy cows.** Brittany A. Casperson<sup>\*1</sup>, Aimee E. Wert-Lutz<sup>2</sup>, and Shawn S. Donkin<sup>1</sup>, <sup>1</sup>Purdue University, West Lafayette, IN, <sup>2</sup>ADM Alliance Nutrition, Quincy, IL.

Nutritive value of crop residues may be improved through prestorage treatment with  $\text{Ca(OH)}_2$ . The objective of this experiment was to determine the effect of maximal substitution of either haylage or corn silage with  $\text{Ca(OH)}_2$ -treated corn stover on feed intake, milk production and milk composition in lactating dairy cows. Corn stover was processed by chopping, rehydrating, and treating with 6.6%  $\text{Ca(OH)}_2$  (DM basis), and stored in bag silos. Six mid-lactation multiparous Holstein cows were assigned to one of 2 groups and randomized within group to a Latin Square design to receive a TMR without any added  $\text{Ca(OH)}_2$ -treated stover (CON), a TMR where  $\text{Ca(OH)}_2$ -stover replaced alfalfa haylage at 15% of the diet (HYLGsub), or a TMR where  $\text{Ca(OH)}_2$ -stover replaced corn silage at 19% of the diet DM (CSsub). Diets were evaluated in a  $3 \times 3$  replicated Latin square consisting of 3 21-d periods. Cows were individually fed in tie stalls. The first 14 d of each period were used for diet adaptation followed by 7 d of data collection. Milk production was not different ( $P = 0.77$ ) among treatments. DMI was reduced ( $P < 0.05$ ) for HYLsub and CSsub diets when compared with CON (24.5, 21.8,  $20.6 \pm 1.0$  kg/d for CON, HYLsub, and CSsub, respectively). Milk fat percent was decreased ( $P < 0.05$ ) with inclusion of  $\text{Ca(OH)}_2$ -treated corn stover ( $3.92$ ,  $3.66$ ,  $3.69 \pm 0.15\%$ , CON, HYLsub, and CSsub, respectively) and milk fat yield tended to decrease ( $P = 0.09$ ). Milk protein percentage was reduced ( $P < 0.05$ ) by  $\text{Ca(OH)}_2$ -treated corn stover inclusion ( $3.27$ ,  $3.16$ , and  $3.11 \pm 0.06\%$  for CON, HYLsub, and CSsub, respectively) but milk protein yield was unaffected ( $P = 0.32$ ). Energy-corrected milk production per unit of DMI (kg/kg) was greater ( $P < 0.05$ ) for cows fed diets containing  $\text{Ca(OH)}_2$ -treated corn stover (1.12, 1.26, and  $1.30 \pm 0.05$  CON, HYLsub, and CSsub, respectively). Results show that  $\text{Ca(OH)}_2$ -treated corn stover can replace haycrop forages or up to 31% of the corn silage in diets for dairy cows without negatively impacting milk production while simultaneously improving the efficiency of conversion of feed to milk.

**Key Words:** corn stover, alternative forage, feed conversion efficiency

**M374 Composition of rumen microbiota alters following diet-induced milk fat depression in dairy cows.** Elnaz Azad<sup>1</sup>, Daniel E. Rico<sup>2</sup>, Hooman Derakhshani<sup>\*1</sup>, Kevin J. Havartine<sup>2</sup>, and Ehsan Khafipour<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Manitoba, Winnipeg, MB, Canada, <sup>2</sup>Department of Animal Science, Penn State University, University Park, PA.

An experiment was conducted to explore the effect of diet-induced milk fat depression (MFD) on the global profile of rumen microbiota. Eight ruminally cannulated cows were subjected to time-course induction of and recovery from diet-induced MFD in a replicated design with 3 periods of 21 d. Briefly, MFD induction followed control and recovery followed MFD. A high-fiber, low-oil diet was fed during the control and recovery periods, and a low-fiber, high-oil (LFHO) diet was fed during the induction period. Whole ruminal digesta samples were collected and subjected to high-throughput Illumina sequencing of the V3-V4 hypervariable regions of bacterial 16S rRNA gene. On average, 40,562 high-quality sequences were generated per sample. Taxonomic classification of sequences unraveled the presence of 15 different bacterial phyla across all samples, among which Bacteroidetes (46.96%), Firmicutes (35.95%), Proteobacteria (7.5%), Spirochaetes (2.64%), and Fibrobacteres (2.10%) were identified as predominant members of rumen microbiota. When compared with control and recovery-associated communities, the diversity of rumen microbiota was significantly decreased following consumption of LFHO diet ( $P < 0.001$ ). Moreover, PERMANOVA analysis of weighted UniFrac distances of microbial communities also revealed distinct clustering pattern for LFHO-associated microbiota compared with control and recovery diet ( $P = 0.002$  and  $P = 0.007$ , respectively). Based on linear discriminant analysis effect size (LEfSe), the proportions of several members of rumen microbiota were also found to be significantly ( $P < 0.05$ ) altered in response to dietary-induced MFD; the proportion of family Succinivibrionaceae (Proteobacteria) and genera *Shuttleworthia* and *Catonella* (Firmicutes) were found to be significantly increased in response to LFHO diet, while fibrolytic genera, such as *Fibrobacter* (Fibrobacteres) and *Ruminococcus* (Firmicutes) were found to be relatively more abundant during the control/recovery periods. Here, we demonstrated the complex dynamics of rumen microbiota that underlie diet-induced MFD.

**Key Words:** milk fat depression, 16S rRNA sequencing, rumen microbiota

**M375 Effects of *Saccharomyces cerevisiae* fermentation product on rumen fermentation during heat stress.** Kristy L. Dorton<sup>\*</sup>, Tracy Werner, Jason Lin, Abigail Souder, Adam M. Brainard, Joan Butler, and Ilkyu Yoon, *Diamond V, Cedar Rapids, IA.*

Effects of *Saccharomyces cerevisiae* fermentation product (SCFP; Original XPC) on rumen volatile fatty acid (VFA) and lipopolysaccharide (LPS) concentrations during heat stress were measured in a crossover experimental design consisting of 2 28 d-periods. Eight cannulated Jersey cows (nonlactating, non-pregnant) were housed in tie stalls containing individual feed bins and an automatic watering system. Cows were fed a mixture of chopped grass hay and a grain mix twice daily and 1 of 2 treatments. Treatments consisted of 14 g Control (grain mix) or 14 g SCFP and were fed before the morning feeding. The THI of the barn was maintained at 80 to 83 during the trial. Rumen fluid samples were collected from 5 locations within the rumen (cranial dorsal, cranial ventral, central rumen, caudal dorsal, and caudal ventral) every 3 h from 0900 h to 2100 h on d 26, 27 and 28, composited within cow, and strained. Samples were analyzed for VFA and LPS concentrations. Data were analyzed using the fit model procedure of JMP. Cows supplemented with SCFP had greater ( $P < 0.0001$ ) concentrations of

total VFA, acetate, propionate, and butyrate than control cows (103.46 vs. 95.19; 69.14 vs. 63.78; 18.75 vs. 17.21; 12.16 vs. 10.80 mM, respectively). Molar proportions of acetate and propionate were not affected by treatment. Molar proportion of butyrate was higher ( $P < 0.002$ ) for cows supplemented with SCFP than control cows (11.72 vs. 11.32%, respectively). Molar proportions of valerate, isobutyrate and isovalerate were lower ( $P \leq 0.007$ ) for cows supplemented with SCFP (1.01 vs. 1.08; 1.18 vs. 1.27; 1.11 vs. 1.20%, respectively). Although not significantly different, cows supplemented with SCFP tended to have lower trend ( $P = 0.16$ ) LPS concentrations than control cows (17,327 vs. 20,156 endotoxin units). Results show that SCFP can maintain better rumen function in cows subjected to heat stress as indicated by higher rumen VFA concentrations. This effect could be the result of stabilized rumen microbial populations, as indicated by the reduced tendency of LPS concentrations in the rumen.

**Key Words:** *Saccharomyces cerevisiae* fermentation product, VFA, LPS

**M376 Peroxisome proliferator-activated receptor  $\beta/\delta$  regulates glucose uptake in bovine mammary epithelial cells.** Jayant Lohakare\*<sup>1,2</sup>, Johan Osorio<sup>2</sup>, and Massimo Bionaz<sup>2</sup>, <sup>1</sup>College of Animal Life Sciences, Kangwon National University, Chuncheon, South Korea, <sup>2</sup>Oregon State University, Corvallis, OR.

A previous study showed that Peroxisome Proliferator-Activated Receptor  $\beta/\delta$  (PPAR $\beta/\delta$ ) activation inhibits glucose uptake in bovine aortic endothelial cells. We hypothesize that inhibition of PPAR $\beta/\delta$  can increase glucose uptake in immortalized bovine mammary alveolar cells (MACT) and increases lactose synthesis. To test our hypothesis, we treated MACT cells with PPAR $\beta/\delta$  synthetic agonist (GW501516) and antagonist (GSK3787) and assessed PPAR $\beta/\delta$  activation, live and dead cell count, and glucose uptake. MACT cells plated at 10,000 cells/well in 96 well plates were transfected with a PPAR Response Element (PPRE X3-TK-luc) plasmid using 0.3  $\mu$ L/well of TransIT-X2 Dynamic Delivery System (Mirus) in Opti-MEM media without fetal bovine serum (FBS). Cells were treated 24h after transfection in triplicates with 10 and 1,000 nM of GW501516, GSK3787, or a 1:1 combination of them plus ethanol as control in high-glucose DMEM medium with 10% FBS. After 24 h of treatment, a nuclear staining (NucBlue Live) was added and 2 images/well were obtained using an inverted fluorescent microscope (DMI6000B, Leica Microsystems, Germany). Luciferase activity was measured via a luminometer and normalized by the number of viable cells measured using CellProfiler software. Glucose concentration in the medium was measured using a Blood Glucose Meter kit (Safeway) and glucose uptake/viable cell was estimated. Data were analyzed using GLIMMIX of SAS. Significance was declared with  $P < 0.05$ . More than 2-fold increase of luciferase compared with control ( $P < 0.001$ ) was observed with both doses of GW501516 and a dose-dependent decrease of luciferase was observed with GSK3787. The number of viable cells was negatively affected by 10 nM of GSK3787 ( $P < 0.05$ ) but was positively affected with 1,000 nM of GSK3787 (16,990 vs. 20,966  $\pm$  521 cells/well). No effect on cell viability was observed with GW501516. There was a tendency ( $P = 0.06$ ) for an overall effect of treatments on glucose uptake. Cells treated with GW501516 had a lower glucose uptake (7.2 vs. 10.4  $\pm$  1.6 ng/cell) compared with cells treated with GSK3787. In conclusion, the use of 1,000 nM of GSK3787 successfully inhibited PPAR $\beta/\delta$  activity and increased glucose uptake in MACT cells. It remains to be determined if the increase in glucose uptake observed results in higher lactose synthesis.

**Key Words:** PPAR $\beta/\delta$ , mammary alveolar cells (MACT), glucose uptake

**M377 Estimation of biohydrogenation in dairy cows fed unsaturated fatty acids by use of reticular and omasal sampling as an alternative to sampling from abomasal cannula.** José Esler Freitas Jr.\*<sup>1</sup>, Tiago Dell Vale<sup>2</sup>, Vitor Pereira Bettero<sup>2</sup>, Marjorye Kametani<sup>2</sup>, Pablo Gomes Paiva<sup>2</sup>, Rodrigo Gardinal<sup>2</sup>, Caio Seiti Takiya<sup>2</sup>, Filipe Zanferari<sup>2</sup>, Thiago T. H. A. Vendramini<sup>2</sup>, Elmeron Ferreira Jesus<sup>2</sup>, Gustavo Delfino Calomeni<sup>2</sup>, and Francisco Palma Renno<sup>2</sup>, <sup>1</sup>Department of Animal Science, Federal University of Bahia, Salvador, Bahia, Brazil, <sup>2</sup>Department of Nutrition and Animal Production, Faculty of Veterinary Medicine, University of São Paulo, Pirassununga, São Paulo, Brazil.

The aim of this study was to evaluate the biohydrogenation in dairy cows fed unsaturated fatty acid by use of reticular and omasal sampling as an alternative to sampling from the abomasal canal. Four Holstein dry cows cannulated in the rumen and abomasum (602  $\pm$  21 kg of body weight; mean  $\pm$  SD) were assigned randomly into a 4  $\times$  4 Latin square design experiment, fed the following diets: (1) control (C), without unsaturated fatty acid supplementation; (2) soybean oil (SO), addition of 3% of refined soybean oil in DM basis of total diet; (3) whole raw soybean (WS), addition of 16% of whole raw soybeans in DM basis of total diet; and (4) calcium salts of unsaturated fatty acids (CSFA), addition of 3% of CSFA in DM basis of total diet. Nutrient flow was calculated using the reconstitution system based on 3 markers (cobalt-EDTA, ytterbium chloride and indigestible NDF). Large and small particles and the fluid phase were recovered from digesta collected of different sites. Samples were collected 8 times during 20 4 h and were combined into one pool sample for further reconstitution. Data were analyzed using PROC MIXED of SAS 9.1 using the Tukey test to test pair wise comparisons. There were not effects of sampling sites, on biohydrogenation of 18:3 (85.6; 97.9 and 93.2% of intake for to digesta reticular, omasal and abomasal respectively). However, the biohydrogenation of C18:2 (84.2; 93.4 and 85.1% of intake for to digesta reticular, omasal and abomasal respectively) ( $P < 0.01$ ) and C18:1 (63.2; 83.7 and 62.9% of intake for to digesta reticular, omasal and abomasal respectively) were overestimated by use sampling omasal in relation to use reticular sampling and abomasal ( $P < 0.01$ ). The reticular sampling technique provided reliable estimates for biohydrogenation of 18:2 and 18:1. The omasal sampling technique can overestimate to biohydrogenation.

**Key Words:** dairy cow, fatty acid, linoleic acid

**M378 Milk odd- and branched-chain fatty acid profile is affected by lactation stage in dairy cows.** Eric Baumann\*, P. Yvan Chouinard, Yolaine Lebeuf, and Rachel Gervais, Université Laval, Quebec, QC, Canada.

The odd- and branched-chain fatty acid (OBCFA) profile of milk fat has emerged as an interesting, non-invasive tool for evaluating rumen fermentation. These fatty acids are synthesized by different rumen microbial populations, absorbed in the intestine, and taken up by the mammary gland to be incorporated in milk fat. It is well known that milk fat composition is influenced by stage of lactation; proportion of short chains (de novo synthesis) being low initially and increasing with days in milk. However, literature is scarce about the specific effects of stage of lactation on milk OBCFA concentrations. Seven Holstein dairy cows were followed during the experiment. Five data collection and sampling periods were conducted on d 60 to 70, d 120 to 130, d 210 to 220, and d 300 to 310 of a lactation, and on d 5 to 15 of the subsequent lactation. During that period, cows were fed total mixed rations based on grass/legume and corn silages, cracked corn, soybean meal, and corn gluten meal, and formulated to meet their energy and nutrient requirements according to NRC (2001). The results indicate that stage of lactation do



affect expression of various milk OBCFA; these effects would need to be considered in the development of models aiming to predict rumen parameters based on milk OBCFA.

**Table 1 (Abstr. 378).** Milk fat concentrations (mg/g) of individual OBCFA

	5-15 DIM	60-70 DIM	120-130 DIM	210-220 DIM	300-310 DIM	P-value
Fatty acid						
11:0	0.20 <sup>c</sup>	0.46 <sup>bc</sup>	0.49 <sup>b</sup>	0.86 <sup>a</sup>	0.69 <sup>ab</sup>	<0.01
13:0	0.41 <sup>d</sup>	0.93 <sup>c</sup>	0.99 <sup>bc</sup>	1.38 <sup>a</sup>	1.20 <sup>ab</sup>	<0.01
15:0	5.47 <sup>d</sup>	9.86 <sup>c</sup>	11.17 <sup>b</sup>	12.50 <sup>a</sup>	12.32 <sup>ab</sup>	<0.01
17:0	3.79 <sup>a</sup>	2.41 <sup>b</sup>	2.31 <sup>bc</sup>	2.07 <sup>c</sup>	2.22 <sup>bc</sup>	<0.01
c9 17:1	1.85 <sup>a</sup>	0.85 <sup>b</sup>	0.76 <sup>b</sup>	0.77 <sup>b</sup>	0.79 <sup>b</sup>	<0.01
Odd	12.04 <sup>d</sup>	14.77 <sup>c</sup>	16.05 <sup>b</sup>	17.90 <sup>a</sup>	17.52 <sup>a</sup>	<0.01
ai 13:0	0.15	0.21	0.22	0.16	0.24	0.60
ai 15:0	2.20 <sup>b</sup>	4.74 <sup>a</sup>	5.22 <sup>a</sup>	4.88 <sup>a</sup>	4.85 <sup>a</sup>	<0.01
ai 17:0	4.26 <sup>a</sup>	3.99 <sup>ab</sup>	4.10 <sup>a</sup>	3.60 <sup>b</sup>	3.67 <sup>ab</sup>	<0.01
Anteiso	6.60 <sup>b</sup>	8.94 <sup>a</sup>	9.54 <sup>a</sup>	8.65 <sup>a</sup>	8.79 <sup>a</sup>	<0.01
i 14:0	0.62 <sup>b</sup>	1.54 <sup>a</sup>	1.82 <sup>a</sup>	1.45 <sup>a</sup>	1.66 <sup>a</sup>	<0.01
i 16:0	2.06 <sup>c</sup>	3.42 <sup>ab</sup>	3.92 <sup>a</sup>	2.98 <sup>b</sup>	3.28 <sup>ab</sup>	<0.01
i 18:0	3.29 <sup>c</sup>	5.25 <sup>ab</sup>	5.99 <sup>a</sup>	4.64 <sup>b</sup>	5.17 <sup>ab</sup>	<0.01
Even iso	3.29 <sup>c</sup>	5.25 <sup>ab</sup>	5.99 <sup>a</sup>	4.64 <sup>b</sup>	5.17 <sup>ab</sup>	<0.01
i 13:0	0.24 <sup>b</sup>	0.33 <sup>a</sup>	0.39 <sup>a</sup>	0.34 <sup>a</sup>	0.35 <sup>a</sup>	<0.01
i 15:0	5.01 <sup>c</sup>	10.03 <sup>b</sup>	12.12 <sup>a</sup>	10.60 <sup>a</sup>	10.76 <sup>a</sup>	<0.01
i 17:0	3.32 <sup>a</sup>	2.88 <sup>b</sup>	3.03 <sup>ab</sup>	2.74 <sup>b</sup>	2.70 <sup>b</sup>	<0.01
Odd iso	8.57 <sup>c</sup>	13.24 <sup>b</sup>	15.54 <sup>a</sup>	13.68 <sup>ab</sup>	13.80 <sup>ab</sup>	<0.01

**Key Words:** milk fat synthesis, odd- and branched-chain fatty acids, stage of lactation

**M379 Ruminal and production effects of supplementing high and low forage dairy rations with a live yeast culture.** Maegan E. Weatherly<sup>1</sup>, Amanda M. Gehman<sup>2</sup>, Amanda M. Lisembee<sup>2</sup>, Joey D. Clark<sup>1</sup>, Laurel L. Ball<sup>2</sup>, and Jeffrey M. Bewley<sup>1</sup>, <sup>1</sup>University of Kentucky, Lexington, KY, <sup>2</sup>Alltech Inc., Nicholasville, KY.

The objective of this study was to assess the effect of yeast supplementation in high and low forage diets on rumen and production parameters. Four, ruminally fistulated, multiparous, mid-lactation, Holstein cows were housed in a tie-stall barn at the University of Kentucky Coldstream Dairy from October 29, 2013 to February 7, 2014. A 4 × 4 Latin square design with a 2 × 2 factorial arrangement of treatments was used. Cows were assigned to 1 of 4 treatments each period including (1) low forage (LF), (2) low forage with 10 g/d yeast (Yea-Sacc; Alltech Inc., Nicholasville, KY; LFY), (3) high forage (HF), or (4) high forage with 10 g/d yeast (HFY). Periods 1 to 3 consisted of 21 d and period 4 was 18 d. Treatment periods were followed by a 7-d washout period where cows were gradually adjusted to the next ration. Dry matter intake was recorded daily. Daily rumination was recorded using HR Tags (SCR Engineers Ltd., Netanya, Israel). Rumen papillae were biopsied from each cow once per feeding period and analyzed for expression of enzymatic genes and transcriptional regulators. The GLM procedure of SAS (Version 9.3 SAS Institute, Inc., Cary, NC) was used to evaluate the fixed effects of cow, period, forage, yeast, and the interaction of forage and yeast on each parameter. Rumen papillae gene expression data were analyzed using a MIXED model in SAS. Rumination time and DMI were the only production parameters significantly influenced by treatment ( $P < 0.01$ ). Dry matter intake was 17.05, 13.41, 19.44, and 20.29 ± 1.40 kg/d for cows on the LF, LFY, HF, and HFY treatments, respectively. Rumination time was 442.88, 323.09, 433.34, and 475.50

± 21.93 min/d for cows on the LF, LFY, HF, and HFY treatments, respectively. Expression of peroxisome proliferator-activated receptor  $\gamma$ , sterol regulatory element-binding transcription factor 1, and oxoglutarate dehydrogenase ornithine carbamoyl transferase were significantly upregulated by yeast supplementation ( $P \leq 0.05$ ). The upregulation of genes that affect metabolism of VFA during yeast supplementation may suggest the importance of this product on rumen stabilization.

**Key Words:** yeast, rumen papillae gene expression, rumination

**M380 Effects of milk replacer and multivitamin-mineral supplementation on metabolism and rumen development in heat-stressed dairy calves.** Steven J. Blair<sup>1</sup>, Cathleen C. Williams<sup>1</sup>, Bruce F. Jenny<sup>1</sup>, Ashley H. Dolejsiova<sup>1</sup>, and Thomas J. Earleywine<sup>2</sup>, <sup>1</sup>Louisiana State University, Baton Rouge, LA, <sup>2</sup>Land O'Lakes Animal Milk Products, Shoreview, MN.

Seventy-one neonatal Holstein calves (40 female; 31 male) were used in a randomized block design with a 2 × 2 factorial arrangement of treatments to evaluate the effects of milk replacer (MR) feeding management alone or in combination with a multivitamin and electrolyte supplement on growth performance and mitigation of heat stress in southeast Louisiana. Milk replacer treatments consisted of Land O'Lakes Herdmaker Supreme (20% CP, 20% fat; CON) and Land O'Lakes Warm Front (27% CP, 10% fat; WF). Supplemented calves received either 0 or 20 mL of Palamountains Calf Boost (CB) in MR once daily. Calves were offered MR treatments and water and calf starter (20% CP) ad libitum beginning on d 2. All milk replacer was mixed at 15% solids. Calves consuming CON were fed 2.28kg MR twice daily. Calves on WF were fed 2.72kg MR twice daily for the first 3 weeks of life, and 3.86kg twice daily until weaning. Beginning on d 42, MR feeding was reduced to 1 time per day for all treatment groups to decrease MR intake by 50%. On d 49 calves were weaned. Calves remained in their hutches until d 56 to determine immediate post weaning performance. Blood was collected on d 14, 28, 42, and 56 for analysis of plasma urea nitrogen (PUN), glucose, and  $\beta$ -hydroxybutyrate (BHBA), as well as rumen fluid for analysis of volatile fatty acids (VFA) and pH. Data were analyzed using the PROC MIXED procedure in SAS. A main effect of milk replacer composition on PUN was observed, with calves fed WF having greater concentrations ( $P < 0.05$ ) than CON. Glucose concentrations decreased ( $P < 0.05$ ) as calves aged. There was no treatment effect ( $P > 0.05$ ) on plasma BHBA, but concentrations increased ( $P < 0.05$ ) as calves aged. Likewise, there was no treatment effect ( $P > 0.1$ ) on rumen acetate, propionate, butyrate, and total VFA concentrations; however, concentrations increased ( $P < 0.05$ ) as calves aged. No effects of treatment or time were observed ( $P > 0.05$ ) for rumen pH. These data indicate that milk replacer composition and feeding management and multivitamin mineral supplements do not affect negatively metabolism or rumen development in young dairy calves.

**Key Words:** calf milk replacer, multivitamin-mineral supplement, heat stress

**M381 Validation of a radio frequency system for monitoring feeding behavior and intake of feed and water in young cattle.** Baltazar Ruas de Oliveira Júnior<sup>1</sup>, Marcelo Neves Ribas<sup>2</sup>, Fernanda Samarini Machado<sup>3</sup>, Juliana Aparecida Mello Lima<sup>1</sup>, Luigi Francis Lima Cavalcanti<sup>2</sup>, Mario Luiz Chizzotti<sup>4</sup>, Rafael Alves de Azevedo<sup>1</sup>, and Sandra Gesteira Coelho<sup>1</sup>, <sup>1</sup>Federal University of Minas Gerais, Belo Horizonte, MG, Brazil, <sup>2</sup>CNPq, RHAÉ-SEVA Engenharia, Projeto Intergado, Contagem, MG, Brazil, <sup>3</sup>EMBRAPA Dairy Cattle,

Juiz de Fora, MG, Brazil, <sup>4</sup>Federal University of Viçosa, Viçosa, MG, Brazil.

The objective was to validate a radio frequency system for monitoring individual feeding behavior, water and feed consumption in young cattle housed in group. Thirty 5 Holstein-Gyr crossbred heifers, fitted with an ear tag containing a unique passive transponder, were distributed in 3 groups of 12, 12 and 11 animals per period and had free access to 12 electronic feed bins and 2 electronic water bins (Intergado, Contagem, Brazil). The system documented the visit duration and feed and water intake by recording animal's identification tag, bin number, initial and final times of visits, and the difference of feed/water weight at start and end for each bin visit. Feed bins were monitored by time-lapse video recording over 4 d and the water bins over 6 d. Video data on animal behavior were compared with those generated by the system. Feed and water consumption were measured using an external scale. For each feed bin, 2 feeding events were monitored using manual weighing's immediately before and after the animal's visit and the difference between them was assumed as feed intake (n = 24 observations). For water bins there were made 60 manual weighing's. These data were compared with those recorded by the system. Video and manual weighing data were regressed on the electronic feeding behavior and feed and water intakes data to evaluate system's precision and accuracy. The system showed a high specificity (98.98 and 98.56% for the feed and water bins, respectively) and sensitivity (99.25 and 98.74%, respectively) for identifying animal's presence or absence. Duration of feed and water bins visits, and feed and water consumption per visit estimated by the system were highly correlated and precise ( $R^2 = 0.917, 0.963, 0.973$  and  $0.986$ , respectively) when compared with observed video and manual weighing data. Feed daily intake per visit registered electronically and manual weighing differed by less than 150 g. It was concluded that Intergado system is a useful tool for monitoring feeding behavior, water and feed intakes in young cattle housed in group.

**Key Words:** electronic monitoring, heifer, precision farming

**M382 Evaluation of two techniques used to dislodge bacteria from particles contained in rumen digesta.** Jared V. Judy\*, Chad J. R. Jenkins, Samodha C. Fernando, and Paul J. Kononoff, *University of Nebraska-Lincoln, Lincoln, NE.*

The objective of this study was to estimate the concentration of bacterial crude protein (BCP) in pellets isolated from ruminal digesta using a preparatory step of either blending or shaking to dislodge bacteria from rumen particles. Using a completely randomized design, 2 multiparous, lactating Holstein cows (DIM 229 ± 7d, DMI 36.1 ± 2.5 kg/d, milk yield 37.7 ± 5.6 kg/d) (mean ± SD), fitted with ruminal cannulas were fed the same diet once daily at 0930 h. Two hours post feeding, approximately 2.5 kg of rumen contents were collected from each cow, then thoroughly mixed and separated into 2 aliquots (blend or shake) then samples were strained through 4 layers of cheesecloth. Particle associated bacteria were separated from the solid portion of rumen contents by adding and equal amount of McDougal's buffer as was collected in the filtrate and physically shook or blended in a commercial blender for 1 min., followed by straining through 4 layers of cheesecloth. Fluid collected after shaking or blending, as well as fluid retained from the initial straining were combined together. Each sample underwent differential centrifugation which yielded bacterial pellets consisting of fluid associated bacteria and particle associated bacteria. DNA was then extracted from bacterial pellets and from the non-centrifuged samples of rumen content particles. The DNA from the bacterial pellets and samples of rumen content were subjected to real-time PCR using the TaqMan assay. Primers and a probe were designed from the DNA encoding part of the 16S rRNA for bacteria.

The concentration of BCP using these 2 methods to dislodge bacteria did not differ ( $P = 0.42$ ) ( $13.9 \pm 5.0$  and  $7.5 \pm 1.9$  mg BCP/g DM for shake vs. blend, respectively). Results suggest that BCP concentration is not different between shaking or blending to dislodge bacteria, however, further research should examine and attempt to identify the large amount of analytical variation observed in both techniques.

**Key Words:** bacteria, bacterial crude protein, rumen

**M383 Effect of an exogenous fibrolytic enzyme on the performance of dairy cows consuming a diet with a high proportion of bermudagrass silage.** Andres A. Pech Cervantes\*, Kathy G. Arriola, Jorge E. Zuniga, Ibukun M. Ogunade, Yun Jiang, Thiago F. Bernardes, Charles R. Staples, and Adegbola T. Adesogan, *Department of Animal Sciences, University of Florida, Gainesville, FL.*

We previously reported that milk production by dairy cows was increased by adding specific xylanase-rich (XYL) and xylanase-cellulase enzymes to corn silage-based diets containing 0 or 10% bermudagrass silage. This study examined effects of adding XYL on the intake and performance of lactating dairy cows consuming a TMR formulated with a greater proportion of bermudagrass silage. Endoglucanase and xylanase activities of XYL were 3,283 and 46,281  $\mu\text{mol}/\text{min}/\text{mL}$ , respectively. Forty lactating Holstein cows (16 multiparous and 24 primiparous;  $21 \pm 3$  DIM; BW  $589 \pm 73$  kg) were stratified by milk production and parity and assigned randomly to Control and XYL diets. The TMR (CP of 16.2% of DM, NDF of 36.4% of DM, and  $\text{NE}_L$  of 1.65 Mcal/kg of DM) contained 20% bermudagrass silage, 25% corn silage, and 55% concentrate (DM basis). Immediately before the a.m. (0700 h) and p.m. (1300 h) feedings, the enzyme was sprayed on the XYL diet at the rate of 1 mL/kg of TMR DM in a Calan data ranger and mixed. A second data ranger was used to feed control cows. Cows were fed experimental diets for 70 d after they were fed a common diet for a 9-d covariate period. The experiment had a randomized complete block design. The statistical model included effects of enzyme, parity, week, and their interactions as well as covariate milk production or DMI. The random effect was cow nested within treatment. Application of XYL did not ( $P > 0.10$ ) affect milk yield (35.1 vs. 36.2 kg/d), DM intake (24.0 vs 23.7 kg/d for XYL and Control), fat-corrected milk (FCM) (36.1 vs. 36.9 kg/d), yields of milk fat (1.29 vs. 1.31 kg/d) and protein (1.07 vs. 1.08 kg/d), milk fat concentration (3.65 vs. 3.61%), and body weight change (0.26 vs. 0.33 kg/d) compared with control cows. However, cows fed the diet treated with XYL had greater milk protein concentration ( $P = 0.01$ ; 3.02 vs. 2.95%) and tended to have less feed efficiency ( $P = 0.06$ ; 1.52 vs. 1.57 kg of FCM/kg of DMI) compared with cows fed the control diet. Adding XYL to a diet containing 20% bermudagrass silage and 25% corn silage did not improve DM intake or milk production.

**Key Words:** bermudagrass silage, milk, enzyme

**M384 Effects of intensive whole-milk feeding in calves on subsequent feeding behavior of dairy heifers.** Camila Flávia de Assis Lage<sup>1</sup>, Mariana Magalhães Campos<sup>2</sup>, Fernanda Samarini Machado<sup>2</sup>, Paulo Campos Martins<sup>1</sup>, Luigi Francis Lima Cavalcanti<sup>3</sup>, Marcelo Neves Ribas<sup>3</sup>, Luiz Gustavo Ribeiro Pereira<sup>2</sup>, Thierry Ribeiro Tomich<sup>2</sup>, Rafael Alves de Azevedo\*<sup>1</sup>, and Sandra Gesteira Coelho<sup>1</sup>, <sup>1</sup>Federal University of Minas Gerais, Belo Horizonte, MG, Brazil, <sup>2</sup>EMBRAPA Dairy Cattle, Coronel Pacheco, Minas GG, Brazil, <sup>3</sup>CNPq, RHAÉ-SEVA Engenharia, Projeto Intergado, Contagem, MG, Brazil.

This study aimed to evaluate the effects of intensive whole milk feeding in calves on subsequent feeding behavior of 58 Holstein-Gyr females. Up to 56 d of age, calves received 6 L/d of 4 different liquid diets consisting of whole milk with the increasing addition of milk replacer (Sprayfo Violet SSP) to adjust the concentration of total solids (TS) to 13.5 (n = 15), 16.1 (n = 15), 18.2 (n = 13), 20.4% (n = 15). After weaning, animals were randomly housed in 4 paddocks, each one equipped with 3 electronic feed bins and one electronic water bin (INTERGADO, Brazil) in Embrapa Dairy Cattle facilities, Brazil. The diet (70% corn silage and 30% concentrate, 195 g of CP/kg, DM basis) was fed ad libitum, twice a day, until 210 d of age. Only events with registered intake were used, and the following results were calculated: Ingestion rate (IR, g/s), average bunk visit duration (AVD, min), daily visit duration (DVD, h) and daily visit frequency (VF, events). Due to the natural right skewness of IR and AVD distributions, their daily median, instead of mean, were used to represent their trends across study period. All variables were analyzed as a completely randomized design with repeated measures using linear mixed model approach. Age and TS were evaluated as fixed effects, while animals as random. The necessity to model error dependence and heteroscedasticity was evaluated by monitoring Schwarz criterion. The effect of TS was decomposed into orthogonal polynomials of linear and quadratic degrees. Significance was declared at  $P < 0.05$ . All variables were consistently influenced by animal's age, where IR was increased as animals become older ( $0.837 \pm 0.075$  g/s at 100 d old versus  $1.617 \pm 0.072$  g/s at 210 d old). An interaction effect between TS and age was detected for IR, where TS linear increased IR for animals older than 190 d of age. None of visit variables were affected by TS levels (AVD =  $3.70 \pm 0.37$  min, DVD =  $2.50 \pm 0.12$  h, VF =  $35.16 \pm 3.21$  events). Intensive whole-milk feeding in calves caused minor effects on subsequent feeding behavior of dairy heifers.

**Key Words:** milk replacer, intake, precision farming

**M385 Comparison of the RQUICKI estimate of insulin sensitivity with glucose and insulin tolerance in periparturient dairy cows.** Sina Saed Samii\*, J. Eduardo Rico, Alice T. Mathews, Cassandra L. Orndorff, Amanda N. Davis, and Joseph W. McFadden, *West Virginia University, Morgantown, WV.*

The revised quantitative insulin sensitivity check index (RQUICKI) has been utilized to evaluate insulin resistance in dairy cows; however, discrepancies between RQUICKI and direct measurements of insulin sensitivity are documented. Our objective was to compare RQUICKI with glucose and insulin tolerance in non-fasted periparturient dairy cows. Multiparous Holstein cows were grouped by BCS at d -28 prepartum: lean (BCS  $2.91 \pm 0.13$ ; n = 7) or overweight (OVER; BCS  $4.03 \pm 0.21$ ; n = 7). Diets were formulated to meet nutrient requirements. An intravenous insulin challenge (0.1 IU/kg BW; ITT) was performed on d -26 and -13, relative to expected calving, and 5 DIM. An intravenous glucose challenge (0.3 g/kg BW; GTT) was performed 24 h post-ITT. Blood and milk were collected routinely. Data were analyzed using a mixed model with repeated measures (fixed effects of BCS and day). Effects are presented as changes relative to lean cows, unless described otherwise. OVER had lower DMI, and lost more BCS and BW postpartum ( $P < 0.05$ ). Adiposity had no effect on milk yield, milk protein yield, and SCC; however, milk fat yield was greater in OVER ( $P < 0.05$ ). OVER had increased plasma NEFA and BHBA ( $P < 0.05$ ). Prepartum plasma insulin levels were higher in OVER ( $P < 0.05$ ). Although plasma glucose levels declined with time ( $P < 0.01$ ), BCS did not modify plasma glucose. RQUICKI values were lower for OVER pre- and postpartum ( $P < 0.05$ ). Postpartum cows had lower insulin-stimulated glucose disposal, relative to prepartum cows ( $P < 0.01$ ). Following insulin-

stimulated glucose disappearance, return to basal glucose in OVER was delayed by 60 min ( $P < 0.05$ ). BCS had no effects on GTT; however, post-glucose challenge area under the curve (AUC) for 180 min and clearance rate (%/min) for the first 30 min were lower for postpartum cows, relative to prepartum cows ( $P < 0.05$ ). Before and after calving, OVER experienced greater glucose-stimulated reductions in NEFA and AUC for 180 min following glucose challenge ( $P < 0.05$ ). Observed inconsistencies between RQUICKI and tolerance testing may be due to direct measurements in the fed state.

**Key Words:** glucose tolerance, insulin resistance, transition cow

**M386 Evolving the plasma free AA dose-response technique to determine bioavailability of Met in RP-Met supplements.** Devan L. Chirgwin\*<sup>1</sup>, Nancy L. Whitehouse<sup>1</sup>, Andre F. Brito<sup>1</sup>, Charles G. Schwab<sup>2</sup>, and Brian K. Sloan<sup>3</sup>, <sup>1</sup>University of New Hampshire, Durham, NH, <sup>2</sup>Schwab Consulting, LLC, Boscobel, WI, <sup>3</sup>Adisseo, Alpharetta, GA.

The plasma free AA dose-response technique has been proposed as the standard approach for arriving at estimates of efficacy for rumen-protected Lys supplements. Results of the first replicate of a  $5 \times 5$  Latin square study, reported last year [J. Dairy Sci. (97(E-Suppl. 1):763], confirmed that a positive relationship also exists between increasing amounts of absorbed Met and plasma Met and total sulfur AA (TSAA) concentrations. The objective of adding a second replicate was to complete the study, and using the combined data set, to determine if using plasma Met or TSAA concentrations ( $\mu\text{M}$ ) is the more precise response parameter and whether expressing either as a % of total AA (TAA) or total essential AA (TEAA) would reduce the error of calculated estimates of Met-bioavailability. Experimental protocol was as previously described: namely, 5 rumen-cannulated Holstein cows (74–222 DIM) were fed a Met-deficient basal diet with identical treatments. Combined data were analyzed using PROC MIXED and PROC REG of SAS. Outlier analysis, using  $\pm 2.0$  SD away from the mean for plasma Met and TSAA concentrations, resulted in removal of all data for one cow. Plasma Met and TSAA concentrations ( $\mu\text{M}$ ), and both expressed as %TAA and %EAA, were regressed on 0, 12, and 24 g of infused Met and 0, 15, and 30 g of fed Met. Slopes (and associated CV, %) for infused and fed Met were 1.40 and 1.04 (3.68 and 1.85) for Met, 0.067 and 0.048 (4.92 and 3.18) for Met as %TAA, and 0.152 and 0.112 (4.48 and 3.22) for Met as %TEAA. Corresponding values for total TSAA were 2.00 and 1.64 (1.58 and 2.57) for  $\mu\text{M}$  concentrations, 0.108 and 0.079 (2.79 and 2.48) for %TAA, and 0.234 and 0.184 (2.67 and 3.48) for %TEAA, respectively. Estimates of bioavailability (and 95% CI) of the RP-Met supplement for the 6 respective methods of expression were 74.4 (2.1), 71.6 (1.9), 73.7 (1.3), 81.8 (3.3), 72.7 (1.3) and 78.6 (2.3). We conclude the plasma free AA dose-response technique is precise, and because Met is a precursor to other sulfur AA, TSAA ( $\mu\text{M}$ ) is the most appropriate response parameter for estimating Met bioavailability of RP-Met supplements.

**Key Words:** methodology, methionine, bioavailability

**M387 Effects of intensive whole-milk feeding in calves on subsequent performance and feed efficiency of crossbred dairy heifers.** Camila Flávia de Assis Lage<sup>1</sup>, Mariana Magalhães Campos<sup>2</sup>, Fernanda Samarini Machado<sup>2</sup>, Paulo Campos Martins<sup>1</sup>, Luigi Francis Lima Cavalcanti<sup>3</sup>, Marcelo Neves Ribas<sup>3</sup>, Luiz Gustavo Ribeiro Pereira<sup>2</sup>, Thierry Ribeiro Tomich<sup>2</sup>, Rafael Alves de Azevedo\*<sup>1</sup>, and Sandra Gesteira Coelho<sup>1</sup>, <sup>1</sup>Federal University of Minas Gerais, Belo Horizonte, MG, Brazil, <sup>2</sup>EMBRAPA Dairy Cattle, Coronel Pacheco,



MG, Brazil, <sup>3</sup>CNPq, RHAÉ – SEVA Engenharia, Projeto Intergado, Contagem, MG, Brazil.

This study aimed to evaluate the effects of intensive whole-milk feeding in calves on subsequent performance and feed efficiency in growing heifers. Up to 56 d of age, Holstein-Gyr calves received 6 L/d of 4 different liquid diets consisting of whole milk with the increasing addition of milk replacer (Sprayfo Violet SSP) to adjust the concentration of total solids (TS) to 13.5 (n = 15), 16.1 (n = 15), 18.2 (n = 13), 20.4% (n = 15). After weaning, animals were housed in 4 paddocks, each one equipped with 3 electronic feed bins and one electronic water bin (Intergado, Brazil) in the experimental farm of Embrapa Dairy Cattle, Brazil. The same diet (70% corn silage and 30% concentrate, dry matter basis; 195 g of CP/kg of DM) was fed in ad libitum, twice a day, until 210 d of age. Daily feed intake (DFI) and water intake (WI) were registered by the electronic system. Average daily gain (ADG) was determined from regression of weekly BW measurements so feed conversion (FC) could be calculated. Longitudinal data was analyzed as a completely randomized design with repeated measures using linear mixed models, where animal's age and TS were fixed effects while animal was considered a random effect. The necessity to add random components to model error dependence and heteroscedasticity was evaluated based on Akaike's Information Criterion. Initial body weight, air temperature and humidity were added as co-variables. Average and cumulative results were analyzed by linear regression, where only the fixed effect of TS was evaluated ( $\alpha = 0.05$ ). DFI and WI were only influenced by age, however in both cases an interaction effect was observed, although none supplementation level caused a steady superiority across period for any variable. This result was corroborated by cumulative DFI and WI that were not influenced by TS. ADG was decreased by increasing nutritional management (e.g.,  $916 \pm 30$  and  $833 \pm 29$  g/d for animals with 13.5 and 20.4%, respectively), what also reflected in a higher FC to animals in higher TS ( $13.19 \pm 0.45$  vs  $14.55 \pm 0.44$  in animals that received 13.5 and 20.4% of FD, respectively).

**Key Words:** milk replacer, feeding, performance

**M388 Immediate and long-term effects of niacin feeding to fresh dairy cows. 1. Ketosis and fertility.** J. M. Havlin\*<sup>1</sup>, P. H. Roberson<sup>1</sup>, and J. E. Garrett<sup>2</sup>, <sup>1</sup>University of California, Davis, Davis, CA, <sup>2</sup>Qualitech, Chaska, MN.

During the fresh period after calving through ~21 d postpartum, dairy cows are often in negative energy balance (NEB) due to high energy demands to support rapidly rising milk output at a time of relatively low dry matter intake (DMI). This NEB makes cows susceptible to ketosis, fatty liver, metritis, and displaced abomasum, which can lead to decreased performance and eventual culling. A possibility to reduce the extent of NEB is to feed niacin (Ni) as nicotinic acid (NA) to reduce milk fat production, thereby minimizing body weight loss to reduce ketogenesis, all to reduce the extent of NEB thereby creating a more successful lactation. Multiparity Holstein cows (672) on a California dairy farm were used from 14 d pre-calving through 150 d in milk (DIM). While in the close-up dry pen (-14 to 1 DIM), cows were comingled and fed the same total mixed ration (TMR), in the fresh pens (1 to ~22 DIM) cows were fed the same TMR, except for inclusion of ruminally protected (RP) Ni (rumen escape estimate = 66%), in separate pens at 0, 3.5, 7 or 14 g NA/cow/d. Cows were comingled in the high pen (~23 to 150 DIM) and fed the same TMR. DMI was tabulated by treatment in the fresh pens, blood samples were collected for NEFA and BHBA analysis during the dry and fresh periods, and fertility data was tabulated through 150 DIM. Feeding 3.5 g/d RPNi increased DMI from 19.3 to 21.5 kg/d in the fresh period, but RPNi at 14 g/d reduced it to below

Control cows (Quadratic  $P = 0.07$ ). Ketosis prevalence (% cows with BHBA  $\geq 1.44$  mg/dl) decreased from 36 to 20% in the fresh period with 3.5 g/d, but RPNi at 14 g/d did not differ from levels of Control cows (Quadratic  $P = 0.06$ ). Niacin feeding had no effect on any fertility measure, with averages for 1st service conception (%) being  $44.6 \pm 4.38$ , pregnancy (%) being  $76.2 \pm 3.70$  and services/conception being  $1.58 \pm 0.082$ . Short-term fresh period RPNi feeding at the 3.5 g/d level reduced the incidence of ketosis caused by NEB, but feeding higher levels removed those benefits. No RPNi feeding level during the fresh period affected fertility parameters through 150 DIM.

**Key Words:** niacin, ketosis, fertility

**M389 Effects of supplementation with a rumen-protected lysine product on production in high-producing dairy cows.**

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The objective of this study was to determine the effects of supplementation and withdrawal of a rumen-protected lysine product, AjiPro-L 2nd generation (2G), on production performance of high-producing dairy cows. Ninety-six multiparous Holstein cows averaging  $2.9 \pm 1.3$  lactations and  $83.6 \pm 36.7$  DIM at the commencement of the trial were assigned at random to one of 12 pens that received either a negative control ration (n = 4), positive control ration with added metabolizable lysine from an animal protein (n = 4), or AjiPro-L 2G added metabolizable lysine from a rumen-protected amino acid (n = 4). The 8wk experimental period consisted of 2 periods; a 4-wk treatment period followed by a 4-wk carryover period. During the treatment period, animals received negative, positive or AjiPro-L 2G treatments, whereas during the carryover period all animals received the positive control treatment. Milk yield, energy-corrected milk yield, water consumption and DMI were recorded daily and milk composition was measured weekly. Data from only cows that completed the entire duration of the study were used in the analysis. Days in milk categories of either high ( $114 \pm 17.7$ ) or low ( $53 \pm 21.3$ ) were also analyzed. Results showed that water consumption and DMI was significantly increased compared with baseline ( $P < 0.05$ ) and MUN levels were significantly decreased compared with baseline ( $P < 0.01$ ) for animals receiving AjiPro-L 2G during the treatment period. When comparing DIM categories, results revealed a significant difference ( $P < 0.05$ ) in change of milk production, ECM, pounds of fat and pounds of lactose in comparison to baseline across treatments. For these parameters, high DIM animals receiving AjiPro-L 2G had greater milk yield, and the least reduction of ECM, pounds of fat, and pounds of lactose from baseline compared with the positive and negative treatments. During the carryover period all parameters measured, except for MUN ( $P < 0.05$ ), were not different between treatments compared with baseline. Under the conditions of this experiment, supplementation of AjiPro-L 2G had the greatest effect on cows averaging 114 DIM in which milk production and milk components were maintained in post-peak cows.

**Key Words:** rumen-protected lysine, dairy cow, production performance

**M390 Immediate and long-term effects of niacin feeding to fresh dairy cows. 2. Body condition and milk production.** J. M. Havlin\*<sup>1</sup>, P. H. Roberson<sup>1</sup>, and J. E. Garrett<sup>2</sup>, <sup>1</sup>University of California, Davis, Davis, CA, <sup>2</sup>Qualitech, Chaska, MN.

During the fresh period after calving through ~21 d postpartum, dairy cows are often in negative energy balance (NEB) due to high energy

demands to support rapidly rising milk output at a time of relatively low dry matter intake (DMI). Multiparity Holstein cows (672) on a California dairy farm were used from 14 d pre-calving through 150 d in milk (DIM). While in the close-up dry pen (-14 to 1 DIM), cows were comingled and fed the same total mixed ration (TMR), in the fresh pens (1 to ~22 DIM) cows were fed the same TMR, except for inclusion of ruminally protected (RP) Ni (rumen escape estimate = 66%) in separate pens at 0, 3.5, 7 or 14 g nicotinic acid/cow/d. Cows were comingled in the high pen (~23 to 150 DIM) and fed the same TMR. Milk production and body condition (BCS) was measured every 2 wk in the fresh pen and every 4 wk in the high pens. At 7 ± 3.9 DIM, there was no effect of RPNi on milk (39.3 ± 0.89 kg) or component yields. However at 21 ± 3.9 DIM there were quadratic trends ( $P < 0.20$ ), with highest outputs at 3.5 g/d feeding and lowest at 14 g/d. However at the 1st high pen milk sampling after cessation of RPNi feeding (48 ± 8.0 DIM), milk and component yields slumped for cows previously fed 3.5 g/d resulting in lower yields vs Control (53.2 vs 51.8 kg milk/d; 1.75 vs 1.67 kg fat/d), but over the next 3 mo they converged with Control. In contrast, milk and component yields of cows previously fed 14 g/d rebounded on the 1st high pen milk sampling after cessation of feeding RPNi resulting in higher yields vs Control (53.2 vs 55.3 kg milk/d; 1.75 vs 1.82 kg fat/d). There was no effect of RPNi feeding on BCS during the fresh period, but BCS of cows previously fed 3.5 g/d decreased more than Control cows in high pens, maintaining a lower BCS through ~80 DIM, but by 150 DIM there was no difference with Control. In contrast the BCS of cows previously fed 14 g/d decreased from the fresh period through 150 DIM. RPNi had small effects on animal performance while supplemented, but cessation of feeding caused short-term rebound carry-over effects. However, performance of cows on all RPNi levels had largely converged by 150 DIM.

**Key Words:** niacin, milk, BCS

**M391 Lactational performance of cows fed extruded linseed on commercial dairy herds.** Amélie Beauregard<sup>1,2</sup>, Marie-Pierre Dallaire<sup>1</sup>, Rachel Gervais<sup>1</sup>, and P. Yvan Chouinard<sup>1,2</sup>, <sup>1</sup>Université Laval, Quebec, QC, Canada, <sup>2</sup>Institute of Nutrition and Functional Foods, Quebec, QC, Canada.

The objective of this study was to determine the effect of feeding extruded linseed (EL) on milk yield and milk composition, including fatty acid profile, and enteric methane output on commercial settings. Thirty dairy herds averaging 55 cows and 30.1 kg of milk/d with 4.0% fat and 3.3% protein were recruited for this study. Twenty-eight herds had Holstein cows, while 2 herds had Jersey and Holstein cows, and all of them were located in the province of Quebec, Canada. A first group of 15 herds was randomly selected to be used as control (CTL). Cows from the remaining 15 herds were fed between 200 and 900 g of EL (mixture of linseed:wheat bran, 70:30; Valorex, Combournillé, France) per cow per day (average 700 g/d) according to their lactation stage and their level of production. Diet compositions were then adjusted to cover the animals' nutrient requirements. The feeding trial was 6 mo in length, and data collected during a 2-mo interval before the experimental period were used as covariates. Actual milk yield was higher in herds fed EL as compared with CTL (28.5 vs. 27.4 L/d;  $P = 0.01$ ). Milk fat content was similar between treatments (4.08 kg/hL;  $P = 0.20$ ), but milk fat yield tended to be higher in herds fed EL as compared with CTL (1.15 vs. 1.12 kg/d;  $P = 0.09$ ). Feeding EL decreased milk protein content (3.30 vs 3.37 kg/hL;  $P < 0.01$ ), but had no effect on milk protein yield (0.94 kg/d;  $P = 0.17$ ) when compared with CTL. Milk fat contents of *cis*-9,*cis*-12,*cis*-15 18:3 (6.72 vs 4.79 mg/g;  $P < 0.01$ ), and other fatty acids of the n-3 family (18:4, 20:4, 20:5, 22:5, 22:6;  $P \leq 0.05$ ) were higher in herds

fed EL as compared with CTL. Methane output, as estimated using a proprietary equation (WO 2009 156453 A1) based on milk yield and fatty acid profile, was 9.4% lower in dairy herds fed EL as compared with CTL (12.9 vs. 14.2 g/L milk;  $P < 0.01$ ). In conclusion, feeding moderate amount of EL (700 g/d, providing 200 g of oil) was efficient to mitigate methane emission while maintaining animal performance and increasing the n-3 FA content in milk fat.

**Key Words:** cow, extruded linseed, methane

**M392 Bacterial communities in the gastrointestinal tract of preruminant dairy calves.** Janet E. Williams\*, William I. Loucks, Elizabeth D. Benda, Nicola F. Beatty, Katelyn M. Steinkamp, Matthew E. Doumit, and Mark A. McGuire, University of Idaho, Moscow, ID.

Nutritional factors are known to influence the development of the gastrointestinal (GI) tract in preruminants. However, the effect of different lipid sources on the bacterial communities in the GI tract of young calves has not been well described. Therefore, the aim of this study was to utilize high-throughput sequencing to investigate the GI microbiota of preruminant calves fed supplemental polyunsaturated fatty acids. Calves ( $n = 8$ ) were fed milk (4 L per d) plus 3% supplemental oil containing either palm oil or a combination of conjugated linoleic acid and flaxseed oil in a ratio of 1:2 (CLA/FLAX) for 50 d beginning 3–6 d after birth. After euthanasia, digesta samples from the rumen, omasum, abomasum, duodenum, cecum, and large intestine (LI) were collected. DNA was extracted using the Qiagen DNA Stool Mini kit and bacterial DNA amplified using primers targeting the V1-V3 hypervariable region of the 16S rRNA gene. Amplicons were sequenced using an Illumina MiSeq v3 paired-end 300-bp protocol for 600 cycles. Sequences were processed and classified using the custom python application dbcAmplicons. Principal coordinate analysis at the genus level revealed 3 clusters: (1) rumen, omasum, abomasum; (2) duodenum; (3) cecum and LI. All 3 clusters had large proportions of reads that could not be classified at the genus level: 1) 39.9% ± 1.9; 2) 76.2% ± 5.4; 3) 56.4% ± 2.6. Clusters 1 and 2 were enriched in *Prevotella* (24.4% ± 1.6 and 7.2% ± 2.7, respectively) and *Succinivibrio* (8.7% ± 1.4 and 2.8% ± 2.1, respectively) while Cluster 3 had similar proportions of *Prevotella* (6.2% ± 2.2), *Bacteroides* (6.2% ± 1.9), and *Oscillibacter* (4.3% ± 0.6). Using generalized linear mixed models, rumen, omasum, and abomasum bacterial communities from calves fed palm oil had greater ( $P < 0.05$ ) abundances of *Succinivibrio* (13.10% ± 1.9 vs 4.1% ± 1.0), *Oscillibacter* (2.7% ± 0.4 vs 1.1% ± 0.3), and *Paraprevotella* (2.5% ± 0.4 vs 1.4% ± 0.3), and lower ( $P < 0.05$ ) abundances of *Prevotella* (20.4% ± 1.8 vs 28.4% ± 2.1) as compared with those from calves fed CLA/FLAX. The implications of different ruminal bacterial composition on growth and tissue development in preruminant calves warrant further research.

**Key Words:** preruminant, microbiome, gastrointestinal

**M393 Application of tri-axial accelerometers to determine the grazing behavior of dairy cows in a commercial dairy herd.** Pieter J. M. Raedts\*, Rajneet S. Sohi, Indunil Kulatililke, and Markandeya Jois, La Trobe University, Melbourne, Victoria, Australia.

In grazing dairy farming systems measurement of feed intake is problematic, especially due to lack of practical methods to determine the quantity of pasture that cows graze. Our aim was to measure relative DM intake by grazing behavior using tri-axial accelerometers. Tri-axial accelerometers (ActiGraph) were set to continuous recording at 60 Hz and attached to collars around the neck of 10 lactating dairy HF and

HF-Jersey cross cows (HF 82.5% to 100%) on a commercial dairy herd in Northern Victoria, Australia. After 16 full days of recording, the sensors were removed from the collars and recorded RAW data downloaded from the sensors. The 16 d RAW data were used to determine grazing time, bites and intensity. Occasional observational data of cow behavior was used to validate behavior of cows as determined from the activity counts. All cows in the herd were offered the same ration (flat-feeding), consisting of approximately 1/3rd of DM as concentrate and 2/3rd of DM as forage. Forage was predominantly grazed pasture. Herd test results during the trial provided data regarding milk production and somatic cell count (SCC) of individual cows. The data set (n = 10) showed AVG per day grazing time of 340 min (265 to 397), 21,390 grazing bites (16,409 to 25,810) and 541,957 counts for grazing intensity (345,094 to 641,808). Two cows had a high SCC (>2,500,000/mL), indicating subclinical mastitis, and were excluded from the data set used for correlations. The cows (n = 8) had an AVG daily milk yield of 30.6 L (17.7 to 39.1) and 108 d in lactation (62 to 134). The correlation between number of daily bites and daily milk yield was  $R^2 = 0.80$ , while grazing time (minutes per day) had a correlation to milk yield of  $R^2 = 0.69$ . These results suggest that tri-axial accelerometer sensors are useful in determining grazing behavior of lactating dairy cows.

**Key Words:** grazing, bites, lactating

**M394 Effect of acetate and *trans*-10,*cis*-12 CLA on milk production in lactating dairy cows.** Natalie L. Urrutia\*, Michel Baldin, Jackie Y. Ying, and Kevin J. Harvatine, *The Pennsylvania State University, University Park, PA.*

During CLA-induced milk fat depression (MFD) acetate and glucose are spared from milk fat synthesis and are available for other metabolic uses. Acetate is the major carbon source spared and although acetate deficiency does not cause milk fat depression the effect of acetate supply on lactation and the effect of spared acetate during MFD is not clear. The objective of this study was to compare the effect of CLA and acetate equivalent to that spared during MFD on milk production. Nine multiparous, lactating, ruminally cannulated Holstein cows ( $244 \pm 107$  DIM; mean  $\pm$  SD) were randomly assigned to treatments in a  $3 \times 3$  Latin square design. Experimental periods were 14 d in length and included 4 d for treatment and 10 d for washout period. Cows received the following treatments: control (CON), acetate [ACE; continuous infusion of 7 M/d acetate pH 6.1 (rumen)], or CLA [10 g/d *trans*-10,*cis*-12 CLA (abomasal)]. Milk samples were collected on the last 2 d of treatment for determination of fat, lactose and protein concentration, and milk fatty acid profile. Data were analyzed using the fit model procedure of JMP Pro. The model included the random effects of cow nested in sequence, sequence and period and the fixed effect of treatment. Dry matter intake, protein and lactose yield and percentage were not affected by treatments. Milk yield tended to be increased 11% in ACE ( $P = 0.09$ ) compared with CON (22.8, 23.4 and 25.4 kg/d for CON, CLA and ACE, respectively). Milk fat yield increased 20% in ACE ( $P = 0.04$ ) and decreased 23% in CLA ( $P < 0.02$ ) and milk fat percent was decreased 30% by CLA ( $P < 0.001$ ), but was not affected by ACE compared with CON. Concentration and yield of de novo FA ( $P < 0.001$ ), while concentration of preformed FA (>C16) was increased by CLA ( $P < 0.001$ ), compared with CON. Yield of de novo FA, palmitic acid and total C16 FA was increased by ACE ( $P < 0.05$  for all) and concentration of palmitic acid was higher ( $P < 0.05$ ) in ACE, compared with CON. In conclusion, acetate supply has an effect on milk production and milk fat synthesis, and spared acetate during MFD may improve energy status.

**Key Words:** acetate, CLA, milk fat synthesis

**M395 Exogenous fibrolytic enzyme in dairy cows diets: Milk yield and composition.** Thiago Henrique da Silva\*<sup>1</sup>, Caio Seiti Takuya<sup>1</sup>, Thiago Henrique Anibale Vendramini<sup>1</sup>, Filipe Zanferari<sup>1</sup>, Elmeson Ferreira de Jesus<sup>2</sup>, and Francisco Palma Rennó<sup>1</sup>, <sup>1</sup>*University of São Paulo, Pirassununga, São Paulo, Brazil*, <sup>2</sup>*São Paulo State University, Jaboticabal, São Paulo, Brazil.*

Exogenous fibrolytic enzymes can be a feature to improve fiber digestion and performance of dairy cows. This study was aimed to evaluate the effect of exogenous fibrolytic enzymes on milk yield and composition. Twenty-four Holstein cows ( $180.2 \pm 54.3$  DIM;  $662.6 \pm 88.2$  kg BW) were assigned in a replicated  $4 \times 4$  Latin square design, with 21-d periods. Dietary treatments were: 0 (control), 8 (low), 16 (middle) and 24 (high) g of enzyme/cow/day (Fibrozyme, Alltech Inc., Nicholasville, KY) of total mixed ration based on corn silage. Milk yield was recorded by a computer twice daily at 0600 and 1600 h. Milk sample was collected proportionately to a.m. and p.m. milking, and analyzed for fat, protein and lactose by near infrared reflectance spectroscopy. Data were subjected to ANOVA and simple polynomial regression using the SAS software, version 9.0. There was no effect of fibrolytic enzyme supplementation on milk yield (30.24 vs. 30.22 vs. 30.52 vs. 29.89 kg/d) and 3.5% fat-corrected milk (FCM) yield (30.96 vs. 31.70 vs. 31.33 vs. 30.71 kg of 3.5% FCM/d) for control, low, middle and high supplementation, respectively. Also, milk composition was not affected ( $P > 0.05$ ) by enzyme supplementation just like yield fat, protein and lactose. A possible explanation for these results is that animals were not with energy challenge. Results of present study indicated that the use of exogenous fibrolytic enzymes did not improve performance of mid-lactation dairy cows.

**Key Words:** digestion, fiber, energy

**M396 Milk urea:allantoin ratio is a useful marker of efficiency of protein utilization in dairy cows.** Pieter J. M. Raedts\*, Devin A. Benheim, Ashlee J. Hammond, Jargal Menghe, and Markandeya Jois, *La Trobe University, Melbourne, Australia.*

Dietary protein is extensively degraded in the rumen into ammonia some of which which is in turn used by rumen bacteria to synthesize microbial protein. Excess ammonia is converted to urea in the liver and excreted in urine and milk. Microbial protein (MCP) is digested and absorbed as amino acids and nucleic acids. Nucleic acids are degraded in the liver into allantoin and excreted in the urine and milk. The ratio of urea to allantoin therefore is an indicator of efficiency of utilization of protein in the feed. Our aim was to investigate the efficiency of protein utilization by determining the urea:allantoin ratio in the milk of cows in the 1, 2, 3 and 6+ months of lactation. Herd test milk was collected from 44 Holstein Friesian (HF) and HF-Jersey cross cows (HF 75% to 100%) on a commercial dairy herd in Northern Victoria, Australia. The herd consisted of 2 cohorts, one in early lactation and one in the second half of lactation. All cows in the herd were offered the same ration (flat-feeding), consisting of approximately one-third of DM as concentrate and two-thirds of DM as grass based forage. Part of the forage was grazed pasture consisting predominantly of perennial English ryegrass (*Lolium perenne*) with a scattering of clover and some weeds. The milk was analyzed for urea concentration by an enzymatic method. Allantoin concentration was determined using high performance liquid chromatography. The MUN:MAC ratio was significantly different ( $P < 0.05$ ) between cows in 1st month of lactation ( $27.92 \pm 2.28$ ) and cows in 2nd, 3rd or 6+ months in lactation ( $34.84 \pm 1.68$ ,  $43.89 \pm 5.35$ ,  $45.60 \pm 3.42$ , respectively). This ratio was also significantly different ( $P < 0.05$ ) between cows in 2nd month of lactation and 6+ months of lactation. Cows in the first month of lactation had a significantly higher MAC



and lower MUN ( $P < 0.05$ ) than cows 3 or more months in lactation. These results indicate that cows in early lactation are more efficient in the utilization of dietary protein

**Key Words:** milk, urea, MUN

**M397 Use of chloride concentration to identify ration sorting by dairy cattle.** Heidi Rossow\*, *University of California, Davis, Davis, CA.*

Currently differences in proportions of particle sizes between the ration fed and the residual ration indicates if dairy cattle are sorting their feed. However, results using the Penn State Particle Sorter (PSPS) can be variable depending on the dry matter of the ration and how vigorously and consistently PSPS is shaken for each sample. Examining differences in chloride concentration (CC) of the ration between the ration fed and residual ration may be an easier and more accurate method to assess ration sorting by dairy cattle. Therefore, the objective of this study is to examine if CC in the ration fed compared with the residual ration could also be used to assess ration sorting by dairy cattle. Ten samples each of the ration fed and the residual ration were collected from 2 pens, including cows close to calving and cows 30–200 d in milk, from 5 dairies in Tulare County CA. Total CC, and CC from each tray of the PSPS was measured by soaking 30 g of sample in 200 mL of de-ionized water for 2 h and then measuring CC using an Oakton waterproof SaltTestr meter (Oakton Instruments, Vernon Hills IL) with a range of 0 to 1% chloride. Overall CC increased with decreasing particle size with means (standard deviations) of CC of 0.195% (0.048), 0.254% (0.041), 0.264% (0.055) and 0.277% (0.080) for top, middle, screen and bottom trays of the PSPS, respectively. Statistics were performed using Proc GLM (SAS Institute, 2013) with CC and PSPS % from each tray regressed on sample type (ration fed or residual ration) for each dairy and pen and if different ( $P < 0.05$ ), then the ration was sorted. For 8 pens representing 4 dairies, results for sorting (fed vs residual) were consistent between PSPS and CC. However for 1 dairy (2 pens), CC indicated sorting and PSPS did not. Therefore, results from CC and PSPS indicate that CC maybe an equivalent method to identify ration sorting but more research needs to be done to determine why results were not consistent on 1 dairy.

**Key Words:** chloride concentration, ration sorting, Penn State particle sorter

**M398 Pre- and postweaning performance of nursery calves offered texturized calf starters with varying protein levels for 56 days.** Bruce Ziegler<sup>1</sup>, David Ziegler<sup>2</sup>, Hugh Chester-Jones<sup>2</sup>, Daniel Schimek<sup>1</sup>, and Sarah Schuling<sup>1</sup>, <sup>1</sup>Hubbard Feeds, Inc., Mankato, MN, <sup>2</sup>University of Minnesota Southern Research and Outreach Center, Waseca, MN.

One hundred and four (2 to 5 d old) individually fed Holstein heifer calves ( $39.5 \pm 0.69$  kg BW) were randomly assigned to 1 of 4 treatments to evaluate pre- (d 1–42) and postweaning (d 43–56) calf performance when fed texturized calf starters varying in crude protein (CP) level. Texturized calf starter (CS) treatments were 1) 15% CP; 2) 18% CP; 3) 21% CP, and 4) 24% CP, as-fed. All calves were fed 0.28 kg of milk replacer (MR; 20% protein:20%fat) in 1.99 L water twice daily for the first 35 d and once daily from d 36 to 42. From d 1 to 14, 1:1 neomycin:oxytetracycline was added to the MR solution to provide 22 mg/kg BW/d. Calf starter and water were fed free choice throughout the trial. Linear (L) and quadratic (Q) contrasts were used to differentiate effects of CS CP level on growth performance. There were no differences among treatments in average daily gain pre- or postweaning. There was

a Q effect of treatment on daily gain for d 1 to 56 ( $P < 0.01$ ). Daily gains were 0.71, 0.76, 0.76, and 0.70 kg for the 15%, 18%, 21% and 24% CP CS treatment, respectively. There was a Q effect for hip height gain ( $P = 0.02$ ). Hip height gains were 10.2, 11.1, 11.0 and 9.9 cm for the 15%, 18%, 21% and 24% treatments, respectively. Preweaning CS intake was similar across treatments. Postweaning CS intake linearly decreased with increasing CP levels, d 43 to 56 ( $P < 0.01$ ) and overall ( $P < 0.03$ ). Preweaning gain/feed increased with CS CP level up to 21% CP then decreased (Q;  $P = 0.04$ ). A similar overall 56 d response in gain/feed occurred (Q;  $P = 0.05$ ). Gain/feed d 1 to 56 was 0.53, 0.55, 0.56, and 0.55 kg for 15%, 18%, 21% and 24% CP treatments, respectively. Fecal scores, scouring days and treatment costs were similar among treatments. b-Hydroxybutyrate levels at 42 d were the lowest in calves fed the 15% CP CS. Calf starter intake decreased with increasing CP levels but did not directly relate to calf performance. Under the conditions of this study, there was no benefit of feeding CS CP levels above 21%.

**Key Words:** calf performance, calf starter, crude protein

**M399 Pre- and postweaning performance and health of dairy calves fed milk replacers vs. pasteurized waste milk.** David Ziegler<sup>\*1</sup>, Hugh Chester-Jones<sup>1</sup>, David Cook<sup>2</sup>, and Julian Olson<sup>2</sup>, <sup>1</sup>University of Minnesota Southern Research and Outreach Center, Waseca, MN, <sup>2</sup>Milk Products, Chilton, WI.

The objectives of this study were to compare pre- (d 1 to 49) and post weaning (d 50 to 56) performance of calves fed a milk replacer (MR) formulated with similar crude protein (CP) and fat (F) concentrations to pasteurized waste milk (PWM) and a combination of PWM and a low (F) and high CP MR. One hundred and five (2 to 5 d old) individually fed Holstein heifer calves ( $38.8 \pm 0.73$  kg) were randomly assigned to 1 of 4 milk treatments. Milk treatments included 1) all-milk, non-medicated MR 20% CP: 20% F fed at 0.34 kg in 2.38 L of water 2x daily from d 1 to 42 and 1x daily from d 43 to weaning at d 49 (CON); 2) all-milk, non-medicated MR 26% CP: 31% F supplemented with additional fatty acids fed as in CON (MRS); 3) pasteurized waste milk 28.4% CP: 30.1% F fed as in CON, feeding rate was adjusted daily based on measured solids (PWM); 4) PWM fed 2x daily with 0.22 kg solids supplemented with 0.12 kg of an all milk non-medicated 24% CP: 7% F MR as in CON, adjusted for solids as in PWM (WMS). Calf starter (CS;18%CP) and water were fed free choice d 1 to 56. Waste milk was collected twice a week from one farm then sampled, cooled, and pasteurized before each feeding. Calves fed PWM and WMS avg. 0.85 kg/d gain vs. 0.72 kg/d for calves fed CON and MRS ( $P < 0.05$ ) for the 56 d study. Hip height gain avg. 13 cm for PWM and WMS vs. 11 cm for CON and MRS ( $P < 0.05$ ). There were no differences in intake of milk solids, avg. 29.7 kg for 49 d. Intake of CS, d 1 to 56, was highest for WMS (48.2 kg) with CON and PWM being intermediate (avg. 38.2 kg) and MRS the lowest (31.9 kg;  $P < 0.05$ ). Gain/feed was highest ( $P < 0.05$ ) for PWM (0.69 kg) with MRS (0.65 kg) being intermediate and CON and WMS the lowest ( $P < 0.05$ ; avg. 0.64 kg). There were no differences in daily fecal scores across treatments. Days with fecal scores = 4 and health costs were higher ( $P < 0.05$ ) for MRS vs. CON, PWM and WMS. From d 57 to d 84 there were no differences in ADG across treatments. Under conditions of this study calves fed WMS had greater CS intake than CON, MRS and PWM. Calves fed MRS did not enhance performance over CON.

**Key Words:** calf performance, milk replacer, pasteurized waste milk.

**M400 Evaluation of the effects of direct-fed microbials, microbial fermentation products, and digestive enzymes on milk yield and milk components in dairy cattle in the tropics.** Karen Espino-Mercado, Coral Castillo-Caballero, Jaime Curbelo-Rodríguez, and Guillermo Ortiz-Colón\*, *University of Puerto Rico at Mayaguez, Mayaguez, PR, Puerto Rico.*

The effect of direct-fed microbials, fermentation products (FP) and digestive enzymes on milk yield and milk composition was evaluated in dairy cattle. Lactating Holstein dairy cows (n = 40) from a commercial dairy herd were divided in 4 groups (n = 10 each) and blocked by days in milk (DIM; < 100 DIM or ≥100 ≤ 171 DIM) and balanced by parity number. Animals had a 2-week adaptation period. The balanced groups were randomly assigned to one of the following treatments: Control (C): 21g wheat middling; Treatment 1 (T1): 21g of a commercial mixture of *A. oryzae* FP, *B. subtilis* FP, *L. acidophilus* FP, yeast and amylase. Treatment 2 (T2): 18g Wheat middlings + 3g of a commercial mixture of *A. niger* FP, *A. oryzae* FP, α amylase, pectinase, endo- glucanase, β- glucanase, xylanase, and mannanase. Treatment 3 (T3): 11g Wheat middlings + 10g of a commercial mixture of *B. subtilis*, amylase and α amylase. Each treatment was top-dressed on the pelleted feed offered at the parlor during every morning milking. Milk production by cow was collected twice a day for 14 weeks using the AfiLab AfiMilk system. Milk urea nitrogen (MUN) and ketone body concentrations in milk by cow was collected once a week for 14 weeks using the Porta BHB milk ketone test and MUN was determined by the Teco Diagnostic Vet- MUN Reagent strips. There was no interaction between treatment, parity and/or period ( $P > 0.10$ ). Average milk production (kg/cow/per day) during the experiment was C 21.4 ± 2.12; T1 19.7 ± 2.11; T2 19.8 ± 2.23; T3 21 ± 2.11 ( $P > 0.05$ ). Average milk BHB concentration (μmol/L) per treatment was C 90.29 ± 13.08; T1 100.39 ± 12.04; T2 89.71 ± 13.12; T3 99.17 ± 12.57 ( $P = 0.2121$ ). Treatment neither had an effect on MUN. Mean MUN (mg/dL) per treatment was C 8.42 ± 1.3; T1 8.38 ± 1.20; T2 8.58 ± 1.44; T3 8.63 ± 1.26 ( $P = 0.8533$ ). In conclusion, in this experiment under tropical conditions, DFM, FP and digestive enzymes, resulted in no change in milk production, MUN and BHB concentrations in milk.

**Key Words:** direct-fed microbial, digestive enzyme, fermentation product

**M401 Phosphorous excretion and digestibility in Jersey and Holstein consuming corn milling co-products.** Gabriel Garcia Gomez\*, Alison Foth, and Paul Kononoff, *University of Nebraska-Lincoln, Lincoln, NE.*

Excess dietary phosphorous (P) in dairy cows diet may result in increased excretion of this mineral. Additionally, P accumulation in the soil may be a result of high concentrations of P when manure is applied to cropland. The objective of this study was to evaluate P intake, digestibility and excretion when dairy cows consumed rations containing reduced fat distillers grains (RFDDGS). Data from this study originated from an energy balance study in which RFDDGS was included at 28.8% of the ration DM. In this study, corn was reduced from 22.9 to 8.95% and soybean meal was reduced from 14.8 to 0% of the ration DM in the control and co-product (Co-P) diet, respectively. The study included 8 Holstein (BW = 693.8 ± 12.9 kg) and 8 Jersey (BW = 429.1 ± 13.0kg) multiparous, lactating cows (93 ± 20 DIM) in a repeated switchback design. The concentration of P in the test treatments were 0.44% and 0.59% ± 0.01% DM for the control and Co-P diet, respectively. The intake and excretion of P was estimated through feed sampling and total collection of feces. All feed and fecal samples were analyzed for P. Concentration of P in feces was lower in control diet compared with Co-P (0.97 vs. 1.27 ± 0.05%, respectively;  $P < 0.01$ ). Excretion of P was

less for cows fed the control diet compared with the Co-P diet (62.34 vs. 89.70 ± 3.82 g/d, respectively;  $P < 0.01$ ). The excretion of P per kg of milk yield was higher in cows fed Co-P diet compared with control diet (21.7 and 15.8 ± 1.29 g/kg, respectively;  $P < 0.01$ ). There was no difference between Holstein and Jersey in concentration of P in the feces (1.16 vs. 1.08 ± 0.07%, respectively;  $P = 0.36$ ), digestibility (32.3 versus 29.0 ± 2.83%, respectively;  $P = 0.40$ ) and P efficiency (19.4 vs. 18.0 ± 1.63g/kg, respectively;  $P = 0.55$ ) across treatments. Results of this study suggest that rations formulated containing RFDDGS should be adjusted for P to reduce P excretion by dairy cows.

**Key Words:** phosphorus, excretion, digestibility

**M402 Variability in diets of lactating dairy herds .** Maria P. Turiello\*<sup>1</sup>, Marco Sambataro<sup>1</sup>, Agustin Turiello<sup>1</sup>, Claudina Vissio<sup>1</sup>, and Alejandro Relling<sup>2,3</sup>, <sup>1</sup>*Universidad Nacional de Rio Cuarto, Facultad de Agronomia y Veterinaria, Cordoba, Argentina,* <sup>2</sup>*Universidad Nacional de La Plata, Facultad de Ciencias Veterinarias, Buenos Aires, Argentina,* <sup>3</sup>*IGEVET CCT CONICET, Buenos Aires, Argentina.*

The objective of this study was to determine daily ration and feed bunk variability in physical composition of TMR offered to lactating cows in dairy herds. Four commercial herds (12 pens) in the south of Cordoba province, Argentina, were visited during 3 consecutive days on February. Fresh TMR offered in the morning was sampled to assess particle size distribution with a Penn State Particle Separator. Two samples were taken at the beginning and 2 at the end of the feed bunk to determine TMR variability for each wagon. Daily ration variability was expressed as CV, and calculated by dividing the SD of each particle length % over the 3 d period by the average of them over the same period. The effect of place at the feed bunk on particle size distribution was analyzed with linearized mixed models using the mixed procedure of InfoStat. Place at the feed bunk (beginning vs end) was included as a fix effect. Herd and mixer wagon within herd were included in the model as random effects. The greatest daily CV was found with long and fine particles (17.4 and 17.6% respectively) but all of them were greater than 8%. We observed a great variability and a significant difference between places of TMR samples in the short particle size sieve (Table 1). These results may be indicating heterogeneity on the feed delivered due to procedures of loading and mixing the TMR. It is very important to determine factors of management and infrastructure associated with variability of TMR to improve consistency and then to increase profitability of dairy farms.

**Table 1 (Abstr. M402).** Distribution (mean ± SD) of particles

Particles (%)	Mixer wagon		P-value
	Beginning	End	
Long	17.0 ± 2.84	17.6 ± 2.85	0.52
Medium	34.8 ± 3.80	34.1 ± 3.80	0.34
Short	35.7 ± 2.01	34.4 ± 2.01	0.05
Fine	12.5 ± 2.16	13.9 ± 2.16	0.11

**Key Words:** TMR variability, particle size distribution

**M403 Mineral blood serum status of Holstein cows during the warm and cold seasons.** Pedro Meda-Alducin\*, Maximino Huerta-Bravo, Gustavo De la Torre-López, Baldomero Alarcón-Zúñiga, and Raymundo Rangel-Santos, *Posgrado en Producción Animal, Departamento de Zootecnia, Universidad Autónoma Chapingo, Texcoco, México.*

Heat stress decreases productive and reproductive performance of dairy cows, and it may be related to mineral status. The aim was to quantify serum concentrations of Cu, Zn, Fe, Ca, Mg and K in Holstein cows during warm (WS, ITH = 72) and cold seasons (CS, ITH = 56). Cows (n = 240) with 2 to 6 lactations and 30–60 DIM, milked 3 times per day were selected from 3 commercial dairy farms (F1-F3) in an arid zone of México. Cows were fed similar diets during both seasons, within farm, with a total mixed ration (corn silage, flaked corn, soybean meal, wheat bran, concentrated, alfalfa and wheat hay, molasses and minerals, according to NRC (2001) requirements. Blood samples were obtained from coccygeal vein with Vacutainer tubes, serum was separated by centrifugation at 3000 rpm during 15 min, and stored at -20°C until their analysis. Mineral content was quantified using an atomic absorption spectrophotometer. Data were analyzed using a lineal model with a 2x3 season x farm arrangement. The interaction season x farm effected ( $P < 0.001$ ) all minerals studied. Concentrations of Cu, Zn, K and Mg in blood serum of cows were not dependent on season. However, Fe was higher ( $P < 0.05$ ) during CS compared with WS in the cows from the 3 farms, while the reverse was true for Ca. Overall, Cu, Zn, Fe, Ca, Mg and K concentrations in blood serum were below the normal range in 65, 35, 26, 0, 35 and 38% of cows during the WS, while corresponding values during the CS were 42, 45, 4, 40, 52 and 38%. It is concluded that the dairy cows studied have several mineral problems that depend more of the interaction season x farm than season. It is recommended to study water quality.

**Table 1 (Abstr. M403).** Means of minerals (mg/L) in blood serum of Holstein cows in warm and cold seasons

Season	Farm	Mineral (mg/L)					
		Cu	Zn	Fe	Ca	Mg	K
Warm	1	0.92 <sup>a</sup>	0.94 <sup>a</sup>	1.60 <sup>c</sup>	105.53 <sup>a</sup>	18.23 <sup>b</sup>	173.58 <sup>c</sup>
Cold	1	0.84 <sup>a</sup>	0.69 <sup>a</sup>	1.97 <sup>b</sup>	94.95 <sup>b</sup>	20.30 <sup>a</sup>	234.81 <sup>a</sup>
Warm	2	0.77 <sup>b</sup>	0.80 <sup>a</sup>	1.68 <sup>c</sup>	108.38 <sup>a</sup>	19.02 <sup>b</sup>	194.11 <sup>b</sup>
Cold	2	0.78 <sup>b</sup>	0.92 <sup>a</sup>	2.01 <sup>b</sup>	60.27 <sup>c</sup>	10.96 <sup>c</sup>	128.62 <sup>d</sup>
Warm	3	0.68 <sup>c</sup>	0.87 <sup>a</sup>	1.55 <sup>c</sup>	112.64 <sup>a</sup>	20.49 <sup>a</sup>	126.81 <sup>d</sup>
Cold	3	0.88 <sup>a</sup>	0.89 <sup>a</sup>	2.64 <sup>a</sup>	89.16 <sup>b</sup>	18.36 <sup>b</sup>	222.67 <sup>a</sup>
<i>P</i> > <i>F</i>		<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
SEM <sup>1</sup>		0.03	0.08	0.08	1.83	0.33	3.88
Normal range		0.8–1.5	0.8–1.4	1.3–2.5	80–110	18–30	160–215

<sup>a-d</sup>Means without a common letter in the same column are different.

**Key Words:** mineral diagnosis, heat stress, Holstein cows

**M404 Effects of rumen-protected methionine or choline supplementation on vaginal discharge and uterine cytology of Holstein cows.** Cassandra S. Skenandore\*<sup>1</sup>, Diego A. Velasco Acosta<sup>1,2</sup>, Zheng Zhou<sup>1</sup>, Maria I. Rivelli<sup>1</sup>, Marcio N. Corrêa<sup>2</sup>, Daniel Luchini<sup>3</sup>, and Felipe C. Cardoso<sup>1</sup>, <sup>1</sup>University of Illinois, Urbana, IL, <sup>2</sup>Federal University of Pelotas, Pelotas, Brazil, <sup>3</sup>Adisseo S.A.S., Alpharetta, GA.

Fertility in dairy cows has been declining in recent years. Supplementation with methionine has been shown to improve reproductive health.

Seventy-one pregnant Holstein cows entering their 2nd or greater lactation were fed the same basal diet and randomly assigned to 4 treatments from 21 d before calving to 30 DIM. From -21 d to calving cows were fed a close-up diet. From calving to 30 DIM cows were fed a fresh cow diet, and from 30 to 72 DIM a high cow diet. Treatments were: CON (n = 16, fed the basal diets with a Lys:Met = 3.5:1), MET (n = 20, fed the basal diets + Smartamine M to a Lys:Met = 2.9:1), CHO (n = 16, fed the basal diets + 60 g/d Reashure), and MIX (n = 19; fed the basal diets plus Smartamine M to a Lys:Met = 2.9:1 and 60 g/d Reashure). Starting at d 31 cows were randomly re-assigned to 2 treatments: (CON; n = 36, fed the basal diet with a Lys:Met = 3.4:1) or (SM; n = 36, fed the basal diet + Smartamine M to a Lys:Met = 2.9:1). Cows were evaluated at 4, 7, 10, 13, 15, 17, and 30 d after calving for the presence of secretion by inserting the Metrichick device into the cow's vagina. Sample appearance was scored from 0 to 3 and smell was scored 0 or 3 according to Sheldon et al. (2006), and combined in a final score (S). On 15, 30, and 72 d after calving, the uterine endometrium of all cows was sampled using an endocervical brush (cyto-brush) and streaked onto slides. Each slide was examined and counted by the same person for the presence of endometrial polymorphonuclear (PMN) cells. Statistical analysis was performed using the MIXED procedure of SAS. There was no treatment effect ( $P = 0.16$ ) for S up to 17 DIM. Cows receiving MIX had a lower ( $0.38 \pm 0.3$ ,  $P = 0.03$ ) S at 30 DIM than CON ( $1.15 \pm 0.3$ ), MET ( $1.08 \pm 0.4$ ), or CHO ( $2.11 \pm 0.4$ ). There were no treatment differences ( $P = 0.93$ ) for the percentage of PMN cells at 15 or 30 DIM. At 72 DIM, cows in SM had lower ( $5.33 \pm 3.6\%$ ,  $P = 0.01$ ) PMN cells than CON ( $10.17 \pm 3.6\%$ ). In conclusion, supplementing cows with Smartamine M after 30 DIM seems to have beneficial effect on cows' uterine health.

**Key Words:** methionine, endometritis, PMN

**M405 Milk yield and composition in cows fed calcium salts of polyunsaturated fatty acids of different particle sizes.** Maxime Leduc\*<sup>1,2</sup>, Rachel Gervais<sup>1</sup>, Yoline Lebeuf<sup>1,2</sup>, and P. Yvan Chouinard<sup>1,2</sup>, <sup>1</sup>Université Laval, Québec, QC, Canada, <sup>2</sup>Institut de Nutrition and Functional Foods, Québec, QC, Canada.

Feeding unsaturated fatty acids (FA) as Ca salts (CS) has been proposed as a way to protect ruminal microbes from the adverse effects of dietary oils. However, ruminal dissociation of CS may limit the efficiency of this protection. Industrial processes used to obtain CS of FA lead to the production of commercial feeds with a range of different particle sizes. We hypothesized that feeding CS as large particles will improve their inertness in the rumen and improve animal performance. CS of unsaturated FA were obtained from Virtus Nutrition LLC (Corcoran, CA). On a FA basis, the preparation contained 22.4% 18:1 n-9, 14.7% 18:2 n-6, 31.9% 18:3 n-3. The product was sieved through a 1.9-mm screen, and the retained particles were identified as coarse CS (CCS). The small particles were ground through a 0.864-mm sieve, and identified as fine CS (FCS). A mixture of unprotected FA, as triglycerides, with a composition similar to that of the CS served as control. Eight Holstein cows were used in a 4 x 4 Latin square design. Treatments were: 1) ruminal dosing of unprotected FA (negative control; N-CTL); 2) ruminal dosing of FCS; 3) ruminal dosing of CCS; and 4) abomasal dosing of unprotected FA (positive control; P-CTL). Treatments were adjusted to provide 600 g FA per day, and were offered daily in 2 equal boluses for 14 d, followed by 14-d washout intervals. Pre-planned contrasts were used to compare CCS with i) N-CTL; ii) FCS; and iii) P-CTL. Milk yield was 30.0 kg/d for CCS, and was similar in cows fed N-CTL (29.4 kg/d;  $P = 0.70$ ) or FCS (29.6 kg/d;  $P = 0.82$ ), and lower in cows fed P-CTL (25.8 kg/d;  $P = 0.01$ ). Milk fat content was 3.48% for CCS, and was lower in milk from cows fed N-CTL (3.01%;  $P < 0.01$ ) or FCS



(3.15%;  $P = 0.03$ ), and higher in milk from cows fed P-CTL (3.84%;  $P < 0.01$ ). Concentrations of trans-10, cis-12 18:2 in milk was 0.41 mg/g of fat for CCS, and was higher for N-CTL (0.69 mg/g;  $P < 0.01$ ) or FCS (0.69 mg/g;  $P < 0.01$ ), and was similar for P-CTL (0.35 mg/g;  $P = 0.29$ ). In conclusion, feeding CCS prevented ruminal production of trans-10, cis-12 18:2 and maintained a higher milk fat content as compared with unprotected FA or FCS.

**Key Words:** dairy cow, Ca salt, fatty acid

#### **M406 Using the NRC (2001) model to examine the relationships between predicted supplies of metabolizable Met and Lys and actual yields of milk and milk protein: A subject revisited.**

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We previously reported relationships between predicted supplies of MP, MP-Met, and MP-Lys (NRC, 2001) and yields of milk and milk protein. The original database consisted of model evaluations of 464 diet treatments from experiments published in the *Journal of Dairy Science* (JDS) from 1976 through 2003. Results indicated yields of milk and milk protein were more accurately predicted by supplies of the first limiting AA rather than by supplies of MP. To establish a more robust database, an additional 550 diet treatments from experiments published in JDS in 2004–2014 were evaluated. For the current analysis, the following criteria were imposed:  $NE_L$  allowable milk  $\geq$  MP allowable milk, MP allowable milk within  $\pm 6$  kg of actual milk yield, MP balance between  $-250$  to  $100$  g/d, Lys  $\leq 7.2\%$  of MP, and Met  $\leq 2.4$  of MP. To develop the regression equations for MP-Met, and to help ensure that Met was more limiting than Lys, the Lys/Met ratio in MP had to be  $\geq 3.20/1$ . To generate the regression equations for MP-Lys, the Lys/Met ratio in MP had to be  $\leq 3.15/1$ . It was preferred this ratio be  $\leq 3.00/1$ , but that was too restrictive for the available data set. The PROC REG procedure of SAS was used to generate the regression equations. The resulting regression equations describe the relationship between measured milk yields and MP, MP-Met and MP-Lys supplies: MP ( $n = 455$ ):  $y = 0.01397x + 1.76875$ ,  $R^2 = 0.74$ ; MP-Met ( $n = 260$ ):  $y = 0.84054x - 1.18403$ ,  $R^2 = 0.78$ ; and MP-Lys ( $n = 29$ ):  $y = 0.25722x - 1.48198$ ,  $R^2 = 0.70$ . The equations describing the relationship between milk protein yields and MP, MP-Met and MP-Lys were MP ( $n = 455$ ):  $y = 0.47131x - 67.02527$ ,  $R^2 = 0.82$ ; MP-Met ( $n = 260$ ):  $y = 28.47206x - 178.08057$ ,  $R^2 = 0.86$ ; and MP-Lys ( $n = 29$ ):  $y = 9.07155x - 217.19663$ ,  $R^2 = 0.85$ . In contrast to the previous equations, these are linear in nature and biologically more correct. We conclude these updated Met and Lys regression equations more accurately predict relationships between MP-Met and Lys supplies and milk and milk protein yields, and can be used by NRC users to determine if milk and milk protein yields are potentially being limited by these 2 AA.

**Key Words:** methionine, lysine

**M407 Milk fatty acid profile in cows fed calcium salts of polyunsaturated fatty acids of different particle sizes.** Maxime Leduc<sup>1,2</sup>, Rachel Gervais<sup>\*1</sup>, Yolaine Lebeuf<sup>1,2</sup>, and P. Yvan Chouinard<sup>1,2</sup>, <sup>1</sup>Université Laval, Québec, QC, Canada, <sup>2</sup>Institut of Nutrition and Functional Foods, Québec, QC, Canada.

Feeding unsaturated fatty acids (FA) as Ca salts has been proposed as a way to protect them against ruminal biohydrogenation. However, dissociation of Ca salts in the rumen limits the efficiency of this protection. Industrial processes used to obtain Ca salts of FA lead to the

production of commercial feeds with a range of different particle sizes. We hypothesized that unsaturated FA in large particles are physically protected against ruminal biohydrogenation. Calcium salts of polyunsaturated FA were obtained from Virtus Nutrition LLC (Corcoran, CA). On a FA basis, the preparation contained 22.4% 18:1n-9, 14.7% 18:2n-6, 31.9% 18:3n-3. The product was sieved through a 1.9-mm screen. The retained particles were saved, and identified as coarse Ca salts. The particles less than 1.9-mm were ground through a 0.864-mm sieve, and identified as fine Ca salts. A mixture of unprotected FA, as triglycerides, with a composition similar to that of the Ca-salts served as control. Eight Holstein cows were used in a  $4 \times 4$  Latin square design. Treatments were N-CTL) ruminal dosing of unprotected FA, used as negative control; FCS) ruminal dosing of fine Ca salts; CCS) ruminal dosing of coarse Ca salts; and P-CTL) abomasal dosing of unprotected FA, used as positive control. Treatments were adjusted to provide 600 g FA per day, and were offered in 2 equal boluses at 1000 and 1600h for 14 d, followed by 14-d washout intervals. Pre-planned contrasts were used to compare CCS with i) N-CTL; ii) FCS; and iii) P-CTL. Milk fat content of 18:2n-6 was 25.4 mg/g for CCS, and was lower in cows fed N-CTL (16.5 mg/g;  $P = 0.01$ ) or FCS (15.0 mg/g;  $P < 0.01$ ); and higher in cows fed P-CTL (35.2 mg/g;  $P < 0.01$ ). The concentrations of 18:3n-3 in milk fat was 28.9 mg/g for CCS, and was lower in cows fed N-CTL (7.0 mg/g;  $P = 0.01$ ) or FCS (14.6 mg/g;  $P = 0.08$ , tendency); and higher in cows fed P-CTL (92.2 mg/g;  $P < 0.01$ ). In conclusion, feeding CCS appeared to have partially prevented ruminal biohydrogenation, and increased milk fat content of polyunsaturated FA as compared with dietary unprotected oil or FCS.

**Key Words:** dairy cow, Ca salt, unsaturated fatty acids

**M408 The effect of linseed oil supplementation on rumen microbiota composition in lactating dairy cows.** H. M. Tun<sup>\*1</sup>, E. Khafipour<sup>1</sup>, and C. Benchaar<sup>2</sup>, <sup>1</sup>Department of Animal Science, University of Manitoba, Winnipeg, MB, Canada, <sup>2</sup>Agriculture and Agri-Food Canada, Dairy and Swine Research and Development Centre, Sherbrooke, QC, Canada.

The effect of linseed oil (LO) supplementation to red clover silage (RCS)- or corn silage (CS)-based diets on rumen microbiota composition was studied in 12 rumen-cannulated lactating dairy cows in a  $4 \times 4$  Latin square design (35-d periods) with a  $2 \times 2$  factorial arrangement of treatments. Rumen liquid and solid samples were collected on d 18 of each period. DNA was extracted and V4 region of 16S rRNA was amplified and subjected to illumina paired-end sequencing. In both rumen liquid and solids, the LO supplementation reduced bacterial  $\alpha$ -diversity in CS-based diet but not in RCS ( $P < 0.05$ ). The LO supplementation altered the  $\beta$ -diversity of rumen microbiota in both CS- and RCS-based diets though the magnitude of shift was greater in CS. The LO supplementation reduced the abundances of several bacterial phyla including Actinobacteria, Chloroflexi, Fibrobacteres, Firmicutes, Plantomyces, SR1, Spirochetes and Tenericutes, and increased the abundances of Bacteroidetes, Elusimicrobia, Lentisphaerae, Proteobacteria and Synergistetes in the CS-based diet ( $P < 0.05$ ) but did not change the proportion of the abovementioned phyla in the RCS-based diet. The LO supplementation reduced methanogenic *Methanobrevibacter* in both diets, but the magnitude of depression found to be greater in CS compared with RCS. The LO supplementation increased *Methanospaera* population with a greater magnitude in the CS-based diet compared with RCS ( $P < 0.05$ ). The LO supplementation also negatively affected the proportion of several beneficial rumen bacteria including *Bifidobacteria*, *Fibrobacter* and *Mollicutes* with greater magnitude in the CS-based diet compared with RCS ( $P < 0.05$ ). Data mechanistically explains how

LO supplementation differentially affected CH<sub>4</sub> emission and animal production in CS- vs. RCS-based diets observed in the parallel study to this. The LO supplementation more effectively reduced the CH<sub>4</sub> emission (26%) in CS vs. RCS (9%), which might be due to greater reduction in *Methanobrevibacteres*. Also, LO supplementation reduced DMI, fiber digestion, and yields of milk fat and protein only in the CS-based diet, which might be due to its negative effect on fibrolytic populations.

**Key Words:** linseed oil, rumen microbiota, methane emission

**M409 The effects of linseed oil supplementation on fecal microbiota in lactating dairy cows.** H. M. Tun\*<sup>1</sup>, E. Khafipour<sup>1</sup>, and C. Benchaar<sup>2</sup>, <sup>1</sup>Department of Animal Science, University of Manitoba, Winnipeg, MB, Canada, <sup>2</sup>Agriculture and Agri-Food Canada, Dairy and Swine Research and Development Centre, Sherbrooke, QC, Canada.

The effect of linseed oil (LO) supplementation to red clover silage (RCS)- or corn silage (CS)-based diets on rumen microbiota composition was studied in 12 rumen-cannulated lactating dairy cows in a 4 × 4 Latin square design (35-d periods) with a 2 × 2 factorial arrangement of treatments. Fecal samples were collected on d 24–29 of each period and pooled. DNA was extracted and V4 region of 16S rRNA was amplified and subjected to Illumina paired-end sequencing. DNA was extracted and V4 region of 16S rRNA was amplified and subjected to illumina paired-end sequencing. The feces from cows fed RCS-based diets had greater microbial α-diversity compared with CS, but LO supplementation did not affect α-diversity of microbial community. In contrast, the LO supplementation altered the β-diversity of fecal microbota only in the CS-based diet ( $P = 0.001$ ). Among 17 bacteria phyla found in fecal samples, the LO supplementation reduced both Actinobacteria and Bacteroidetes populations and increased Fimicutes under the CS-based diet ( $P < 0.05$ ). However, under the RCS-based diet, the LO supplementation showed no significant effect on any of bacterial phyla. In CS-based fed cows, LO supplementation suppressed abundances of 2 genera from Bacteriodales family, unclassified RF16 and unclassified Rikenellaceae, as well as the genus SHD231 in Anaerolinaceae family ( $P < 0.05$ ). The 16S rRNA sequencing generated taxa belonged to Euryarchaeota in which no archaeal genera were found to be associated with LO supplementation. Data suggest that LO supplementation only affected fecal microbiota in the CS-based diet and that the source of the basal forage diet influences the effectiveness of LO supplementation and composition of microbiota even in the hindgut of dairy cows.

**Key Words:** linseed oil, fecal microbiota, methane emission

**M410 Effects of acetate, and propionate infusion and pH on VFA production.** Sandip Ghimire\*<sup>1</sup>, Benjamin A. Wenner<sup>2</sup>, Richard A. Kohn<sup>3</sup>, Jeffrey L. Firkins<sup>2</sup>, and Mark D. Hanigan<sup>1</sup>, <sup>1</sup>Virginia Polytechnic Institute and State University, Blacksburg, VA, <sup>2</sup>The Ohio State University, Columbus, OH, <sup>3</sup>The University of Maryland, College Park, MD.

Four continuous culture fermenters were used to determine the effect of varying VFA concentrations and pH on VFA production. The treatments were applied in the fermenters over 4 periods in a 4 × 4 Latin square design. The 4 treatments were: control, 20 mmol/d acetate infusion (INFAC), 7 mmol/d propionate infusion (INFPR), and low pH (LOWPH). For the LOWPH, buffer composition was adjusted to lower pH by 0.5 units compared with control (ranging from 6.62 to 6.97). The fermenters were fed 40 g of a pelleted 50:50 alfalfa: concentrate diet once daily. After 7 d of adjustment, filtered liquid effluent (4 mL) was

sampled at 0, 2, 4, 6, 8, 12, 16, and 22 h after feeding for VFA concentration analysis. Acetate and butyrate production were not affected by treatments. Production of propionate was higher in LOWPH ( $P < 0.05$ ) compared with control and INFPR (Table 1). The hourly production of propionate was not different between treatments ( $P > 0.1$ ) except at 2 h when it was higher in LOWPH compared with INFPR ( $P > 0.05$ ). The acetate to propionate ratio (AP ratio) in INFPR was higher than other treatments ( $P < 0.05$ ). The AP ratio of LOWPH was lower than control. These results reveal that propionate production was increased by low pH, and higher concentration of propionate increased the AP ratio in continuous culture, thus affecting VFA recycling.

**Table 1 (Abstr. M410).** Effect of acetate infusion, propionate infusion, and low pH on VFA production<sup>1</sup>

VFA (mmol/d)	INFAC	INFPR	LOWPH	Control	SE	P-value
Acetate	88	93	89	91	5.2	0.56
Propionate	34.7 <sup>ab</sup>	30.8 <sup>b</sup>	38.4 <sup>a</sup>	33.4 <sup>b</sup>	1.8	0.01
Butyrate	15.3	15.4	14.1	14.7	0.9	0.12
Acetate: Propionate	2.55 <sup>ac</sup>	3.03 <sup>a</sup>	2.32 <sup>c</sup>	2.73 <sup>b</sup>	0.1	<0.01

<sup>1</sup>Production was calculated after subtracting infused VFA from entry rate.

**Key Words:** fermenter, VFA, pH

**M411 Effect of linseed meal on animal performance and oxidative stability of omega 3 enriched milk in Holstein dairy cows.** Daniel E. Rico\*, Rachel Gervais, Lauriane Schwebel, Yolaine Lebeuf, and Yvan Chouinard, Département de Sciences Animales, Université Laval, Quebec, QC, Canada.

Linseed meal antioxidants could help prevent oxidative degradation of omega-3 enriched milk. Six Holstein dairy cows (120 ± 30 DIM, 36.3 ± 6.5 kg milk/d; Mean ± SD) were used in a replicated 3 × 3 Latin Square design (20-d periods; 14 d of adaptation) investigating the effect of linseed meal on animal performance and oxidative stability of omega-3 enriched milk. Linseed oil was abomasally infused continuously to all cows at 243 ± 23 g/d, and dietary treatments were: 1) Linseed meal (16.5% of DM; LS), Canola meal (16.5% of DM) + 7000 units of vitamin E/kg DM (VE), Canola meal (16.5% of DM; CON). Milk yield was recorded and sampled for composition analyses on the last 3 d of each period. Oxidation measurements were done in fresh milk collected on d 17. Data were analyzed using the MIXED procedure of SAS (SAS 9.3, The SAS institute, Cary, NY) including the random effects of period and cow, and the fixed effects of square and treatment. Preplanned contrasts were CON vs. LS and VE vs. LS. Milk yield and fat corrected milk were not affected by treatment and averaged 34.2 ± 2.3 and 32.8 ± 2.3 (mean ± SE), respectively. Milk fat concentration tended to be lower in LS (3.76%) relative to CON (4.00%;  $P = 0.06$ ), but was no different from VE (3.73%). Milk protein concentration was higher in LS (3.38%) compared with CON (3.30%  $P = 0.01$ ), and was not different from VE (3.38%). The concentration of lactose and the yields of fat, protei, and lactose were not different among treatments. The concentration of C18:3 n-3 was not affected by treatment and averaged 5.1 ± 0.79% of total milk fatty acids. Treatments had no effect on the concentration of dissolved oxygen, redox potential or conjugated diene hydroperoxides of fresh milk, which averaged 5.5 ± 0.17 mg/L of milk, 148.5 ± 24.1 mV and 0.92 ± 0.06 mmol/L, respectively. However, VE reduced milk concentrations of the volatile lipid oxidation products propanal, hexanal, and 1-octen-3-one by >73% ( $P < 0.05$ ) and tended to reduce hept-cis-4-enal concentrations by 90% ( $P = 0.06$ ) relative to

LS, whereas there were no differences between LS and CON. Vitamin E may help prevent oxidative degradation of milk moderately enriched in omega-3, whereas linseed meal has no effect.

**Key Words:** dairy cow, omega-3, linseed meal.

**M412 Effect of potassium carbonate and soybean oil supplementation on lactational performance in early-lactating dairy cows fed a high-concentrate diet.** A. Rene Alfonso Avila\*<sup>1</sup>, Edith Charbonneau<sup>1</sup>, P. Yvan Chouinard<sup>1</sup>, Gaëtan F. Tremblay<sup>2</sup>, and Rachel Gervais<sup>1</sup>, <sup>1</sup>Université Laval, Quebec, QC, Canada, <sup>2</sup>Agriculture and Agri-Food Canada, Quebec, QC, Canada.

Research suggests that the decrease in milk fat synthesis observed in dairy cows fed rations high in concentrates or supplemented with vegetable oils could be prevented by increasing dietary cation-anion difference (DCAD) and/or potassium supply. The objective of this study was to evaluate the effect of potassium carbonate (K<sub>2</sub>CO<sub>3</sub>) on lactational performance of early-lactating dairy cows fed diet supplemented with soybean oil (SBO). Eight primiparous and 20 multiparous Holstein cows averaging 39 ± 9 DIM (Mean ± SD) were used in a randomized complete block design (7 blocks) based on DIM and number of calving with a 2 × 2 factorial arrangement of treatments. Within each block, cows were fed a basal diet formulated to achieve 40% forage (58% corn silage) and 60% concentrate (47% non-fibrous carbohydrates), with 0 (DCAD: +95 mEq/kg) or 1.5% K<sub>2</sub>CO<sub>3</sub> (DM basis; DCAD: +316 mEq/kg), and 0 or 2% SBO. Effects of K<sub>2</sub>CO<sub>3</sub>, SBO and their interaction were evaluated. Treatment period was 28 d in length, plus 1 wk pretreatment collection period, used as covariate and the last 5 d used for data and sample collection. Dry matter intake was not affected by treatments (24.8 ± 1.2 kg/d; *P* = 0.56), but milk yield was increased when SBO was added to the diet (41.8 vs 38.6 kg/d; *P* = 0.01). Milk protein content was decreased by SBO (3.02% vs. 3.23%; *P* = 0.01), but a similar milk protein yield was observed among treatments (1.26 ± 0.01 kg/d; *P* = 0.19). Milk fat percentage (3.30 ± 0.07%; *P* = 0.21) was not affected by treatments. Feeding SBO tended to decrease milk fat yield, exclusively when cows were fed a diet without K<sub>2</sub>CO<sub>3</sub> (0% K<sub>2</sub>CO<sub>3</sub>: 1.26 vs. 1.33 kg/d, 1.5% K<sub>2</sub>CO<sub>3</sub>: 1.31 vs. 1.34 kg/d; interaction: *P* = 0.09). 4% fat-corrected milk was increased with K<sub>2</sub>CO<sub>3</sub> supplementation in cows fed SBO diets (37.4 vs. 35.0 kg/d), whereas the opposite effect was observed for cows receiving diets without SBO (35.4 vs. 36.9 kg/d; interaction: *P* = 0.06). Milk urea nitrogen was decreased by SBO (14.2 vs. 16.2 mg/dL; *P* = 0.03) and K<sub>2</sub>CO<sub>3</sub> (13.9 vs. 16.5 mg/dL; *P* = 0.01). In conclusion, the effect of K<sub>2</sub>CO<sub>3</sub> on milk production and composition is affected by dietary unsaturated fatty acid supplementation.

**Key Words:** DCAD, milk fat synthesis, potassium carbonate

**M413 Ratio between plasma sphingolipids reveals acyl-chain specific changes during the transition from pregnancy to lactation in Holstein cows.** Sina Saed Samii\*, J. Eduardo Rico, Alice T. Mathews, and Joseph W. McFadden, West Virginia University, Morgantown, WV.

The ratio between sphingolipids is a means to understand sphingolipid biology, an analysis utilized in biomedicine to study ceramide-mediated insulin resistance. Our objective was to evaluate whether the ratio between plasma sphingolipids in dairy cows is modified during the peripartum. Multiparous Holstein cows were grouped by BCS at d -28 prepartum: lean (BCS 2.91 ± 0.13; n = 7) or overweight (OVER; BCS 4.03 ± 0.21; n = 7). Diets were formulated to meet nutrient requirements. Blood was collected routinely from d -21 to 21. LC/MS was used to

profile 37 Cer, monohexosylceramides (GlcCer), lactosylceramides (LacCer), and SM in plasma. Log-transformed data were analyzed using a mixed model with repeated measures (fixed effects of BCS and day). Nonparametric correlations were analyzed. NEFA mobilization increased during transition, more so in OVER (*P* < 0.01). Ratio of C16:0-SM to C16:0-Cer (C16:0 SM: Cer) increased during transition (*P* < 0.01) and tended to be lower in OVER (*P* < 0.1). C18:0 SM: Cer reached a nadir at calving (*P* < 0.01). In contrast, C24:0 and C22:1 SM: Cer progressively decreased with time (*P* < 0.01). C16:0, C22:0, and C26:0 Cer:GlcCer decreased during transition (*P* < 0.01). C18:0 Cer:GlcCer tended to display a biphasic response (increased then declined; *P* = 0.08). Neither BCS nor day modified C24:0 Cer:GlcCer. C16:0 and C18:0 GlcCer: LacCer increased and decreased, respectively, as calving approached (*P* < 0.01). After calving, C24:0 GlcCer: LacCer declined until d 21 (*P* < 0.05). C18:0 and C24:0 SM: Cer, C16:0 and C20:0 Cer:GlcCer, and C18:0 and C24:1 GlcCer: LacCer were negatively correlated with NEFA (*r* = -48 to -0.22; *P* < 0.01). In contrast, C16:0 GlcCer: LacCer was positively correlated with NEFA (*r* = 0.57; *P* < 0.01). C18:0 and C26:0 Cer:GlcCer were negatively correlated with estimated insulin sensitivity (RQUICKI; *r* = -0.35 to -0.29; *P* < 0.01), whereas, C20:0 Cer:GlcCer and C18:0 GlcCer: LacCer were positively correlated with RQUICKI (*r* = 0.19 to 0.29; *P* < 0.05). Research will need to determine whether changes in plasma fatty acids are related to the ratio between acyl-chain specific sphingolipids.

**Key Words:** ceramide, dairy cow, sphingomyelin

**M414 Effects of feeding protected unsaturated fatty acids (Persia Fat) on Insulin resistance parameters of fresh Iranian Holstein dairy cows.** Hamed Khalilvandi-Behroozyar<sup>1</sup>, Mehdi Dehghan-Banadaky\*<sup>2</sup>, Mohammad Ghaffarzadeh<sup>3</sup>, Kamran Rezayazdi<sup>2</sup>, and Essa Dirandeh<sup>4</sup>, <sup>1</sup>Department of Animal Science, Urmia University, Urmia, West Azerbaijan, Iran, <sup>2</sup>Department of Animal Science, University of Tehran, Karaj, Alborz, Iran, <sup>3</sup>Chemistry and Chemical Engineering Research center of Iran, Tehran, Iran, <sup>4</sup>Department of Animal Science, Sari University of Agriculture and Natural resources, Sari, Mazandaran, Iran.

The onset of insulin resistance (IR) in fresh dairy cows, will promote sparing of glucose, but the ability of insulin to inhibit HSL and suppress NEFA release from adipose may be impaired. The aim of this study was to evaluate the effects of feeding different FA in dairy cows on IR parameters. Twenty-four multiparous Iranian Holstein cows were used -30 d to 50 DIM. Dietary treatments consisted of (1) Prilled Palm (PO) [Energizer RP10, 2 and 2.25% DM in pre- and postpartum]; (2) Ca-salts of sunflower oil [Persia Fat-SO]; (3) Ca-salts of fish oil [Persia Fat-FO] and (4) equal amounts of Persia Fat-FO and Persia Fat-SO (2.2 and 2.5% of dietary DM in pre- and postpartum). Cows were weighed on 2 consecutive days to determine the doses of glucose for IVGTT (20 DIM, by administering 0.25 g/kg BW glucose i.v.) and insulin for insulin challenge (by 0.1 IU/kg BW insulin i.v. followed by saline). Blood samples were collected at -30 to 180 min relative to administration of glucose and 120 min to insulin. The areas under the curve (AUC) of glucose and insulin during IVGTT and IC were calculated using the trapezoidal method. The parameters obtained from IVGTT and IC, were analyzed using PROC MIXED of SAS. The model included the fixed effects of treatment and sequence, and the random effects of period and cow within sequence. A covariate was used to adjust for differences in glucose concentration before the challenges. Statistically significant lower glucose and higher Insulin concentration in PO fed cows, in line with higher TNF- $\alpha$  and glycerol levels can be a hint to lower insulin sensitivity compared with Persia Fat fed animals. Accordingly, PO



impaired glucose and NEFA CR (%/min) during IVGTT and glucose, insulin and NEFA during IC, reflecting lower responsiveness to insulin. The highest and lowest AUC (mg/dL) for glucose, NEFA and insulin during IVGTT, determined for PO and Persia Fat- Fish fed animals, respectively. After IC data support lower response to insulin in the case of glucose, NEFA and Insulin according to lower clearance rate for PO fed animals than Persia Fat. NEFA AUC30 (mEq/L) values were -13817 for PO vs. -16338, -18275 and -19462 for SO, Mix and FO. Overall, data support the idea that feeding rumen protected PUFA can modulate insulin resistance in fresh dairy cows.

**Key Words:** inflammation, palm oil, fish oil

**M415 A novel method to determine rumen biohydrogenation kinetics of alpha-linolenic acid (18:3 n-3).** Michel Baldin\*<sup>1</sup>, Natalie L. Urrutia<sup>1</sup>, Daniel E. Rico<sup>2</sup>, Kelsie Baxter<sup>1</sup>, Yun Ying<sup>1</sup>, and Kevin J. Harvatine<sup>1</sup>, <sup>1</sup>Penn State University, University Park, PA, <sup>2</sup>Université Laval, Québec, QC, Canada.

Biohydrogenation (BH) of unsaturated fatty acids (FA) has been extensively studied in vitro allowing inferences on BH pathways and kinetics. However, BH rates and intermediates formed in vitro may not parallel BH pathways in vivo. The objective was to develop an in vivo method to determine the rate of  $\alpha$ -linolenic acid (18:3 n-3) BH and identify intermediates formed. Eleven rumen cannulated high-producing Holstein cows [40  $\pm$  6 kg milk/d (Mean  $\pm$  SD)] were fed at a rate of 6%/h of expected total DMI a diet balanced to 29% NDF and 5.9% EE (1.5% soybean oil).

A single bolus consisting of 200 g of flaxseed oil (53% 18:3) and 15 g of tridecanoic acid (13:0) was mixed with rumen contents and rumen digesta was collected at -1, 0.1, 0.5, 1, 2, 3, 4, 6 and 8 h relative to the bolus. Samples were immediately placed in dry ice, stored at -20°C, freeze-dried, methylated and analyzed by gas chromatography using 17:1 and 19:0 as internal standards. Data were first analyzed using PROC Mixed with repeated measures for time point comparison. Second, the disappearance of 13:0 and 18:3 was fit to a single exponential decay model using the nonlinear procedure of JMP Pro. The bolus increased total fat in the rumen from 4.3 to 6.0% and enriched 13:0 concentration from 0.04 to 2.2% of FA and 18:3 concentration from 2.0 to 11.3% of FA. The fractional rate of disappearance of 13:0 was 0.4%/min ( $r^2 = 0.98$ ) and of 18:3 was 2.5%/min ( $r^2 = 0.99$ ), with 18:3 reaching pre-bolus concentration within 4 h. Assuming that 13:0 disappeared only by passage, 18:3 disappeared by passage and biohydrogenation, and the rate of passage of 13:0 and 18:3 are the same, the extent of bolused 18:3 BH was 85%. The concentration of *cis*-9,*trans*-11,*cis*-15 18:3 peaked at 1.2% of FA at 1 h (8-fold increase), *trans*-11,*cis*-15 18:2 peaked at 3.9% of FA at 2 h (13-fold increase), and *trans*-11 18:1 peaked at 6.6% FA at 3 h (43% increase). The in vivo method resulted in the expected extent of biohydrogenation and biohydrogenation intermediates, but the rate of ruminal biohydrogenation of 18:3 was much higher than that commonly observed in vitro.

**Key Words:** biohydrogenation, fatty acid

# Ruminant Nutrition: General I

**M416 Samples dried with commercial dry matter techniques differ in volatile compound contents.** Donald Meyer<sup>\*1</sup>, Lynn Nagengast<sup>1</sup>, Dustin Sawyer<sup>1</sup>, and John Goesser<sup>1,2</sup>, <sup>1</sup>Rock River Laboratory, Watertown, WI, <sup>2</sup>University of Wisconsin-Madison, Madison, WI.

On-farm feed dry matter techniques determine DM by difference between original feed weight and a dried weight. Oven DM techniques, though, have been documented to volatilize more than water, leading to underestimated feed DM measures. The objective of our work was to evaluate if feeds, dried through commercially adopted DM techniques, differ in total volatile compound content relative to undried. Corn (n = 14), grass (n = 5), legume (n = 15) and small grain silages (n = 14) were collected, divided into equal subsamples using a riffle-splitter, vacuum-sealed, and frozen until analyzed. Subsamples were thawed and then handled according to 5 different drying treatments; undried (CTL), on-farm type forced-air oven dry, 60 min (KOS), 50C for 48h forced-air oven dry (OV), freeze-dry (FD) or sequential microwave-NIR (LAB). Following treatment, and to assess DM technique non-water losses, undried and corresponding dried samples were analyzed for volatile fermentation products by HPLC (lactic, acetic, propionic, butyric, succinic, and formic acids, and ethanol). Each constituent was expressed as a % of DM, using sequential microwave-3 h 105°C oven dry as a standard DM. Fermentation products were then summed to determine total volatile compounds (TV). Technique difference from CTL represents significant non-water loss and a DM measure error. The resulting data were not normally distributed and were log-transformed before being evaluated using the Fit Model procedure in SAS JMPv11.0. Feed, drying treatment and their interaction were treated as fixed effects and assessed using backward-elimination. Significance was declared at  $P < 0.05$ . Feed type and drying treatment were significantly related to TV. Results presented here are converted back to % of DM. The TV means were compared using Tukey's test, finding legume and corn silage (5.1 and 4.9) differed from small grain (3.6), which differed grass silage (1.80). The TV was the greatest for CTL (4.43) and was not significantly different from LAB, KOS, or FD (4.18, 3.67, and 3.13, respectively). The CTL differed ( $P < 0.05$ ) from OV (2.7) while KOS and FD did not differ from OV. Results suggest OV underestimates feed DM.

**Key Words:** silage, dry matter, volatiles

**M417 Comparison of in situ and in vitro methods for predicting in vivo fiber digestion.** David E. Cook<sup>\*1</sup>, John P. Goesser<sup>1,2</sup>, Lynn Nagengast<sup>2</sup>, and David K. Combs<sup>1</sup>, <sup>1</sup>Department of Dairy Science, University of Wisconsin-Madison, Madison, WI, <sup>2</sup>Rock River Laboratory Inc., Watertown, WI.

Our objectives were to compare potentially digestible NDF (pdNDF) and pdNDF digestion rate ( $k_d$ ), using in situ (IS) or traditional in vitro (TR) assays, and compare subsequent estimates of total-tract NDF digestibility (TTNDFD) to in vivo (IV) TTNDFD measurements. High and low digestible corn silages (HDCS and LDCS) and alfalfa silage (AS) previously characterized for pdNDF,  $k_d$  and TTNDFD by in vivo analysis were utilized. For the IS and TR analyses, samples were incubated for 6, 12, 24, 30, 48, 72, 96, 120, and 240h. For TR, 0.5g dried, 1mm ground feed was weighed into flasks and digested with Van Soest buffer. Rumen fluid was collected from 2 cannulated, lactating cows consuming a high forage diet. Rumen fluid was held under CO<sub>2</sub>, processed and used to immediately inoculate samples. For

IS, 0.5g dried, 2mm ground feed was weighed in Ankom F57 bags. TR samples were digested in each of 2 separate runs and IS in each of 3 cannulated, lactating cows consuming a high forage diet. The NDFD data from both methods were modeled using nonlinear option of SAS JMP (v11.0) to determine pdNDF and associated  $k_d$  for each digestion method. The  $k_d$  (%/h) and concentration of pdNDF (% of NDF), using IS were 2.39 and 88.4 for HDCS, 2.24 and 83.5 for LDCS, and 5.54 and 65.5 for AS, respectively. The  $k_d$  and pdNDF using TR were 4.64 and 74.93 for HDCS, 9.82 and 56.29 for LDCS, and 5.28 and 69.84 for AS, respectively. Total-tract NDF digestibility (TTNDFD) was predicted using the following equation:  $\text{pdNDF}(k_d/(k_d+k_p))$ . The  $k_p$  (2.67%/h) was derived from a meta analysis of in vivo passage rates for pdNDF. HDCS, LDCS, and AS TTNDFD were 46.4, 42.3, and 49.2 for IS; and 52.8, 51.5, and 49.2 for TR. Weighted averages of the feed TTNDFD values were used to predict TMR TTNDFD for comparison with in vivo observations. Resulting TMR TTNDFD were then compared across techniques using a linear model within JMP. Main effects were IS, TR and IV. Means for each technique were compared using student's *t*-test. The TTNDFD determined from TR (50.7) differed ( $P < 0.01$ ) from IS (46.3) and IV (43.6). Estimates of total-tract NDF digestion based on TR,  $k_d$ , and pdNDF overestimated TTNDFD measured in vivo.

**Key Words:** total tract, NDF, digestion

**M418 Response to iso-alpha acids from *Humulus lupulus* (hops) extract on fermentation by rumen microbes in continuous culture fermenters.** Isaac J. Salfer<sup>\*</sup>, Samuel W. Fessenden, and Marshall D. Stern, University of Minnesota, St. Paul, MN.

Iso- $\alpha$  acids from hops (*Humulus lupulus*) derived from the brewing industry have been shown to exhibit bacteriostatic properties against gram-positive bacteria. Previous research using whole or ground hops has shown promising results for decreasing hyper-ammonia producing bacteria in the rumen. However, hops contain additional fermentable substrate and other metabolites including tannins,  $\beta$ -acids and xanthohumol that confound the direct effects of iso- $\alpha$ -acids on rumen fermentation. Research using strictly iso- $\alpha$  acids in rumen culture is limited. The objective of this study was to examine the direct effects of iso- $\alpha$  acids on fermentation by rumen microbes using a dual-flow continuous culture system. Eight fermenters were used in 2 consecutive 10-d periods consisting of 7 d of adaptation followed by 3 d of sampling. Fermenters were provided with a basal diet consisting of 44% corn silage, 14% alfalfa hay, 13% ground corn, 11% protein mix, 10% corn gluten feed, 5% cottonseed and 3% liquid vitamin and mineral supplements on a DM basis. This diet provided substrate for ruminal microbes maintained in continuous culture at a rate of 75 g of DM/L of fermenter volume/day. Iso- $\alpha$  Extract (IE) solution was added to the artificial saliva buffer to supply 0 (CON), 600 (LOW), 1200 (MED) and 1800 (HIGH) mg of IE/kg of diet DM/day. There was no effect ( $P > 0.05$ ) on DM, OM, NDF or ADF digestion (%). Volatile fatty acid (VFA) metabolism was not affected by IE treatment ( $P > 0.05$ ), with total VFA concentrations of 105.5, 93.4, 87.9 and 103.6 mM for the CON, LOW, MED and HIGH treatments, respectively. Similarly, N metabolism was not affected ( $P > 0.05$ ) by IE level, with the CON, LOW MED, and HIGH treatments resulting in nitrogen concentrations of 7.4, 5.3, 7.6 and 6.8 mg N/dL of rumen fluid, respectively. No significant effects ( $P > 0.05$ ) on fermenter pH were observed. In conclusion, administration of IE had no effects

on measurements of fermentation by ruminal microbes maintained in continuous culture fermenters.

**Key Words:** rumen, continuous culture, hops

**M419 Sodium salicylate depresses fermentation by ruminal microbes in vitro.** Abigail J. Carpenter\*, Claudio F. Vargas-Rodriguez, Jacob A. B. Jantz, and Barry J. Bradford, *Kansas State University, Manhattan, KS.*

The administration of the anti-inflammatory medication sodium salicylate (SS) after calving has been shown to increase whole-lactation milk production in multiparous cows; however, treatment with SS is associated with hypoglycemia following its administration in some circumstances. We hypothesized that decreased glucogenic substrate supply from fermentation may contribute to decreased blood glucose concentrations in SS-treated cattle. We performed a 24-h batch culture experiment to determine the effects of SS on rumen microorganisms in vitro. Strained and pooled fluid from 3 heifers was combined in a 2:1 ratio with McDougall's buffer, and 150 mL of the inoculum was added to each flask (n = 5/treatment). Blank flasks (n = 5) contained inoculum alone, while each treated and control flask contained 2.5 g of fermentation substrate. Before inoculum was added to the flasks, 1 mL of premixed treatment mixtures were added to achieve the desired final amount of SS (CON = 0 mg, LOW = 125 mg, MED = 250 mg, HI = 375 mg). Gas production was measured with the ANKOM<sup>RF</sup> Gas Production System. Dry matter disappearance (DMD) was significantly depressed by inclusion of SS ( $P < 0.05$ ), with HI having a lower DMD than LOW ( $P < 0.05$ ), and MED intermediate (CON = 48%, LOW = 37%, MED = 30%, HI = 23% of DM; SEM = 2%). Final pH was not different between LOW and CON, but MED and HI had higher final pH than CON (CON = 6.31, LOW = 6.36, MED = 6.42, HI = 6.45; SEM = 0.01,  $P < 0.05$ ). No differences were observed due to treatment for volume, rate, or lag in gas production ( $P \geq 0.28$ ). These results indicate that SS may have an inhibitory effect on rumen microorganisms in vitro, which is counterintuitive considering previous findings that SS administration in early lactation increases 305-d milk production, but consistent with previous observations of decreased blood glucose concentrations in SS-treated cows.

**Key Words:** sodium salicylate, fermentation, rumen modification

**M420 Comparison of different four methods for determining in vitro digestibility of annual ryegrass.** Mariano Alende\*<sup>1,2</sup>, Louisa Bowen<sup>1</sup>, Prabha Ranasinghe<sup>1</sup>, Gabriela Volpi-Lagrecia<sup>1,2</sup>, Gustavo Lascano<sup>1</sup>, and John Andrae<sup>1</sup>, *<sup>1</sup>Clemson University, Clemson, SC, <sup>2</sup>INTA, Anguil, Argentina.*

Multiple in vitro methods have been developed to assess forage digestibility but little is known regarding agreement in disappearance among them. This study compared 3 different rumen in vitro apparent digestibility (AD) methods (Daisy<sup>II</sup> [D], Batch Culture [BC] and the Ankom Gas Production System [G]) at 4 incubation times ([IT]; 12, 24, 36 and 48 h). Additionally, results obtained at 24 h were compared with those obtained from dual-flow continuous fermenters [CF]. Annual ryegrass (*Lolium multiflorum*; 33.8% NDF, 16.6% CP, 91.1% OM, 27.3% WSC) was clipped from an ungrazed pasture, dried (<60°C) and ground in a Wiley mill (1 mm). Three runs of each method were conducted using rumen fluid from a cannulated Holstein cow in mid-lactation fed a 34% corn silage, 6% grass hay and 60% grain mix diet. Ankom F57 acetone pre-rinsed bags containing  $0.5 \pm 0.01$  g of sample were used for D, BC and G. Digestibility coefficients in CF were estimated in 3 periods (7-d

adaptation, 3-d collection) ran simultaneously with the other methods. The same buffer was used for all 4 methods. Data were analyzed using Mixed procedure of SAS in a model including method, IT and period as a random factor, with IT as repeated measure. Means within each IT were compared by PDIF function. Results are presented in Table 1. Results indicate that D predicts higher AD than G and BC at IT greater than 12 h. Digestibility estimated using CF was similar to the obtained with BC and G at 24 h. We conclude that different in vitro digestibility methods could yield different results.

**Table 1 (Abstr. M422).** Dry matter digestibility (%) data of annual ryegrass samples assessed by 4 in vitro methods at 4 incubation times

In vitro method	Incubation time (h)			
	12	24	36	48
Daisy <sup>II</sup>	58.35	70.92 <sup>a</sup>	75.93 <sup>a</sup>	79.85 <sup>a</sup>
Batch culture	52.88	61.29 <sup>b</sup>	68.14 <sup>b</sup>	71.82 <sup>b</sup>
Gas Production System	51.23	58.89 <sup>b</sup>	64.01 <sup>b</sup>	66.44 <sup>b</sup>
Continuous fermenter		58.97 <sup>b</sup>		

<sup>a,b</sup>Means within each column with a different letter indicate significant differences ( $P < 0.05$ ).

**Key Words:** rumen in vitro DMD, methods comparison

**M421 The effect of dietary inclusion of sugar and type of sugar on ruminal short-chain fatty acid and glucose uptake across the ovine ruminal epithelium.** Katie M. Wood\*<sup>1</sup>, Christine L. Rosser<sup>1</sup>, Matthew E. Walpole<sup>1</sup>, Rodrigo Kanafany Guzmán<sup>1</sup>, Beth Mason<sup>2</sup>, Timothy Mutsvangwa<sup>1</sup>, and Gregory B. Penner<sup>1</sup>, *<sup>1</sup>Dept of Animal and Poultry Science, University of Saskatchewan, Saskatoon, SK, Canada, <sup>2</sup>Saputo Dairy Products Canada Ltd., Saint-Léonard, QC, Canada.*

The objective was to determine whether sugar inclusion and the type of sugar (lactose; LAC or sucrose; SUC) as a replacement for starch influences ruminal glucose and short-chain fatty acid (SCFA) uptake by the rumen epithelium. Eighteen Suffolk × Arcott wethers were randomly assigned to 1 of 3 diets: control (CON), LAC (whey permeate), and SUC (molasses). Sugar content was 6% of DM for LAC and SUC and partially replaced barley grain ensuring NSC was balanced among treatments. After 31 d, lambs were killed and the caudal-ventral blind sac was collected and washed using a pre-heated buffer solution. The tissue was then stripped of the underlying muscular layer and mounted in Ussing chambers under short-circuit conditions. The uptake of <sup>14</sup>C-butyrate, <sup>14</sup>C-propionate and <sup>3</sup>H-acetate under non-inhibited conditions and conditions that inhibit protein-mediated uptake were measured. The uptake of <sup>14</sup>C-glucose was evaluated without inhibition and when phlorizin (0.5 mM; inhibitor of sodium-linked glucose transporter; SGLT-1), or phloretin (0.5 mM; inhibitor of facilitated glucose transporters) were included. Data were analyzed using PROC MIXED in SAS and contrasts used to evaluate the effect of sugar (CON vs. sugar) and type (LAC vs. SUC). Acetate uptake was reduced ( $P = 0.04$ ) for lambs fed sugar due to a reduction in passive diffusion ( $P = 0.02$ ), whereas protein-mediated uptake did not differ ( $P = 0.24$ ). Propionate uptake was not affected by sugar inclusion; however, lambs fed LAC tended ( $P = 0.098$ ) to have greater propionate uptake and had greater ( $P = 0.043$ ) protein-mediated propionate uptake than those fed SUC. Lambs fed LAC tended ( $P = 0.10$ ) to have greater protein-mediated and passive uptake of butyrate than those fed SUC. Glucose uptake was increased 1.7 times for lambs fed sugar compared with those fed CON ( $P = 0.012$ ). The SGLT-1 uptake tended to be greater for lambs fed sugar than those fed CON ( $P = 0.09$ ).



The results of this study indicate that the type of sugar included in diets may influence SCFA and glucose uptake by the rumen epithelium.

**Key Words:** rumen, absorption, glucose

**M422 Evaluation of three rumen-protected lysine sources produced in two different batches using a modified three-step in vitro procedure.** Haley E. Larson<sup>\*1</sup>, Izuru Shinzato<sup>2</sup>, Makoto Miura<sup>3</sup>, Samuel W. Fessenden<sup>1</sup>, Isaac J. Salfer<sup>1</sup>, and Marshall D. Stern<sup>1</sup>, <sup>1</sup>University of Minnesota, Saint Paul, MN, <sup>2</sup>Ajinomoto Heartland, Inc., Chicago, IL, <sup>3</sup>Research Institute for Bioscience Products & Fine Chemicals, Ajinomoto Co., Inc., Kawasaki, Japan.

A standardized 3-step in vitro procedure for evaluating rumen-protected lysine (RPL) sources was modified by Ajinomoto Co. Inc. and the University of Minnesota to estimate bioavailability of lysine within RPL products. The objective of this study was to evaluate variation between batch and source of RPL products AjiPro-L (A) and 2 other commercial products (B and C). Each RPL product was replicated 3 times in 3 runs to evaluate consistency of results within and between runs using the procedure. One gram of product from each RPL source, taken from one of 2 batches (2013, 2014) was individually weighed into a Dacron polyester bag. Bags were incubated in media bottles in a shaking water bath at 39°C for 30 h. Simulation of ruminal (20 h), abomasal (2 h) and intestinal (8 h) digestion was accomplished by immersing solutions at 20, 22 and 30 h. At each time point, a 10-mL aliquot of buffer was collected and analyzed for lysine concentration using a Bioflow BF-7 biosensor (Oji Scientific Instruments Co., Ltd., Japan). Statistical differences ( $P < 0.05$ ) were identified using a one-way ANOVA. Differences ( $P < 0.001$ ) were observed between the 3 product sources for rumen insoluble lysine (RIL) and abomasal insoluble lysine (AIL). Intestinally available lysine (IAL) for product A differed from B and C ( $P < 0.001$ ). Results showed variation between 2013 and 2014 batches of product sources A and B for RIL and IAL, and product sources B and C for AIL ( $P < 0.001$ ). The release efficiency (RE) of RIL for each product source differed between years ( $P < 0.001$ ). Within each run of the procedure, there was no variation observed between replicate samples ( $P > 0.05$ ). Variation was observed between runs for RIL ( $P < 0.002$ ), AIL ( $P < 0.001$ ) and RE ( $P < 0.03$ ), with no variation between IAL ( $P > 0.05$ ). The modified 3-step procedure demonstrated differences between product types and batches within the same product source. The procedure was consistent between replicates of a run demonstrating its ability to compare differences in bioavailability of RPL products.

**Key Words:** in vitro, rumen-protected lysine

**M423 Duodenal infusion of casein but not glutamic acid increases nitrogen retention in cattle provided continuous duodenal infusion of cornstarch.** Ethan J. Blom<sup>\*1</sup>, Derek W. Brake<sup>1</sup>, and David E. Anderson<sup>2</sup>, <sup>1</sup>South Dakota State University, Brookings, SD, <sup>2</sup>University of Tennessee Knoxville, Knoxville, TN.

Our objective was to quantify effects of increases among small intestinal starch digestion in response to greater postruminal flows of glutamic acid (Glu) on N retention. Five steers (351 ± 11 kg BW) were fed (5.1 ± 0.06 kg DM/d) a soybean hull-based diet formulated to provide adequate ruminally available N, moderate amounts of RUP, and small amounts of starch (0.8% DM). Cattle were placed in a 5 × 5 Latin square with 12-d periods. Cattle received (DM basis) continuous duodenal infusion of raw cornstarch (1.5 ± 0.08 kg/d), and either 0 (control), 30.9 ± 0.59, 62.4 ± 1.16, 120.4 ± 3.39 g/d Glu or 407 ± 18.3 g/d casein (a positive control). Data were analyzed with Mixed procedures of SAS; linear and

quadratic effects of Glu were determined and the positive control was compared with the negative control by a *t* test. Nitrogen intake from feed was not different ( $P \geq 0.23$ ). Infusate N increased from 0 to 13 g/d with greater amounts of Glu (Linear  $< 0.01$ ) and casein provided 61 g N/d ( $P < 0.01$ ). Similarly, total N intake was greater when cattle were provided casein ( $P < 0.01$ ), but was not affected by Glu (Linear = 0.75). Increases in postruminal Glu did not affect urinary N excretion ( $P \geq 0.30$ ), but casein increased urinary N excretion ( $P < 0.01$ ). Fecal N excretion was not different ( $P \geq 0.55$ ) despite reduced ileal flow of starch in response to greater postruminal flows of Glu (Linear = 0.04) or casein ( $P = 0.07$ ). Glutamic acid had little impact on N retention ( $P \geq 0.95$ ) despite increases in small intestinal starch digestion (Linear = 0.02), but casein increased N retention ( $P < 0.01$ ). It is possible that increases in energy available for gain from increased small intestinal starch digestion in response to greater postruminal flow of Glu are used for purposes other than protein gain; however, it is likely that increases in energy available for gain exceeded capabilities for N deposition under conditions of our experimental model, because N retained as a proportion of N intake was not different when cattle were provided Glu ( $P \geq 0.83$ ) or casein ( $P = 0.38$ ).

**Key Words:** nitrogen retention, glutamic acid, cattle

**M424 Rumen fermentation responses to phytogetic medicinal oils.** Barbara J. M. Lemos<sup>\*</sup>, Flavia M. Souza, Edemilson C. Conceição, Victor R. M. Couto, and Juliano J. R. Fernandes, Universidade Federal de Goiás, Goiania, Goiás, Brazil.

The effects of oils extracted from the trunk of *Copaifera langsdorffii* (Copaiba; 21.3% β-caryophyllene, a essential oil), and from the fruit of *Pterodon emarginatus* (Sucupira; 5.3% β-caryophyllene) on rumen fermentation were evaluated in 2 independent in vitro experiments at doses CONTROL, LOW, MEDIUM and HIGH (0, 30, 300, and 3000 mg/L of buffered rumen fluid, respectively). The oils were dissolved in 2.5 mL of ethanol, also added to CONTROL. The incubation was repeated 4 times in each experiment. The diet (50:50 concentrate:roughage ratio, 90% DM, 20% CP, 2.2% EE, 36% NDF) was ground (1 mm) and incubated using filter bags for 3, 6, 12, 24, 48, 72 and 96 h. Rumen fluid was collected from a cannulated Holstein steer fed the same concentrate:roughage ratio diet. Samples of culture fluid collected after 96 h were analyzed for N-NH<sub>3</sub> and VFA concentrations. The disappearance data of DM were used to calculate the degradability of DM (Orskov et al., 1980). The HIGH dose of *C. langsdorffii* oil, showed degradability of parameter b (potential degradability of the DM assumed to be degraded over time) 29% lower than CONTROL (63.4 vs 45.3%;  $P = 0.002$ ) and the potential degradability (a + b, amount of DM which can be degraded within the rumen given sufficient time) was 15% lower than CONTROL (87.9 vs 75.2%;  $P = 0.001$ ). No responses of *C. langsdorffii* oil was observed ( $P > 0.05$ ) on effective degradability (at rates of passage of 5 and 8%/h) and on constituents of culture fluid (N-NH<sub>3</sub>, total VFA, acetate, propionate, isobutirate, butirate, isovalerate, valerate and acetate:propionate ratio). In the *P. emarginatus* oil study, the response on parameter b was 45% lower for HIGH dose compared with CONTROL dose (60.4 vs 32.1%;  $P < 0.001$ ). The potential degradability was affected by *P. emarginatus* doses ( $P = 0.013$ ), it was similar to CONTROL and LOW doses (highest values, 83.0 and 83.4%, respectively), intermediate to MEDIUM (73.3%) and lower to HIGH dose (64.8%; 22% lower than CONTROL dose,  $P = 0.023$ ). There were no effects ( $P > 0.05$ ) of *P. emarginatus* oil on effective degradability and constituents of culture fluid. In conclusion, these phytogetic oils were able to modify microbial fermentation in the rumen environment.

**Key Words:** degradability, feed additive, in vitro

#### M425 Effect of thyme (*Thymus vulgaris*) and peppermint (*Mentha piperita*) on digestibility of a finishing diet in lambs.

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This experiment was conducted to study the effect of addition of 2 medicinal plants, peppermint or thyme, on digestibility of a finishing diet in lambs. Eighteen male lambs (initial body weight  $21.6 \pm 1.5$  kg) were randomly assigned to one of 3 diets: control (with no additive), peppermint (control diet plus 3% of peppermint) or thyme (control diet plus 3% of thyme), for a 90-d finishing period. Lambs were fed individually and digestibility of nutrients were determined using insoluble ash method during 5 d of fecal sampling from all lambs. The ratio of concentrate:forage was 70:30. Diets were isocaloric and isonitrogenous. Data were analyzed as a completely randomized design with means compared using Duncan's test (SAS v9.2). Addition of peppermint or thyme to diets had no effect on digestibility of CP, ether extract, NDF and ADF but improved DM intake and digestibility of calcium and phosphorus ( $P < 0.05$ ). Supplementation with peppermint or thyme could increase performance of finishing lambs through improved feed intake. Results are given in Table 1.

**Table 1 (Abstr. M425).** Effect of peppermint and thyme on DMI (g/d) and in vivo nutrient digestibility (%) of a finishing diet in lambs

Item	Peppermint	Thyme	Control	SEM	P-value
DM intake	1,494.4 <sup>a</sup>	1,488.5 <sup>a</sup>	1,185.5 <sup>b</sup>	40.6	0.001
Nutrient digestibility					
Crude protein	77.65	75.17	76.96	0.667	1.14
Ether extract	77.93	75.43	76.58	0.074	1.28
NDF	58.04	59.50	65.19	0.424	2.20
ADF	49.76	45.96	50.13	0.183	2.56
Calcium	53.98 <sup>a</sup>	44.21 <sup>a</sup>	37.30 <sup>b</sup>	2.99	0.05
Phosphorus	66.65 <sup>a</sup>	53.61 <sup>a</sup>	50.66 <sup>b</sup>	2.99	0.03

**Key Words:** Sanjabi sheep, peppermint, thyme

#### M426 Abomasal infusion of glucose increases intramuscular lipid content and acetate incorporation into fatty acids in subcutaneous adipose tissue relative to ruminal acetate infusion.

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We hypothesized that abomasal infusion of glucose, which would provide more glucose for absorption from the small intestine, would promote intramuscular (i.m.) adipose tissue development to a greater extent than ruminal infusion of acetate, propionate, or glucose. At 22 mo of age, Angus crossbred steers ( $n = 24$ ) were fitted with ruminal cannulas and adapted to a standard, corn/sorghum finishing diet over a 2-wk period. Subsequently, the steers were infused with isocaloric amounts (3.76 Mcal/d) of glucose, propionate, or acetate. Glucose was infused either into the rumen (control group) or into the abomasum, whereas propionate and acetate were infused into the rumen. Relative to abomasal glucose infusion, acetate infusion decreased DM, OM, DE, and GE intake ( $P \leq 0.05$ ). USDA marbling scores were greater in acetate-infused steers than in than in ruminal glucose-infused steers ( $P = 0.04$ ) and abomasal glucose-infused steers ( $P = 0.08$ ). Ruminal propionate-infused steers had lower subcutaneous (s.c.) fat thickness over the 12th thoracic rib ( $P \leq 0.10$ ) and lower USDA yield grades ( $P \leq 0.05$ ) than

ruminal acetate-infused steers and ruminal glucose-infused steers ( $P = 0.05$ ). The lowest proportions of palmitic acid and palmitoleic acid in s.c. adipose tissue was observed in ruminal glucose-infused steers. Acetate infusion decreased the lipid content of i.m. adipose tissue ( $P = 0.09$ ) and decreased the in vitro incorporation of acetate into fatty acids in s.c. adipose tissue relative to abomasal glucose infusion, ruminal glucose infusion, or propionate infusion ( $P \leq 0.02$ ). Carcass data, lipogenesis, and fatty acid composition were analyzed using the General linear mixed models of SPSS statistics. The rate of glucose incorporation into fatty acids was greater in i.m. adipose tissue of propionate-infused steers than in abomasal or ruminal glucose-infused steers ( $P \leq 0.07$ ). In summary, abomasal infusion of glucose did not promote higher marbling scores but did cause the greatest amounts of i.m. adipose tissue lipid, whereas propionate infusion promoted the greatest rates of fatty acid synthesis from glucose in i.m. adipose tissue.

**Key Words:** glucose, beef quality, lipogenesis

#### M427 Effects of increased inclusion of algae meal with differing fatty acid profiles on lamb total-tract digestibility.

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Heterotrophic microalgae can be manipulated to contain specific fatty acids; however, it is unknown how these differences may affect nutrient digestibility in ruminants. Algae meal (ALG) contains delipidated algae (57% DM basis) and soyhulls (43%). To determine the impact of replacing corn with ALG of differing fatty acid profiles, high oleic acid (HE) or high lauric acid (HL), on nutrient digestibility, 10 whiteface wethers ( $27.9 \pm 0.87$  kg) were used in a replicated  $5 \times 5$  Latin square. There were 5 periods, with 10 d of adaptation and 5 d of fecal and urine collection. Sheep ( $n = 2$  sheep·diet<sup>-1</sup>·period<sup>-1</sup>) received one of 5 finishing diets containing corn, 35% corn silage, 5% hay, 10% soybean meal, and 5% micro ingredients: a corn-based control (CON), 18% HE ALG (18HE), 36% HE ALG (36HE), 18% HL ALG (18HL), and 36% HL ALG (36HL). Both the HE and HL ALG were similar in nutrient composition except for fat (15.8 and 8.19%, respectively) and K (1.6 and 5.4%, respectively). Pooled SEM and LSMEANS are reported. Intake of DM linearly ( $P < 0.01$ ) increased as either ALG increased in the diet; however, DMI was less ( $P < 0.01$ ) for 18HE-fed lambs vs. 18HL-fed lambs. Urine output linearly ( $P < 0.001$ ) increased as ALG increased in the HL-fed lambs and was less ( $P < 0.001$ ) for 36HE-fed lambs vs. 36HL-fed lambs. Digestibility of DM linearly ( $P \leq 0.04$ ) decreased as both HE and HL ALG increased in the diet (75.2, 73.9, 70.1, 73.2, and  $72.7 \pm 0.82\%$  for CON, 18HE, 36HE, 18HL, and 36HL, respectively). Lambs fed 36HE had less ( $P = 0.03$ ) DM digestibility than those fed 36HL. Digestibility of NDF linearly ( $P = 0.04$ ) increased in lambs fed HL diets (47.7, 51.6,  $54.5 \pm 2.18$  for CON, 18HL, and 36HL, respectively). Fat digestibility linearly ( $P \leq 0.02$ ) increased as either ALG increased in the diet; however, digestibility was less ( $P < 0.001$ ) for 36HE-fed lambs than 36HL-fed lambs. Both ALG are highly digestible and could serve as viable feedstuffs in feedlot diets. Due to varying inclusions of fat and K within the strains of algae it was difficult to ascertain if the fatty acid profile had any effect on nutrient digestibility.

**Key Words:** algae, digestibility, sheep

**M428 Urine pH, serum calcium, and dry matter intake evaluated in Jersey cows fed anionic salts or Animate.** Tyler J. Schell<sup>\*1</sup>, Shelby A. Armstrong<sup>1</sup>, Derek J. McLean<sup>1</sup>, Ken P. Zanzalari<sup>1</sup>, James D. Chapman<sup>1</sup>, and Lane O. Ely<sup>2</sup>, <sup>1</sup>Phibro Animal Health Corporation, Quincy, IL, <sup>2</sup>University of Georgia, Athens, GA.

Negative DCAD diets have been proven beneficial for reducing incidence of periparturient diseases in dairy cows, however maintaining dry matter intakes (DMI) have been an issue with these strategies. Twelve non-pregnant, nonlactating Jersey cows (464 kg ± 19 kg BW) were used in a crossover design study to evaluate the effect of diets fully acidified (-15 mEq/100g DM) with either an anionic salt mix (SM) or Animate (AN) on urine pH, serum calcium and DMI when fed continuously for 21 or 24 d. Cows were randomly assigned to diet (6 h/diet) at the onset of the study. The first feeding period was 24 d (P1) and the second was 21 d (P2), with a 7-d wash-out non-anionic (NA) diet period between P1 and P2. In P1, P2 and NA urine pH and DMI were recorded daily. Urine samples were collected mid-stream, 4–6 h post-feeding and pH measured. Anionic diets were offered in grain mixes and adjusted daily to maintain a urine pH between 5.5 and 6.0. Blood samples were taken on d 0 (diet assignment), 7, 14, 21, 24 in P1 and 7, 14 and 21 in P2. Diets were balanced using NDS Professional, fed as TMRs 1x/d in Calan gates and orts recorded. To insure adaptation to diets, data recorded in the last 4 d in P1, P2 and NA were averaged and compared across periods using PROC GLM and significance tested to  $P < 0.05$ . Urine pH in P1 and P2 for cows fed the SM and AN were  $5.8 \pm 0.14$  and  $5.66 \pm 0.14$ , respectively, and were different from the NA ( $6.52 \pm 0.14$ ,  $P = 0.001$ ). Serum calcium (mg/dL) was similar for cows fed SM or AN in P1 ( $9.32 \pm 0.19$  vs.  $9.37 \pm 0.2$ ) and P2 ( $9.33 \pm 0.21$  vs.  $9.2 \pm 0.24$ ) and not different from the NA ( $9.18 \pm 0.12$ ). Prior to start of P1, cows averaged  $8.95 \pm 0.18$  kg DM. During P1 and P2, cows fed AN had greater DMI ( $+0.79$  kg/d) than when fed the SM diets ( $P = 0.027$ ). Comparing the last 4 d of P1 and P2, cows fed the AN diets consumed more DM ( $+1.16$  kg/d,  $+1.02$  kg/d, respectively) than when fed the SM diets ( $P = 0.001$ ) but DMI was not different from the NA period ( $8.94 \pm 0.19$  kg). In conclusion, Animate was equally effective as anionic salts for reducing urine pH and maintaining serum calcium levels in Jersey cows; however, DMI were significantly improved.

**Key Words:** dry matter intake, Animate, Jersey cow

**M429 Nonlinear models to describe the transit of particles through the ruminant digestive tract: Evaluation of models and theoretical implications.** Ricardo Augusto Mendonça Vieira<sup>\*</sup>, Marcelo Cabral da Silva, Tadeu Silva de Oliveira, and Alberto Magno Fernandes, *Universidade Estadual do Norte Fluminense, Campos dos Goytacazes, RJ, Brazil.*

Fecal profiles (n) of the particulate markers Cr (n = 52) and La (n = 30) and the fluid marker complex Co-EDTA (n = 57) were obtained from digestion kinetic studies with cows, steers, and sheep with the aim of evaluating mathematical models designed to interpret marker excretion profiles in feces. The models evaluated were the multicompartmental model of Dhanoa et al. (1985), the Gamma model described as GN by Pond et al. (1988), and the GNG1 model proposed by Matis (1972). Models were fitted by ordinary nonlinear least squares and evaluated on the basis of the Akaike information criterion and derived measures (information-likelihood criteria). Despite remarkable differences in terms of information-likelihood measures, the models were quite similar in terms of visual adherence to observed time profiles and presented overlapping interval estimates for compartment mean retention times. The major relative numerical discrepancies observed among the models were related to the mean and variance of the transit time for the first

appearance of the marker in feces. The model with the best performance in mimicking the marker profiles was the GN model; the GNG1 model and the multicompartmental model were almost equivalent in terms of information-likelihood. Therefore, we derived a mathematical model to account for digestibility (D) and fill (Q) of the ruminoreticular digesta whenever the best model used to interpret marker studies, chosen based on information-likelihood criteria, was the GN model. Therefore, the GN model solutions for D and Q are as follows:  $D = [k/(\lambda + k)] \sum_i [\lambda/(\lambda + k)]^{i-1}$ , and  $Q = AF/(\lambda + k) \sum_i [\lambda/(\lambda + k)]^{i-1} + UFN/\lambda$ , for  $i = 1, 2, \dots, N$ . In the models, k,  $\lambda$ , A, and U are the fractional rate of the pool of escapable particles in the rumen, the fractional escape of particles from the rumination pool to the escapable pool, the fiber fraction available for digestion, and the unavailable fiber fraction, respectively. F is the average daily intake rate, and N is the order of time dependency. Funded by CNPq, CAPES, and FAPERJ.

**Key Words:** ruminant digestive tract, kinetics, mathematical model

**M430 Supplementation of grazing cow's diet with corn oil and palm kernel oil: ruminal fermentation, milk production and fatty acids profile.** Jair Esteban Parales, Martha Lucia Pabón, and Juan Evagenlista Carulla<sup>\*</sup>, *Universidad Nacional de Colombia, Bogotá, Cundinamarca, Colombia.*

The effects of supplementing corn oil (CO) alone or combined with palm kernel oil (PKO) to grazing cows on ruminal fermentation, milk production and its fatty acid (FA) profile were studied. Six multiparous Holstein cows ( $597 \pm 11.5$  kg BW,  $160 \pm 29$  d in milk) were assigned to a double Latin square design (3 cows × 3 diets × 3 periods × 2 squares). Three diets were evaluated: a control (C) without oil and 2 with oil addition (720 g/cow/day), one with CO, and the other with CO:PKO (75:25; COKP). Each trial period lasted 28 d (21 for adaptation and 7 for sample collection). Cows strip grazed a *Pennisetum clandestinum* pasture (3 kg DM/100 kg LW/d) and received daily corn silage (0.9 kg DM), concentrate (4.2 kg DM) and chromium oxide (9 g). Rumen fluid was collected on d 28th, milk samples on d 15, 18 and 21 of each period and feces from d 15 to 21. Milk production was recorded daily. Milk FA and ruminal VFA were determined by GC and Cr in feces and concentrate by AA. Methane production was estimated using ruminal VFA concentration. Data were subjected to ANOVA using GLM procedure of SAS and comparisons among means using Tukey's test. Voluntary intake and digestibility of the diet were similar among treatments as well as the molar proportions of acetate, propionate, and butyrate. Total VFA (m/L) was lower (C = 94.7 vs CO = 72.2 and COKP = 74.0,  $P < 0.001$ ) for the diets with oils resulting in lower estimated methane production ( $P < 0.01$ ). Addition of oils increased milk production (l/d) (C = 21.4 vs CO = 23.6 and COKP = 23.9;  $P < 0.01$ ) and milk fat (%) (C = 3.15 vs CO = 3.40 and COKP = 3.40;  $P < 0.05$ ). De novo synthesized FA (g/100 g FA) in milk was higher for the control treatment (C = 68.5 vs. CO = 52.2 and COKP = 58.6;  $P < 0.01$ ). Conjugated linoleic acid (CLA) (g/100g FA) in milk was higher for the oil diets but decreased by addition of CPK (C = 0.68 vs CO = 1.56, and COKP = 1.01;  $P < 0.01$ ). Supplementation with CO or COKP in diets of grazing dairy cows increased milk production without affecting voluntary intake or the digestibility of the diet but CO addition resulted in higher milk CLA.

**Key Words:** methane, CLA, *Pennisetum clandestinum*



**M431 Effect of rumen-protected carbohydrate supplementation on blood and plasma metabolites in finishing steers during heat stress.** Juan P. Russi\*<sup>1,3</sup>, Elias Peruzzo<sup>1</sup>, Nicolas DiLorenzo<sup>2</sup>, and Alejandro E. Relling<sup>1</sup>, <sup>1</sup>Facultad de Cs Veterinarias, UNLP, Buenos Aires, Argentina, <sup>2</sup>University of Florida, Marianna, FL, <sup>3</sup>RUPCA LLC, Merced, CA.

Finishing steers during the summer can be challenging due to the effects of high temperatures and humidity on DMI. The objective of this study was to evaluate the inclusion of a rumen-protected carbohydrate (RUPCA) (US Patent # 8,507,025) on blood metabolites of finishing steers during heat stress. Temperature humidity Index average measured every day during the experiment was  $72 \pm 4.9$ . Crossbred steers ( $n = 135$ ;  $355 \pm 20$ kg) were used in a 62-d experiment. Steers were blocked by initial BW and placed into 15 pens. Steers within blocks were randomly assigned to 3 treatments. T0 fed 91.4% of a basal diet (% DM), 22.3% corn silage, 65.9% dry corn, 0.6% sunflower meal, 0.5% urea, 2% minerals and vitamins and 8.6% of a supplement containing (% DM) 58.1% soybean meal, 38.9% soluble carbohydrates, 2% urea and 1% minerals salts, T1 fed the basal diet plus 4.3% supplement and 4.3% RUPCA and T2 fed basal diet plus 8.6% RUPCA. The supplement and RUPCA consisted of the same ingredients, differing on the processing of the carbohydrate (i.e., protected or not from ruminal degradation). Blood samples were taken from jugular vein prior morning feeding on d 0, 15, 39, and 62 and analyzed for glucose, insulin, urea and NEFA concentrations. Data were analyzed as a randomized complete block design with repeated measures using a mixed model of SAS. Initial body weight was used as a covariate. Treatment  $\times$  day interactions were found for insulin ( $P = 0.01$ ) and urea ( $P = 0.02$ ) plasma concentrations. There were no differences on plasma concentration of insulin, NEFA or urea among treatments ( $P > 0.10$ ). T0 showed higher blood glucose concentration ( $P = 0.05$ ). The results suggest that including RUPCA might help to mitigate the negative effects of heat stress on blood metabolites, potentially improving animal performance. Results are shown in Table 1.

**Table 1 (Abstr. M431).**

Item	T0	T1	T2	SEM	P-value		
					Trt	Day	$\times$ day
Glucose, mg/dL	89.4 <sup>a</sup>	81.9 <sup>b</sup>	83.5 <sup>b</sup>	2.73	0.05	<0.0001	0.35
Insulin, $\mu$ g/dL	0.56	0.73	0.68	0.072	0.35	0.0014	0.01
NEFA, mM	184.4	191.3	160.8	17.15	0.43	<0.0001	0.49
UREA, mg/dL	25.8	26.1	22.0	0.029	0.21	<0.0001	0.02

<sup>a,b</sup>Means without common superscript differ ( $P < 0.05$ ).

**Key Words:** carbohydrate, heat stress, bypass energy

**M432 Effect of rumen-protected carbohydrate supplementation on performance, blood and plasma metabolites in growing heifers.** Juan P. Russi\*<sup>1</sup>, Elias Peruzzo<sup>1</sup>, Nicolas DiLorenzo<sup>2</sup>, and Alejandro E. Relling<sup>1</sup>, <sup>1</sup>Facultad de Cs Veterinarias, UNLP, Buenos Aires, Argentina, <sup>2</sup>University of Florida, Mariana, FL.

The objective of this study was to evaluate the inclusion of a rumen-protected carbohydrate (RUPCA) (US Patent # 8,507,025) on performance, blood and plasma metabolites in growing heifers. Crossbred heifers ( $n = 135$ ;  $136 \pm 14$ kg) were used in an 84-d experiment. Heifers were blocked by initial BW, placed into 15 pens and fed a diet comprised of (DM basis) 38.8% corn silage, 41.5% dry corn, 2% minerals and vitamins mix, and 17.7% supplement or RUPCA, which varied depending on treatments. The supplement and RUPCA consisted of the same

ingredients (58.1% soybean meal, 38.9% soluble carbohydrates, 2% urea and 1% mineral salt), differing in the processing of the carbohydrate (i.e., protected or not from ruminal degradation). Heifers within blocks were randomly assigned to 3 treatments: T0) 17.7% supplement (100% unprotected carbohydrate), T1) 8.85% supplement and 8.85% RUPCA, and T2) 17.7% RUPCA (100% protected carbohydrate). Body weight was measured on d 0, 21, 42, 63, and 84. Pen DMI was measured weekly from d 21 to 84. Blood samples were taken on d 0, 42, 63, and 84 from jugular vein prior morning feeding and analyzed for glucose, insulin, urea and NEFA concentrations. Data were analyzed as a randomized complete block design with repeated measures using a mixed model of SAS. Treatment  $\times$  day interaction were found for DMI ( $P = 0.02$ ), ADG ( $P < 0.0001$ ) and G:F ( $P < 0.0001$ ) and with T1 having the lowest DMI ( $P < 0.05$ ) and the greatest G:F ( $P < 0.05$ ). No differences were found in the concentrations of blood glucose ( $P > 0.91$ ), plasma insulin ( $P = 0.82$ ), plasma NEFA ( $P = 0.802$ ) or plasma urea ( $P = 0.336$ ). Feeding RUPCA to growing heifers improved G:F through lower DMI without altering ADG, blood or plasma metabolites. Results are shown in Table 1.

**Table 1 (Abstr. M432).**

Item	T0	T1	T2	SEM	P-value		
					Trt	Day	$\times$ day
DMI, kg/d	6.9 <sup>a</sup>	5.9 <sup>b</sup>	6.8 <sup>a</sup>	0.06	<0.0001	<0.0001	0.02
ADG, kg	1.18	1.13	1.19	0.027	0.21	<0.0001	<0.0001
G:F	0.161 <sup>b</sup>	0.202 <sup>a</sup>	0.177 <sup>ab</sup>	0.0103	0.0003	<0.0001	<0.0001
Glucose, mg/dL	90.2	91.6	91.2	3.52	0.91	<0.0001	0.92
Insulin, $\mu$ g/dL	0.26	0.26	0.24	0.053	0.82	0.016	0.72
NEFA, mM	200.8	187.9	181.3	18.74	0.54	0.0025	0.42
Urea, mg/dL	14.9	13.4	14.8	1.03	0.36	<0.0001	0.39

<sup>a,b</sup>Means without common superscript differ ( $P < 0.05$ ).

**Key Words:** carbohydrate, rumen, bypass energy

**M433 Nonlinear parameter estimation in R and SAS: Similarities and discrepancies of both statistical programs based on a case study of digestion kinetics and animal growth curves.** Ricardo Augusto Mendonça Vieira\*<sup>1</sup>, Leonardo Siqueira Glória<sup>2</sup>, and Fabyano Fonseca e Silva<sup>2</sup>, <sup>1</sup>Universidade Estadual do Norte Fluminense, Campos dos Goytacazes, RJ, Brazil, <sup>2</sup>Universidade Federal de Viçosa, Viçosa, MG, Brazil.

Growth, digestion, and passage kinetics in ruminants were studied as reference problems of nonlinear phenomena in animal science to be analyzed using nonlinear models. The statistical packages R and SAS were compared in terms of their nonlinear parameter estimation by ordinary nonlinear least squares and maximum likelihood algorithms. The assessed programs included the nls2, nlsLM, and nlme functions of R, and the NLIN, %NLINMIX macro of SAS. The quality of fit of the models was evaluated through likelihood criteria. The NLIN, nls2, and nlsLM functions yielded nonlinear parameter estimates that were almost equal in terms of scale; nevertheless, the interval estimates obtained with nls2 and nlsLM were within those estimated with PROC NLIN, despite the fact that the approximate confidence intervals are estimated using the same Student's *t*-test in both R and SAS. The degradation of fiber and passage kinetics of particulate markers were predicted with very small

numerical differences both in terms of scale and dispersion estimates. For the nonlinear mixed-effects models used to interpret growth data, the nlme (R) and %NLINMIX macro (SAS) algorithms differed in terms of the value of the likelihood function whenever heterogeneous variances, correlations, and weighting variances were fitted; nonetheless, when assuming independence and homoscedasticity, the results of the log-likelihood function were identical. The number of possible models fitted to the growth profiles was 28 with nlme function of R, whereas with %NLINMIX macro of SAS, only 13 possible models were fitted. Fortunately, the conclusions reached by fitting growth models to lamb growth data with either R or SAS were the same. Nonetheless, because each fitting experience is unique, there is no guarantee that the same conclusions would be achieved because the programs do not behave equally in the case of fitting nonlinear mixed models with different correlation and variance structures combinations. Funded by CNPq, CAPES, and FAPERJ.

**Key Words:** nonlinear phenomena, R-project, SAS software

**M434 Effect of crude glycerin in supplement on rumen microbial profile of Nelore steers consuming low quality pasture during the dry season.** Elias San Vito\*, Pablo Castagnino, Erick E. Dallantonia, Yury T. Granja-Salcedo, Lutti M. Delevatti, and Telma T. Berchielli, *University Estadual Paulista-UNESP, Jaboticabal, São Paulo, Brazil.*

The effect of crude glycerin (CG) - 80% of glycerol - inclusion as a substitute to corn grain in supplements on rumen fluid protozoa numbers, and relative proportion of cellulolytic bacteria and methanogenic archaea of rumen-cannulated Nelore steers (n = 10; BW = 408.8 ± 38.5 kg) on low quality pasture, in the dry season was studied. Treatments were constituted by 5 levels of CG in the supplement: 0, 7, 14, 21 and 28% DM of CG. Animals were supplemented, daily at 1000 h in a ratio of 700 g/100 kg of BW. Supplement consisted of corn grain, soybean meal, urea, gluten meal and mineral mix. Bacteria and protozoa samples were collected on the d 11 of each experimental period, 3 h after supplementation, in the solid and liquid extracts in different parts of the rumen. Real-time PCR was used to quantify microbial population. Methanogens and cellulolytic bacteria were expressed as a proportion of total rumen bacterial 16S rDNA. Ciliate protozoa species were identified and quantified (#/mL) in a Sedgewick-Rafter chamber. Data of ciliated protozoa were log<sub>10</sub>-transformed and analyzed in a replicated Latin square design using the MIXED procedure of SAS. Bacterial proportions were analyzed using the software R, with data compared between treatments (with or without 28% DM of CG in the supplement) using the Wilcoxon test, considered significant effects at  $P < 0.05$ . The inclusion of CG in supplement did not affect ( $P > 0.05$ ) rumen protozoa number (6.52, 4.46, 3.02, 2.95, 1.73, 2.16 and 1.67 for the genera *Entodinium*, *Dasytricha*, *Isotricha*, *Eremoplastron*, *Eudiplodinium*, *Elytroplastron*, and *Polyplastron*). The inclusion of CG had no effect in the rumen relative proportion of *Ruminococcus albus* ( $P = 0.237$ ), *Ruminococcus flavefaciens* ( $P = 0.129$ ), and methanogens ( $P = 0.151$ ) with mean values of 0.0046, 0.0044, and 0.0169. However, increased ( $P = 0.003$ ) *Fibrobacter succinogenes*, with mean values of 0.0033 and 0.0296 for 0% DM and 28% DM of CG in the supplements, respectively. Inclusion up to 28% DM of CG in the supplement did not interfere negatively on rumen microbial profile, increased relative proportion of *F. succinogenes* of Nelore steers in low quality pasture.

**Key Words:** pasture, cellulolytic, glycerol

**M435 Effect of crude glycerin in supplement on rumen microbial profile of Nelore steers grazing tropical grass during the rainy season.** Telma T. Berchielli\*, Elias San Vito, Pablo Castagnino, Yury T. Granja-Salcedo, and Erick E. Dallantonia, *University Estadual Paulista-UNESP, Jaboticabal, São Paulo, Brazil.*

The effect of crude glycerin (CG) - 80% of glycerol - inclusion as a substitute to corn grain in supplements on rumen fluid protozoa numbers, and relative proportion of cellulolytic bacteria and methanogenic archaea of rumen-cannulated Nelore steers (n = 10; BW = 490 ± 47 kg) grazing tropical grass, during the rainy season was studied. Treatments were constituted by 5 levels of CG in the supplement: 0, 7, 14, 21 and 28% DM of CG. Animals were supplemented daily at 1000 h in a proportion of 300 g/100 kg of BW. Supplement consisted of corn grain, soybean meal, urea, gluten meal and mineral mix. Bacteria and protozoa samples were collected on the d 11 of each experimental period, 3 h after supplementation, in the solid and liquid extracts in different parts of the rumen. Real-time PCR was used to quantify microbial population. Methanogens and cellulolytic bacteria were expressed as a proportion (%) of total rumen bacterial 16S rDNA. Ciliate protozoa species were identified and quantified (#/mL) in a Sedgewick-Rafter chamber. Data of protozoa were log<sub>10</sub>-transformed and analyzed in a replicated Latin square design using the MIXED procedure of SAS. Bacterial proportions were analyzed using the software R, with data compared between treatments (with or without 28% DM of CG in the supplement) using the Wilcoxon test, the effects of treatments were considered significant at  $P < 0.05$ . The inclusion of crude glycerin in supplement did not affect ( $P > 0.05$ ) rumen protozoa number (6.8, 5.6, 5.2, 2.4, 2.0, and 3.6 for the genera *Entodinium*, *Dasytricha*, *Isotricha*, *Eremoplastron*, *Diploplastron*, and *Polyplastron*). The inclusion of CG decreased the relative proportion of *Ruminococcus albus* ( $P = 0.047$ ) and *Ruminococcus flavefaciens* ( $P = 0.036$ ), with mean values of 0.0702 and 0.0174; 0.0411 and 0.0073 respectively for 0% DM and 28% DM of CG in the supplements. However, had no effect on *Fibrobacter succinogenes* ( $P = 0.420$ ), and methanogens ( $P = 0.150$ ) with mean values of 0.0199 and 0.1791, respectively. Inclusion of CG at the level of 28% of DM in the supplement negatively affect the rumen cellulolytic bacteria *R. albus* and *R. flavefaciens* of Nelore steers grazing tropical grass.

**M436 Improvement in saccharification yield of mixed rumen enzymes by identification of recalcitrant cell wall constituents using enzyme fingerprinting.** Ajay Badhan<sup>1</sup>, Yuxi Wang\*<sup>1</sup>, Robert Gruninger<sup>1</sup>, Justin Powlowski<sup>2</sup>, Adrian Tsang<sup>2</sup>, and Tim McAllister<sup>1</sup>, <sup>1</sup>*Agriculture and Agri-Food Canada, Lethbridge Research Centre, Lethbridge, AB, Canada,* <sup>2</sup>*Centre for Structural and Functional Genomics, Concordia Univ, Montreal, QC, Canada.*

Identification of factors that limit plant cell wall digestion of forages and the development of enzymatic approaches that improve hydrolysis could play a key role in improving the production efficiency of ruminants fed these feeds. Enzyme fingerprinting and FTIR analysis of barley silage and total-tract indigestible fiber residue (TIFR) in heifers' feces were used to identify cell wall components resistant to digestion. The results identified acetyl xylan esterases as key to enhance ruminal digestion, and that polysaccharide-lignin complexes are principal components in TIFR. Enzyme pre-treatment to enhance glucose yield from hydrolyses of barley straw and alfalfa hay by mixed rumen enzymes (MRE) was subsequently developed. The enzymes for pre-hydrolysis were recombinant acetyl xylan esterases (AXE16B\_ASPNG and AXE16A\_ASPNG), polygalacturonase (PGA28A\_ASPNG),  $\alpha$ -arabinofuranosidase (ABF54B\_ASPNG) all from *Aspergillus niger*, feruloyl esterase (FAE1a) from *Anaeromyces mucronatus* (expressed in

*E. coli*), endoglucanase GH7 (EGL7A\_THITE) from *Thielavia terrestris* (produced in *A. niger*). Lyophilized cell-free rumen fluid from cows fed forage diet was used as source of MRE. The study was conducted using micro assays in combination with simplex lattice mixture models that were designed using Design of Experiment statistical software. Fungal hemicellulases and auxiliary enzymes initiated degradation of structural polysaccharides upon application and improved the in vitro saccharification of alfalfa and barley straw by MRE. The analysis model predicted 75% (SEM: 2.8%) higher relative glucose yield from alfalfa pretreated with PGA28A\_ASPNG and ABF54B\_ASPNG in 1:1 ratio. Whereas, prehydrolysis of barley straw with a mixture of 50% EGL7A\_THITE and 50% FAE1a increased glucose release by 100% (SEM: 1.8%) upon incubation with MRE. The results showed that microassays in combination with simplex lattice statistical experimental design can be used to predict effective enzyme pretreatments that can enhance plant cell wall digestion by MRE, and strengthen the rationale of developing specific enzyme pretreatments for forages, depending on their structure and composition.

**Key Words:** enzyme fingerprinting, saccharification, plant cell wall

**M437 Effect of dietary protein level and vigna hay supplementation on production and efficiency of lactating dairy cows.** Elmer Edgardo Corea-Guillén<sup>1</sup>, J. M. Aguilar-Aguilar<sup>1</sup>, N. P. Alas-Avelar<sup>1</sup>, E. A. Alas-García<sup>1</sup>, J. M. Flores-Tensos<sup>1</sup>, and Glen A. Broderick<sup>\*2</sup>, <sup>1</sup>Universidad de El Salvador, San Salvador, El Salvador; <sup>2</sup>Broderick Nutrition & Research LLC, Madison, WI.

This trial assessed whether dietary content of CP or supplementation with vigna hay (*Vigna sinensis*) would improve milk production or efficiency of lactating cows. Thirty-two multiparous Holstein cows were blocked by DIM into 8 squares of 4 cows in a replicated 4 × 4 Latin square with a 2 × 2 arrangement of treatments: 15.5 or 17% CP, and with or without vigna hay (16.6% CP), which replaced 12.5% of dietary DM from sorghum silage (8.4% CP). Soybean meal was fed to adjust dietary CP; diets contained about 50% forage and 50% concentrate, and 38% NDF. Cows within squares were randomly assigned to treatment sequences and fed diets for 3-wk periods; data from the last week were analyzed using the mixed procedures of SAS. Table 1 reports LS-means. Diet did not alter yield of milk or milk components. Higher dietary CP increased DMI but also increased MUN and reduced N efficiency without improving yield. Supplementing dietary sorghum silage with vigna hay maintained production, reduced DMI and MUN, and increased milk/DMI, N efficiency and apparent NDF digestibility. Daily income over feed cost (IOC) averaged \$0.85/cow more on the vigna hay diets. Results from this trial indicated that replacing a portion of dietary forage with vigna hay improved nutrient and economic efficiency of milk production.

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**Table 1 (Abstr. M437).** Effect of dietary CP (15% and 17%) and vigna hay supplementation (0 and 12.5%) on production

Item	15.5% CP		17% CP		SEM	Probability		
	0	12.5	0	12.5		CP	Vig	CP × Vig
DMI, kg/d	19.5	19.0	20.4	19.8	0.34	<0.01	<0.01	0.73
Milk yield, kg/d	28.0	29.3	29.1	29.1	0.77	0.35	0.19	0.23
Milk/DMI	1.45	1.55	1.43	1.48	0.047	0.08	0.01	0.33
Fat yield, kg/d	0.96	0.99	0.98	1.02	0.035	0.38	0.17	0.78
Protein yield, kg/d	0.88	0.91	0.92	0.92	0.024	0.17	0.31	0.30
MUN, mg/dL	17.4	16.1	19.6	18.6	0.33	<0.01	<0.01	0.54
Milk N/NI, %	28.2	30.0	25.8	27.4	0.88	<0.01	<0.01	0.81
NDF digestibility, %	42.4	52.1	41.5	52.5	0.68	0.68	<0.01	0.31
IOC, USD/cow/d	9.40	10.47	9.23	10.06	0.438	0.28	<0.01	0.67

**Key Words:** dietary protein, vigna hay, income over feed cost

**M438 In vitro effects of a commercial blend of functional oils on rumen fermentation, methane production, and methanogenic archaea.** Ahmad Reza Seradj<sup>1</sup>, Joan Torrent<sup>\*2</sup>, Gabriel de la Fuente<sup>1</sup>, and Joaquim Balcells<sup>1</sup>, <sup>1</sup>University of Lleida, Lleida, Catalonia, Spain; <sup>2</sup>Oligo Basics, Cary, NC.

A complete randomized block design trial with 4 in vitro incubation sets were prepared to evaluate the effect of a commercial blend of functional oils (FO) containing cashew nut shell liquid and castor oil as active ingredients (Essential, Oligo Basics Agroind. Ltda., Cascavel, Brazil) on rumen fermentation, methane production and methanogenic archaea. Bottles of 120 mL were filled with 600 mg of concentrate (same as given to 4 rumen liquid donor steers), and 80 mL of an incubation solution including rumen inoculum, mineral, buffer and reducing solutions under a CO<sub>2</sub> stream. Sealed bottles were incubated at 39 ± 1°C for 24 h and either dosed with 500 µg of FO/g DM of basal diet or not (control). The headspace pressure was measured and sampled (0.1 mL) at 2 h intervals, up to 12 h and then 24 h post incubation to determine gas and then methane concentration using GC. The pattern of cumulative gas/methane production ( $y$ ) was fitted to the model:  $y = a(1 - e^{-b(t-c)})$ , being  $a$  the potential cumulative gas/methane production (mL);  $b$  the production rate (mL/h) and  $c$  the lag time (h). After 12 and 24 h, 2 bottles per treatment per set were sampled for NH<sub>3</sub>-N, volatile fatty acid (VFA) concentration and molecular analyses. The DNA was extracted using a QIAamp Kit. Specific primers were used to determine absolute abundance (Log<sub>10</sub> gene copy number/ g fresh sample) of total bacteria and hydrogenotrophic methanogenic archaea (HMA) and the relative abundance ( $2^{(-\Delta Ct)}$ ) of HMA in relation to total archaea using qPCR (CFX96 Touch). Bottles supplemented with FO showed a tendency (14.26 vs. 13.50 SEM 0.284;  $P = 0.06$ ) to decrease methane production (mL/g DM substrate) and reduced the methane ratio (CH<sub>4</sub>/gas; v/v; 0.093 vs. 0.089 SEM 0.0011;  $P = 0.049$ ), where the discrete lag time for gas production increased (0.04 vs. 0.12 SEM 0.024;  $P = 0.047$ ). Addition of FO improved rumen fermentation, increasing molar proportion of propionate (33.5 vs. 34.3 SEM 0.24;  $P = 0.024$ ) and decreasing NH<sub>3</sub>-N concentration (305.3 vs. 284.4 SEM 5.95;  $P = 0.022$ ) and relative abundance of HMA (17.8 vs. 14.7 SEM 0.96;  $P = 0.032$ ).

**Key Words:** fermentation, functional oil, methanogenic archaea



**M439 The influence of feeding oscillating dietary crude protein contents on milk production and nitrogen utilization in lactating dairy cows.** Jolet Köhler\* and Timothy Mutsvangwa, *Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, SK, Canada.*

There is increasing public pressure on intensive dairy operations to reduce nitrogen (N) excretion into the environment, which can be achieved by adopting feeding practices that enhance the efficiency of N utilization. One such feeding strategy is feeding diets with oscillating crude protein (CP) contents, and studies with finishing beef cattle and growing sheep have reported improvements in N retention when oscillating CP diets are fed compared with static CP diets. This experiment was conducted to determine: 1) the effects of feeding oscillating CP diets on N balance and milk production in dairy cows; and 2) the optimum frequency of oscillating dietary CP concentration (i.e., 24, 48, or 72 h). Eight Holstein cows ( $714 \pm 36$  kg of BW;  $114 \pm 15$  DIM) were used in a replicated  $4 \times 4$  Latin square design with 30-d periods (consisting of 18 d of dietary adaptation and 12 d of sample and data collection). Treatments were a diet containing 17% CP fed on a continuous basis (designated STATIC), and diets containing 14% and 20% CP that were fed on an oscillating basis at 24 (OSC-24), 48 (OSC-48), or 72 (OSC-72) h. Diets were fed twice per day as TMR. The actual CP contents were 17.8% for the STATIC, and 14.9 and 20.3% for the oscillating CP diets, which deviated from the target CP concentrations due to variations in forage CP content. Dry matter intake (mean = 26.6 kg/d) and milk production (mean = 36.4 kg/d) were not affected ( $P \geq 0.57$ ) by diet. Milk contents and yields of fat, protein, and lactose were unaffected ( $P \geq 0.33$ ) by diet. Although N intakes were similar across dietary treatments, retained N was greater ( $P = 0.02$ ) in cows fed the OSC-48 diet compared with those fed the STATIC and OSC-24 diets. Apparent total-tract DM, organic matter, CP, NDF, and ADF digestibilities did not differ ( $P > 0.05$ ) among diets; however, cows fed the OSC-72 diet had greater ( $P = 0.02$ ) apparent total-tract fat digestibility compared with those fed the STATIC and OSC-24 diets. Our results demonstrate that feeding oscillating dietary CP diets on a 48-h basis improves N efficiency by enhancing N retention when compared with feeding a static CP diet or an oscillating dietary CP on a 24-h basis.

**Key Words:** milk production, nitrogen utilization, oscillating crude protein

**M440 Effect of guanidinoacetic acid on metabolism of cattle.** Mehrnaz Ardalan\*<sup>1</sup>, Erick D. Batista<sup>1,2</sup>, Cheryl K. Armendariz<sup>1</sup>, and Evan C. Titgemeyer<sup>1</sup>, *<sup>1</sup>Department of Animal Sciences and Industry, Kansas State University, Manhattan, KS, <sup>2</sup>Departamento de Zootecnia, Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.*

Guanidinoacetic acid (GAA) is the precursor of creatine, which is produced by hepatic methylation of GAA. We evaluated the metabolic response of cattle to post-ruminal supplementation of GAA with and without methionine (Met) supplementation as a source of methyl groups. Six ruminally cannulated Holstein heifers ( $520 \pm 49$  kg initial BW) were fed twice daily a diet containing 6 kg/d rolled corn, 4 kg/d alfalfa hay, and 50 g/d trace-mineralized salt. The experiment used a split-plot design. All treatments were infused continuously to the abomasum. The main plot treatments were 0 or 12 g/d of L-Met arranged in a completely randomized design; 3 heifers received each treatment throughout the entire experiment. Subplot treatments were 0, 10, 20, 30, and 40 g/d of GAA, with treatments provided in sequence from lowest to highest level using 6-d periods for each level. Blood and urine samples were collected on d 6 of each period. Met supplementation increased plasma Met ( $P < 0.01$ ). GAA supplementation linearly increased plasma Arg

(% of total AA), suggesting a sparing of Arg for GAA synthesis. GAA supplementation linearly increased plasma concentrations of GAA and creatinine ( $P < 0.001$ ) and increased plasma creatine at all levels of GAA except when 40 g/d of GAA was supplemented with no Met (GAA quadratic  $\times$  Met,  $P = 0.07$ ). Plasma homocysteine was not affected by GAA supplementation when heifers received 12 g/d Met, but it was increased in response to 30 or 40 g/d of GAA when no Met was supplemented (GAA linear  $\times$  Met,  $P = 0.003$ ); increases did not suggest a dangerous hyperhomocysteinemia. Urinary GAA and creatine concentrations were increased by all levels of GAA when 12 g/d Met was provided and by GAA supplementation up to 30 g/d when no Met was provided, but 40 g/d GAA did not increase urinary concentrations of GAA or creatine when no Met was supplemented (GAA quadratic  $\times$  Met,  $P \leq 0.06$ ). Data suggest that post-ruminal GAA supplementation increased creatine supply to cattle. An apparent methyl group deficiency, demonstrated by increases in plasma homocysteine, developed when 30 or 40 g/d of GAA was supplemented, but it was ameliorated by 12 g/d Met.

**Key Words:** creatine, guanidinoacetic acid, methionine

**M441 Canola meals produced in different years have similar contents of rumen-undegraded protein.** Glen A. Broderick\*<sup>1</sup>, Stefania Colombini<sup>2</sup>, and Sara Costa<sup>2</sup>, *<sup>1</sup>Broderick Nutrition & Research LLC, Madison, WI, <sup>2</sup>University of Milan, Milan, Italy.*

When evaluating protein degradability in canola meal (CM) produced in 2011 and 2012 we observed that CM from different processing plants differed in rumen-degraded protein (RDP) and rumen-undegraded protein (RUP), estimated using the Michaelis-Menten inhibitor in vitro method (MMIIV). However, these analyses were conducted in 2 different years and may have been influenced by methodological changes over time (e.g., degradative activity of rumen inoculum may have altered between years). Three CM samples were collected from each of 12 Canadian production plants in each of 2 years (2013 and 2014; total = 72). Meals were analyzed chemically and for protein degradation rate, RDP and RUP by MMIIV, assuming passage rates of 0.16 and 0.06/h for soluble and insoluble proteins, respectively (Colombini et al., *J. Dairy Sci.* 94:1967–1977, 2011). Half of the meals from both years were analyzed in each of 3 MMIIV incubations; a total of 6 incubations were conducted. Differences among plants and years were assessed with the Proc Mixed procedure of SAS, and LSM over all plants for each year are in Table 1. As seen in 2011 and 2012, there were large differences in RUP among plants (data not shown). There were differences by year ( $P \leq 0.04$ ) in DM and protein fraction degraded at  $t = 0$  (FD<sub>0</sub>); CP in CM from 2013 was 1.8 percentage units greater than 2014 ( $P < 0.01$ ). Theta (unadjusted degradation rate) was not different between years ( $P = 0.50$ ). However, adjusting Theta for FD<sub>0</sub> resulted in differences in degradation rate by year ( $P = 0.03$ ) and trends ( $P = 0.06$ ) for differences in RDP and RUP. The results suggest that apparent differences among production years in RUP content of CM may be due to small differences in protein already degraded before incubation.

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**Table 1 (Abstr. M441).** Composition and degradability of canola meals from 2 years

Trait <sup>1</sup>	2013	2014	SEM	P-value
DM, %	90.9	90.4	0.15	0.04
CP, % of DM	42.4	40.6	0.22	<0.01
Insoluble-N, % of TN	71.5	72.7	0.55	0.14
Soluble-N, % of TN	28.5	27.3	0.55	0.14
FD <sub>0</sub> , % of TN	2.4	1.8	0.13	0.01
Theta, /h	0.154	0.156	0.0078	0.50
Adjusted degradation rate, /h	0.143	0.147	0.0078	0.03
RDP, % of CP	64.5	65.0	1.18	0.06
RUP, % of CP	35.5	35.0	1.18	0.06

<sup>1</sup>TN = total N; FD<sub>0</sub> = protein degraded at t = 0; Theta = unadjusted degradation rate.

**Key Words:** canola meal, inhibitor in vitro, rumen-undegraded protein

**M442 Enteric methane emissions of crossbred heifers fed mixtures of *Pennisetum purpureum* grass and *Leucaena leucocephala*.** A.T. Piñeiro-Vázquez<sup>1</sup>, J.R. Canul-Solis<sup>1</sup>, J.A. Alayón-Gamboa<sup>2</sup>, A.J. Ayala-Burgos<sup>1</sup>, F.J. Solorio-Sánchez<sup>1</sup>, C.F. Aguilar-Pérez<sup>1</sup>, and J.C. Ku-Vera<sup>1</sup>, <sup>1</sup>Faculty of Veterinary Medicine and Animal Science, University of Yucatán, Mérida, Yucatán, México, <sup>2</sup>The College of the Southern Frontier, Campeche, México.

The aim of the work was to assess feed intake, apparent digestibility and methane (CH<sub>4</sub>) emissions of crossbred heifers fed a basal ration of *Pennisetum purpureum* grass mixed with increasing amounts of chopped forage of the tropical legume *Leucaena leucocephala*. Five crossbred (*Bos indicus* × *Bos taurus*) heifers with an average live weight of 295 ± 6 kg were used. Heifers were randomly allotted to 5 treatments (0, 20, 40, 60 and 80% *L. leucocephala* on a DM basis) in a 5 × 5 Latin square design. The DM intake, DM digestibility and CH<sub>4</sub> production (L/day) were determined in 24 h periods, while heifers were housed in open-circuit respiration chambers. Data were analyzed with PROC GLM of SAS. Dry matter intake (DMI) and organic matter intake (OMI) were similar ( $P > 0.05$ ) between treatments registering an average of 7.0 and 6.5 kg/day and an intake of 98.7 g DM/kg<sup>0.75</sup>/day. Dry matter digestibility (DMD) was similar ( $P > 0.05$ ) between treatments registering an average of 492.3 g/kg. Organic matter digestibility (OMD) was similar between the control treatment (without *L. leucocephala*) and treatments with incorporation of 20 and 40% of *L. leucocephala*, however, with 60 and 80% incorporation, OMD was significantly different ( $P \leq 0.05$ ) compared with the control treatment. Molar proportions of acetic and propionic acids in the rumen were significantly ( $P \leq 0.05$ ) affected, observing a difference between treatments with incorporation of *L. leucocephala* and the control treatment. A linear ( $P = 0.0005$ , 0.0004 and 0.0022) reduction was observed in enteric CH<sub>4</sub> emissions when expressed as DM, OM, and NDF (L/kg) respectively, as the level of *L. leucocephala* was increased, additionally a reduction of 61.3, 61.7 and 53.1% was observed in CH<sub>4</sub> emissions (L/kg) when expressed per unit of DMI, OMI and NDFI respectively, with 80% incorporation of the legume in ration DM compared with the control treatment. These data point out toward the potential of *L. leucocephala* to reduce CH<sub>4</sub> emissions in cattle fed a basal ration of *P. purpureum* grass.

**Key Words:** methane emissions, respiration chamber, feed intake

**M443 Effect of dietary crude protein content on milk yield and composition in dairy cows fed diets based on rehydrated corn silage and sugar cane silage.** Marcos André Arcari, Cristian Marlon de Magalhães Rodrigues Martins, Juliano Leonel Gonçalves, Danylo Oliveira Sousa, Bruna Gomes Alves\*, Alessandra Módena Orsi, and Marcos Veiga dos Santos, Universidade de São Paulo, Pirassununga, SP, Brazil.

The aim was to evaluate the effect of dietary crude protein (CP) content on milk yield and composition for dairy cows fed with rehydrated corn silage (RCS) and sugar cane silage (SS). Fifteen Holstein cows were distributed in 3 contemporary 3 × 3 Latin square design and diets consisted of 600g / kg dry matter (DM) of SS, 400g / kg DM of RCS and CP levels (135, 161 and 186 g CP / kg DM) contained 89.9, 120.8 and 180.5 g soybean meal; 21.2, 40.4 and 50.5 g corn meal gluten and 6.7, 8.8 and 7.9 g urea / kg DM. The dry matter intake was 18.7 kg, 21.0 kg and 20.5 kg DM respectively. The data were analyzed using the MIXED procedure of SAS, where treatment effect was decomposed into 2 orthogonal polynomial contrasts (linear and quadratic). The CP content linearly increased the milk yield ( $P = 0.0005$ ) (27.65, 29.88 and 30.32 L / day) [ $Y = 20.6593$  (SE = 2.3976) + 0.5509 (SE = 0.1350) × CP] and 3.5% fat-corrected milk ( $P = 0.0002$ ) (27.90, 30.63, 31.46 L / day) [ $Y = 18.6007$  (SE = 2.8809) + 0.7275 (SE = 0.1643) × CP]. There was a linear increase in the milk concentrations of fat ( $P = 0.045$ ) (3.53, 3.64 and 3.71 g / 100mL) [ $Y = 3.0605$  (SE = 0.2827) + 0.03627 (SE = 0.01675) × CP], protein ( $P = 0.025$ ) (2.82, 2.87 and 2.9g / 100mL) [ $Y = 2.6042$  (SE = 0.1305) + 0.01728 (SE = 0.007107) × CP], casein ( $P < 0.0001$ ) (2.08, 2.17 and 2.32g / 100mL) [ $Y = 1.4634$  (SE = 0.06818) + 0.04644 (SE = 0.003836) × CP], total solids ( $P = 0.011$ ) (11.91, 12.24 and 12.23%) [ $Y = 11.0888$  (SE = 0.4023) + 0.06661 (SE = 0.02284) × CP], milk urea nitrogen (12.71, 18.44 and 18.95 mg / dL) [ $Y = -3.6595$  (SE = 2.4423) + 1.2998 (SE = 0.1440) × CP] and casein percentage:milk protein ratio ( $P < 0.0001$ ) (73.78, 75.61 and 79.88%) [ $Y = 58.0580$  (SE = 3.4810) + 1.1727 (SE = 0.2044) × CP] for cows fed 135, 161 and 186 g CP / kg DM, respectively. And, there was a quadratic effect on the lactose content ( $P = 0.005$ ) [ $Y = 1.8836$  (SE = 0.8580) + 0.3406 (SE = 0.1127) × CP - 0.01091 (SE = 0.003650) × CP<sup>2</sup>]. The increase in dietary CP content when cows were fed with RCS and SS caused an increase on milk yield and its main compounds solids.

**Key Words:** milk, protein, rehydrated corn silage

**M444 Effects of levels of whole cottonseed and soybean oil on intake and ruminal fermentation in Nellore steers.** Vinicius N. Gouvea<sup>1</sup>, Marcos V. Biehl<sup>2</sup>, Marcos V. C. Ferraz Junior<sup>1</sup>, Jose A. Faleiro Neto<sup>1</sup>, Elizangela M. Moreira<sup>1</sup>, Marcelo H. Santos<sup>1</sup>, Renan G. Silva<sup>1</sup>, Mariana F. Westphalen<sup>2</sup>, Alexandre A. Miszura<sup>1</sup>, Daniel M. Polizel<sup>1</sup>, and Alexandre V. Pires<sup>2,1</sup>, <sup>1</sup>University of Sao Paulo, Pirassununga, SP, Brazil, <sup>2</sup>University of Sao Paulo, Piracicaba, SP, Brazil.

In this study 6 ruminally cannulated Nellore steers (407 ± 24 kg BW) were used in a 6 × 6 Latin square design to evaluate 5 levels of whole cottonseed (0, 8, 16, 24 and 32% DM basis respectively treatments WC0; WC8; WC16; WC24 and WC32) plus a negative control diet – WCO + soybean oil (treatment SO; to reach the WC0 diet at the same fat content of WC32). Isonitrogenous (14% CP) diets containing 20% roughage 80% concentrate were formulated to provide the same amount of ruminally degradable protein. The 26-d experimental periods consisted 21-d for adaptation followed by 5-d for collection. Increasing WC linearly decreased DMI ( $P < 0.01$ ; 7.9; 7.21; 6.77; 6.06; 6.18 kg/d respectively for WC0; WC8; WC16; WC24 and WC32).

No differences were observed between S0 and WC0 ( $P = 0.23$ ; 7.17 vs 7.90 kg/d respectively) neither between S0 and WC32 ( $P = 0.08$ ; 7.17 vs 6.18 kg respectively). Increasing WC increased quadratically the pH ( $P < 0.001$ ; 6.24; 6.44; 6.54; 6.65; 6.56 respectively for WC0; WC8; WC16; WC24 and WC32). The SO inclusion increased the pH compared with WC0 diet ( $P < 0.05$ ; 6.37 vs 6.24 respectively). Even with the same fat content WC32 diet presented higher pH than SO diet ( $P < 0.001$ ; 6.56 vs 6.37 respectively). Total volatile fatty acids (VFA) linearly decreased with WC inclusion ( $P < 0.001$ ; 107; 94.4; 87.0; 79.8 and 79.0 mM respectively for WC0; WC8; WC16; WC24 and WC32). The SO inclusion did not change total VFA compared with WC0 diet ( $P = 0.22$ ; 102 vs 107 mM respectively). The SO inclusion increased the total VFA compared with WC32 diet ( $P < 0.001$ ; 102 vs 79 mM respectively). Acetate:propionate (A:P) ratio linearly decreased with WC inclusion ( $P < 0.001$ ; 2.60; 2.62; 2.22; 2.23; 1.88 respectively for WC0; WC8; WC16; WC24 and WC32). The SO inclusion did not change the A:P ratio compared with WC0 diet ( $P = 0.73$ ; 2.66 vs 2.60 respectively) but compared with WC32 the SO increased the A:P ratio ( $P < 0.005$ ; 2.66 vs 1.88 respectively). Intake and rumen fermentation parameters were negatively affected by the levels of WC inclusion in the diets but the main reasons for that not seems only related with the increased fat content.

**Key Words:** beef, fatty acid, feedlot

**M445 Effect of lipid sources with different fatty acid profiles on rumen metabolites of feedlot Nellore steers.** Juliana Duarte Messana\*, Giovanni Fiorentini, Pablo S. Castagnino, Roberta C. Canesin, and Telma T. Berchielli, *UNESP - Univ. Estadual Paulista, Jaboticabal, SP, Brazil.*

The aim of this study was to investigate the effect of diets containing lipid sources with different fatty acid profiles on ruminal pH,  $\text{NH}_3\text{-N}$ , volatile fatty acids (VFA) and microbial protein synthesis. Ten Nellore steers with initial body weight of  $268 \pm 27$  kg, ruminally cannulated, were used in a double  $5 \times 5$  Latin square design (20 d of each period). Dietary treatments were: without fat (WF), palm oil (PO), linseed oil (LO), protected fat (PF; Lactoplus), and whole soybeans (WS). The roughage feed was corn silage (600 g/kg on a DM basis) plus concentrate (400 g/kg on a DM basis). To evaluate rumen fermentation parameters, rumen fluid samples (approximately 80 mL) were collected manually on the last day of the experimental period (20 d), both before supplying the diet (time zero) and 1, 2, 4, 6, 8, 10, 12, and 14 h after feeding. Microbial protein synthesis was calculated via urinary total excretion of purine derivatives (allantoin + uric acid). The diet with PF and WF increased the concentration of  $\text{NH}_3\text{-N}$  ( $P < 0.001$ ); however, the diet did not change in VFAs ( $P > 0.05$ ), such as the molar percentage of acetate (ACE), propionate (PROP), butyrate and the ACE:PROP ratio. There was a tendency ( $P = 0.06$ ) to change ruminal pH values and total VFA ( $P = 0.092$ ) with lipid diets. The higher  $\text{NH}_3\text{-N}$  and tendency to reduce pH values in the WF and PF diets could be linked to the higher DMI (nitrogen and organic matter), and consequently higher ruminal fermentation. The higher production of microbial N ( $P = 0.030$ ) was in the animals fed WF, LO and WS, whereas animals of the PO diet had the lowest production. The higher microbial protein synthesis was found in animals on the diet with LO and WS ( $P = 0.040$ ). Diets with some type of protection (PF and WS) caused less disturbance on ruminal fermentation.

**Key Words:** pH, ruminal fermentation, volatile fatty acid

**M446 The effect of carbohydrate source in a urea-based liquid supplement on ruminal fermentation and methane production of wintering beef cows fed low-quality forage.** A. C. Conway<sup>1</sup>, J. J. Michal<sup>1</sup>, J. S. Chang<sup>2</sup>, B. Carter<sup>3</sup>, M. E. Benson<sup>1</sup>, T. Bodine<sup>4</sup>, and K. A. Johnson\*<sup>1</sup>, <sup>1</sup>Department of Animal Sciences, Pullman, WA., <sup>2</sup>Korea National Open University, Seoul, Korea, <sup>3</sup>Performix Nutrition Systems, Nampa, ID, <sup>4</sup>Northwest Research & Nutrition, LLC, Yakima, WA.

The objectives of this study were to investigate the effects of 2 readily fermentable carbohydrate (RFC) sources in a urea-based liquid supplement on ruminal fermentation characteristics. Four ruminally cannulated Angus cows were fed 11.8 kg of bluegrass straw (BGS), 0.78 kg chopped triticale (TRIT), and 0.56 kg supplement on a DM basis. The experiment was performed in a Latin square design with repeated measures. Treatments were alfalfa hay (CON), a molasses-urea liquid supplement (MOL), a glycerol-urea liquid supplement (GLY) and a mixture of MOL and GLY (50/50). Ruminal fluid samples were collected at 0, 2, 4, 8, 12, 16, 20, and 24 h after feeding to analyze pH, VFA, and ruminal ammonia ( $\text{NH}_3\text{-N}$ ) concentration. Ruminally incubated samples of BGS and TRIT were removed at 0, 6, 12, 24, 48, and 72 h after feeding to assess digestibility. Ruminal fluid was collected for bacterial speciation. Disappearance of BGS DM, OM and CP were unaffected ( $P < 0.05$ ). Disappearance of NDF (48.2, 48.5, 50.3 versus  $44.2 \pm 2.0$ , respectively) and ADF (47.1, 47.5, 49.0 versus  $43.9 \pm 3.4$  respectively) in BGS increased 4–6% with MOL, GLY, and 50/50 ( $P = 0.03$ ) compared with CON. There was no treatment effect for digestibility of TRIT ( $P < 0.05$ ). Total VFA concentration, VFA ratios, and ruminal pH were unaffected ( $P < 0.05$ ), but MOL, GLY, and 50/50 increased  $\text{NH}_3\text{-N}$  concentration ( $P = 0.007$ ). Average  $\text{CH}_4$  emissions ( $192.13 \pm 25.6$  g/d) and methane yield (g  $\text{CH}_4$ /g gross energy intake; 5.2–5.5%) were unaffected ( $P < 0.05$ ). Microbial species varied over time ( $P < 0.01$ ) but not with RFC source. The improved fiber degradation in BGS indicates a GLY-based supplement is as effective as the standard MOL-based liquid supplement for wintering cows.

**Key Words:** liquid supplement, glycerol, methane

**M447 Effect of varying type of forage and feeding times of rumen degradable nitrogen sources on the production, digestibility, feeding behavior and rumen metabolites of lactating dairy cows.** Mustafa Hajilou\*<sup>1</sup>, Hamid Reza Mirzaei Alamouti<sup>1</sup>, Mehdi Ganjkanlou<sup>2</sup>, Hamid Amanlou<sup>1</sup>, and Mehdi Dehghan Banadacki<sup>2</sup>, <sup>1</sup>Department of Animal Science, University of Zanjan, Zanjan, Iran, <sup>2</sup>Department of Animal Science, Campus of Agriculture and Natural Resources, University of Tehran, Karaj, Iran.

This study was designed to evaluate effects of varying type of forage and feeding times of rumen degradable nitrogen sources on milk production, digestibility, feeding behavior and rumen metabolites of lactating dairy cows. Twelve multiparous ( $100 \pm 10$  d in milk) Holstein dairy cows were used in incomplete Latin square design (five 21-d periods) with a  $2 \times 3$  factorial arrangement of treatments. All diets had a 50:50 forage-to-concentrate ratio, contained 16% CP, and were formulated to be equal in rumen-degradable protein. Treatments include different alfalfa hay-to-corn-silage ratios (35:15 and 15:35) and different feeding times of the rumen degradable nitrogen sources: 1- total mixed ration (TMR) was offered once daily in the morning (0900); 2- part of soybean meal was offered at 2100; 3- part of urea was offered at 2100. Data were analyzed using the mixed procedures of SAS. Diets based on corn silage and feeding soybean meal at night increased milk and ECM yield ( $P < 0.05$ ) (ALFA1 = 32.06, ALFA2 = 33.95, ALFA3 = 32.25, CORN1 =



32.69, CORN2 = 34.17, CORN3 = 33.91; SEM = 0.55). Dry matter, organic matter and neutral detergent fiber digestibility numerically increased in cows feed corn silage diets ( $P < 0.09$ ) and not affected by feeding times of rumen degradable nitrogen sources ( $P < 0.25$ ). Feeding urea at night increased ruminating and chewing time (min/d and min/kg of NDF intake) in alfalfa hay based diets ( $P < 0.05$ ), but not in corn silage based diets. Standing ruminating (min/d) was higher for alfalfa hay fed cows ( $P < 0.01$ ) and resting ruminating (min/d) was higher for corn silage fed cows ( $P < 0.01$ ). Total volatile fatty acids and molar proportions not affected by treatments ( $P > 0.15$ ). Corn silage based diets improved milk production and digestibility.

**Key Words:** forage, Holstein dairy cow, nitrogen source

#### **M448 Effects of intravenous infusion of olive oil, safflower oil, and flaxseed oil on milk fatty acid composition in dairy cows.**

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The objective of this study was to determine the effects of intravenous infusion of olive oil, safflower oil and flaxseed oil in various combinations on milk fat composition in lactating dairy cows. Eight Chinese Holstein cows ( $101 \pm 11$  DIM) were used in a replicated  $4 \times 4$  Latin square design. Each period lasted 12 d with 5 d of infusion and 7 d of adaptation. Treatments were jugular vein infusion of 1) 170 g/d of oil mixture contained olive and safflower oil (43.10% *cis*-9 C18:1, 42.58% *cis*-6 C18:2 and 1.08% *n*-3 C18:3, OS); 2) 167 g/d of oil mixture contained olive and flaxseed oil (43.69% *cis*-9 C18:1, 13.89% *cis*-6 C18:2 and 29.94% *n*-3 C18:3, OF); 3) 161 g/d of oil mixture contained safflower and flaxseed oil (15.12% *cis*-9 C18:1, 44.72% *cis*-6 C18:2 and 30.50% *n*-3 C18:3, SF); and 4) 224 g/d of oil mixture contained olive, safflower and flaxseed oil (33.02% *cis*-9 C18:1, 32.94% *cis*-6 C18:2 and 22.63% *n*-3 C18:3, OSF). Treatment emulsions were consisted of water, oil mixture, 10 g/L of soy lecithin and 25 g/L of glycerol. The volume of the emulsion was 2 L/d per cow. Infusion process lasted for 6 h per day. Milk samples were collected twice daily during the last 2 d of each 5 d infusion period. Data were analyzed by using the Mixed procedure of SAS. Significance was declared at  $P < 0.05$ . Milk fatty acid from OF had greater concentration of *cis*-9 C18:1 (26.29%) compared with 21.80% from SF and 22.25% from OSF (SEM = 1.04). The concentration of *cis*-6 C18:2 from OF (4.36%) had the lowest level ( $P < 0.0001$ ) compared with 6.77%, 7.07% and 7.00% from OS, SF and OSF (SEM = 0.21), respectively. The concentration of *n*-3 C18:3 in OS (0.66%) was lower ( $P < 0.0001$ ) than in OF (3.57%), SF (3.26%) and OSF (3.34%), SEM = 0.13. The concentrations of C18:0 and short- and medium-chain fatty acids were not changed through treatments, whereas C16:0 was lower ( $P = 0.0202$ ) in treatment OF (30.70%) than in treatment OS (32.62%), SEM = 0.74. Infusion of different oil combinations increased target fatty acids in milk and markedly altered milk fat composition.

**Key Words:** intravenous infusion, milk fatty acid, dairy cow

**M449 Pelleting-induced changes at different conditioning temperatures and times on metabolic characteristics of the proteins and feed milk value of co-products from bio-oil processing.** Xuewei Huang, Tom Scott, Colleen Christensen, Yajing Ban, Xinxin

Li, and Peiqiang Yu\*, *Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, SK, Canada.*

The dramatic increase in bio-oil production in Canada has resulted in millions of tonnes of different types of co-products: carinata meal and canola meal. Little research has been conducted to determine pelleting induced changes at different conditioning temperature and time on the metabolic characteristics of the proteins in co-products from bio-oil processing in dairy cattle. The objectives of this study were to investigate the effects of conditioning temperature (70, 80 and 90°C), time (50 and 75 s), and interaction (temperature  $\times$  time) during the pelleting process on the metabolic characteristics of the proteins, feed milk value, the total truly absorbed protein supply (DVE), and degraded protein balance (DPB) to dairy cattle and to compare between unprocessed mash and pellets in true protein supply to small intestine of dairy cows. The DVE/OEB system was applied in determining metabolic characteristics of the proteins, feed milk value, DVE and DPB values. The data were analyzed with a randomized complete block design (RCBD) with a  $3 \times 2$  factorial arrangement. Statistical analyses were performed through MIXED procedure of SAS 9.3. The results showed that the unprocessed co-product from bio-oil processing is a good source of the truly digested protein in small intestine (DVE: 170 g/kg DM) with DPB of 115 g/kg DM. It was unexpectedly found that the pelleting process under current conditions did not increased but reduced DVE and increased DPB values. Within pelleting processed treatments, there was no interaction ( $P = 0.64$ ) between conditioning temperature and time on the metabolic characteristics of the proteins. However, increasing conditioning temperature tended to decrease DPB ( $P = 0.051$ ) values of co-product pellets. Feed milk value of the co-products was reduced after pelleting process as well (3.4 vs. 3.0 kg milk per kg feed). In conclusion, pelleting with relative low temperature (70–90°C) decreased DVE, feed milk value, and increased DPB of co-products compared with raw co-products. Increasing conditioning temperature during pelleting tended to decrease the potential N loss of pelleted co-products.

**Key Words:** feed technology, bio-oil processing co-product, protein

#### **M450 Ruminal fermentation of Nellore steers fed different sources of forage in diets with crude glycerin in feedlot.**

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This study investigated the effect of alternative forages for corn silage such as sugar cane and sugar cane bagasse included in a similar forage NDF level (fNDF) in diets with crude glycerin (80.64% of glycerol) on ruminal fermentation of Nellore feedlot steers. Nine ruminally cannulated Nellore steers ( $300.0 \pm 30$ kg of BW and  $18 \pm 2$  mo of age) were used in a  $3 \times 3$  Latin Square experimental design with 3 treatments and 3 animals in 3 simultaneous replicates to evaluate the effect of different sources of forage in diets with crude glycerin (80.64% of glycerol) on ruminal pH and ammonia-N concentration. Experimental periods were 15 d (14 d for adaptation and 1 d for ruminal sampling). The treatments were different sources of forage (fixed 15% of NDF from forage; fNDF): corn silage (CS), sugar cane (SC) and sugar cane bagasse (SB), in diets with 10% (DM) of crude glycerin. Ruminal contents were obtained at 0, 2, 4, 6, 8, 10, and 12 h after feeding. Data were analyzed as a triple Latin Square design and repeated measurements on time using the PROC MIXED procedure of SAS. The least squares means were generated and

compared ( $P < 0.05$ ) using Tukey's test. The pH values of animals fed with SB (6.40) were greater compared with CS (6.08;  $P < 0.05$ ), which did not differ from animals fed with SC (6.27;  $P > 0.05$ ). Animals fed with SC presented lower values of  $\text{NH}_3\text{-N}$  (11.54;  $P < 0.05$ ) compared with animals fed with CS (15.42) and SB (15.40). Sugar cane and sugar cane bagasse included in 15% of fNDF in diets with crude glycerin (10% DM) altered ruminal parameters, however, maintained adequate conditions for animal performance.

**Key Words:** byproduct, corn silage, sugar cane

#### **M451 Comparison of ruminal microbial diversity and richness in whole rumen content, rumen liquid and solid fractions.**

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Whole rumen content, fluid or solid fractions have been used often to assess the rumen microbial diversity and richness; however, the suitability of using each fraction assessing the ruminal microbial diversity and richness has not been evaluated. To compare the bacterial profiles in each fraction, samples of rumen content from 6 cows were collected via fistula and squeezed through 4 layers of cheesecloth to get liquid and solid fractions before analyzing with the next generation sequencing technique. Results showed that the Operational Taxonomic Units (OTUs) numbers ( $4897 \pm 582$ ,  $5253 \pm 855$  and  $4860 \pm 615$  for whole content, liquid and solid fraction, respectively) and Simpson indices

( $0.99 \pm 0.008$ ,  $0.99 \pm 0.006$  and  $0.99 \pm 0.004$  for whole content, liquid and solid fraction, respectively) among whole content, liquid and solid fractions were similar ( $P > 0.05$ ). No statistical difference was found among the inner- or inter-group similarities for each kind of samples using Bray-Curtis metric ( $P > 0.05$ ). At Phylum level, Bacteroidetes and Firmicutes were the predominant bacteria and accounted for more than 90% of the microbes in all samples in our study. However, richness of Bacteroidetes in liquid fraction was  $64.29 \pm 0.03\%$  which was higher than that in solid fraction as  $48.25 \pm 0.04\%$  ( $P < 0.05$ ), while that of Firmicutes was  $31.72 \pm 0.03\%$  in liquid fraction which was lower compared with that of solid fraction as  $44.71 \pm 0.05\%$  ( $P < 0.05$ ). At genus level, on analyzing the top 9 bacteria (accounting for more than 43% in quantity in all samples), the fold change values of liquid fraction versus whole content (liquid/whole) were 0.51, 0.73, 0.76, 0.82, 1.07, 1.19, 1.20, 1.35, and 1.38 for *Coprococcus*, *Succiniclasticum*, *Butyrivibrio*, *Shuttleworthia*, YRC22, *Prevotella*, *Ruminococcus*, CF231, and *Oscillospira*, respectively. These findings indicate that using whole, fluid, or solid fractions to assess the microbial diversity generate similar results. However, the richness of predominant bacteria in phylum and genus may differ depending on the sample fraction type.

**Key Words:** rumen microbial diversity, rumen microbial richness, rumen content fraction

## Small Ruminant I

**M452 Chemical composition and quality of fresh lamb from rationally grazed hair and wool x hair sheep lambs as influenced by soy hull supplementation.** Beruk B. Lemma<sup>\*1</sup>, Jung Hoon Lee<sup>1</sup>, Stephan A. Wildeus<sup>2</sup>, Govind Kannan<sup>1</sup>, and Brou Kouakou<sup>1</sup>, <sup>1</sup>Fort Valley State University, Fort Valley, GA, <sup>2</sup>Virginia State University, Petersburg, VA.

This study was conducted to evaluate the effects of soy hull supplementation on the quality characteristics of fresh lamb from hair and wool x hair sheep lambs. Forty-seven lambs (5-mo old), 23 purebred hair sheep (Barbados Blackbelly or BB, BW = 16.2 ± 1.9 kg; St. Croix or SC, BW = 19.5 ± 2.0 kg) and 24 crossbred wool (Dorset, D) x hair (BB; BW = 21.7 ± 2.3 kg or SC; BW = 21.7 ± 2.9 kg) sheep lambs of both sexes rotationally grazed predominately tall fescue with or without soy hull supplementation during summer. Soy hull was provided at 2.0% of BW at individual feeding stations. After 90 d of grazing, lambs were harvested using standard procedures. After 24 h cooler storage (2°C), longissimus muscle (LM) pH was measured in individual carcasses. Each carcass was fabricated to obtained 2.5-cm thick loin chops for meat quality analysis. All data were analyzed as a completely randomized design with a 2 x 2 x 2 factorial treatment arrangement: breed type (pure- or cross-bred), supplement (with or without soy hull), and sex (male or female). The CIE L\* (lightness), a\* (redness), and b\* (yellowness) values of lamb chops were significantly influenced by supplementation, sex, and their interaction. Chops from supplemented lambs had higher ( $P < 0.001$ ) CIE a\* values than those from grazed-only lambs. The LM from supplemented lambs had higher ( $P < 0.001$ ) protein (22.4 vs 21.4%) and fat contents (2.88 vs 2.47%) than that from grazed-only lambs. The percent metmyoglobin and thiobarbituric acid reactive substances (TBARS) values of LM from lamb chops were significantly affected by sex, breed type, and breed type x sex. The LM from purebred lambs had a higher ( $P < 0.05$ ) level of TBARS (0.32 vs 0.27 mg MDA/kg) than that of crossbred lambs. Chops from supplemented lambs had lower ( $P < 0.01$ ) shear values than those from grazed-only lambs. The results indicate that soy hull supplementation can improve color and texture property of fresh hair sheep lambs grazing fescue pasture regardless of breed type.

**Key Words:** hair and wool sheep, meat quality, soy hull

**M453 Effects of feeding entrapped fish oil in a chemically treated protein matrix on milk composition of lactating goats.** Jung Hoon Lee<sup>\*1</sup>, Christina R. Alfred<sup>1</sup>, Beruk B. Lemma<sup>1</sup>, Brou Kouakou<sup>1</sup>, and Byung J. Min<sup>2</sup>, <sup>1</sup>Fort Valley State University, Fort Valley, GA, <sup>2</sup>Tuskegee University, Tuskegee, AL.

Because of the potential benefit to human health, there is considerable interest in increasing omega-3 fatty acid contents in milk fat. However, efforts to increase the levels of eicosapentaenoic (EPA; C20:5n-3) and docosahexaenoic (DHA; C22:6n-3) acids had limited success because they undergo biohydrogenation in rumen. This study investigated the effect of entrapped fish oil (EFO) containing EPA and DHA acids on milk and milk fat compositions of lactating goats. The EFO were prepared with fish oil, defatted soy flour, and acetaldehyde. Nine Saanen lactating goats (4 yr old; BW = 48.3 ± 2.26 kg) were divided into groups of 3 and randomly assigned to 3 diets using a 3 x 3 Latin square design with 14-d periods. The 3 diets consisted of 95% basal diet containing alfalfa meal, yellow corn and soybean meal, plus 5.0% lipid from either poultry fat (PF), fish oil (FO) or EFO. Each period consisted of a 10-d adjustment

to assigned diets followed by 4-d of milking collection. The designated diet was provided once daily ad libitum at individual feed stations. The collected milk samples from each goat were analyzed for basic nutrient,  $\alpha$ -tocopherol and fatty acid composition. All data were analyzed as a 3 x 3 Latin square design. Lactating goats fed a PF containing diet had a higher ( $P < 0.01$ ) content of milk fat compared with goats fed either a FO- or an EFO-containing diet (3.98 vs 3.34 or 3.22%). No significant differences were found in the amount of  $\alpha$ -tocopherol (0.87–0.90  $\mu\text{g/mL}$ ) in the milk samples from the 3 different diets. Compared with lactating goats fed PF-diet, goats fed either FO- or EFO- diets had higher ( $P < 0.01$ ) concentrations of EPA (C20:5n-3, 0.30 vs. 0.81 or 1.66%), docosapentaenoic (C22:5n-3, 0.24 vs. 0.77 or 0.98%), and DHA (C22:6n-3, 0.32 vs. 1.21 or 1.86%) in milk. Furthermore, the concentration of EPA in milk from goats fed EFO-diet was higher ( $P < 0.01$ ) than that from goats fed a FO-diet. The results indicate that feeding lactating goats with entrapped fish oil in a chemically treated protein matrix might increase the deposition of omega-3 fatty acids in their milk.

**Key Words:** lactating goat, milk fat, eicosapentaenoic and docosahexaenoic acids

**M454 Fatty acid composition and  $\alpha$ -tocopherol content of blood serum from lactation goats fed entrapped fish oil in a chemically treated protein matrix.** Christina R. Alfred<sup>\*1</sup>, Jung Hoon Lee<sup>1</sup>, Beruk B. Lemma<sup>1</sup>, Brou Kouakou<sup>1</sup>, and Byung J. Min<sup>2</sup>, <sup>1</sup>Fort Valley State University, Fort Valley, GA, <sup>2</sup>Tuskegee University, Tuskegee, AL.

This study evaluated the effects of feeding entrapped fish oil containing eicosapentaenoic (EPA; C20:5n-3) and docosahexaenoic (DHA; C22:6n-3) acids in chemically treated protein on fatty acid compositions and  $\alpha$ -tocopherol concentrations of blood serum in lactating goats. The entrapped fish oil (EFO) were prepared with fish oil, defatted soy flour, and acetaldehyde. Nine Saanen lactating goats (4 yr old; BW = 48.3 ± 2.26 kg) were divided into groups of 3 and randomly assigned to 3 diets using a 3 x 3 Latin square design with 3 14-d periods. Experimental diets consisted of 95% basal diet, containing alfalfa meal, yellow corn and soybean meal, with 5.0% of oil/fat from either poultry fat (PF), fish oil (FO) or EFO. The designated diet was provided once daily ad libitum at individual feed stations. Blood samples were collected from individual goats at the end of each feeding period which were analyzed for  $\alpha$ -tocopherol and fatty acid composition. All data were analyzed as a 3 x 3 Latin square design. Average daily intake of diet containing EFO was lower ( $P < 0.01$ ) than diets containing either PF or FO (1.04 vs 2.56 or 1.90 ± 0.300 kg). Consequently, the ADG of goats fed EFO diet was lower ( $P < 0.05$ ) than the other 2 diets. The blood serum from goats fed EFO diet tended ( $P = 0.08$ ) to have a higher amount of  $\alpha$ -tocopherol (2.31 vs 1.89 or 1.90  $\mu\text{g/mL}$ ) than did those from goats fed either PF or FO diet. Compared with goats fed PF-diet, goats fed either FO- or EFO- diet had higher ( $P < 0.01$ ) concentrations of EPA (C20:5n-3, 1.76 vs 10.57 or 13.23%), docosapentaenoic (C22:5n-3, 1.32 vs 2.02 or 1.93%), and DHA (C22:6n-3, 1.95 vs 4.58 or 5.38%) acids in blood serums. Furthermore, the concentrations of EPA and DHA in blood serums from either EFO- or FO-diet fed goats were similar even with a limited intake of EFO-diet. Thus, feeding lactating goats with entrapped fish oil in chemically treated protein might protect n-3 fatty



acids and  $\alpha$ -tocopherols from ruminal degradation and subsequently increased those contents in the blood serum of lactating goats.

**Key Words:** lactating goat, blood serum, eicosapentaenoic and docosahexaenoic acids

**M455 Fatty acid composition of different fat depots from meat goats supplemented with tannin-rich pine bark.** Beruk B. Lemma\*<sup>1</sup>, Jung Hoon Lee<sup>1</sup>, Byeng R. Min<sup>2</sup>, Govind Kannan<sup>1</sup>, and Brou Kouakou<sup>1</sup>, <sup>1</sup>Fort Valley State University, Fort Valley, GA, <sup>2</sup>Tuskegee University, Tuskegee, AL.

This research assessed the effect of feeding ground pine bark (PB, *Pinus* spp.), containing up to 13% condensed tannins (CT) on a DM basis, on different fat depots in meat goats. Twenty-four intact male Kiko goats (8 mo of age; BW = 39.7 ± 2.55 kg) were grazed in a winter rye grass-dominant pasture, and supplemented either bermudagrass hay (BG) or PB pellet. Each supplementation (n = 12 goats/treatment) consisted of alfalfa pellet (16.9% CP, 3.30% ether extract or EE, 48.7% NDF), molasses, and mineral mixtures with either BG (20.5% CP, 4.44% EE, 40.9% NDF) or PB powder (9.10% CP, 3.35% EE, 59.0% NDF), which was provided at 1.5% of BW at individual feeding stations. After 50 d grazing, goats were harvested. Intramuscular, subcutaneous, and kidney fats were obtained from each carcass. Total lipids from each fat depot sample were extracted by the chloroform-methanol method. Extracted lipids were prepared for the fatty acid methyl esters (FAME) and then analyzed by a gas chromatography. All data were analyzed as a completely randomized design. Palmitic (C16:0), stearic (C18:0), oleic (C18:1n9), and linoleic (C18:2n6) acids were the major fatty acids in the intramuscular fats from goats supplemented with either BG-hay or PB-pellet (85.9 vs 86.0% of total fatty acid). The subcutaneous and kidney fats consisted mainly of myristic (C14:0), C16:0, C18:0, and C18:1n9 acids, which accounted for 79.1 and 81.9% vs 85.3 and 84.7% of total fatty acids in the BG-hay and PB-pellet supplemented goats, respectively. No significant differences were found in these major fatty acids in the 3 different fat depots from goats fed either BG-hay or PB-pellet. However, compared with goats fed BG-hay, goats fed PB-pellet had lower ( $P < 0.05$ ) concentrations of margaric (C17:0), margaroleic (C17:1n9), and conjugated linoleic (C18:2, CLA) acids in intramuscular fats; a higher ( $P < 0.05$ ) concentration of eicosapentaenoic (C20:5n3) acid in subcutaneous and kidney fats. The results indicate that the supplementation of PB did not change the major fatty acids in the different fat depots in meat goats.

**Key Words:** goat, pine bark, fatty acid profile

**M456 Influence of dietary condensed tannins from pine bark and/or sericea lespedeza on chemical composition and quality of goat meat.** Travet Witherspoon<sup>1</sup>, Jung Hoon Lee\*<sup>2</sup>, Beruk B. Lemma<sup>1</sup>, Byeng R. Min<sup>3</sup>, Govind Kannan<sup>2</sup>, and Brou Kouakou<sup>2</sup>, <sup>1</sup>Thomasville High School, Thomasville, GA, <sup>2</sup>Fort Valley State University, Fort Valley, GA, <sup>3</sup>Tuskegee University, Tuskegee, AL.

Even though pine bark (PB) and sericea lespedeza (SL) contain a high amount of condensed tannins (CT), they have been recognized as an economical source of nutrients for goats. Yet their effect on the quality of goat meat (chevon) has not been intensively studied. This study was conducted to determine the quality characteristics of chevon from meat goats fed CT-containing PB and/or SL diets. Twenty-four Kiko × Boer male goats (8 mo of age; BW = 37.3 ± 2.56 kg) were randomly assigned to a feeding trial. Goats were randomly divided into 4 groups with 6 goats in each group. Each group was assigned randomly 1 of 4

diets: 30% bermudagrass (BG) hay (control, BG-diet); 30% PB pellet (PB-diet); 30% SL pellet (SL-diet); and 15% PB + 15% SL pellet (PS-diet) with the remainder of each diet made up of 70% alfalfa pellets mixed with a commercial molasses-based sweet feed. After a 50 d feeding trial, goats were slaughtered using standard procedures. After 24 h cooler storage (2°C), longissimus muscle (LM) pH was measured from individual carcasses and each carcass was fabricated to obtain 2.5-cm thick loin chops for meat quality analysis. All data were analyzed as a completely randomized design. The LM pH values of goats fed the 4 different diets were not significantly different. Chops from goats fed a PB-diet had higher ( $P < 0.05$ ) CIE L\* (lightness) and a\* (redness) values than those from goats fed the other 3 different diets. The LM from goats fed a PS-diet had higher ( $P < 0.05$ ) moisture and ash contents than that from goats fed a PB- and a BG-diet, respectively. Furthermore, the LM from goats fed a PB-diet had higher ( $P < 0.05$ ) percent metmyoglobin and thiobarbituric acid reactive substances (TBARS) values than that from goats fed the other diets. Compared with goats fed BG-diet, goat fed either a SL- or a PB-diet had lower ( $P < 0.05$ ) shear values (3.74 vs 3.26 or 3.03 ± 0.13 kg) in cooked loin chops. The results indicate that a diet containing higher amounts of dietary tannins from PB might improve the texture property and color of goat meat, whereas the resulting chevon might be more susceptible to the lipid oxidation.

**Key Words:** chevon quality, pine bark, sericea lespedeza

**M457 Fatty acid composition of different fat depots from meat goats supplemented with either tannin-rich pine bark and sericea lespedeza alone or in combination.** Christina R. Alfred<sup>1</sup>, Jung Hoon Lee\*<sup>1</sup>, Travet Witherspoon<sup>2</sup>, Beruk B. Lemma<sup>1</sup>, Byeng R. Min<sup>3</sup>, Govind Kannan<sup>1</sup>, and Brou Kouakou<sup>1</sup>, <sup>1</sup>Fort Valley State University, Fort Valley, GA, <sup>2</sup>Thomasville High School, Thomasville, GA, <sup>3</sup>Tuskegee University, Tuskegee, AL.

In vitro studies showed that dietary tannins inhibit the growth of microorganisms that are responsible for ruminal biohydrogenation of fatty acids. Both pine bark (PB) and sericea lespedeza (SL) contain high amounts of condensed tannins (CT). However, their effect on the fatty acid profile of different fat depots in meat goats has not been studied. This study was conducted to determine the fatty acid composition of different fat depots from meat goats fed CT-containing either PB and SL alone or in combination diets. Twenty-four crossbred (Kiko × Boer) goats (BW = 37.3 ± 2.56 kg) were randomly assigned to one of 4 diets: 30% bermudagrass (BG) hay (control, BG-diet); 30% PB pellet (PB-diet); 30% SL pellet (SL-diet); and 15% PB + 15% SL pellet (PS-diet) with the remainder of each diet made up of 70% alfalfa pellets mixed with a commercial molasses-based sweet feed. After 50 d feeding, goats were harvested. Intramuscular, subcutaneous, and kidney fats were obtained from each carcass. Total lipids from each fat sample were extracted, and prepared for the fatty acid methyl esters (FAME). The FAME were analyzed by a gas chromatography. All data were analyzed as a completely randomized design. Compared with goats fed BG-diet, goats fed either PB- and SL-diet had a lower ( $P < 0.05$ ) concentration of stearic (C18:0) acid (16.93 vs 14.93 or 15.01%) in intramuscular fat. Goats fed PB-diet had higher concentrations of linoleic (C18:2n-6; 5.55 vs 8.21%) and linolenic (C18:3n-3; 0.57 vs 0.91%) acids in intramuscular fat compared with goat fed BG-diet. In kidney fat, goats fed either PB- or SL-diet had a lower ( $P < 0.05$ ) concentration of margaroleic (C17:1n-9) acid (0.45 vs 0.33 or 0.36%), but goats fed PB-diet had higher a concentration of C18:2n-6 (2.50 vs 5.48%) acid compared with goats fed BG-diet. Goats fed PS-diet had a higher concentration of C18:2n-6 acid (1.48 vs 2.75%) in subcutaneous fat compared with those fed BG-diet. The

results indicated that goats fed PB might increase the deposition of C18 polyunsaturated fatty acids in fresh goat meats.

**Key Words:** pine bark, sericea lespedeza, fatty acid profile of goat

**M458 Influence of reproductive stage and breed on the hemogram of sheep.** Leilson R. Bezerra\*<sup>1,2</sup>, Jacira N. C. Torreão<sup>1</sup>, Carlo A. T. Marques<sup>1</sup>, Marcos J. Araujo<sup>1</sup>, and Ronaldo L. Oliveira<sup>2</sup>, <sup>1</sup>Federal University of Piauí, Bom Jesus, Piauí, Brazil, <sup>2</sup>Federal University of Bahia, Salvador, Bahia, Brazil.

The objective with this study was to evaluate the influence of reproductive stages (pregnancy, puerperium) and breed (Santa Inês and Morada Nova) on the hemogram. We used 20 Santa Inês sheep and 20 Morada Nova sheep diagnosed with positive pregnancy and distributed in a completely randomized design, factorial arrangement 2 × 2 (reproductive stage × breed) in a split-plot arrangement over time (blood collection). During the experimental period, the animals were kept in paddocks containing *Andropogon gayanus*, which was enclosed in the late afternoon for supplementation consisting of cracked corn (70%), soybean meal (25%) and mineral supplement (5%), based on the dry matter. Blood samples were collected by jugular venipuncture every 14 d in the morning before the animals were released to pasture, starting from 1st day of gestation up to the 90th day of lactation (when the lambs were weaned). The hemogram analysis consisted of global erythrocyte counts, packed cell volume or hematocrit, hemoglobin, mean corpuscular volume (MCV), mean corpuscular hemoglobin concentration (MCHC), total plasma proteins (TPP) and total leukocyte counts. The means were compared with the Student-Newman-Keuls test, and was considered significant effect when  $P < 0.05$ . There was no influence of breed in erythrocyte count ( $P = 0.4718$ ) and TPP ( $P = 0.2609$ ). The Morada Nova breed showed higher hemoglobin ( $P < 0.0001$ ) and hematocrit ( $P = 0.0024$ ) and total leukocyte count ( $P < 0.0001$ ) than Santa Inês breed. There was increased in global erythrocyte counts ( $P < 0.0001$ ) in pregnancy and a reestablishment during the puerperium period. The pregnancy period reduced hemoglobin ( $P < 0.0001$ ), hematocrit ( $P = 0.0024$ ), MCV ( $P < 0.0001$ ) and MCHC ( $P < 0.0001$ ) and puerperium period increased hematocrit ( $P = 0.0356$ ) and total leukocyte count ( $P = 0.0243$ ). The TPP also increased ( $P < 0.0001$ ) during pregnancy period to compensate for the high nutritional requirements of the fetus, and these levels remained high until the end of the puerperium phase when the lambs were weaned. There is recuperation of blood values such as TPP in puerperium, which increase during this period to compensate for the high nutritional demands of the fetus in pregnancy.

**Key Words:** hemoglobin, indigenous sheep, parturition

**M459 Production and chemical composition of milk from goats fed different levels of buriti oil.** Leilson R. Bezerra\*<sup>1,3</sup>, Jasiel S. Morais<sup>1,2</sup>, Ronaldo L. Oliveira<sup>3</sup>, Aderbal M. A. Silva<sup>2</sup>, and Ricardo L. Edvan<sup>1</sup>, <sup>1</sup>Federal University of Piauí, Bom Jesus, Piauí, Brazil, <sup>2</sup>Federal University of Campina Grande, Patos, Paraíba, Brazil, <sup>3</sup>Federal University of Bahia, Salvador, Bahia, Brazil.

Buriti (*Mauritia flexuosa*) oil can be a great energy source for goats in lactation. This study aimed to determine the most effective level of buriti oil to use in supplements for lactating goats. Eight Anglo-Nubian crossbreed goats in 50 d of lactation were grouped into 2 4 × 4 double simultaneous Latin squares being 4 experimental periods of 21 d each, with 16 adaptation days and 5 d data collection. The treatments were 4 diets containing 0–15, 30 or 45 g·kg<sup>-1</sup> of buriti oil in the total diet. The diets, a total mixed ration, were composed of corn silage and con-

centrate made with: corn, soybean, mineral supplement and buriti oil and were formulated according to NRC (2007) recommendations to lactating goats with production of 2 kg<sup>-1</sup>·goat<sup>-1</sup>·day<sup>-1</sup> and 4% milk fat. Animals were fed twice a day immediately after milking, at 8:00 and 16:00 h. The data were submitted to ANOVA and regression, using the MIXED model procedure from the statistics program SAS 9.1.2. The dry matter intake was reduced linearly ( $Y = -0.16x + 2.1$ ,  $r^2 = 0.914$ ) by the addition of buriti oil ( $P < 0.05$ ). The milk production and milk production corrected for 4% of fat were not affected by the addition of buriti oil, but the corrected milk production of total solids ( $Y = 0.033x + 1.152$ ,  $r^2 = 0.898$ ) were increased linearly ( $P = 0.0378$ ). In regards to milk constituents, the fat milk ( $Y = 0.237x + 4.465$ ,  $r^2 = 0.985$ ) was increased linearly ( $P < 0.0001$ ) by the buriti oil levels in the total diet. The mean concentrations of total protein (37.63 g·day<sup>-1</sup>) casein (31.80 g·day<sup>-1</sup>) lactose (45.09 g·day<sup>-1</sup>) and urea (18.74 mg·dL<sup>-1</sup>) were not affected by the addition of buriti oil in diet ( $P > 0.05$ ). The inclusion of buriti oil at 45 g·kg<sup>-1</sup> in a total mixed ration increases corrected milk production of total solids and milk fat of Anglo-Nubian crossbreed goats.

**Key Words:** fat milk, nutrition, palm tree

**M460 The effects of administering a fibrolytic probiotic made from moose rumen bacteria to neonatal lambs.** Suzanne L. Ishaq\*<sup>1</sup>, Christina J. Kim<sup>1</sup>, and André-Denis G. Wright<sup>2</sup>, <sup>1</sup>University of Vermont, Burlington, VT, <sup>2</sup>University of Arizona, Tucson, AZ.

The present study investigated the effect of a fibrolytic probiotic using bacteria isolated from the rumen of the North American moose (*Alces alces*). Bacteria from moose were chosen for their ability to digest a variety of plant polysaccharides and lignin. Twenty 5-d old lambs (mean 5.9 ± 0.2 kg) were divided into probiotic and control groups, with probiotic receiving 5 bacterial isolates orally through weaning (9 wk), after which the lambs were put on pasture. Neither weight gain nor wool quality was improved in lambs given a probiotic, however, dietary efficiency was increased as evidenced by the reduced feed intake (and rearing costs) without a loss to weight gain. Total VFAs were not significantly different between groups. Acetate, propionate, butyrate, and ethanol were significantly ( $P < 0.05$ ) higher in experimental lambs at certain weeks. The acetate to propionate ratio was statistically lower in the experimental group at wk 9, 11, and 15, which was previously shown to indicate increased dietary efficiency. Sampling coverage decreased over time, while Shannon, Inverse Simpson, CHAO, and ACE increased over time, which is a function of the increasing diversity of microbiota as the rumen develops. The experimental group had a higher diversity at the beginning of the experiment. Bacteroidetes was the most prevalent phylum (38–73% of total sequences) in both groups for the duration of the study, with the exception of the first sampling of the control group. Firmicutes was the second most prevalent (23–59%). In the control group, Bacteroidetes increased while Firmicutes decreased for the first 3 sampling points, while the experimental had a general trend of decreasing Bacteroidetes and increasing Firmicutes over time. *Prevotella* was the most prevalent genus, followed by *Butyrivibrio* and *Ruminococcus*. Protozoal densities increased over time and stabilized, but methanogen densities varied greatly in the first 6 mo of life for lambs. This is likely due to the changing diet and bacterial populations in the rumen.

**Key Words:** lamb, probiotic, moose

**M461 Factors affecting feed efficiency in dairy goats.** Tadeu Silva de Oliveira\*<sup>1</sup>, Ricardo Augusto Mendonça Vieira<sup>1</sup>, Danielle Ferreira Baffa<sup>2</sup>, Aberto Magno Fernandes<sup>1</sup>, and José Carlos Pereira<sup>2</sup>, <sup>1</sup>Universidade Estadual do Norte Fluminense-Darcy Ribeiro, Rio

de Janeiro, Brazil, <sup>2</sup>Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.

The objective of this study was to present some factors affecting feed efficiency in dairy goats. For our study, individual and average data from performance experiments with lactating goats were used. The following variables were evaluated: gross feed efficiency, adjusted feed efficiency, dry matter intake, milk-yield, 3.5% fat-corrected milk yield, dry matter digestibility, dietary neutral detergent fiber content, different roughage-to-concentrate ratios and body weight. The statistical analyses involved the application of descriptive and dispersion measures besides Pearson correlation coefficient PROC CORR and linear regression analysis PROC MIXED of SAS. However, because average daily gain (ADG) and dry matter intake (DMI) are variables of normal distribution, and because their division generates a Cauchy distribution variable it is more appropriate to generate a normal distribution variable from the transformation of the 2 variables according PROC TRANSREG of SAS. The analyzed variables were highly correlated with feed efficiency. The dry matter intake, dietary NDF content, roughage to-concentrate ratio and live weight had a negative correlation ( $P < 0.05$ ) with feed efficiency, indicating that increase in these variables inversely affecting FE, reducing it. In contrast, milk yield, 3.5% fat-corrected milk yield and dry matter digestibility positively affected ( $P < 0.05$ ) FE. Fat corrected milk yield was positively correlated with FE ( $R = 0.89$ ;  $P < 0.001$ ) as was milk yield ( $R = 0.83$ ;  $P < 0.001$ ). Feed efficiency (FE) increased by 0.061 points with every 1-L increase in milk yield. Fat corrected milk is a better indicator of feed efficiency because of the difference in energy represented by milk fat percentage. In conclusion, among these factors, standardization of the milk fat appeared to be the most efficient in describing the feed efficiency in lactating goats. Funded by FAPERJ, CAPES, and CNPq.

**Key Words:** dry matter intake, goat, milk yield

**M462 Variation of masses of body fat and protein and visceral organs of alpine goats in the first eight weeks of lactation.** Tadeu Silva de Oliveira<sup>\*1</sup>, Ricardo Augusto Mendonça Vieira<sup>1</sup>, Marcelo Teixeira Rodrigues<sup>2</sup>, Aberto Magno Fernandes<sup>1</sup>, and Danielle Ferreira Baffa<sup>2</sup>, <sup>1</sup>Universidade Estadual do Norte Fluminense-Darcy Ribeiro, Rio de Janeiro, Brazil, <sup>2</sup>Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.

The use constant values to represent the loss of energy due to mobilization of fat and protein as considered by INRA (1989), AFRC (1993) and Morand-Fehr and Hervieu (1999) does not reflect in a reliable dynamic mobilization. It should be considered that most data used for that inference were based on short-term calorimetric studies. Thus, it is essential to quantitatively know low body energy reserves are transformed into milk energy, and low energy mobilized and consumed concur for energy balance in the lactation course. This study aimed to measure changes of body fat and protein masses, and also quantify mass-changes of visceral organs of Swiss Alpine does throughout the first 8 weeks of lactation. Fifty-one multiparous does were allotted in a completely randomized design with 6 replications and 8 treatments (weeks of lactation). Animals were slaughtered sequentially to measure the mass of fat and protein retained in the body, as well as the weight of the visceral organs. The data were analyzed by using the MIXED procedure of SAS, in which 2 covariance structures (homogeneous and heterogeneous variances) and the likelihood of linear, quadratic, and cubic models were evaluated. The empty body weight (EBW) decreased ( $P < 0.05$ ) over the 8 weeks of lactation (from 56.8 to 35.6 kg) and carcass fat also decreased ( $P < 0.05$ ) from 5.16 to 2.10 kg and from 9.1 to 5.9% of EBW. Body fat reduced from 24.13 to 13.51%, as well as protein (from 14.18 to

14.75%), mostly because of the mobilization of noncarcass fat (from 28.58 to 15.73%) and protein (from 12.42 to 13.20%). The visceral fat mass was mobilized with greater intensity from the third to the eighth week of lactation, i.e., 4.33 kg reduced to 1.40 kg in the period or 7.57% reduced to 3.01% of EBW. There was no effect ( $P > 0.05$ ) on liver mass, rumen, omasum, abomasum, small intestine, and large intestine during the first 8 weeks of lactation, but there was a decrease ( $P < 0.05$ ) in the uterus (from 1.63 to 0.14 kg) and mammary gland masses (from 2.84 to 1.48 kg). In conclusion, Swiss Alpine does in early lactation mobilized energy reserves not only from carcass and internal fat but also from visceral organs. Funded by FAPERJ, CAPES and CNPq.

**Key Words:** adipose tissue, goats, mobilization

**M463 Meat composition and yield of carcass and non-carcass components in crossbred lambs fed frying soybean oil.** F. O. Scarpino-van Cleef\*, E. H. C. B. van Cleef, M. T. C. Almeida, A. P. D' Aurea, H. L. Perez, and J. M. B. Ezequiel, São Paulo State University, Jaboticabal, São Paulo, Brazil.

Twenty-four crossbred uncastrated male Santa Ines × Dorper lambs (24.9 ± 2.4 kg BW) were used to evaluate meat composition and yield of carcass and non-carcass components when fed diets containing supplementary oil sources. Treatments consisted of a control diet (CON) containing 40% corn silage, 10.8% corn grain, 9.8% soybean hulls, 37.45% sunflower meal, 0.55% urea, 1.4% mineral premix, and 2 diets containing additional 6% soybean oil (SOY) or 6% residual soybean frying oil (FRY). Lambs were blocked by initial body weight (BW), and randomly assigned to 24 individual feedlot pens. Animals were harvested when they reached 35 kg BW. Carcass and edible non-carcass components were separated and weighed. Total edible non-carcass components yield (TENC) was obtained as the sum of weights of blood, tongue, lungs + trachea, liver + gall bladder, heart, kidneys, gastrointestinal tract (reticulum, rumen, omasum, abomasum and intestines), and abdominal and kidney fats. Total yield of usable products (TUP) was calculated as the sum of hot carcass weight and TENC. A sample of *Longissimus* muscle was collected between 12th and 13th ribs, and evaluated for moisture, crude protein, ether extract and ash contents. Data were analyzed as a randomized block design using a mixed model, and the contrasts control × oil treatments and soybean oil × frying oil were evaluated. TENC values were 6.62, 6.56, and 6.35 kg; and TUP values were 22.27, 21.58, and 22.19 kg for CON, SOY and FRY, respectively ( $P > 0.10$ ). The weight of the liver of oil-fed lambs was greater than the ones fed CON ( $P = 0.04$ ). The weight of the spleen tended to be greater in lambs fed FRY ( $P = 0.06$ ), while the other individual non-carcass components were not affected by treatments. The ether extract of meat tended to be greater in lambs fed diets containing oil, regardless the source ( $P = 0.07$ ). The meat moisture, mineral matter, or crude protein contents were unaffected by dietary oil supplementation. Data indicate soybean-frying oil is a potential alternative energy ingredient in diets for feedlot lambs and that the inclusion of 6% of this oil does not depreciate carcass and edible non-carcass components yields, and it increases meat intramuscular fat slightly.

**Key Words:** byproduct, lipid, non-carcass component

**M464 Milk production, quality, and components measured in lactating dairy goats supplemented with OmniGen-AF.** Angela D. Rowson<sup>\*1</sup>, Shelby A. Armstrong<sup>1</sup>, Lane O. Ely<sup>2</sup>, and Derek J. McLean<sup>1</sup>, <sup>1</sup>Phibro Animal Health Corporation, Quincy, IL, <sup>2</sup>University of Georgia, Athens, GA.



OmniGen-AF (OG) is a nutritional supplement that supports immune function in ruminant species. Forty-four, second parity lactating dairy goats were used to determine the effects of feeding OG on milk production, quality and components. Goats ( $63.5 \pm 4.8$  kg BW) were housed on a commercial goat dairy in Wisconsin. Does were randomly assigned to 1 of 2 diets: 1) Control (CT;  $n = 22$ ), and 2) OG ( $n = 22$ ). CT does were fed a complete commercially available feed pellet (dry pellet: CP 14%, fat 3%, fiber 11.2%; lactating pellet: CP 16%, fat 3.4%, fiber 8.1%) twice a day and had ad libitum access to alfalfa hay. OG does were fed the same diet but with 6 g/h/d of OG added to the pellet. Both diets met the nutrient requirements of the does including the diet without OG. Diets were initiated at dry-off (67–94 d prepartum) and continued for the entire lactation. Does were crossbreds (Saanan, Nubian, Alpine and LaMancha) with all breeds equally represented in both groups. DHIA milk testing was performed every 28 d during lactation, resulting in a total of 10 tests. Milk production, % fat, % protein, fat corrected milk (FCM), energy corrected milk (ECM), SCC, and somatic cell score (SCS) data were collected at each test. Mean milk produced per day was significantly greater ( $P < 0.05$ ) for OG does than CT does (3.65 vs. 3.3 kg/h/d, respectively). Mean FCM was 3.59 kg/h/d for OG goats vs. 3.15 kg/h/d for CT goats ( $P < 0.01$ ). Mean ECM was 3.82 kg/h/d for OG goats and 3.39 kg/h/d for CT goats ( $P < 0.02$ ). Mean milk % fat was significantly greater ( $P < 0.05$ ) for OG does (3.93%) compared with CT does (3.66%). The mean SCC for OG does was  $847,060 \text{ mL}^{-1}$  vs.  $1,167,100 \text{ mL}^{-1}$  for CT does ( $P < 0.1$ ). Further analysis of the last 2 test dates (mean days in milk = 239.7 and 266.8) indicated OG does had lower SCC [ $1,348,090 \text{ mL}^{-1}$  ( $P < 0.1$ ) and  $2,003,630 \text{ mL}^{-1}$  ( $P < 0.05$ )] than CT does ( $2,471,090 \text{ mL}^{-1}$  and  $5,057,630 \text{ mL}^{-1}$ ). There were no differences ( $P > 0.1$ ) in mean % protein or mean SCS between the 2 groups. These results, including improved milk production, quality and select components, suggest enhanced mammary health in lactating dairy goats supplemented with OG.

**Key Words:** goat, milk, OmniGen-AF

**M465 Effect of increasing levels of concentrate on nutrient digestibility and growth performance in lambs.** Michelle de Oliveira Maia Parente<sup>1</sup>, Ruan Mourão da Silva Gomes\*<sup>1</sup>, Wesclley Jesus dos Santos Sodré<sup>1</sup>, Henrique Nunes Parente<sup>1</sup>, Miguel Arcaño Moreira Filho<sup>1</sup>, Nágila Maria de Carvalho Almeida<sup>1</sup>, Rosane Cláudia Rodrigues<sup>1</sup>, Jocélio dos Santos Araújo<sup>1</sup>, and Daniel Louçana da Costa Araújo<sup>2</sup>, <sup>1</sup>Universidade Federal do Maranhão, Chapadinha, Maranhão, Brazil, <sup>2</sup>Universidade Federal do Piauí, Teresina, Piauí, Brazil.

Quality and quantity of feed are the major factors in increasing ruminant productivity under tropical conditions. Increase in energy density in diets by providing larger quantities of concentrate feeds may improve feed efficiency and animal performance contributing to higher overall efficiency of utilization of dietary energy for body weight gain. Therefore, the objectives were to determine the effects increasing levels of concentrate on performance, intake and digestibility of lambs. Fifteen crossbred lambs (initial BW of  $18.2 \pm 3.2$  kg and 100 d old) were used in a randomized complete block design according to initial BW and age. Lambs were penned individually during 45 d and fed an isonitrogenous ( $16.6 \pm 0.55$  CP, DM basis) diet. Increasing levels of concentrate were 40, 60, or 80% corresponding to the experimental diets C40, C60 and C80, respectively. Data were analyzed using SAS PROC Mixed procedure and means compared by Tukey Test. Effects were declared significant at  $P \leq 0.05$ . No effect were observed on Dry matter (DM) intake (0.76, 0.87 and 0.85 kg/d for C40, C60 and C80) and crude protein (CP) intake (0.14, 0.16 and 0.15 kg/d for C40, C60 and C80). The DM digestibility was greater ( $P \leq 0.01$ ) for lambs fed C60 and C80 diets (80.18, 87.53

and 88.16% for C40, C60 and C80, respectively), while CP digestibility was unaffected ( $P \geq 0.05$ ) by concentrate levels (91.03, 92.21 and 91.56% for C40, C60 and C80, respectively). The Neutral Detergent Fiber (NDF) intake decreased for lambs fed C80 diet ( $P \leq 0.05$ ) while NDF digestibility was unaffected ( $P \geq 0.05$ ). ADG and G:F were greater ( $P \leq 0.01$ ) with increasing concentrate levels. Average daily gain was 60.1, 126.1 and 232.6 g/d while G:F was 0.08, 0.14 and 0.23 for C40, C60 and C80, respectively. It is concluded that supplementation of concentrate between 60 to 80% improves DM digestibility and performance of crossbred lambs without affect DM intake.

**Key Words:** average daily gain, energy density, feed efficiency

**M466 Feeding behavior of lambs fed increasing levels of concentrate in the diet.** Michelle de Oliveira Maia Parente<sup>1</sup>, Wesclley Jesus dos Santos Sodré<sup>1</sup>, Ruan Mourão da Silva Gomes\*<sup>1</sup>, Miguel Arcaño Moreira Filho<sup>1</sup>, Grazieli Silva Oliveira<sup>1</sup>, Alayne Andrade Cutrim<sup>1</sup>, Arnaud Azevêdo Alves<sup>2</sup>, and Viviany Lúcia Fernandes dos Santos<sup>3</sup>, <sup>1</sup>Universidade Federal do Maranhão, Chapadinha, Maranhão, Brazil, <sup>2</sup>Universidade Federal do Piauí, Teresina, Piauí, Brazil, <sup>3</sup>Universidade Federal do Rio Grande do Norte, Natal, Rio Grande do Norte.

The study of ingestive behavior is a highly important tool in the evaluation of diets, because through it is possible to acquire knowledge of the possible relationships existing between foods and animal, allowing us to adjust the feeding management of animals to obtain better productive performance. The objective of this trial was to determine the effects increasing levels of concentrate on feeding behavior of lambs. Fifteen crossbred lambs (initial BW of  $18.2 \pm 3.2$  kg and 100 d old) were used in a randomized complete block design according to initial BW and age. Lambs were penned individually during 45 d and fed an isonitrogenous ( $16.6 \pm 0.55$  CP, DM basis) diet. Increasing levels of concentrate were 40, 60, or 80% corresponding to the experimental diets C40, C60 and C80, respectively. Animals were monitored every 5 min during 24 h, on the 23th day of the experiment, according to the activities: eating, rumination, idle and other activities. The feed and rumination efficiencies, expressed as g DM/hour were obtained by dividing the average daily intake of dry matter by the total time spent eating and/or ruminating in 24 h, respectively. Orthogonal polynomials for diet responses were determined by linear and quadratic effects. Effects were declared significant at  $P \leq 0.05$ . There was a quadratic response ( $P \leq 0.05$ ) for time spent in eating. The levels of concentrate did not affect ( $P \geq 0.05$ ) the eating efficiency (149.3, 182.3 and 226.1 g DM/h for C40, C60 and C80), time spent in idle (505.0, 493.0 and 589.0 min/d for C40, C60 and C80) and water intake (1.8, 2.5 and 2.5 kg/d for C40, C60 and C80). The time spent in other activities (133.0, 185.0 and 220.0 min/d for C40, C60 and C80) and rumination efficiency (92.4, 116.5 and 132.4 g DM/h for C40, C60 and C80) tended to linearly increase ( $P = 0.07$ ) with high concentrate levels. However, the time spent in rumination (495.0, 462.0 and 391.0 min/d for C40, C60 and C80) tended to linearly decrease ( $P = 0.06$ ) with high concentrate levels. It is concluded that high concentrate levels (C80) changed the feeding behavior of lambs, especially for time spent for eating

**Key Words:** eating efficiency, idle, rumination efficiency

**M467 Efficacy of Rumatel (morantel tartrate) against gastrointestinal nematode infections in lactating dairy goats.** Angela D. Rowson\*<sup>1</sup>, Shelby A. Armstrong<sup>1</sup>, Brian P. Schnell<sup>2</sup>, and Lane O. Ely<sup>3</sup>, <sup>1</sup>Phibro Animal Health Corporation, Quincy, IL, <sup>2</sup>University of Wisconsin, Madison, WI, <sup>3</sup>University of Georgia, Athens, GA.

Rumatel (morantel tartrate) is an FDA-approved anthelmintic safe for use in goats and has no milk withhold. The objective of this study was to evaluate the efficacy of Rumatel on the removal of gastrointestinal nematodes (GIN) in lactating dairy goats. Twenty second-lactation crossbreed does (59–68 kg; mean days in milk = 128.5) from a south-central Wisconsin commercial herd were used in this study. Goats were housed in a pole barn with roll-up curtain sides on a bedded pack with continual access to pasture. A complete feed pellet was provided to the does twice a day and alfalfa hay was fed ad libitum. Goats had not been dewormed for 7 mo. Fecal pellets were collected from the rectum of each doe at 2 different time points, once immediately before the administration of Rumatel and again 14 d later. Fecal samples were analyzed for GIN using the McMaster's technique for fecal egg counts. Eligible samples [ $\geq 25$  *Trichostrongyle* spp. eggs/gram of feces (EPG)] were further analyzed using the Peanut Agglutination test. Conjunctional examinations were conducted on all does using the FAMACHA card as a color reference (scored 1–5) at each fecal collection. Goats were dewormed once using Manna Pro Positive Pellet Goat Dewormer (morantel tartrate: 880 g/ton) immediately following the first fecal collection. Does were group-fed the product with enough pellet provided for each goat to consume 0.68 kg (0.66 g of morantel tartrate). Means were compared with a Student's *t*-test. Feeding Rumatel resulted in significantly lower ( $P < 0.01$ ) EPG of total nematodes, *Trichostrongyle* spp., and *Haemonchus contortus*. Fecal egg counts were 176.3, 167.5, and 166.1 EPG, respectively, before deworming and 20.0, 20.0, and 14.7 EPG after deworming. Rumatel treatment also lowered the percentage of goats with *H. contortus* infections from 74.7% to 29.6% ( $P = 0.001$ ) and decreased FAMACHA scores from 3.5 to 2.65 ( $P = 0.001$ ). Results indicate that Rumatel is an efficacious anthelmintic for lactating dairy goats for GIN like *Trichostrongyle* spp., including *H. contortus*.

**Key Words:** goats, *Haemonchus contortus*, Rumatel

**M468 Determining the critical time to measure fasting heat production of Saanen goats.** Ana Rebeca Castro Lima\*, Kleber Tomás de Resende, Márcia Helena Machado da Rocha Fernandes, Izabelle Auxiliadora Molina de Almeida Teixeira, Thiago Henrique Borghi, José Mauricio dos Santos Neto, and Carolina Isabel Soriano Oporto, *Unesp, Jaboticabal, São Paulo, Brazil*.

The objective of this study was to establish the physiological parameters of Saanen goats at absorptive and post absorptive statuses, to determine the required period of fasting for these animals. Gas exchange was recorded by an open-circuit face-mask respirometry. Six nonlactating and non-pregnant Saanen (49.2  $\pm$  3.2 kg) goats were used in this experiment, allocated to individual pens and fed the same total mixed ration (19.5% of crude protein, 10 MJ per kg DM of metabolizable energy, ME). Gas exchange was measured in groups of 3 animals each and each data collection period lasted 6 d. During feeding period (3 d), each group was submitted to a double Latin square (3  $\times$  3) with 3 animals and 3 h window in the morning (7.00–9.00h; 9.00–11.00h; 11.00–13.00h) and evening (13.00–15.00h; 15.00–17.00h; 17.00–19.00h). After the 3-d feeding period, animals were subjected to fasting (no feed, only water) and gas exchange measurement was performed for 30 min periods at 12, 20, 36, 44, 60, and 68 h after fasting. To evaluate the decay of methane during fasting, methane production was fitted to a nonlinear logistic model. The critical time to reach the asymptote, or to measure fast heat production (FHP), was obtained when the upper limit of the standard error of mean of minimum methane produced and when CH<sub>4</sub> production became equivalent. During the feeding period, Saanen goats drank 3.34  $\pm$  0.53 L/d of fresh water and consumed 848.6  $\pm$  40.2 g/d of dry matter, the daily heat production, averaged 557.4  $\pm$  38.7 kJ/kg<sup>0.75</sup> BW and their

FHP was 236.0  $\pm$  19.7 kJ/kg<sup>0.75</sup> BW. After fasting, methane production decreased exponentially and the critical time to measure FHP was 47 h of fasting for Saanen goats when methane production was statistically equal to zero. Taken together, our results suggest that the ideal period to measure FHP should be between 40 and 60 h of fasting for goats fed at maintenance levels. The results presented herein are relevant for future energy metabolism studies in goats.

**Key Words:** heat production, indirect calorimetry, methane

**M469 Calcium and phosphorus accretion rate in Saanen goat kids of different genders.** José Mauricio dos Santos Neto\*, Kléber Tomás de Resende, Márcia Helena Machado da Rocha Fernandes, Izabelle Auxiliadora Molina de Almeida Teixeira, Julian André Castillo Vargas, Ana Rebeca Castro Lima, Fernanda Oliveira de Miranda Figueiredo, Rafael Fernandes Leite, Paula Fernanda Varella dos Santos, and Carolina Isabel Soriano Oporto, *São Paulo State University (Unesp), Jaboticabal, São Paulo, Brazil*.

Calcium (Ca) and phosphorus (P) constitute an essential part of goat diets, being necessary for adequate growth and physiological function; however, studies about their accretion rate in goats are still scant, especially regarding the differences among genders. In this sense, we evaluated and compared the Ca and P accretion rate in intact males, castrated males and females, weighting from 15 to 30 kg of body weight (BW). We used 58 Saanen goat kids (20 intact males, 20 castrated males and 18 females) fed ad libitum and housed in individual pens, with an initial BW of 15.85  $\pm$  0.11 kg. The diet consisted of 45% forage (dehydrated corn plant) and 55% concentrate. The animals of different genders were assigned in a completely randomized design and slaughtered at 16.6  $\pm$  0.40 kg BW, 23.1  $\pm$  1.33 kg and 31.2  $\pm$  0.58 kg BW. Allometric equations used to calculate the relationships between macromineral quantities and empty body weight (EBW) were generated and compared among genders by NLINMIX procedure (SAS Institute Inc., Cary, NC 9.4). The Ca and P accretion rate were estimated by the first derivative of these allometric equations. The Ca and P accretion rate did not differ among genders ( $P > 0.05$ ) and decreased from 11.04 to 11.01 g of Ca and from 6.96 to 5.98 g P per kg of EBW gain, when kids grown from 15 to 30 kg BW, respectively. Because Ca and P mostly concentrate in skeletal tissue, absence of difference may indicate that bone development in Saanen goats are similar among genders, and presumably, those animals have reached bone maturity early in life.

**Key Words:** body composition, comparative slaughter, macromineral

**M470 Effects of milk replacer feeding time on growth performance, nutrient digestibility and serum profiles in early-weaned lambs.** Jian-min Chai, Hai-chao Wang, Qi-yu Diao, Tao Ma, Min-li Qi, Yan Tu, and Nai-feng Zhang\*, *Feed Research Institute, Chinese Academy of Agricultural Sciences, Key Laboratory of Feed Biotechnology of the Ministry of Agriculture, Beijing, China*.

This study was conducted to evaluate the effect of milk replacer feeding time on growth performance, nutrient digestibility and serum profiles in early-weaned lambs. Forty-eight newborn Hu lambs (2.53  $\pm$  0.13 kg; 24 male and 24 female) were divided into 4 treatments: ewe-reared (ER) or weaned at 10, 20, or 30 d post birth (EW10, EW20, and EW30) and fed milk replacer (MR). All liquid feed were eliminated at 60 d of age (d 60). Creep feed was offered ad libitum to all lambs from d 15 to 90. Lambs were weighed on d 60 and 90. The data of liveweight (LWT), average daily gain (ADG), feed (including creep and MR) intake, nutrient utilization and serum parameters were analyzed as completely

randomized design using one-way ANOVA. Duncan's method for multiple comparisons was used if the overall F test for the measurement was significant ( $P < 0.05$ ). The results indicated that lambs of EW10, EW 20, and EW 30 had higher LWT (22.1, 21.4, and 20.2 vs 18.5 g), ADG (225.1, 219.6, and 208.6 vs 177.6 g/d), and creep feed intake (512.9, 412.1, and 413.0 vs 370.3 g/d) ( $P < 0.001$ ) than those in ER group. There were no differences between treatments in the apparent digestibility of dry matter (DM), organic matter (OM), nitrogen (N), phosphorus (P) ( $P > 0.05$ ) from d 50 to 60. The apparent digestibility of DM, OM, N and P of EW20 and EW30 groups were significantly higher ( $P < 0.05$ ) than that of EW10 and ER groups from d 80 to 90. Lambs in the EW10 group had higher concentration of albumin, serum urea nitrogen, and immunoglobulin G than those of other groups ( $P < 0.05$ ). In conclusion, early weaning could improve creep intake, growth performance, and nutrient utilization of Hu lambs. Time of early weaning significantly increased the growth of lambs.

**Key Words:** early weaning, milk replacer, lamb

**M471 Seasonal weight loss tolerance biomarkers to in dairy goats: An approach for breed selection.** Mariana Palma<sup>1</sup>, Lorenzo Hernández-Castellano<sup>2,3</sup>, Noemi Castro<sup>2</sup>, Anastasio Argüello<sup>2</sup>, Juan Capote<sup>4</sup>, Manolis Matzapetakis<sup>1</sup>, and Andre M. de Almeida\*<sup>5</sup>, <sup>1</sup>Instituto de Tecnologia Química e Biológica António Xavier, Universidade Nova de Lisboa, Oeiras, Portugal, <sup>2</sup>Department of Animal Science, Universidad de Las Palmas de Gran Canaria, Arucas, Canary Islands, Spain, <sup>3</sup>Veterinary Physiology, Vetsuisse Faculty, University of Bern, Bern, Switzerland, <sup>4</sup>Instituto Canario de Investigaciones Agrarias, Valle Guerra, Canary Islands, Spain, <sup>5</sup>Instituto de Biologia Experimental e Tecnológica, Oeiras, Portugal.

Goat milk and dairy products are an important nutritional and economic resource in Tropics. However, the yearly production of milk is affected by the scarcity of pastures during the dry season, which leads to seasonal weight loss (SWL) in ruminants. The aim of the work is to identify physiological markers indicative of SWL tolerance in 2 dairy goat breeds, with different SWL tolerance levels. The results will help to define breed selection strategies in drought prone regions. Nuclear magnetic resonance (NMR) was used to compare the observable metabolome present in aqueous extracts of mammary glands and milk serum from 2 dairy goat breeds; tolerant to dry environment (Majorera) and susceptible (Palmera). Ten Palmera and 9 Majorera dairy goats in mid lactation were used in this study. Animals were divided in 4 experimental groups (Majorera control, Palmera control, Majorera restricted and Palmera restricted). Milk samples were collected daily, after milking from the whole available milk, and mammary gland biopsies were collected after 23 d. Aqueous fractions were obtained by tissue aqueous/organic extraction and milk serum was obtained by ultra-centrifugation. We collected <sup>1</sup>H NMR spectra (1D-NOESY and CPMG) from the aqueous extract of the mammary gland, and the milk serum. <sup>1</sup>H NMR Spectrum deconvolution was used for metabolite profiling analysis and has led to the identification of 47 compounds in the aqueous fraction of mammary gland extracts. Lactose, glutamate, glycine, lactate and glucose were found to be the most abundant. Statistical evaluation using principal component analysis (PCA) and partial least squares (PLS) revealed differences between control and restricted animals, although no differences between breeds were observed. Preliminary profiling analysis of milk serum samples resulted in the identification of 20 metabolites.

**Key Words:** goat, undernutrition, nuclear magnetic resonance metabolomics

**M472 An NGS-based gene expression profile study in the goat mammary gland: Effect of undernutrition in two breeds with different levels of adaptation to nutritional stress.** José Parreira<sup>1,2</sup>, Joana R. Lérias<sup>1</sup>, Lorenzo E. Hernández-Castellano<sup>3,4</sup>, Mariana Palma<sup>2</sup>, Noemi Castro<sup>4</sup>, Anastasio Argüello<sup>4</sup>, Juan Capote<sup>5</sup>, Susana S. Araújo<sup>1,2</sup>, and André M. de Almeida\*<sup>1</sup>, <sup>1</sup>Instituto de Biologia Experimental Tecnológica, Oeiras, Portugal, <sup>2</sup>Instituto de Tecnologia Química e Biológica (I.T.Q.B.), Universidade Nova de Lisboa, Oeiras, Portugal, <sup>3</sup>Veterinary Physiology, Vetsuisse Faculty, University of Bern, Bern, Switzerland, <sup>4</sup>Department of Animal Science, Universidad de Las Palmas de Gran Canaria, Arucas, Canary Islands, Spain, <sup>5</sup>Instituto Canario de Investigaciones Agrarias, Valle Guerra, Canary Islands, Spain.

Goat milk has a high relevance worldwide, particularly in the tropics. Consequently, dairy goat productivity performance is very important for animal production and the dairy industry. Undernutrition during the dry season causes Seasonal Weight Loss (SWL), a relevant constraint for animal production. Knowledge on the genetic factors regulating milk production is limited and the impact of feed restriction, in this mechanism has been poorly addressed. A Systems Biology approach can provide a new and comprehensive insight on the molecular mechanisms underlying milk production, under feed restriction, as well as the regulation of the several biological processes involved. With such strategy, the identification of specific biomarkers and metabolic pathways involved could be used to develop new strategies to overcome this constraint. The objective of this work is to understand the molecular mechanisms by which goat breeds are adapted to SWL. We report a preliminary study of a quantitative differential analysis of mammary glands transcriptome using the Illumina high-throughput transcriptome sequencing (RNA-Seq). Two Canary Island (Spain) goat breeds were used; Majorera (SWL tolerant) and Palmera (SWL susceptible), with different productivity performances. We studied the impact of nutritional restriction on the transcriptome of mammary gland secretory tissue. Results show that a concerted reprogramming of genes expression occurs as result of the stress imposed, irrespective to the breed. Also suggesting a different behavior of both breeds in response to the treatment applied. Moreover, an enrichment analysis of the differentially expressed genes provided some insight into what biological processes are related with the response to SWL during lactation. Finally, a validation of the RNA-Seq assay using RT-qPCR was performed on candidate genes differentially expressed, for instance glycerol kinase (GK) and adrenoreceptor  $\beta$  2 (ADRB2). The knowledge obtained through this project will be useful to release new genomic resources as well as shed some light into the genes and regulatory networks underlying milk production under nutritional stress.

**Key Words:** dairy goat, undernutrition, transcriptomics

**M473 Digestibility of diets containing calcium salts of fatty acids in Saanen goats.** Bruna Susan de Labio Molina, Claudete Regina Alcalde\*, Bruna Hygino, Ludmila Couto Gomes, Caroline Isabela da Silva, Ana Paula Silva Possamai, and Rodrigo de Souza, Universidade Estadual de Maringá, Maringá, Paraná, Brazil.

According on the fat source included in the diets for goats, the availability of nutrients can be improved. The objective was to evaluate the inclusion of calcium salts of fatty acids (CSFA) in diets of Saanen goats on the digestibility of dry matter and nutrients. Five Saanen goats (63.23  $\pm$  9.08 kg body weight and  $\pm$  105 d of lactation) were distributed in a Latin square design (5  $\times$  5). The treatments were diets: control (without CSFA) and the other with: 6.25, 12.50, 18.75, and 25.0 g of CSFA g/kg DM, respectively. The ingredients that were: ground corn, soybean meal, protected fat (calcium salt linked with long chain fatty acids containing



1,760 g/kg total digestible nutrients, 820 g/kg ether extract, 100 g/kg calcium, 260 g/kg oleic acid, and 420 g/kg linoleic acid), and mineral-vitamin supplement. The oat hay was utilized in a proportion of 570 g/kg DM for all the treatments. The rations were adjusted to provide 700 g of total digestible nutrients/kg DM and 155 g of crude protein/kg DM. The internal marker used to estimate the digestibility was the indigestible acid detergent fiber. After 15 d for the animals to adapt to the diets, samples of feces from the exit of the rectum were collected during 6 consecutive days to obtain a composite sample per animal for each period. Calcium salts of fatty acids showed a quadratic effect on digestibility of dry matter; organic matter; neutral detergent fiber and total carbohydrate with improvement from the minimum points of 13.2; 13.3; 16.9 and 14.6 g of CSFA/kg of dry matter, respectively. Crude protein digestibility and non-fiber carbohydrates digestibility were not influenced by treatments, however, for the ether extract digestibility there was a positive linear response as function of the calcium salts of fatty acids in diets. A quadratic effect was observed on total digestible nutrients with the minimum point of 9.8 g of CSFA/kg DM. In conclusion, the inclusion of 25.0 g calcium salts of fatty acids/kg DM in diets of Saanen goats shows a better response for the total digestible nutrients.

**Key Words:** digestion, goat, indigestible ADF

**M474 Loin characteristics of feedlot lambs fed different sources of non-protein nitrogen and carbohydrate.** Ricardo D. Kliemann\*, Jean C. S. Lourenço, Larissa Goltz, Juliana M. Lima, Dayanna Pastal, Tiago M. Santos, Sergio R. Fernandes, and Américo F. Garcez Neto, *Federal University of Paraná, Palotina, Paraná, Brazil.*

The formulation of balanced diets has been a way to optimize the nutrient use by different animals and to get carcasses with high proportion of muscles and proper fat content to attract potential consumer markets. The proportion of these tissues in the carcass can be estimated by measurements in the *Longissimus dorsi* (*L. dorsi*) muscle. Characteristics of *L. dorsi* in feedlot lambs fed different sources of carbohydrates (CHO) and non-protein nitrogen (NPN) were evaluated. It was used 20 non-castrated crossbred lambs with 4 mo of age and 26 kg of body weight. The diets were composed by 57% of grass hay (*Brachiaria* hybrid) and 43% of concentrate. The hay was harvested at late mature stage with high (72.4%) neutral detergent fiber and low (4.9%) crude protein. Four concentrates were formulated from the combination of 2 CHO (ground corn: starch; whey permeate: lactose) and 2 NPN (fast and slow release urea) sources. After cooling, loin of the left half of carcasses was separated and measurements in the *L. dorsi* were made. The measurements were loin eye width (LEW), loin eye depth (LED), loin eye area (LEA), minimum fat thickness (MinFT), maximum fat thickness (MaxFT). The LEW:LED ratio was also calculated. It was used a completely randomized design with 4 treatments and 5 replicates. Data were analyzed by ANOVA following a 2 × 2 factorial scheme. LED (2.57 ± 0.06 cm), LEA (10.37 ± 0.31 cm<sup>2</sup>), MinFT (0.76 ± 0.09 mm), MaxFT (1.69 ± 0.12 mm) and LEW:LED ratio (2.12 ± 0.06) were not ( $P > 0.05$ ) affected by the diets. It was found an interaction ( $P < 0.05$ ) between CHO and NPN for LEW. The LEW was smaller ( $P < 0.05$ ) for feedlot lambs fed whey permeate and fast release urea (4.98 cm) than for those fed whey permeate and slow release urea (5.78 cm). The mean LEW was 5.39 ± 0.10 cm. Differences in the LEW did not change the LEA, which may be inferred that the association of whey permeate with slow and fast release urea resulted in the same muscle development. The association of CHO and NPN with concentrates of closest potential degradation

rates does not influence the proportions of muscle and fat of lambs fed low quality forage at finishing.

**Key Words:** fat, muscle, sheep

**M475 Blood metabolites of feedlot lambs fed different sources of non-protein nitrogen and carbohydrate.** Ricardo D. Kliemann\*, Larissa Goltz, Jean C. S. Lourenço, Eduardo M. Nascimento, Dayanna Pastal, Tiago M. Santos, Sergio R. Fernandes, and Américo F. Garcez Neto, *Federal University of Paraná, Palotina, Paraná, Brazil.*

The levels of blood metabolites of feedlot lambs fed low quality forage and different sources of carbohydrate (CHO) and non-protein nitrogen (NPN) in the concentrate feed was evaluated. Twelve non-castrated male lambs with 4 mo of age and 23.5 kg BW were used. The experimental design was 4 × 3 incomplete Latin square replicated 3 times, with 4 treatments (diets) and 3 periods of 21 d. The diets were composed by 57% of grass hay (*Brachiaria* hybrid) and 43% of concentrate. The hay was harvested at late mature stage with high fiber (72.4% NDF) and low protein (4.9% CP). Four concentrate feeds were formulated from the combination of 2 CHO (ground corn: starch; whey permeate: lactose) and 2 NPN (fast and slow release urea) sources. The diets were isoenergetic (2.32 Mcal/kg ME) and isoproteic (11% CP), and fed at 1.5 times the ME requirement. At the end of each period, blood samples were collected after fasting (feed for 14 h) to assess the serum levels of total proteins (TP), albumin (Alb), creatinine (Creat), urea, glucose (Glu), cholesterol (Chol), triglycerides (TG) and β-hydroxybutyrate (BHBA). Data were analyzed by ANOVA and means were compared by LSD test ( $P < 0.05$ ). Serum levels of TP (61.7 g/L), Alb (31 g/L), Creat (0.91 mg/dL), Glu (64 mg/dL) and BHBA (0.12 mg/dL) were not affected by the diets. Serum urea was higher in lambs fed corn and fast release urea (29.9 mg/dL), and lower in those fed whey permeate independent from NPN source (25.4 mg/dL). High levels of serum urea can be related to high production of ammonia in the rumen due to the differences in degradation rates between corn and fast release urea, and by the higher CP of corn (9%) compared with whey permeate (3%). Maybe the whey permeate was fermented faster than corn resulting in greater MP production and less ammonia in the rumen. Serum levels of Chol and TG were higher in lambs fed corn (45.2 and 24.6 mg/dL) compared with those fed whey (34.6 and 17.1 mg/dL), which was caused by the lipid fraction of corn (5.4%). The inclusion of corn or whey permeate in the concentrate feed affects the protein and energy metabolism of lambs fed low quality forage.

**Key Words:** cholesterol, metabolism, urea

**M476 Multi-scale straightness index analysis of goat behavior.** Terry A. Gipson\*<sup>1</sup>, Kenneth M. Andries<sup>2</sup>, Terry Hutchens<sup>2</sup>, and Myron E. Evans<sup>3</sup>, <sup>1</sup>American Institute for Goat Research, Langston University, Langston, OK, <sup>2</sup>Kentucky State University, Frankfort, KY, <sup>3</sup>University of Kentucky Cooperative Extension, Grayson, KY.

Multi-scale straightness index (MSSI) has been proposed for determining behavioral states such as resting, grazing, or commuting in wildlife but its suitability in livestock is not known. Therefore, the objective of this study was to apply MSSI to a herd of grazing goats. On a random sample of 13 mature Boer-cross females from a herd of 120, GPS collars that recorded a fix every 5 min were fitted for 3 consecutive days in August, 2012. The study area was a 40-ha unimproved hill pasture. The fix records were post-processed and imported into ArcMap. Boundaries of the pasture, including a 7-m external buffer, were constructed as shapefiles. Only fixes (average of 804 ± 15.2 fixes/goat) within the

boundary and buffer shape-files were exported to a spreadsheet for the calculation of MSSSI using granularity (g) from 1 to 12 and window (w) from 1 to 36 with the constraint that w/g must be an integer, yielding a total of 110 g-w combinations. Within daytime (D) and nighttime (N) periods, MSSSI were calculated for each g-w combination yielding 868,707 MSSSI for D and 541,599 MSSSI for N. LSmeans for each g-w combination were estimated using mixed model analysis with collar as random effect and day (1, 2, or 3) as repeated measure. A linear-linear-linear grafted polynomial analysis was conducted to ascertain ridge points for g-w combinations (between 2 break points). The first linear segment for both D and N represented targeted travel and accounted for 11% of the MSSSI; however, the g/w ratio was 1 so this was straight line travel between 2 points. The last linear segment represented highly tortuous travel indicating most probably grazing or resting. This segment was 81% and 85% of the MSSSI with an average MSSSI of  $0.17 \pm 0.083$  and  $0.08 \pm 0.050$  for D and N, respectively. The middle linear segment represented targeted travel and accounted for 8% and 4% of the MSSSI with an average MSSSI of  $0.53 \pm 0.108$  and  $0.41 \pm 0.119$  for D and N, respectively. For the targeted travel segment, g averaged 1 for both D and N and w averaged 5 and 3 for D and N, respectively. Even in a herd of goats familiar with the terrain, targeted travel account for a small percentage of behavior and was only for a short duration.

**Key Words:** goat, GPS, multi-scale straightness index (MSSSI)

**M477 Effects of high heat load on BW, DMI, rectal temperature, and respiration rate of Katahdin sheep and Boer and Spanish goat wethers.** Mengistu Urge<sup>1,2</sup>, Ryszard Puchala<sup>1</sup>, Terry A. Gipson<sup>1</sup>, Tilahun Sahlu<sup>1</sup>, and Arthur L. Goetsch<sup>\*1</sup>, <sup>1</sup>*American Institute for Goat Research, Langston University, Langston, OK*, <sup>2</sup>*School of Animal and Range Sciences, Haramaya University, Dire Dawa, Ethiopia*.

Thirty-three yearling Katahdin sheep (K) and Boer (B) and Spanish (S) goat wethers were used to determine conditions such as an appropriate heat load index (HLI) for a method of evaluating differences in resilience to high HLI among different small ruminant species, breeds, ecotypes, and individuals. Grass hay was consumed ad libitum with concentrate (80% corn, 20% soybean meal) at 0.5% BW. After a baseline period (1) with thermo-neutral conditions, target HLI were 80, 90, 95, and 100 (e.g., 42.0°C, 40% relative humidity) during the day and 70, 77, 81, and 85 at night in subsequent 1-wk periods (2–5). Actual values averaged 66, 80, 92, 97, and 101 during day (0700 to 1900 h) and 66, 75, 84, 86, and 89 at night in periods 1–5, respectively. Final period BW was not markedly affected by HLI (100.2, 99.7, 103.1, and 101.3% of baseline; SE = 0.74), and hay DMI also was generally similar to the baseline (95.0, 72.4, 93.6, and 96.4% in periods 2–5, respectively; SE = 3.69). Rectal temperature at 0600, 1300, and 1700 h was lowest for Katahdin in periods 3 (39.4, 39.2, and 39.6°C) and 4 (39.9, 39.6, and 40.0°C for Boer, Katahdin, and Spanish, respectively; SE = 0.07) but similar at other times. There was an interaction in respiration rate between animal type and period (71, 105, and 105 in period 2, 93, 101, and 104 in period 3, 121, 139, and 129 in period 4, and 105, 126, and 109 breaths/min in period 5 for B, K, and S, respectively; SE = 4.75). Rectal temperature and respiration rate were much lower at 0600 (38.7, 38.9, and 38.8°C and 34, 77, and 56 breaths/min) than at 1300 (39.7, 39.6, and 39.9°C and 127, 137, and 135 breaths/min) and 1700 h (40.1, 39.7, and 40.2°C, SE = 0.07, and 131, 139, and 144 breaths/min for B, K, and S, respectively, SE = 4.4). In conclusion, in some periods K exhibited greater ability than B and S to increase respiration rate and minimize rectal temperature at high HLI; periods longer than 1 wk at high HLI are required for evaluating changes in BW and DMI; and variables such as rectal temperature

and respiration rate may be more meaningful when measured in the afternoon than morning.

**Key Words:** goat, sheep, heat load

**M478 Effects of mixing different breeds to evaluate electric fence strand additions to barbed wire fence to contain growing meat goat kids.** Yoko Tsukahara\*, Terry A. Gipson, Jerry Hayes, Ryszard Puchala, Tilahun Sahl, and Arthur L. Goetsch, *American Institute for Goat Research, Langston University, Langston, OK*.

A method of evaluating electric fence strand addition to cattle barbed wire fence for goat containment would be useful to promote co-grazing. Therefore, 38 Boer (B) wethers (6.3 mo of age and 22.1 kg BW initially), 41 B doelings (6.3 mo and 21.4 kg), 40 Spanish (S) wethers (6.7 mo and 17.8 kg), and 40 S doelings (6.8 mo and 18.4 kg), were used to evaluate effects of grouping, single breed (SGL) and breeds combined (COM), on behavior (e.g., pen exit and shock) when exposed to barbed wire fence with different electric strand additions. Five 2.4 × 3.7 m evaluation pens had 1 side of barb wire strands at 30, 56, 81, 107, and 132 cm from the ground. Fence treatments (FT) were electrified strands (6 kV) at 15 and 43 (LH), 15 and 23 (LM), 15 (L), 23 (M), and 43 cm (H). For adaptation, kids were exposed in evaluation pens to no electric strands (NES), NES, LH at 0 kV, LH at 6 kV, and NES in wk 1, 2, 3, 4, and 5, respectively. Then kids were divided into 2 replication sets per grouping (2 B-SGL, 2 S-SGL, 2 B-COM, and 2 S-COM); each of the 5 pens consisted of 4 or 3 animals for 1 h exposure to FT while observing behavior visually and with video surveillance. There were no main effects of grouping. Fence treatment affected ( $P < 0.01$ ) the percentage of animals receiving a shock (59.4, 44.5, 34.4, 22.8, and 6.2%; SE = 8.53), exiting with shock (37.5, 35.8, 31.3, 19.6, and 3.1%; SE = 7.63), and exiting without shock (0.0, 14.9, 50.0, 67.7, and 76.3% for LH, LM, L, M, H, respectively; SE = 7.56). There was an interaction ( $P = 0.01$ ) between FT and grouping in pen exit (50.0, 25.0, 75.0, 85.7, and 42.9% with B-COM, 12.5, 77.8, 87.5, 75.0, and 100% with B-SGL, 62.5, 62.5, 75.0, 87.5, and 75.0% with S-COM, and 25.0, 37.5, 87.5, 100.0, and 100.0% with S-SGL for LH, LM, L, M, and H, respectively; SE = 14.83). In conclusion, in contrast to previous findings with mature does, these results do not provide clear evidence supporting notable effect of method of grouping growing meat goat kids for evaluating effectiveness of various electric strand additions to barbed wire fence for goat containment, indicating appropriateness of either method.

**Key Words:** behavior, fence, goat

**M479 Diet selection patterns and ingestive behavior of feedlot lambs fed different sources of non-protein nitrogen and carbohydrate.** Jean C. S. Lourenço\*, Ricardo D. Kliemann, Larissa Goltz, Caroline Dell'Agnolo, Dayanna Pastal, Tiago M. Santos, Sergio R. Fernandes, and Américo F. Garcez Neto, *Federal University of Paraná, Palotina, Paraná, Brazil*.

The mixture of carbohydrates (CHO) and non-protein nitrogen (NPN) with different ruminal degradation may affect the feeding behavior and feed selection by ruminants. This study was carried out to evaluate the ingestive behavior and diet selection by feedlot lambs fed different sources of CHO and NPN. It was used 20 non-castrated crossbred lambs with 4 mo of age and 26 kg of body weight. The diets were composed by 57% of grass hay (*Brachiaria* hybrid) and 43% of concentrate. The hay was harvested at late mature stage with high (72.4%) neutral detergent fiber and low (4.9%) crude protein content. Four concentrate feeds were formulated from the combination of 2 CHO (ground corn:

starch; whey permeate: lactose) and 2 NPN (fast and slow release urea) sources. Orts samples were taken to determine the proportion of leaves (LVS), stems (STS) and leaf:stem ratio, and activities of eating (EAT), ruminating (RUM) and resting (RES) were recorded. The efficiency of rumination (RE) and dry matter intake (IE) were also calculated. It was used a completely randomized design with 4 diets and 5 replicates. Data were analyzed by ANOVA following a  $2 \times 2$  factorial scheme. LVS ( $56.2 \pm 2.7\%$ ), STS ( $43.8 \pm 2.7\%$ ), leaf:stem ratio ( $1.60 \pm 0.16$ ), EAT ( $318 \pm 8$  min), RUM ( $537 \pm 11$  min), RES ( $585 \pm 14$  min), RE ( $1.82 \pm 0.06$  g DM/min) and IE ( $3.10 \pm 0.11$  g DM/min) were not affected ( $P > 0.05$ ) by diets. The mixture of CHO and NPN with more distinct potential degradation rates (ground corn + fast release urea; dry whey permeate + fast release urea) did not increase ( $P > 0.05$ ) the selection of LVS, neither the mixture of sources with closest potential degradation rates (ground corn + slow release urea; dry whey permeate + fast release urea) reduced the RUM. It is possible that these results were not effective to increase the ruminal degradation of low quality forage. Such hypothesis is supported by the similarity of RE values between diets. In the present study the association of CHO and NPN with similar or distinct degradation rates does not influence the diet selection and the ingestive behavior.

**Key Words:** degradation rate, rumination, sheep

**M480 Carcass characteristics of feedlot lambs fed different sources of non-protein nitrogen and carbohydrate.** Jean C. S. Lourenço\*, Larissa Goltz, Ricardo D. Kliemann, Douglas Bloedorn, Dayanna Pastal, Tiago M. Santos, Sergio R. Fernandes, and Américo F. Garcez Neto, *Federal University of Paraná, Palotina, Paraná, Brazil.*

A balanced supply of nutrients can be regarded a way to support an increase of body weight and a tool to improve the carcass grade at slaughter. In that context carcass weight, yield and fat covering can be changed by the quality of different diets affecting not only the carcass parameters but also the meat value of the lambs. The carcass grade and weight at slaughtering and its parameters were evaluated on feedlot lambs fed different sources of non-protein nitrogen (NPN) and carbohydrates (CHO). It was used 20 non-castrated lambs with 4 mo of age and 26 kg of body weight. Lambs were fed ad libitum for 45 d. The diets were composed by 57% of grass hay (*Brachiaria* hybrid) and 43% of concentrate. The hay was harvested at late mature stage with high (72.4%) neutral detergent fiber and low (4.9%) crude protein. Four concentrates were formulated from the combination of 2 CHO (ground corn: starch; whey permeate: lactose) and 2 NPN (fast and slow release urea) sources. Slaughter weight (SW), body condition score (BCS), hot carcass weight (HCW), cold carcass weight (CCW), fat thickness (FT), hot carcass yield (HCY), cold carcass yield (CCY) and chilling losses (CL) were measured. A completely randomized design was used with 4 diets and 5 replicates. Data were analyzed by ANOVA following a  $2 \times 2$  factorial scheme. SW was higher ( $P < 0.05$ ) for lambs fed corn (31.3 kg) than for those fed whey (29.4 kg) suggesting that the starch intake was more effective in improving low quality forage degradation than lactose. However, BCS ( $3.0 \pm 0.1$ ), HCW ( $12.34 \pm 0.41$  kg), CCW ( $11.87 \pm 0.37$  kg), HCY ( $40.7 \pm 0.5\%$ ), CCY ( $39.2 \pm 0.5\%$ ), FT ( $1.23 \pm 0.10$  mm) were not influenced ( $P > 0.05$ ) by diets. CL was higher ( $P < 0.05$ ) for diets with slow release urea (4.01%) compared with those with fast release urea (3.65%) suggesting higher muscle water retention for diets with urea of faster degradation. For this reason, we can recommend the use of a concentrate diet composed of fast release urea

and corn for finishing lambs. Because these feed ingredients are usually cheaper, this will also reduce the cost of fattening.

**Key Words:** fattening, sheep, slaughter weight

**M481 Weight and cutting yield of feedlot lambs fed different sources of non-protein nitrogen and carbohydrate.** Larissa Goltz, Jean C. S. Lourenço, Ricardo D. Kliemann, Bruna N. Zuffo, Dayanna Pastal, Tiago M. Santos, Sergio R. Fernandes, and Américo F. Garcez Neto\*, *Federal University of Paraná, Palotina, Paraná, Brazil.*

Diets formulated with feed of different ruminal degradation rates can affect the performance and feed efficiency of lambs. In that condition is expected that the carcass weight and its cuts are also influenced changing the potential income for farmers. This work was carried out to evaluate the weights and yield of carcass cuttings of feedlot lambs fed diets of different carbohydrate (CHO) and non-protein nitrogen (NPN) sources. It was used 20 non-castrated crossbred lambs with 4 mo of age and 26 kg of body weight. The diets were composed by 57% of grass hay (*Brachiaria* hybrid) and 43% of concentrate. The hay was harvested at late mature stage with high (72.4%) neutral detergent fiber and low (4.9%) crude protein. Four concentrate feeds were formulated from the combination of 2 CHO (ground corn: starch; whey permeate: lactose) and 2 NPN (fast and slow release urea) sources. The diets were isoenergetic (2.32 Mcal/kg ME) and isoproteic (11% CP). The lambs were slaughtered at the end of trial after 45 d with a mean body weight of  $30.4 \pm 1.0$  kg. After cooling, the left half of carcasses was separated into 6 commercial cuts of meat: neck, breast, ribs, shoulder, loin and leg. It was recorded the weight of each cut and its proportion in relation to the half carcass. A completely randomized design was used with 4 treatments and 5 replicates. Data were analyzed by ANOVA following a  $2 \times 2$  factorial scheme between CHO and NPN. The weights of neck ( $0.57 \pm 0.02$  kg), breast ( $0.63 \pm 0.02$  kg), ribs ( $0.85 \pm 0.03$  kg), shoulder ( $1.19 \pm 0.04$  kg), loin ( $0.64 \pm 0.02$  kg) and leg ( $2.05 \pm 0.07$  kg) were not influenced ( $P > 0.05$ ) by the treatments. The same pattern was found to the yield of neck ( $9.7 \pm 0.2\%$ ) breast ( $10.6 \pm 0.3\%$ ), ribs ( $14.3 \pm 0.2\%$ ), shoulder ( $20.1 \pm 0.2\%$ ), loin ( $10.8 \pm 0.2\%$ ) and leg ( $34.6 \pm 0.3\%$ ). The association of CHO and NPN with concentrates of closest degradation rates (whey permeate + fast release urea; ground corn + slow release urea) do not increase ( $P > 0.05$ ) the weight neither change the yield of cuts of meat in the lamb carcass when low quality forage is added to the diet during the finishing.

**Key Words:** slow release urea, sheep, whey permeate

**M482 Effect of *Acacia mearnsii* tannin extract on intake and digestibility by sheep fed a tropical grass hay plus concentrate.** S. C. Avila\*<sup>1</sup>, G. V. Kozloski<sup>1</sup>, T. Orlandi<sup>1</sup>, M. P. Mezzomo<sup>1</sup>, and D. L. Harmon<sup>2</sup>, <sup>1</sup>*Universidade Federal de Santa Maria, Santa Maria, Rio Grande do Sul, Brazil,* <sup>2</sup>*University of Kentucky, Lexington, Kentucky.*

Tannins are plant polyphenols with the capacity to form complexes mainly with proteins reducing their degradation in the rumen and, thus, have the potential to increase the metabolizable protein supply and/or to decrease the urinary N losses by ruminants. However, tannins might also decrease feed intake and digestibility. Moreover, the nutritional effect of tannins is dependent on the tannin source. Six Texel wethers ( $67.0 \pm 6.53$  kg) fed ad libitum Tifton hay (*Cynodon* sp.) and 12 g/kg body weight (dry matter (DM) basis) of concentrate were used in a crossover design with 2 21 d periods to evaluate the effect of dietary inclusion of a tannin extract from *Acacia mearnsii* on feed intake, digestibility and N excre-



tion. The concentrate was composed of cracked corn grain (0.36), wheat bran (0.36) and soybean meal (0.28) with or without 10 g/kg DM of tannin extract of *Acacia mearnsii*. Total feed, refusals, feces and urinary excretion were measured during the last 7 d of each experimental period. The concentrate represented in average 0.40 of total DM intake. Total intake (g/day) of organic matter (OM,  $1720 \pm 132.0$ ), neutral detergent fiber (NDF,  $1039 \pm 78.8$ ) and N ( $38.2 \pm 3.29$ ), as well as the apparent digestibility of OM ( $0.77 \pm 0.031$ ), N compounds ( $0.82 \pm 0.020$ ) and NDF ( $0.72 \pm 0.038$ ) and urinary N excretion ( $21.4 \pm 2.33$  g/day) were not affected by tannin extract inclusion. Tannin extract reduced ( $P < 0.05$ ) the proportion of fecal N from endogenous origin ( $0.81$  vs  $0.76$ ) and the true digestibility of N compounds ( $0.97$  vs  $0.95$ ). These results indicate that tannin inclusion can affect N digestion. Further studies are needed to optimize the amount of tannin for each diet.

**Key Words:** consumption, digestion, nitrogen compound

**M483 Probiotic supplementation affects resilience to parasites in goats.** Mulumebet Worku\*, Kingsley Ekwemalour, Emmanuel Asiana, Hamid Ismail, and Sara Adjei-Fremah, *North Carolina A&T State University, Greensboro, NC.*

The objective of this study was to evaluate resistance and resilience to gastrointestinal parasites in goats following supplementation with probiotics. Improving resistance and resilience to gastro-intestinal parasites is essential to combating the threat to the small ruminant industry from anthelmintic resistance. Following initial screening for infection, female SpanishXBoer goats were assigned to 4 groups of 4 ( $n = 16$ ), one week post weaning. Goats were drenched daily with 10 mL sterile water containing recommended doses of either CorPET (treatment I)-a probiotic from the mushroom *Coriolus versicolor* (Mycology Research Laboratories, Ltd., Netherlands), Fastrak microbial pack (treatment II) or ruminant gel (treatment III) (Conklin Company, Inc. Shakopee, MN). A control group of age-matched goats received sterile water (treatment IV) for an 8 week period. Samples were collected weekly. Body weight, Body condition and FAMACHA scores were evaluated. Fecal samples were used for Hemonchus and coccidia egg counts to evaluate resistance. Blood samples were used to measure Packed cell volume (PCV) and for isolation of total RNA using Tri-reagent. The Nanodrop spectrophotometer was used to evaluate RNA concentration and purity. Data were analyzed using SAS PROC MIXED for repeated measures analyses. The effect of treatment on fecal egg count was variable. Treatment with commercial probiotics significantly increased body weight in wk 8, improved FAMACHA scores in wk 7, PCV was improved by treatments II and III in wk 5 ( $P < 0.05$ ). There was no effect on body condition score. Transcription of total RNA in blood increased during periods of high parasite egg output and were not affected by treatment ( $P > 0.05$ ). Thus probiotic supplementation may affect resilience to parasite infection in goats in a time and marker specific manner.

**Key Words:** goat, probiotic, resilience

**M484 Determination of the grazing activity energy cost in Boer goat wethers using a portable indirect calorimetry method.** Marie E. Brassard<sup>1,2</sup>, Ryszard Puchala\*<sup>1</sup>, Tilahun Sahlul<sup>1</sup>, and Arthur L. Goetsch<sup>1</sup>, <sup>1</sup>American Institute for Goat Research, Langston University, Langston, OK, <sup>2</sup>Université Laval, Québec, QC, Canada.

Heat energy (HE) of small ruminants in free-moving settings such as grazing is often measured indirectly from heart rate (HR) and the ratio of HE to HR determined when situated once or periodically in a stationary

calorimetry system. Therefore, feasibility of use of a portable indirect calorimetry system with goats while grazing, for a direct estimate of HE based on gas exchange without reliance on HR, was investigated in this experiment. Ten yearling Boer goat wethers ( $44.4 \pm 0.95$  kg) were used to determine HE and the grazing activity energy cost (GAEC) while standing or grazing Sudangrass pasture with a portable indirect calorimetry system. The method entailed use of a partial face mask that allowed unrestricted grazing to measure oxygen consumption and carbon dioxide emission for 30 min while restrained in a stanchion near the grazing area, followed by 60 min of grazing with other members of the group. The face mask was attached to a 15-m tether along with a corrugated plastic hose through which exhaled air was passed to a FlowKit Mass Flow Generator and FoxBox Respirometry System (Sable System, Las Vegas, NV) that were carried by a researcher who allowed unrestricted goat movement. Prior to measurements, animals were trained to become accustomed to presence of personnel and use of the equipment, with observed grazing behavior similar among all animals of the group. Measurement periods were during morning and afternoon grazing bouts. Heat energy while restrained was  $18.7$  kJ/kg  $BW^{0.75}$ /h or  $446 \pm 10.7$  kJ/kg  $BW^{0.75}$ /d. Grazing HE increased to  $35.1$  kJ/kg  $BW^{0.75}$ /h or  $843 \pm 39.3$  kJ/kg  $BW^{0.75}$ /d, implying that the GAEC was  $16.4$  kJ/kg  $BW^{0.75}$ /h. Goats spent 8.5 h/d grazing; therefore, the daily GAEC was  $138 \pm 17.3$  kJ/kg  $BW^{0.75}$ . A very similar GAEC of ( $165 \pm 10.4$  kJ/kg  $BW^{0.75}$ /d) was determined from the difference in HE estimated indirectly from HR between times when grazing a 0.8-ha pasture and confined in nearby  $1.2 \times 1.2$  m pens and fed fresh forage. In conclusion, this method offers promise for relatively simple and direct estimates of the sizable fraction of total HE comprised by GAEC.

**Key Words:** goat, grazing, heat energy

**M485 Effects of two heart rate-based methods of estimating the grazing activity energy cost of Boer goat wethers.** Marie E. Brassard<sup>1,2</sup>, Ryszard Puchala\*<sup>1</sup>, Terry A. Gipson<sup>1</sup>, Tilahun Sahlul<sup>1</sup>, and Arthur L. Goetsch<sup>1</sup>, <sup>1</sup>American Institute for Goat Research, Langston University, Langston, OK, <sup>2</sup>Université Laval, Québec, QC, Canada.

There are different methods to estimate the grazing activity energy cost (GAEC) of ruminants from heart rate (HR), regarding both treatment differences and contribution to total heat energy (HE). The primary objective of this experiment was to investigate differences between 2 such methods. Ten yearling Boer goat wethers ( $44.4 \pm 0.95$  kg) consuming fresh Sudangrass ad libitum while grazing a 0.8-ha pasture or confined in nearby  $1.2 \times 1.2$  m pens were used in a crossover. Heart rate measured over 24 h in 5-min intervals and the ratio of HE to HR previously determined for each animal with a stationary calorimetry system for 24 h while consuming grass hay was used to estimate HE. A GPS collar and leg activity monitor were used when HR was measured to determine HE while resting-lying (L), resting-standing (S), grazing (G), and walking (W); behavior in confinement was L or S. The grazing activity method (GAM) was based on time spent in the different activities multiplied by corresponding HE values, with GAEC assumed the sum of differences between S, G, and W relative to L. The confinement method (COM) entailed subtracting total HE while confined from that when grazing. There were differences ( $P < 0.01$ ) in percentage of the day spent in the 4 activities (33.8, 53.9, 11.4, and 0.9%; SE = 2.44) and the associated daily HE (241, 322, 75, and 6 kJ/kg  $BW^{0.75}$ ; SE = 17.9), although HE per unit time only tended ( $P = 0.099$ ) to vary (707, 598, 472, and 636 kJ/kg  $BW^{0.75}$  on a daily basis for G, L, S, and W, respectively; SE = 65.8). Total daily HE (642 and 482 kJ/kg  $BW^{0.75}$ ; SE = 17.2) and

HE while lying (598 and 450 kJ/kg BW<sup>0.75</sup>; SE = 18.1) were greater when grazing than confined ( $P < 0.01$ ). Daily GAEC was considerably greater ( $P < 0.01$ ) for the COM vs. GAM expressed in kJ/kg BW<sup>0.75</sup> (165 and 46; SE = 14.0) and relative to HE when confined for COM and of L on a daily basis for GAM (35 and 8%; SE = 3.5). In conclusion, method of estimation can have substantial effect on GAEC. Greater L HE per unit time when grazing than confined may contribute to lower GAEC for GAM than for COM, although factors such as dietary and environmental conditions will influence accuracy of COM.

**Key Words:** activity, energy, goat

**M486 The effects of variation in birth weight of littermates on lamb performance.** Jennifer L. Juengel<sup>1</sup>, George H. Davis<sup>1</sup>, Roger Wheeler<sup>2</sup>, and Peter D. Johnstone<sup>1</sup>, <sup>1</sup>*AgResearch, Invermay Agricultural Centre, Mosgiel, New Zealand*, <sup>2</sup>*AgResearch, Woodlands Research Station, Invercargill, New Zealand*.

The objective of this study was to assess the relative importance of lamb birth weight (BW) and variation between BW of littermates (VAR) on lamb performance. Records from lambs born into twin ( $n = 4160$ ) or triplet ( $n = 1279$ ) litters from 1988 to 2005 were analyzed. Data collected for each lamb included dam, sire, sex, BW, birth date, lamb fate, and weaning weight and date. Data were analyzed with REML with BW and VAR grouped into classes (BW (kg):  $\leq 2$ ,  $>2-3$ ,  $>3-3.5$ ,  $>3.5-4$ ,  $>4-5$ ,  $>5$ ; VAR (kg):  $0-0.1$ ,  $>0.1-0.35$ ,  $>0.35-0.5$ ,  $>0.5-0.75$ ,  $>0.75-1.00$ ,  $>-1.3$ ,  $>1.3$ ). The model included BW, VAR, sex and year of birth as fixed effects with sire of lamb included as a random effect. Overall, 85.3% of twins and 74.7% of triplet lambs survived to weaning. For both twin and triplet lambs, BW was associated with survival ( $P < 0.001$ ) with lambs  $< 2$  kg having very low survival rates ( $<40\%$ ). For triplets and the larger of twin lambs, VAR was not associated with survival. However, VAR did affect survival of the smallest lamb in twin litters ( $P < 0.001$ ) with lambs with VAR  $> 1.3$  kg being less apt to survive (67.8% compared with those from litters of smaller VAR (range 80.4–86.2% survival). Growth rate of twin and triplet lambs was affected by BW ( $P < 0.001$ ). As BW increased, growth rate also increased. For triplet born and raised lambs, VAR did not affect growth of the lamb. Surprisingly, twin lambs from litters with high VAR had slightly greater growth rate than those from lambs of low VAR ( $P < 0.05$ ). Given the strong effect of BW on lamb performance, the large data set of twins was examined to determine whether variation in ovulation rate or pattern influenced BW. While year born, lamb birth date and sex and ewe age and weight before mating affected BW of the lamb ( $P < 0.001$ ), neither ovulation rate nor pattern (i.e., both ova released from one ovary or one from each) were associated with BW of the lambs. In summary, the BW was strongly associated with survival and growth of the lamb. The litter VAR had little or no effect on survival and did not negatively affect growth of the lamb. Thus, birth weight, but not variability in birth weight within a litter, appears to be a major driver of lamb performance.

**Key Words:** sheep, lamb survival, lamb growth

**M487 Effect of different rearing systems on growth, nutrient utilization and serum indices of early weaned Hu twin lambs.** Hai-chao Wang, Jian-min Chai, Yan Tu, Nai-feng Zhang, Tao Ma, Bo Wang, and Qi-yu Diao\*, *Feed Research Institute of Chinese Academy of Agricultural Sciences, Key Laboratory of Feed Biotechnology of the Ministry of Agriculture, Beijing, China*.

This experiment was conducted to study the effect of different rearing systems on growth, nutrient utilization and serum indices of early weaned Hu twin lambs. Twenty-four pairs of newborn Hu male twin lambs were equally divided into 2 groups according to paired experiment design. In each pair, one lamb was artificially reared (AR) with milk replacer, while the other one was ewe reared (ER). Body weight was recorded and blood samples were collected every 10 d. Two digestion trials were undertaken during d 45 to 55 and d 75 to 85 with 4 pairs of lambs from each group. On d 60 and 90, another 3 pairs of lambs from each group at each time point were euthanized for weighing gastrointestinal organs. The data of body weight and serum indices (total protein, cholesterol, glucose, triglyceride, albumin, blood urea nitrogen, alkaline phosphatase activity, and immunoglobulin) were analyzed using PROC ANOVA with repeated measure, while the others were analyzed using PROC paired *t*-test of SAS 9.1. The results showed that the BW of lambs in ER group was higher than that in AR group on d 30 ( $P < 0.05$ ). No difference was observed in the final BW of AR group and ER group (21.02 vs 21.92 kg,  $P > 0.05$ ) or the overall average daily gain (205.97 vs 217.01 g/d,  $P > 0.05$ ). On d 60, the proportion of the rumen to 4 stomachs (weight basis) of lambs in AR group was higher ( $P < 0.05$ ) while that of omasum and abomasum were lower ( $P < 0.05$ ) than those in ER group. The proportion of small intestine was higher in AR group than that in ER group ( $P < 0.05$ ). There was no difference in the nutrient digestibility ( $P > 0.05$ ) or the serum concentration of total protein, cholesterol, glucose, triglyceride, albumin, blood urea nitrogen, alkaline phosphatase activity, and immunoglobulin (G, A, and M) ( $P > 0.05$ ). In general, lambs reared with milk replacer achieved the same growth performance as their ewe-reared counterparts.

**Key Words:** lamb, milk replacer, growth performance

**M488 Preliminary results of the feedlot performance of South African Boer goats.** Tertius S. Brand<sup>1,2</sup>, Daniel A. van der Merwe<sup>2</sup>, Louw C. Hoffman<sup>2</sup>, and Emiliano Raffrenato<sup>2</sup>, <sup>1</sup>*Directorate: Animal Sciences, Department of Agriculture, Western Cape Government, Elsenburg, South Africa*, <sup>2</sup>*Department of Animal Sciences, Stellenbosch University, Stellenbosch, South Africa*.

Goats are more commonly reared in extensive production systems in South Africa and are directly marketed from the farm at a weight of less than 30 kg. This study was performed to determine the effect of dietary energy content on the production of South African Boer goats in a feedlot system to be able to increase marketing weight. In this trial, 53 Boer goats were randomly divided into 3 dietary treatments that varied in energy content, giving a low, 11.3 MJ ME/kg feed (18 goats), medium, 12.0 MJ ME/kg feed (16 goats) and high energy diets, 12.7 MJ ME/kg feed (19 goats). The diets were equal in terms of crude protein (17.0%), calcium (0.86%) and phosphorus (0.36%). These goats were reared in individual pens for a period of 8 weeks, where the trial diets were fed on an ad libitum basis and growth and feed intake were monitored. Initial body weight was 22.4 kg and after 8 weeks the goats on the medium diet had gained the most weight, weighing 34.6 kg followed by the low energy (33.4 kg) and the high-energy diets (31.4 kg). The difference between the 3 treatments were significant at  $P = 0.01$ . The average daily gains differed significantly ( $P = 0.036$ ) with goats on the medium energy diet attaining the highest ADG of 0.253 kg/day, while goats on the high-energy diet had the lowest ADG of 0.183 kg/day. This suggests that the energy content of the medium energy diet is probably close to the energy requirements of the goats, while the low diet does not contain sufficient energy to maintain higher growth rates. Goats are also not adapted to digest feeds with high-energy contents

and therefore can probably not use the excess energy for growth. Feed intake differed between the 3 treatments ( $P < 0.001$ ) with the goats on the low energy diet having the highest DMI of 1.456 kg/day which decreased with an increase in dietary energy content (1.112 kg/d for the high energy diet). The feed intake for the medium energy group was intermediate at 1.284 kg/d. The feed conversion ratio (FCR) did not differ significantly for the 3 treatments and was calculated to be 6.61

kg feed/kg weight gain. Goats fed the medium energy diet displayed the best growth at a moderate feed intake, even though the FCR between the diets did not differ significantly.

**Key Words:** Boer goat, dietary energy, feed intake



## Swine Species

**M489 Long-term effects of dietary calcium montmorillonite on swine productivity in a conventional farm in Japan.** Fang Chi\*<sup>1</sup>, Munetaka Oi<sup>2</sup>, Tomohiro Furuichi<sup>2</sup>, San Ching<sup>1</sup>, and LeAnn Johnston<sup>1</sup>, <sup>1</sup>Amlan International, Chicago, IL, <sup>2</sup>Toyoura Veterinary Clinic, Kanagawa, Japan.

Dietary clay (Calibrin-Z; CZ), has been shown to enhance weaning pig performance due to its barrier function in the intestine. An 18-month trial was conducted to investigate the long-term feeding effects of CZ in swine. Sixteen crossbred sows (3.2 parities) were randomly assigned to 2 TRT. The sows were fed an antibiotic free diet with 0 (CON) or 0.1% CZ during the trial. The farrowings were between Nov 2012 and Mar 2013, and from Apr to Aug 2013. Litter size and weaning weights were recorded (sow was the experimental unit). Weaned pigs from approximately 1/2 of the litters (362 pigs) were divided into 2 TRT in a split-plot arrangement based on sow TRT, and fed diets with 0 or 0.1% CZ until slaughtered (litter was the experimental unit). Pigs were weighed and blood samples collected at weaning, d-60, 90, 140, and end of trial. Feeds were formulated to meet Japanese feeding standards for swine. Total born from the 2 farrowings was not different ( $P > 0.05$ ) averaging 12.7 for the sows fed the CON vs. 14.4 piglets for the sows fed 0.1% CZ. However, total pigs born in the second litter was numerically higher (12.5 vs. 15.8;  $P > 0.05$ ) when CZ was fed. This improvement in the size of the second litter suggests that feeding 0.1% CZ for a longer period may have positive effects on sow reproductive performance. The combined data of 2 cycles of G-F pig performance, showed pigs fed 0.1% CZ diets needed 4.9 fewer days to reach market weight (166.8 vs. 161.9 d;  $P < 0.01$ ) although ADG was not improved (840 vs. 864 g;  $P > 0.05$ ). Total feed consumption was lower in pigs fed the CZ diets (308.6 vs. 287.4 kg;  $P < 0.01$ ), and therefore, an improvement of FCR (2.92 vs. 2.75;  $P < 0.05$ ) was obtained when pigs consumed diets containing 0.1% CZ. There were no differences ( $P > 0.05$ ) in the serum biochemical markers concentrations, such as total protein, albumin, GOT, GTP,  $\gamma$ GTP and IgG etc. In conclusion, pigs fed commercial diets containing 0.1% Calibrin-Z under normal conditions in Japan may increase numbers of total born and born-alive, and improve FCR in growing-finishing phases.

**Key Words:** sow litter size, growing-finishing, calcium montmorillonite

**M490 Effect of dietary micronutrient on the transcriptome of boar semen.** Dianelys Gonzalez-Pena\*, Robmay Garcia, and Robert V. Knox, and Sandra L. Rodriguez-Zas, *University of Illinois at Urbana-Champaign, Urbana, IL.*

Micronutrients are important co-factors or substrates to enzymes that protect and repair DNA against damage or methylation. Micronutrient deficiency could induce genomic instability and changes in DNA leading to cell apoptosis and degenerative diseases. The objective of this study was to compare the sperm transcriptome of boars receiving a diet supplemented with high amounts of methylating micronutrients (methionine, choline, betaine, vitamin B6, folate, and vitamin B12) relative to boars with non-supplemented diet. Semen mRNA from 4 boars was profiled and single-end reads were mapped to the *Sus scrofa* reference genome (UCSC susScr3) using Tophat v2.0.12. In total, 477 transcripts from 446 genes were tested and 73 transcripts from 72 genes were differentially expressed between the supplemented and non-supplemented boars using Cufflink v2.2.1 ( $P$ -value  $< 0.05$ ). Among the differentially expressed transcripts, 89% of those from boars with the supplemented

diet were overexpressed relative to those from non-supplemented boars. The porcine seminal protein II (PSP-II) was under-expressed; meanwhile, the AWN protein (AWN) was overexpressed in boars with the supplemented diet relative to non-supplemented. PSP-II encodes a protein with paradoxical functions: in vitro PSP-II enhances viability, motility and mitochondrial activity of highly-extended boar spermatozoa; in vivo PSP-II triggers the recruitment of polymorphonuclear leukocytes and T cells in the uterus of the sow after mating. AWN encodes a sperm surface-associated protein that participates in sperm adhesion to the egg. Functional analysis of the differentially expressed genes using DAVID identified one category cluster (enrichment score  $> 6$ ) related to cellular metabolism and biosynthetic process, structural constituents of ribosomes and structural molecular activity. The limited differentially expressed transcripts and the solely functional category enrichment suggest that methylating micronutrients in high quantities had no impact on the transcriptome of sperm cells.

**Key Words:** micronutrient, transcriptome, semen

**M491 Effects of supplementing high-fiber diets with a multi-carbohydrase enzyme on net portal flux of essential amino acids and urea-nitrogen in growing pigs.** Atta K. Agyekum\*<sup>1</sup>, Elijah Kiarie<sup>2,1</sup>, and Charles M. Nyachoti<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Manitoba, Winnipeg, MB, Canada, <sup>2</sup>DuPont Industrial Biosciences, Marlborough, Wiltshire, UK.

The present study investigated the effects of adding a multicarbohydrase enzyme (MC; 4000 units/g xylanase, 1000 units/g  $\alpha$ -amylase, 500 units/g protease, 150 units/g  $\beta$ -glucanase) to a high-fiber diet on net portal fluxes of essential AA (EAA) and urea-N in growing pigs. Five female pigs (22.8  $\pm$  1.6 kg BW), with permanent catheters in the portal vein, ileal vein and carotid artery, were fed 3 isocaloric/nitrogenous diets at 4% of their BW once daily at 0900 h for 7 d in a replicated 3  $\times$  3 Latin square design. The diets contained corn and soybean meal with 0% (CTRL) or 30% distillers dried grains with solubles (DDGS; HF) produced from a 1:1 mixture of wheat and corn. The third diet was supplemented with MC in addition to the 30% DDGS (HF+MC). On d 7, para-amino hippuric acid was infused into the ileal vein (to measure flow rate) and blood was sampled from the portal vein and carotid artery for 7 h after feeding to assay EAA and urea-N. Portal absorption of nutrients was derived by multiplying the porto-arterial plasma concentration differences by portal vein plasma flow. Diet had no effect ( $P > 0.10$ ) on postprandial portal vein plasma flow rate and net urea-N flux, but portal urea-N was lower ( $P < 0.05$ ) during the first h and tended to be lower ( $P = 0.07$ ) over the 7 h in pigs fed CTRL. Postprandial portal Arg, Ile, Leu, Trp, and Val were lower ( $P < 0.05$ ) in HF-fed pigs from 30 to 240 min than CTRL-fed pigs and MC supplementation tended ( $P < 0.10$ ) to or improved ( $P < 0.05$ ) portal fluxes of those AA. Further, pigs fed CTRL had higher ( $P < 0.05$ ) net portal fluxes of the above-mentioned AA from 30 to 120 min postprandial. Also, CTRL-fed pigs had higher ( $P < 0.05$ ) net portal fluxes of most EAA and pigs fed HF+MC had higher ( $P < 0.05$ ) Lys and similar Met and Phe ( $P > 0.10$ ) net portal fluxes compared with CTRL-fed pigs. The results showed that supplementing the HF diet with MC improved net portal appearance and fluxes of some EAA in growing pigs.

**Key Words:** fibrous diet, multicarbohydrase, net portal flux

**M492 Lifetime reproductive performance of South African Mukota sows following bio-stimulation protocols.** Dennis O. Ume-siobi\* and Matthew C. Achilonu, *Central University of Technology, Free State, Bloemfontein, South Africa.*

Bio-stimulation is the stimulus evoked by the presence of a male which induces sexual and oestrus expressions in females through genital stimulation, pheromones, and or other external cues. The aim of this study was to comparatively evaluate the effects of 2 bio-stimulation methods on lifetime reproductive performance of sows, arranged in a 2x3 factorial experiment. Thirty sows were assigned per group to each of 2 bio-stimulation protocols, twice daily (08:30 to 09:00 and 16:00 to 16:30) per 30 min duration in a 4-week observation per parity over 3 parities. The bio-stimulation protocols involved (1) fence-line boar exposure (FBE): in which sows were exposed to fence-line boars during oestrus detection and (2) physical boar exposure (PBE): in which sows received physical contact with the boar during oestrus detection. Following the bio-stimulation applications to each sow treatment group, oestrus was synchronised in sows by a single subcutaneous injection of P.G. 600 (400 IU PMSG with 200 IU HCG/5 mL dose/animal; Intervet Inc., Millsboro, DE). After the onset of oestrus, sows on each treatment were artificially inseminated using semen from the same boars and collections. All experimental females received inseminations of  $3.5 \times 10^9$  sperm/80 mL at 24 h after onset of oestrus. Physical boar exposure before oestrus induction (PG600) evoked the highest ( $P \leq 0.01$ ) reproductive performance in sows in parity 3 compared with those in parity 2 and 1, respectively. Sows that received physical boar exposure in parity 3 produced a higher ( $P \leq 0.05$ ) oestrus expressions ( $98 \pm 23.1$  vs.  $77.5 \pm 25.5\%$ ), Non-return rate ( $92.1 \pm 6.3$  vs.  $74.4 \pm 3.5\%$ ), farrowing rate ( $88.2 \pm 16.5$  vs.  $63.8 \pm 7.1\%$ ), litter size ( $12 \pm 0.02$  vs.  $8 \pm 0.05$ ) and live piglets ( $10 \pm 0.05$  vs.  $6.5 \pm 0.02$ ), respectively compared with those from sows that received fence-line boar exposure in parity 3. Results suggest that direct exposure of boars to sows before artificial insemination following oestrus induction progressively enhances the lifetime reproductive performance in South African Mukota sows.

**Key Words:** boar effect, reproductive longevity, Mukota pigs

**M493 Improved piglets performance with protected sodium heptanoate.** M. Puyalto\*<sup>1</sup>, P. Honrubia<sup>1</sup>, M. I. Gracia<sup>2</sup>, and J. J. Mallo<sup>1</sup>, <sup>1</sup>Norel S.A., Spain, <sup>2</sup>IMASDE Agroalimentaria S.L., Spain.

The objective of the present study was to evaluate the effect in piglets productive parameters of sodium heptanoate protected with hydrogenated palm fatty acids distillate sodium salt (HEPT'ON) in a control diet without any additive. Thirty-two 21-d-old piglets ( $6.5 \pm 0.5$  Kg) were randomly divided in 2 groups: control (C) and 70% Na-heptanoate protected (HEPT'ON) 3 kg/t (21–35 d) and 1 kg/t (36–49 d). Every treatment was replicated 4 times and each replicate consisted of 4 piglets in the first 2 weeks and 3 piglets in the second 2 weeks of the trial. Mash feeds and water were offered ad libitum. BW, ADG, ADFI and FCR were recorded for every period. At the end of each period (35 d and 49 d), one piglet per replicate was euthanized and samples from the ileum and cecum were taken to analyze gut microflora. Data were analyzed as a completely randomized design by GLM of SPSS v. 19.0. Piglets receiving sodium heptanoate protected with hydrogenated PFAD sodium salt (HEPT'ON) tended to have higher final body weight ( $15.997$  kg vs  $14.201$  kg;  $P = 0.0686$ ). The use of HEPT'ON resulted in a tendency to improve FCR ( $P = 0.0660$ ). Also the ADG was numerically improved in this group ( $P = 0.0686$ ). No significant differences were observed in count of *Lactobacillus* and *E. coli*. We can conclude that the use of sodium heptanoate protected with hydrogenated PFAD sodium salt may

be able to improve productive parameters in piglets compared with the control treatment. Further research is needed.

**Key Words:** protected sodium heptanoate, piglets

**M494 Effects of dietary supplementation organic medicinal charcoal (Olga Black) on aflatoxin adsorption capacity, digestibility, population of *Lactobacillus* and *E. coli* in feces, and the fecal odor emission by In vitro and In vivo.** Kwang-Sik Kim\*<sup>1</sup>, Jin-Ho Cho<sup>2</sup>, Ki Hyun Kim<sup>1</sup>, and Young Hwa Kim<sup>1</sup>, <sup>1</sup>National institute of animal science, Cheonan, Chungnam, Korea, <sup>2</sup>Chungbuk National University, Cheongju, Chungbuk, Korea.

This study was performed to evaluate effects of dietary supplementation organic medicinal charcoal (OMC: Olga Black) on aflatoxin adsorption capacity, digestibility, population of *Lactobacillus* and *E. coli* of feces and the fecal odor emission by in vitro and in vivo in finishing pig diet. Exp 1. the aflatoxin adsorption capacity of OMC, pyrolygneous charcoal (PC), and coconut tree charcoal (CC) was measured using the ELISA test kit. The aflatoxin adsorption capacity was 100, 10 and 20% in OMC, PL and CC, respectively ( $P < 0.05$ ). The effects of OMC on digestibility of dry- and organic matter were evaluated by in vitro prediction methods (Boisen, 1991). Treatments of in vitro and in vivo included: 1) CON (basal diet contaminated with 1,543ppb of deoxynivalenol and 596ppb of zearalenone); 2) OMC (basal diet + 0.25% Olga Black); 3) PC (basal diet + 0.50% Pyrolygneous Charcoal); 4) CC (basal diet + 0.50% Coconut tree Charcoal). The digestibility of OMC on dry matter was significantly higher than CON, PC, and CC ( $P < 0.05$ ). The digestibility of OMC on organic matter was significantly higher than CON ( $P < 0.05$ ). Exp 2. a 10-d trial with 20 [(Yorkshire  $\times$  Landrace)  $\times$  Duroc] finishing pigs (BW =  $81 \pm 3.3$  kg) was conducted to investigate the population of *Lactobacillus* and *E. coli* in feces, and the fecal odor ( $\text{NH}_3$ ,  $\text{H}_2\text{S}$ , total mercaptans, and acetic acid) in vivo. The fecal  $\text{NH}_3$  and  $\text{H}_2\text{S}$  emissions of OMC were significantly lower than CON, PC, and CC ( $P < 0.05$ ). Several *Lactobacillus* in feces of OMC and CC were significantly higher than CON and PC ( $P < 0.05$ ). Several fecal *E. coli* of OMC and CC were lower than CON and PC ( $P < 0.05$ ). It was thought that organic medicinal charcoal is useful as a feed additive in pig diets because it improves the digestibility of feed and the fecal odor, and has positive effect on the population of microorganism in feces.

**Key Words:** swine, organic medicinal charcoal, fecal

**M495 Effect of a functional feed additive on the stress nervous modulation response—Application on both the feeding behavior and performance of lactating sows and their litters.** Mónika Korondi<sup>1</sup>, Bertrand Medina\*<sup>2</sup>, and Tamás Tóth<sup>1,3</sup>, <sup>1</sup>Adexgo Ltd., Balatonfüred, Hungary, <sup>2</sup>Laboratoires Phodé, Terssac, France, <sup>3</sup>Department of Animal Nutrition, University of West Hungary, Mosonmagyaróvár, Hungary.

Recently, the effects of a 1-week administration of cyclic terpene (d-limonene) on neurotransmitters as well as its expected anti-stress effect were evaluated on mice. Considering this, the preliminary aims of Laboratoires Phodé were to investigate the effect of specific and purified fractions of natural plant extracts (*Citrus* sp.) characterized by a high level of d-limonene (135000 ppm) on the feeding behavior (total feed intake through the lactation phase) and performance of lactating sows and their litters (piglet weaning weight). The trial was performed on sows (n = 98, Topigs genetic stock, ALW =  $262 \pm 26.6$  kg) over 3 repetitions from farrowing (d0) to inseminating period (d36). Control (CTL) and Treated (VéO) groups of sows were completed by setting

lactation pairs under the same housing (individual farrowing pens) and feeding conditions (lactating feed: 17.1% CP, 5.57 Mcal/kg ME with a twice-a-day dispensing). The administration of the functional feed additive (VéOPremium, Laboratoire PHODÉ, France – 250 g/MT) was started with a 4-d pre-feeding period before the sows were moved to the farrowing barn (d-4). Data were analyzed under non-parametric tests (Kolmogorov-Smirnov test, Levene-test, independent-samples *t*-test) using SPSS 13.0 for Windows. There were no significant differences ( $P > 0.05$ ) between groups regarding the body weight loss of lactating sows. The average lactating FI was significantly higher (+ 4.7%,  $P \leq 0.05$ ) in the VéO group compared with the CTL one ( $157 \pm 6.3$  vs  $150 \pm 8.3$  kg, respectively). Regarding the average number of weaned piglet/litter of sow, no significant difference was observed between groups (12.25 vs 12.3 piglets/litter, for both VéO and CTL groups, respectively). The average live-weight (ALW) of piglets at weaning showed significantly heavier value (+6.5%,  $P \leq 0.05$ ) in the VéO group (ALW =  $7.37 \pm 0.72$  kg) although the piglets were weaned after 26.1 d on average, while the average number of days from birth to weaning was 26.6 in the control group (ALW =  $6.92 \pm 0.77$  kg). These results suggest that the administration of these specific and purified fractions of natural plant extracts are able to support higher sow feed intake and piglet weaning weight ( $P \leq 0.05$ ) through lactation phase.

**Key Words:** botanical extract, lactating sow, stress

#### **M496 Improved piglets performance with sodium heptanoate.**

M. Puyalto\*<sup>1</sup>, P. Honrubia<sup>1</sup>, M. I. Gracia<sup>2</sup>, and J. J. Mallo<sup>1</sup>, <sup>1</sup>Norel S.A., Spain, <sup>2</sup>IMASDE Agroalimentaria S.L., Spain.

The objective of the present study was to evaluate the effect in piglets productive parameters of sodium heptanoate in a control diet without any additive. Forty-eight 21-d-old piglets ( $6.5 \pm 0.5$  kg) were randomly divided in 2 groups: control (C) and 98% Na-heptanoate (NaC7) 3 kg/t. Every treatment was replicated 6 times and each replicate consisted of 4 piglets. Mash feeds and water were offered ad libitum. BW, ADG, ADFI and FCR were recorded for every period. At the end of each period (35 d and 49 d), one piglet per replicate was euthanized and samples from the ileum and cecum were taken to analyze gut microflora. Besides, samples of duodenum, jejunum and ileum epitheliums were obtained to determine their development status. Data were analyzed as a completely randomized design by GLM of SPSS v. 19.0. Piglets receiving sodium heptanoate had higher final body weight without significative differences (16 Kg vs 16.2 Kg;  $P = 0.7583$ ). However the use of NaC7 resulted in an improvement ( $P = 0.0431$ ) in the FCR (1.416 vs 1.324) when compared with the control treatment. No significant differences were observed in count of *Lactobacillus* and *E. coli*. Neither were there significant differences in villi height; however, there was a tendency to show deeper crypts in duodenum ( $P = 0.1058$ ) in the animals that received the sodium heptanoate ( $441.1 \mu\text{m}$  vs  $374.3 \mu\text{m}$ ). We can conclude that the use of sodium heptanoate is able to improve productive parameters in piglets compared with the control treatment.

**Key Words:** sodium heptanoate, piglet

#### **M497 Feeder space may affect pig performance in the early growing-finishing period.**

David Solà-Oriol, Patricia Romero, Deborah Temple, Laia Blavi\*, and Josep Gasa, *Animal Nutrition and Welfare Service, Department of Animal and Food Sciences, Universitat Autònoma de Barcelona, Bellaterra, Spain.*

The number pigs fed per feeder space decrease along the whole growing period. However moving pigs from the weaning to the growing-

finishing facilities is a challenging period and feeder space may affect performance, growth and homogeneity of pigs. The aim of the present work was to study the effect feeder type in terms of feeder space (2 or 5) at the start of the growing period. At weaning a total of 264 28-d-old piglets [Pt × (LD × LW)] were distributed into 24 pens (11 piglets/pen) according to sex and initial body weight (BW) for the transition period (T; 28 to 64d). During T period, a commercial INOX lidded hopper was used (3 feeder spaces of 17cm/pen). When pigs moved to the growing facilities, the 24 pens were maintained and split into 2 groups of 12 according sex, BW and feeder type. Two different commercial concrete hoppers differing in feeder space were used (2 feeder space of 23cm or 5 feeder space of 18cm/pen) Pigs were feed the same diets during T and growing (G) period. Feed was offered ad libitum in mash form. Therefore, 2 experimental treatments were performed at the start of the growing period (64 to 92d) according to feeder space and sexes taking into account the same conditions from 28 to 64d. Individual BW was recorded at 28, 64 and 92d of age and average daily gain (ADG) and coefficient of variation (CV) of the pen were calculated. Behavioral measures and lesion scoring were also monitored on d 74 of age. No sex effect was observed for the T and G ( $P > 0.10$ ). Higher BW (33.1 vs 32.5kg;  $P = 0.061$ ) and ADG (583 vs 562g/d;  $P = 0.062$ ) were observed for the animals raised in the 5 feeder space hoppers compared with the 2 feeder spaces. Moreover, the CV of the animals at the end of T was reduced (12%) along G with a 5 feeder spaces while increased (1.2%) with 2 feed spaces. Finally, no differences were observed in terms of aggressions and lesions due to feeder space during G period. It is concluded that feeder space may affect growth performance and body weigh homogeneity in growing pigs.

**Key Words:** feeder space, performance, pig homogeneity

#### **M498 The effect of immunocastration on growth performances and carcass quality of heavy males and gilts.**

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A trial was carried out with 48 Duroc × (Landrace × Large White) pigs intended for dry-cured ham; 24 males and 24 gilts of 46.3 and 42.9 kg BW, respectively. For that end, surgically castrated males are used to avoid sexual odour and minimum levels of fat thickness, measured at Gluteus medius muscle (GM; >16 mm), are required to improve the ripening process and ham quality. Currently, the main problems are the future prohibition in EU of this type of castration and the lack of fat in gilts. The effect of immunization against GnRH on growth performances and carcass quality was evaluated. There were 4 experimental treatments; surgically castrated males (CM), immunocastrated males (IM), entire gilts (EG) and immunocastrated gilts (IG). The CM had been castrated at 5 d of age. Immunization was carried out in both sexes with 2 injections of Improvac (Zoetis) with an interval of 4 weeks (1st: 44.6 kg BW, 2nd: 70.6 kg BW, as average). The replicate was a pen with 3 pigs (n = 4) for performance traits and the animal (n = 12) for carcass traits. A commercial diet, based on barley, wheat, and vegetable meals and containing 13.74 MJ DE/kg, 14.1% CP, and 0.75% Lys, was provided ad libitum through the trial. Pigs were slaughtered at 127 kg BW. Data were analyzed by ANOVA and the statistical model included the sex as main effect. Duncan test was used to compare pairs of means. From 1st to 2nd injection, the EG grew faster than IG with CM and IM being intermediate ( $P < 0.001$ ) but this effect disappeared after. At the end of the trial, the CM had higher feed:gain ratio than the other treatments ( $P < 0.05$ ). No effect of immunocastration was observed on carcass



yield but the IG had wider fat depth at GM and lower lean yield than EG with CM and IM being intermediate ( $P < 0.05$ ). Intramuscular fat content was not affected by sex. It is concluded that the immunization against GnRF might be a good strategy to improve some traits desirable in pigs intended for dry-cured ham because, in gilts, it increased fat depth and, in males, it reduced feed:gain ratio in comparison with those surgically castrated.

**Key Words:** pig immunocastration, growth performance, carcass quality

**M499 Is the lactation period the main variable responsible for reducing the efficiency of the swine production?** Sergi López-Vergé, David Solà-Oriol, Laia Blavi\*, and Josep Gasa, *Animal Nutrition and Welfare Service, Department of Animal and Food Sciences, Universitat Autònoma de Barcelona, Bellaterra, Spain.*

Pig body weight (BW) homogeneity across the entire production cycle plays an important role in the swine industry because it directly affects efficiency and farm occupation time, mainly in regards to the growing-finishing facilities. The aim of this study was to know if the BW variability is equally distributed along the pig productive life or is primarily important at one particular stage. A total of 433 crossbred piglets [Pietrain  $\times$  (Landrace  $\times$  Large White)] from 40 litters were used. All diets were offered ad libitum. During the nursery period, piglets were fed a diet formulated to contain 11.0 MJ/kg NE, 20.2% CP and 1.37 Lys. Thereafter, the animals were moved to a growing-finishing facility. Pigs were fed the same commercial growing and finishing diets (10.0 MJ/kg NE, 16.0% CP, 0.95 Lys). All animals were individually weighed from birth until slaughter at d 2 (CF; cross fostering), d 28 (weaning), d 42 (14 d post-weaning), d 63 (35 d post-weaning) and every 3 weeks until pigs were slaughtered. The relationship between BW at one stage and the next succeeding one along the whole cycle was analyzed by using the CORR procedure of SAS. Moreover, the variability at all periods was calculated taking into account the coefficient of variation (CV, %) by using the MEANS procedure of SAS. A strong relationship was observed between 2 consecutive steps along the whole production cycle (Pearson Correlation Coefficient ( $r$ ) ranging from 0.85 to 0.96 ( $P < 0.001$ ) until the first group of pigs were slaughtered), except for the lactation period (from birth to weaning,  $r = 0.46$ ,  $P > 0.001$ ). At the same time, the CV was higher at the first stages (22.17% until 35 d post-weaning) and then decreased until slaughter (8.42%). Results showed that the events occurred in the first stages of pig life, but especially during the lactation period (in terms of variability), have a huge effect along the subsequent performance of pigs, suggesting a more accurate management for the smallest piglets just after birth.

**Key Words:** correlation, piglet, variability.

**M500 Evaluation of the efficacy of sodium heptanoate or butyrate in front of an enterotoxigenic *Escherichia coli* (ETEC) K88 oral challenge in piglets.** P. López-Colom<sup>1</sup>, L. Castillejos<sup>1</sup>, M. Puyalto<sup>2</sup>, J. J. Mallo<sup>\*2</sup>, and S. M. Martín-Orúe<sup>1</sup>, *<sup>1</sup>Animal Nutrition and Welfare Service, Departament de Ciència Animal i dels Aliments, Universitat Autònoma de Barcelona, Bellaterra, Barcelona, Spain, <sup>2</sup>Norel S.A., Madrid, Madrid, Spain.*

This study evaluated the efficacy of sodium heptanoate (HEPT'ON) or sodium butyrate (GUSTOR BP70; Norel S.A.) against ETEC K88 in weanlings. A total of 72 3-week-old piglets were divided into 24 pens and 3 experimental groups: Plain diet (CTR); supplemented with GUSTOR BP70 (BUT); or supplemented with HEPT'ON (HPT), both

at 3 kg/t and containing 70% of acid salt protected with vegetable fat. Intake and weight were monitored along 15 d. After one week, animals were orally inoculated with ETEC K88 ( $1 \times 10^9$  cfu) and fecal consistency and rectal temperature evaluated afterward. On d 4 and 8 post inoculation (PI) one animal per pen was euthanized to evaluate inflammatory response (TNF $\alpha$  and Pig-MAP) and counts of enterobacteria and *E. coli* in ileal-colonic contents and ileal mucosa scrapes. No significant differences were seen in performance although numerical values were higher for the experimental diets (217, 264 and 243 g ADFI and 117, 123 and 124 g ADG for CTR, BUT and HPT). No significant differences were found in fecal consistency, rectal temperature and inflammatory markers. Regarding microbiological changes, no significant differences were observed between treatments in the colon. Nonetheless, in ileum digesta, there were numerical differences ( $P = 0.126$ ) with higher number of enterobacteria in both acids treatments at d 4 PI, that was correlated to an increase in the number of enterobacteria and *E. coli* recovered from the ileal scrapes (4.30, 5.88 and 5.46 log cfu *E. coli* for CTR, BUT and HPT,  $P = 0.003$ ). This increase, however, was not found at d 8 PI when even a decrease in the numerical values of *E. coli* in ileal digesta was seen for BUT treatment (6.58, 6.01 and 6.93 log cfu for CTR, BUT and HPT,  $P = 0.036$ ) that also showed a tendency to improve the colonic consistency ( $P = 0.099$ ). These results suggest the potential of BUT to improve the recovery of the animals after an ETEC challenge. More studies under field conditions with a higher number of animals would be needed to confirm the numerical differences found in performance.

**Key Words:** *Escherichia coli* K88, sodium heptanoate, sodium butyrate

**M501 The use of multiple imputation for the accurate measurements of individual feed intake by electronic feeders.** Shihui Jiao<sup>\*1</sup>, Christian Maltecca<sup>1</sup>, Yijian Huang<sup>2</sup>, and Kent A. Gray<sup>2</sup>, *<sup>1</sup>North Carolina State University, Raleigh, NC, <sup>2</sup>Smithfield Premium Genetics, Rose Hill, NC.*

Obtaining accurate individual feed intake records is a key first step in achieving genetic progress toward a more efficient pig for nutrient utilization. Feed intake records collected by electronic feeding systems contain errors (extreme values or outliers), which are due to feeder malfunction or animal movements. In this study, we introduce a new feed intake data editing strategy to replace errors and missing observations occurring in feed intake data, based on multiple imputation methods. Compared with the well-established linear mixed model (LMM) approach, multiple imputation either by using conditional distribution (MI) or by chained equation (MICE) results in increased accuracy of data adjustment in simulated phenotypes with artificially introduced errors. Feeder visit records in the simulated data sets were sampled from a data set including individual pig feed intake visits collected by Smithfield Premium Genetics from year 2004 to 2013. Three scenarios were considered in the analysis with 5%, 10% and 20% error visits simulated. Each scenario was replicated 5 times. Accuracy of the error-adjustments was measured as correlation between the true error-free daily feed intake (DFI) or average daily feed intake (ADFI), and the adjusted ones. Multiple imputation methods outperformed the linear mixed model approach in all scenarios with average accuracies of 96.71%, 93.45% and 90.24% obtained with MI and 96.84%, 94.42% and 90.13% obtained with MICE, compared with 91.0%, 82.63% and 68.69% using LMM for DFI with simulated error rate 5%, 10% and 20%, respectively. Similar results were obtained for ADFI. In conclusion, multiple imputation was introduced in this study as a more accurate

and flexible error-adjustment method for feed intake data collected by electronic feeders.

**Key Words:** feed intake, electronic feeder, multiple imputation

**M502 Effect of using nonconventional legumes (narbon vetch, black chickpea, and winter pea) as protein sources in piglet diets.**

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A trial was carried out to study the effect of the partial substitution of soybean meal (SBM) by alternative protein sources: black chickpea (*Cicer arietinum*; BC), narbon vetch (*Vicia narbonensis*; NV), and winter pea (*Pisum sativum*; WP) on growth performance of piglets during starter period. The anti-nutritional factors are the main handicap of including in a diet a high proportion of legumes with no previous treatment. The protease inhibitors (PI) (trypsin inhibitor; TI and chymotrypsin; CI) interfere in the absorption of proteins affecting feed intake and feed efficiency in young pigs. A total of 192 animals (96 barrows and 96 gilts), from PIC L-65 sires x Topigs 20 dams, of 11.6 ± 0.56 kg BW and 42 ± 2 d of age were used in the experiment. Piglets were allocated according to sex and initial BW. Four diets were provided: 1) control with 21% SBM, 2) 24% BC + 14.4% SBM, 3) 20% BC + 5% NV + 13.2% SBM and 4) 24% WP + 5% NV + 11.4% SBM. Diets had a similar nutritional profile (10.24 MJ NE/kg, 18% CP, and 1.25% Lys). Trial lasted 20 d (23.35 ± 1.42 kg BW as average). Data were analyzed as a random factorial design 2 (sexes) × 4 (diets) by GLM of SAS. Each experimental treatment was replicated 4 times considering the box, with 6 piglets, as the replicate. The PI content in the experimental diets was 1.03, 2.41, 1.57, and 1.13 units TI/mg feedstuff, and 1.85, 2.66, 2.24, and 2.60 units CI/mg feedstuff for diet 1, 2, 3, and 4, respectively. No significant difference was detected for any variable studied between barrows and gilts. The average daily gain was not affected by diet ( $P = 0.42$ ) but piglets fed diets 1, 2 and 3 ate more feed than those fed diet 4 (898, 951, 876 vs 836 ± 25.7 g/d, respectively;  $P < 0.001$ ). Diet 4 provided better feed to gain ratio than diets 1 and 2, with diet 3 in an intermediate position (1.54, 1.58, 1.51, and 1.44 ± 0.034 g/g, for diets 1, 2, 3, and 4, respectively;  $P < 0.005$ ). Although more studies are necessary, we can conclude that black chickpea, narbon vetch, and winter pea can substitute partial soybean meal in piglet diets during the starter phase.

**Key Words:** nonconventional legume, piglet, productive performance

**M503 Effects of breed, sex, and birth weight on growth and carcass composition traits in pigs.** L. L. Lo<sup>\*1</sup>, C. C. Tsai<sup>1</sup>, and R. S. Lin<sup>2</sup>, <sup>1</sup>Chinese Culture University, Taipei, Taiwan, Republic of China, <sup>2</sup>National I-Lan University, I-Lan, Taiwan, Republic of China.

Effects of breed, sex, and birth weight on growth and carcass composition traits were investigated using purebred Duroc, Landrace, and Yorkshire, respectively. Twenty-three sets of three littermates, consisting of a boar, gilt, and barrow for each of 3 breeds was used. All pigs were classified into 3 groups based on their birth weights (BWT): small (mean = 1.15 kg), medium (mean = 1.50 kg), and big (mean = 1.75 kg). Gilts and barrows from each littermate were slaughtered at 103.40 kg (SD = 13.98 kg) of body weight. A mixed model that included fixed effects of

breed, sex, birth group, interaction of breed and sex, and random effect of animal was used. Breed significantly affected growth traits. Landrace pigs had the highest ( $P < 0.05$ ) overall average daily gain than in other 2 breeds. The overall average daily gain was 608.93 ± 7.87 g, 649.00 ± 7.82 g, and 603.38 ± 8.61 g for Duroc, Landrace, and Yorkshire, respectively. Heavier pigs at birth showed significant advantage ( $P < 0.05$ ) on overall average daily gain (small = 595.99 ± 12.00 g, medium = 615.64 ± 6.17 g, large = 649.68 ± 7.65 g). Carcasses from Landrace pigs had thinner average backfat and tenth rib backfat thickness than those from Duroc breeds. Duroc pigs however had heavier ( $P < 0.05$ ) ham weight than that of Landrace pigs indicated the carcass characteristics of Duroc breed. Most carcass composition traits did not show the sex effect except for fat percentage and weight of tenderloin ( $P < 0.05$ ). Pigs with heavier birth weight had larger ( $P < 0.05$ ) Boston butt. In conclusion, the results suggest that breed, sex, and birth weight are important sources of variation for growth and carcass composition traits of pigs.

**Key Words:** birth weight, carcass, pig

**M504 Reducing malodorous compounds on swine in vitro fermentation using probiotics.** Yeon Jae Choi, Lovelia L. Mamuad, Seung Hyun Kim, and Sang Suk Lee\*, Suncheon National University, Suncheon, Jeonnam, South Korea.

Bacteria play a significant role in odor production. Thereby, we evaluated different bacteria for their ability to reduce malodorous compounds in swine using in vitro fermentation. Fresh cultures of *Lactobacillus plantarum* KACC 91016 (T1), *L. brevis* ATCC 14869(t) (T2), *Bacillus subtilis* KACC 10112 (T3), *Saccharomyces cerevisiae* KACC 30068 were used in this study. One hundred milliliters of anaerobic salt medium containing 10% fresh swine feces was transferred to serum bottles containing 1g DM of 50:50 soluble starch to casein ratio and inoculated with or without (control) 1mL of different microbial cultures and incubated for 24h. Highest ( $P < 0.05$ ) total gas production was observed in T1 and lowest in control. Opposite was observed in hydrogen sulfide (H<sub>2</sub>S) concentration, wherein lowest ( $P < 0.05$ ) H<sub>2</sub>S was observed in T1 and highest in control with 17.80% and 22.20%, respectively. Also, lower percent methanethiol, dimethyl sulfide and trimethylamine were observed in treated compared with control. Highest ( $P < 0.05$ ) concentration of total fatty acids, propionate, and butyrate were also observed in T1 and the highest ( $P < 0.05$ ) acetate was observed in T3. Among the bacteria used, addition of *L. plantarum* is the most effective for reducing malodorous compounds

**Key Words:** in vitro, malodorous compounds in swine, probiotic

**M505 Relationship between birth weight and subsequent piglet performance: A meta-analytic study.** Eloiza Lanferdini<sup>1</sup>, Ines Andretta<sup>2</sup>, Leonardo da Silva Fonseca<sup>1</sup>, Rennan Herculano Rufino Moreira<sup>1</sup>, Vinicius de Souza Cantarelli<sup>1</sup>, Rony Antônio Ferreira<sup>1</sup>, Alysson Saraiva<sup>3</sup>, Cesar Augusto Pospissil Garbossa<sup>1</sup>, and Márvio Lobão Teixeira de Abreu<sup>\*2</sup>, <sup>1</sup>Universidade Federal de Lavras, Lavras, Minas Gerais, Brazil, <sup>2</sup>Universidade Federal do Rio Grande do Sul, Porto Alegre, Rio Grande do Sul, Brazil, <sup>3</sup>Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.

The aim of this study was to evaluate the effect of birth weight of piglets on subsequent performance through a meta-analysis. The database totaled 2,691 piglets (15 articles published between 2002 and 2014). Average birth weights were 1.785, 1.511 and 1.129 kg for heavy, medium and light piglets, respectively. The selected database publications contained results of experiments with at least 2 weight groups of piglets

at birth and performance results. The meta-analysis was based on 3 sequential analysis: graphic, correlation and variance-covariance. The birth weight of piglets was positively correlated with body weight in the subsequent phases and with weight gain during the lactation period. On the other hand, the age at slaughter to reach 110 kg correlation with birth weight of piglets was negative. The birth weight of piglets affected ( $P < 0.05$ ) body weight and weight gain in the subsequent phases. Heavy piglets at birth showed an increase ( $P < 0.05$ ) of 6.6 kg in the slaughter weight, at 150 d of age, compared with light piglets. Piglets with medium weight at birth reached similar weight at slaughter ( $P > 0.05$ ) compared with heavy piglets and showed a difference ( $P < 0.05$ ) of 5.2 kg compared with light piglets. The different birth weight of piglets had no influence ( $P > 0.05$ ) on feed intake in the subsequent phases of growth. Feed efficiency improved ( $P < 0.05$ ) in medium and heavy piglets at birth in the growing and finishing phases. The slaughter age to reach 110 kg of live weight decreased ( $P < 0.05$ ) in the piglets born with medium and heavy weight, respectively, compared with piglets with low birth weight. Heavy piglets at birth (1.785 kg) performed better by increasing weight gain, and consequently greater final body weight and fewer days (10–12 d) to reach slaughter weight compared with light piglets (1.129 kg). Above 1.50 kg at birth the pig performance until slaughter is not influenced by birth weight.

**Key Words:** animal growth, meta-analysis, pig production

**M506 Meat quality of finishing pigs from sows supplemented with arginine during gestation.** Leonardo da Silva Fonseca<sup>1</sup>, Eloiza Lanferdini<sup>1</sup>, Rennan Herculano Rufino Moreira<sup>1</sup>, Rhuane Fillipe Chaves<sup>1</sup>, Peter Bitencourt Faria<sup>1</sup>, Marianne Kutschenko<sup>2</sup>, Eduardo Terra Nogueira<sup>2</sup>, Alysson Saraiva<sup>3</sup>, Cesar Augusto Pospissil Garbossa<sup>1</sup>, and Márvio Lobão Teixeira de Abreu<sup>\*1</sup>, <sup>1</sup>Universidade Federal de Lavras, Lavras, Minas Gerais, Brazil, <sup>2</sup>Ajinomoto Animal Nutrition, Limeira, São Paulo, Brazil, <sup>3</sup>Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.

The aim of this study was to evaluate carcass traits and meat quality of finishing pigs from sows supplemented with arginine during gestation. Piglets of 48 sows from parities 2 to 6 fed gestation diets with or without 1.0% L-arginine (Ajinomoto) supplementation from 30 to 60 d of gestation and from 80 d of gestation until farrow were used. Twenty 4 pigs (6 males and 6 females per treatment), selected by average weight (96.17 kg  $\pm$  3.74 for the pigs from control sows and 103.67 kg  $\pm$  4.05 for the pigs from arginine fed sows) were slaughtered at 150 d of age. *Longissimus dorsi* temperature and pH at the 12th rib were measured 45 min and 24 h after slaughter on the left half of the carcass. Loin eye area and the fat area were evaluated by drawing the outline of the muscle and the fat at the tenth rib on a paper and then scanning and measuring the area through ImageJ IJ 1.46r. *Longissimus dorsi* samples were collected and cooled to perform the color ratings. Saturation indices ( $C^*$ ) and hue angle ( $h^*$ ) were determined. Pigs from sows fed arginine during pregnancy were 7.80% heavier ( $P < 0.05$ ) at 140 d of age. The drip loss was negatively affected ( $P < 0.05$ ) by arginine supplementation which was 3.40% higher compared with the control group. This feature may negatively influence the product choice by the consumer as it is one of PSE meat characteristics. The loin depth of the control group was 13.70% lower ( $P < 0.05$ ), however the loin eye area and the loin fat area were not affected ( $P > 0.05$ ).  $L^*$ ,  $a^*$ ,  $b^*$ ,  $C^*$ , and  $h^*$  values as well as shear force and cooking loss were not affected ( $P > 0.05$ ) by arginine. Dietary supplementation of gestating sows with arginine increase pig weight and loin depth at slaughter; however, drip loss is also increased.

**Key Words:** amino acid, carcass, nutrition

**M507 Growth curve analysis of placental and fetal growth influenced by adjacent fetal sex status under crowded uterine conditions in pigs.** Brad A. Freking\* and Clay A. Lents, USDA, ARS, US Meat Animal Research Center, Clay Center, NE.

Intrauterine position and sex of adjacent fetuses in litter bearing species have been implicated in physiological and behavioral differences in males and females. Our objective was to establish growth curves for fetal and placental weight gain as influenced by sex status of flanking fetuses under crowded uterine conditions. Gilts were subjected to unilateral-hysterectomy-ovariectomy surgery at 160 d of age and mated at approximately 280 d of age. Gilts were assigned to be harvested at d 45, 65, 85, or 105 of gestation. A total of 297 pregnancies were evaluated in 4 contemporary groups. Position in the uterus relative to the cervix, fetal weight, and placental weight were recorded at harvest. Data were coded to test when each fetus was adjacent to 0, 1, or 2 opposite sex fetuses after removal of the first and last fetuses in each litter to remove bias of ends not being flanked on both sides. After this editing a total of 343, 651, and 303 fetuses were included in the analyses for 0, 1, or 2 opposite sex flanking fetuses, respectively. Nonlinear analyses fitted a logistic function ( $Wt = Ae^{kt}$ ) to the fetal and placental weight data to establish unique growth curves for each flanking sex status code. Two parameters were estimated representing the initial weight at day zero ( $A$ ) and a slope parameter ( $k$ ) with respect to time of gestation ( $t$ ). Gauss-Newton method was used in the NLIN procedure to generate solutions. A 4 df F-test was calculated to compare the ability of code-specific functions to account for more variation than the pooled function. The null hypothesis of a single pooled function to describe fetal weight was rejected ( $F = 2.49$ ;  $P < 0.05$ ) but not for placental weight ( $F = 0.80$ ;  $P > 0.1$ ). Code specific growth curves indicated placental weight separated only in the latter stage (d 105) of gestation whereas fetal weights separated as early as d 65 of gestation. Fetal growth development in pigs is influenced by sex status of adjacent fetuses, with increased growth by those surrounded by the same sex, and could be a potential source of variation in behavioral and reproductive differences later in life.

**Key Words:** pig, fetal growth, survival

**M508 Effect of mineral sources on reproductive performance in sows.** Tsung-cheng Tsai\*<sup>1</sup>, Gary A. Apgar<sup>2</sup>, Mark J. Estienne<sup>3</sup>, Mark Wilson<sup>4</sup>, and Charles V. Maxwell<sup>1</sup>, <sup>1</sup>University of Arkansas, Fayetteville, AR, <sup>2</sup>Southern Illinois University, Carbondale, IL, <sup>3</sup>Virginia Tech, Blacksburg, VA., <sup>4</sup>Zinpro Inc., Eden Prairie, MN.

Sows ( $n = 245$  from 3 stations) were blocked by parity [Pty 1: parity 1; Pty 2: parity 2 and 3; Pty 3: parity  $\geq 4$ ] and initial BW, and then randomly assigned to corn-SBM diets supplemented with either inorganic [IM: 120 ppm Zn (ZnO), 30 ppm Cu (CuSO<sub>4</sub>), and 50 ppm Mn (MnSO<sub>4</sub>)] or the same total level of minerals with AA complex of Zn, Cu and Mn (OM) replacing 50% of the IM in iso-mineral based diets which also provided 120, 165, 0.3, 0.3 ppm of inorganic Ca, Fe, I, and Se, respectively. Gestation (3391 kcal/kg of ME; 0.65% SID lysine) and lactation (3348 kcal/kg; 1.00% SID lysine) diets were formulated to meet 1998 NRC nutrient recommendations. Sow individual BW (at breeding, 110 d, 48 h postpartum, and weaning) and Intake (gestation: breeding to 110 d; lactation: postpartum to weaning) were collected to calculate ADG, ADFI and G:F ratio. During gestation, IM fed sows gained less weight (60.4 vs. 64.6 kg,  $P = 0.054$ ), consumed less feed (263.5 vs. 264.8 kg,  $P = 0.047$ ), and had lower G:F ratio (0.27 vs. 0.29,  $P = 0.044$ ) than OM fed sows. In lactation, sows fed IM had higher ADFI in wk 2 (5.85 vs. 5.19 kg/d,  $P = 0.021$ ), and weaned more light BW piglets per litter (<3.63 kg; 6.55 vs. 3.23%,  $P = 0.047$ ) than those fed OM, whereas number born alive (11.4 vs. 10.9,  $P = 0.238$ ), number weaned (10.2 vs. 9.8,  $P =$



0.184), piglets pre-weaning ADG (0.27 vs. 0.27 kg/d,  $P = 0.767$ ), and the percentage of pre-weaning mortality (13.1 vs. 12.9%,  $P = 0.923$ ) were similar for IM and OM fed sows, respectively. Of all parities, the Pty 1 and 2 sows gained more BW in gestation (66.1 and 67.5 vs. 53.9 kg,  $P < 0.001$ ) than Pty 3. In addition, the Pty 1 sows had lower overall lactation intake (99.0 vs. 124.7 and 126.2 kg,  $P < 0.001$ ) when compared with Pty 2 and 3 sows. Sows of Pty 2 delivered 8.9% heavier average BW piglets [1.67 vs. 1.50 (Pty 1) and 1.52 (Pty 3) kg,  $P < 0.001$ ], and had 11.5% higher piglet ADG [0.29 vs. 0.26 (Pty 1 and 3) kg/d,  $P < 0.001$ ]. Results of current study demonstrate that supplemental organic minerals improved gestation weight gain, G:F ratio, and produced more full value weaned pigs.

**Key Words:** organic and inorganic minerals, sow, reproductive performance

**M509 Transcriptome profile of boar spermatozoa as revealed by RNA-sequencing.** Jean M. Feugang<sup>\*1</sup>, Shengfa S. Liao<sup>1</sup>, William S. Sanders<sup>2,3</sup>, Jingqiao Lu<sup>4</sup>, Mark A. Crenshaw<sup>1</sup>, Scott T. Willard<sup>1,5</sup>, and Peter L. Ryan<sup>1,6</sup>, <sup>1</sup>Department of Animal and Dairy Sciences, Mississippi State University, Mississippi State, MS, <sup>2</sup>Department of Computer Science & Engineering, Mississippi State University, Mississippi State, MS, <sup>3</sup>Institute for Genomics, Biocomputing, and Biotechnology, Mississippi State University, Mississippi State, MS, <sup>4</sup>School of Medicine, Emory University, GA, <sup>5</sup>Biochemistry, Molecular Biology, Entomology and Plant Pathology, Mississippi State University, Mississippi State, MS, <sup>6</sup>Pathobiology and Population Medicine, Mississippi State University, Mississippi State, MS.

High-throughput RNA sequencing (RNA-Seq) overcomes the limitations of the current hybridization-based techniques to detect the actual pool of RNA transcripts in spermatozoa. The application of this technology in livestock can speed the discovery of potential predictors of male fertility. As a first step to identify novel gene products of fertility importance in pigs, we conducted a RNA-sequence analysis of boar spermatozoa. Fresh semen of 8 fertile boars (3 ejaculates/boar) were purchased from a commercial stud and pure motile spermatozoa were obtained through a discontinuous percoll gradient. Total sperm RNA were extracted using commercial kits with an in-column DNase digestion. The purity and integrity of RNA samples were checked and those with high quality parameters were used for deep-RNA sequencing to produce millions of short cDNA reads using Illumina RNA-Seq technology. Resulting reads were aligned to the pig reference genome to produce a genome-scale transcription map that consisted of both the transcript structure and the expression level of each gene (fragments per kilobase of exon per million fragments mapped). Total of 18,357 sequence tags were successfully mapped to all pig chromosomes and mitochondrial genome. Five chromosomes (2, 1, 6, 7, and 13) comprised the highest density of mapped transcripts (42%), while the bottom lowest density (8%) was found in chromosomes 10, 18, 16, 11, and Y. The Y chromosome and mitochondrial genome contained only 0.07% and 0.08% of total mapped sequence tags. Structural annotation revealed a diverse population of sperm transcripts comprising both coding and non-coding RNAs. Approximately 12,355 of sequence tags were annotated with ENSEMBL and rRNAs (e.g., 5s and 7sk), snRNA (e.g., U1 and U6), miRNAs (e.g., mir127 and mir935), and mitochondrial RNA (e.g., ND6 and CO1) constituted the most abundant sequence tags. We further confirmed the presence of selected genes (e.g., AQP11 and AQN-1) through RT-PCR. The findings revealed a large pool of coding and non-coding RNA in mature boar spermatozoa. The full investigation of this RNA

population will allow for the identification of those having critical roles during fertilization and early embryogenesis.

**Key Words:** gamete, reproduction, swine

**M510 Effects of chicken egg anti-F4 antibodies supplementation on performance and diarrhea incidences in enterotoxigenic *Escherichia coli* K88<sup>+</sup> challenged piglets.** Kolawole R. Aluko<sup>\*1</sup>, Deepak Ettungalpadi Velayudhan<sup>1</sup>, Lin Fang<sup>2</sup>, and Charles M. Nyachoti<sup>1</sup>, <sup>1</sup>University of Manitoba, Winnipeg, MB, Canada, <sup>2</sup>Zyme Fast System Inc., Winnipeg, MB, Canada.

This study was conducted to evaluate the effects of feeding diets supplemented with spay-dried whole egg containing anti-F4 antibodies (SDWE) against recombinantly produced F4 antigens to piglets challenged with enterotoxigenic *Escherichia coli* K88 (ETEC). Twenty-seven individually housed piglets [(Yorkshire × Landrace) × Duroc, 7.27 ± 0.47 kg initial BW] weaned at 21 ± 1 d were randomly allotted to 3 dietary treatments (n = 9) consisting of a wheat-soybean meal basal diet containing either 0 (control egg powder; CEP), 0.1% (SDWE1) or 0.4% (SDWE2) SDWE for a 14-d study. After a 7-d adaptation period, all pigs were weighed, blood samples collected and then orally challenged with 6 mL (2 × 10<sup>9</sup> cfu/mL) of freshly grown ETEC inoculum on d 8. Pigs had ad libitum access to feed and water throughout the study. Blood was sampled at 24 h and 48 h post-challenge to determine plasma urea nitrogen (PUN) content and diarrhea incidences and fecal consistency scores were recorded from d 9 to d 12. On d 14, all pigs were weighed and then killed to obtain intestinal tissue samples for villus height and crypt depth measurements. During the pre-challenge period, pigs fed the SDWE2 diet had higher ( $P < 0.05$ ) ADG and G:F compared with CEP but there were no differences among treatments in any of the performance response criteria during the post-challenge period. Incidences of diarrhea were similar among treatments although piglets fed SDWE-containing diets recovered from diarrhea within 48 h (with fecal consistency score of 0.0) of ETEC compared with CEP pigs. Also, fecal shedding of ETEC, PUN content and intestinal histomorphology were similar among treatments. The results show that SDWE at 0.4% supported greater piglet performance before challenge although there was no benefit of SDWE supplementation at either 0.1% or 0.4% evident during the post-challenge period.

**Key Words:** ETEC K88<sup>+</sup>, chicken egg anti-F4 antibodies, recombinant F4 antigens

**M511 Combined effects of chitosan and probiotic supplementation on performance and diarrhea incidences in enterotoxigenic *Escherichia coli* K88<sup>+</sup> challenged piglets.** Kolawole R. Aluko<sup>\*1</sup>, Deepak Ettungalpadi Velayudhan<sup>1</sup>, Aike Li<sup>2</sup>, Yulong Yin<sup>3</sup>, and Charles M. Nyachoti<sup>1</sup>, <sup>1</sup>University of Manitoba, Winnipeg, MB, Canada, <sup>2</sup>Academy of State Administration of Grain, Beijing, China, <sup>3</sup>Institute of Subtropical Agriculture, Chinese Academic of Sciences, Changsha, China.

The aim of this study was to investigate the combined effects of chitosan oligosaccharide (COS) and a microencapsulated *Enterococcus faecalis* probiotic (PRO) on growth performance and diarrhea incidences in enterotoxigenic *Escherichia coli* K88 (ETEC) challenged piglets in a 14-d study. Thirty piglets [(Yorkshire × Landrace) × Duroc], 7.19 ± 0.52 kg initial BW weaned at 21 ± 1 d were allotted to 5 treatment groups (n = 6) consisting of a corn-soybean meal diet with no additive (negative control, NC), the NC + 0.25% chlortetracycline (positive control, PC), NC + 400 mg/kg COS (COS), NC + 100 mg/kg PRO (PRO) and NC +

a combination of COS and PRO (CPRO). The basal diet was formulated to meet the NRC (2012) nutrient specifications for 5 to 10 kg BW pigs. Pigs were individually housed in cages, acclimated to treatments for a 7-d period and had ad libitum access to feed and water throughout the study. On d 8, pigs were weighed, blood samples were collected and then orally challenged with 6 mL ( $1 \times 10^{11}$  cfu/mL) of freshly grown ETEC inoculum. Post-challenge, blood was sampled at 24 h and 48 h to determine plasma urea nitrogen (PUN) and diarrhea incidences and fecal consistency scores were recorded from d 9 to d 12. On d 14, all pigs were weighed and then killed to obtain intestinal tissue samples for histomorphometric measurements. Growth performance responses were similar among treatments during the pre- and post- challenge periods. There were no significant differences in PUN content, incidences of diarrhea and fecal consistency scores among treatment. The intestinal histomorphology results did not differ significantly among treatments except for PC with increased ( $P = 0.0003$ ) villus-crypt ratio compared with the NC. Therefore, under the conditions of the present study, supplementing pig starter diet with 400 mg/kg COS or 100 mg/kg PRO or their combination had no effect against ETEC-induced diarrhea and did not improve growth performance in ETEC challenged piglets compared with control group.

**Key Words:** ETEC K88<sup>+</sup>, chitosan oligosaccharide, *Enterococcus faecalis* probiotic

#### **M512 Yeast cell wall supplementation in the diet of weaned piglets and its effect on performance and diarrhea incidence.**

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The objective of this study was to evaluate effects of yeast cell wall (YCW) supplementation in weaned piglet's diet on performance and diarrhea incidence. Trial 1 was conducted on a swine farm which recently recovered from hog cholera disease, and 120 piglets, 28-d old (avg. 8.37 kg) were distributed in a randomized complete block design (RCBD), with 2 treatments (0 and 2 kg/MT of YCW from *Saccharomyces cerevisiae*), being 8 reps for control group and 7 for YCW group, of 8 piglets in each. The 2nd trial was conducted on a swine farm where the low disease profile was relatively stable for the past 2 years, and 100 piglets, 28-d old (avg. 7.53 kg) were distributed in RCBD, with 2 treatments (same as 1st trial) each having 5 reps with 10 piglets in each. The periods were 20 and 21d, respectively, for 1st and 2nd trials, divided into booster (28–30d) and pre-starter diets (31–47d 1st trial, and 31–48d 2nd trial). The parameters measured in both trials were: body weight (BW, kg), body weight gain (BWG, kg), average daily feed intake (ADFI, kg), feed:gain (F:G) and mortality (M, %). Diarrhea incidence (DI) was observed in 1st trial, and given a corresponding score from zero to 3 in order of increasing severity. The data were analyzed using the GLM (SAS), and means were compared by Kruskal-Wallis test for non-parabolic data. The YCW supplementation in the diet of weaned piglets resulted in no differences ( $P > 0.05$ ) in performance parameters analyzed for both trials. However, the groups supplemented with YCW, numerically had improved BWG (+6.3; +6.4%) and F:G (+7%; 13.2%), respectively, for both trials. There were no differences ( $P > 0.05$ ) between treatments for DI score. For control group, 90.36% of the population has 0 score at the beginning, which suggests a normal type of feces, and at the final week of the experiment, it minimally increased to 93.05%. For YCW group, there was an increase in the number of animals having a 0 score, from 82.58% to 93.79%, at the last week of the experiment. Supplementation of YCW at 2 kg/MT to diets

of weaned piglets had numerical, but no significant, positive benefits when considering BWG, F:G, and DI, compared with control treatment.

**Key Words:** body weight, *Saccharomyces cerevisiae*

#### **M513 Yeast cell wall supplementation in the diet of weaned piglets and its effect on gut health.**

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The objective of this small-scale study was to evaluate effects of yeast cell wall (YCW) supplementation in weaned piglet's diet on gut health and integrity. The trial was conducted with 12 piglets, 28-d old ( $8.5 \pm 1.5$  kg), distributed in a complete randomized design, with 2 treatments (0 and 2kg/MT of YCW from *Saccharomyces cerevisiae*), 6 reps, 1 piglet in each. On d 1 of trial, *E. coli* was inoculated at  $9.8 \times 10^8$  cfu/mL per pig. The experimental period was 14 d, where on d 1, 1 animal of control group (CG) was killed for baseline values; on d 7, 3 piglets from each treatment were killed; and on d 14, the remaining piglets were killed. Intestinal samples were collected from jejunum on d 1, 7, and 14 and measured under the light microscope for villus height (VH,  $\mu\text{m}$ ), villus width (VW,  $\mu\text{m}$ ), mucosal thickness (MT,  $\mu\text{m}$ ), crypt depth (CD,  $\mu\text{m}$ ) and villous surface area (VSA,  $\mu\text{m}^2$ ). Also scanning electron microscopy (SEM) was performed. The jejunal samples were also used to determine *E. coli* (JEC) and total coliform counts (JCC). On d 4, 7, 11, and 14 of the trial, fecal samples of all pigs were also collected for *E. coli* (FEC) and total coliform count (FCC). The data were analyzed using the GLM (SAS), and means were compared by *t*-test ( $P = 0.05$ ). Overall (d 7 and 14), the YCW-treated piglets differed ( $P < 0.05$ ) from baseline piglet for VH, VW and CD. The CG had better results ( $P < 0.05$ ) for VH, VW, CD and VSA at 7th d post-infection. However, at 14 d post-infection, YCW supplementation had improved ( $P < 0.05$ ) VH, CD and VSA. Although there were no statistical differences ( $P > 0.05$ ) between treatments for MT at d 14, it was numerically improved (+16.2 $\mu\text{m}$ ) for piglets supplemented with YCW. For SEM, YCW group appeared to have less erosion in villi apices and better microvilli surface, when compared with CG at d 7 and 14. The JEC, JCC, FEC and FCC average of YCW group and CG showed a significant increase ( $P < 0.05$ ) among the days post-infection, compared with the baseline values; however, the averages of YCW group were significantly lower ( $P < 0.05$ ) than the CG. These results demonstrated that YCW supplementation at 2 kg/MT to weaned piglets decreased jejunal and fecal *E. coli* counts and improved intestinal integrity.

**Key Words:** *Saccharomyces cerevisiae*

#### **M514 Effect of chronic heat stress on mRNA expression of heat shock protein 70, uncoupling protein 3, and cytochrome p450 in pigs.**

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Exposure of pigs to sudden increases in ambient temperature (AT), causing heat stress (HS) provokes cellular and molecular changes to cope with HS and maintain homeostasis. Abundance of mRNA coding for heat shock protein 70 kDa (HSP70) involved in the folding of proteins during heat stress, uncoupling protein 3 (UCP3) involved in heat generating in mitochondria, and cytochrome p450 (Cp450) involved in redox equilibrium, is affected in animals under acute HS. But it is

not clear whether those alterations are maintained in pigs with chronic exposure to HS. An experiment was conducted with 18 pigs (30–35 kg BW) to analyze the effect of chronic HS on mRNA expression of HSP70, UCP3, and Cp450. There were 3 treatments: 1) pigs housed inside an AT controlled room ( $24 \pm 2^\circ\text{C}$ ) and fed ad libitum (TNL), 2) as in 1 but feed restricted (TNR), 3) pigs housed under natural HS conditions (27.1 to  $39.6^\circ\text{C}$ , average  $33.3^\circ\text{C}$ ). Feed intake in HS pigs was restricted to 95% of ad libitum intake, and that of TNR pigs was similar to HS pigs. Body temperature (BT) was recorded every 15 min with a device implanted under the skin of 4 pigs per treatment. At the end of the 21-d study, all pigs were killed and samples of liver, and longissimus (LM) and semitendinosus (SM) muscles were collected to analyze the mRNA expression of HSP70, UCP3 and Cp450. The BT of TNL, TNR, and HS pigs was  $38.4$ ,  $37.7$  and  $39.0^\circ\text{C}$ , respectively. It was higher in HS pigs compared with TNL and TNR ( $P < 0.05$ ). Expression values ( $\times 10^{-4}$ ) for HSP70 in TNL, TNR, and HS pigs were liver, 2.03, 4.08, 7.66; LM, 2.47, 5.2, 4.69; SM, 2.10, 2.31, 2.23, respectively; for UCP3 were LD, 4.58, 4.92, 3.57; SM, 3.46, 2.42, 6.04. Expression values ( $\times 10^{-3}$ ) for Cp450 in liver were 3.59, 1.77, 1.68. Chronic HS did not affect the expression of HSP, UCP3 or Cp450 in pigs. Although BT of pigs exposed to chronic HS remains higher than in TN pigs, the lack of effect on gene expression may suggest that pigs became acclimated at or before 21 d of natural exposure to HS.

**Key Words:** pig, heat stress, gene expression

**M515 Influence of CLA supplementation on body composition of finished pigs.** Kyle J. Stutts\*, Ginger G. Vann, Mark J. Anderson, Jessica L. Leatherwood, Marcy M. Beverly, and Stanley F. Kelley, Sam Houston State University, Huntsville, TX.

Twenty-four pigs (Large White  $\times$  Landrace; 128.2 to 134.1 kg) of similar breeding were utilized in a randomized complete block design to evaluate the effects of dietary supplementation of conjugated linoleic acid on body composition of mature, finished pigs. Pigs were blocked by BW and sex and were randomly assigned and evenly distributed between a treatment ( $n = 12$ ; TRT) or control group ( $n = 12$ ; CON) for an 84-d feeding trial. Diets were formulated to be isocaloric and isonitrogenous and consisted of a CON diet of a commercially available concentrate (Producers Cooperative, Bryan, TX) with an additional 2% of the total diet of soybean oil, or the TRT diet that consisted of the same pelleted concentrate with an additional 2% of the total diet of oil containing CLA. The CLA source (BASF Corp., Florham Park, NJ) being utilized contained 55% CLA (mixture of *cis*-9, *trans*-11; *trans*-10, *cis*-12; and *trans*-9, *trans*-11 isomers) yielding a CLA supplementation of 1.1% of the total diet. Oil was top-dressed and mixed into feed immediately before feeding. Pigs were fed at 12-h intervals at 3.5% BW per day in concentrate. Body weight was obtained every 7 d with the intake of concentrate adjusted accordingly. Fat thickness (cm) was measured at 14-d intervals via ultrasonography at the 10th and last ribs. Longissimus muscle area (LMA) was collected via ultrasonography at d 0 and 84. Data were analyzed using the mixed procedure of SAS. Overall effects were analyzed using repeated measures and data from individual days were analyzed using fat thickness from d 0 as a covariate. CLA supplementation had no effect on body composition. Overall, there was no difference between treatments in BW ( $P = 0.98$ ), 10th rib fat thickness ( $P = 0.07$ ), last rib fat thickness ( $P = 0.17$ ), or LMA ( $P = 0.96$ ). Additionally, there were no treatment  $\times$  time interactions for the variables measured. These data indicate that CLA supplemented at 1.1% of the diet when mature pigs are fed at 3.5% BW has no effect on body composition or BW.

Further studies are needed to fully elucidate dietary CLA supplementation to alter body composition when fed to mature pigs.

**Key Words:** CLA, swine, fat thickness

**M516 Effect of acute water and feed deprivation at weaning and subsequent heat stress on serum stress markers and ileal mucosa gene expression in nursery pigs.** Nathan L. Horn\*<sup>1</sup>, Guy Miller<sup>2</sup>, Frank Ruch<sup>3</sup>, Carrie R. Little<sup>3</sup>, Kolapo M. Ajuwon<sup>1</sup>, and Olayiwola Adeola<sup>1</sup>, <sup>1</sup>Purdue University, West Lafayette, IN, <sup>2</sup>Biomatric, Princeton, MN, <sup>3</sup>JBS United, Sheridan, IN.

The current experiment was conducted to investigate the effect of a feed and water deprivation event at weaning and subsequent heat stress event on serum stress markers and expression of selected genes in the ileum. Mixed-sex pigs were allotted on the basis of IBW ( $6.97 \text{ kg} \pm 0.89$ ) in a RCBD with treatments in a split-plot arrangement and consisting of the whole-plot factor of with or without a 24-h feed + water deprivation event at weaning (deprivation event) and the sub-plot factor of with or without a cyclic 3-d heat stress event starting 27 d post-weaning. On 1, 27, and 30 d post-weaning one pig from each pen was selected for blood and ileal mucosa collection to determine serum cortisol, corticotrophin releasing factor (CRF), and endotoxins; and mucosal gene expression measurement of tumor necrosis factor  $\alpha$  (TNF- $\alpha$ ), interleukin 6 (IL-6), interleukin 8 (IL-8), glucose transporter 2 (GLUT2), claudin 1 (CL-1), occludin (OC), and zonula occludens 2 (ZO-2) by RT-PCR. There was an increase ( $P < 0.05$ ) in serum CRF and endotoxins and a tendency for an increase ( $P = 0.09$ ) in serum cortisol due to the deprivation event 1 d post-weaning. Furthermore, serum endotoxins and CRF tended to increase ( $P < 0.10$ ) due to the deprivation event on 27 and 30 d post-weaning, respectively. Gene expression of GLUT 2 tended to decrease ( $P = 0.07$ ), CL-1 tended to increase ( $P = 0.10$ ), and OC decreased ( $P = 0.05$ ) due to the deprivation event 1 d post-weaning. Expression of IL-8 and OC genes decreased ( $P < 0.05$ ) due to the deprivation event 27 d post-weaning and OC and ZO-2 gene expression tended to decrease ( $P = 0.07$ ) due to the heat stress event 30 d post-weaning. Results from the current experiment show that a post-weaning feed and water deprivation event impacts serum stress markers and mucosal gene expression throughout the nursery period and a subsequent heat stress event affects tight junction gene expression although an interaction of deprivation event by heat stress was not observed.

**Key Words:** feed and water deprivation, heat stress, nursery pig

**M517 The growth of Tamworth  $\times$  Berkshire pigs farrowed outdoors and reared in a hoop structure.** H.-S. Park<sup>1</sup>, N. Whitley<sup>2</sup>, and S.-H. Oh\*<sup>1</sup>, <sup>1</sup>Department of Animal Sciences, North Carolina A&T State University, Greensboro, NC, <sup>2</sup>Cooperative Extension Program, North Carolina A&T State University, Greensboro, NC.

The objective was to investigate the growth of Tamworth- or Berkshire-sired pigs from Berkshire sows at the North Carolina Agricultural and Technical State University Farm. The location features a humid subtropical climate with subtropical summer temperatures and mild winters and an average annual precipitation of approximately 110 cm. Forty-four purebred Berkshire females were used to produce crossbred and purebred Berkshire pigs. Females were given altrenogest (Matrix) for estrus synchronization for 14 consecutive days at 7 ml for gilts and 9 ml for sows. Thirty-three females were detected in heat and artificially inseminated, which resulted in 10 farrowed sows. Each sow farrowed in a 15-m<sup>2</sup> paddock in June 2014. Total number born were 67 in Tamworth  $\times$  Berkshire (TB) and 51 in Berkshire  $\times$  Berkshire (BB), respectively.



Number born alive were 53 in TB, and 35 in BB. Pigs were weaned at 4 weeks of age. Body weights from 43 weaned Tamworth × Berkshire (TB) crossbred pigs were recorded from 6 sows, and weights from 28 weaned Berkshire × Berkshire (BB) purebreds were recorded from 4 sows as a control group. Birth weights (BW) were recorded at 3 d of age, and then body weights were recorded every 4 weeks from their birthdate to 20 weeks of age; weaning weight (WW), 8 weeks of age (W8), 12 weeks of age (W12), 16 weeks of age (W16), and 20 weeks of age (W20). Data were analyzed with repeated measures ANOVA using SAS PROC GLM. Least squares means of weights in TB were 1.90 ± 0.06kg (BW), 8.11 ± 0.29kg (WW), 19.11 ± 0.58kg (W8), 34.80 ± 1.10kg (W12), 54.49 ± 1.51kg (W16), and 79.16 ± 1.96kg (W20), respectively. Least squares means of weights in BB were 1.80 ± 0.08kg (BW), 7.78 ± 0.36kg (WW), 16.99 ± 0.74kg (W8), 31.58 ± 1.39kg (W12), 52.04 ± 1.91kg (W16), and 79.78 ± 2.48kg (W20), respectively. Body weights were significantly greater for TB compared with BB only for W8 and W12 ( $P < 0.05$ ). More research is needed regarding breed types used in alternative production systems.

**Key Words:** Tamworth, Berkshire, growth

**M518 Effect of postcervical artificial insemination with semen supplemented with oxytocin in sows in warm weather.** Juan M.

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To determine the effect of applying semen doses spiked with oxytocin by post-cervical insemination, on the reproductive performance of sows

serviced during the months of June to October (max. registered ambient temperature of 46°C and 28 min) in northwest Mexico, 223 multi parturient sows were inseminated. Sows were assigned randomly to be serviced twice with semen from same boar(s) in 1 of 2 treatments: (1) serviced with a reduced dosage equivalent to  $1.5 \times 10,000,000,000$  viable spermatozoa cell diluted in 40 mL of semen dose (CONT, n = 111) and (2) similar to CONT plus addition of 4 IU oxytocin to semen at service time (OXY, n = 112). Sows were serviced from June to October, using an intrauterine semen delivery device. Total number born and total number born alive were counted at farrowing. Results were compared using  $X^2$  analyses. Treatment had no effect on litter size with 11.0 vs. 10.8 ( $P = 0.577$ ), or on number born alive with 9.8 vs. 9.6 for CONT and OXY, respectively. OXY enhances ( $P = 0.03$ ) 9.6% the farrowing rate, with means values of 84.68 vs. 93.75 for CONT and OXY, respectively. These results suggest that the addition of oxytocin to semen at serviced time improves farrowing of multi parturient sows serviced during summer-autumn in northwest Mexico, without effects on other reproductive performance variables.

**Key Words:** sow, postcervical insemination

# Animal Health: Reproductive health and acute immune responses

**20 Efficacy of nonsteroidal antiinflammatory drugs for the treatment of acute puerperal metritis in dairy cows.** Alina Pohl and Wolfgang Heuwieser\*, *Clinic of Animal Reproduction, Freie Universität Berlin, Berlin, Germany.*

Acute puerperal metritis (APM) in dairy cows is often treated with antibiotics. An increasing antibiotic resistance is well documented and associated with decreasing clinical efficacy, animal welfare and economic consequences. Hence, there is a significant need to encourage prudent use of antibiotics and alternative therapies to antibiotics. The objective of this study was to compare the efficacies of ketoprofen and ceftiofur for the treatment of APM. Between June 2013 and February 2015, a total of 610 dairy cows from 6 farms in Germany were enrolled. Inclusion criteria was a rectal temperature (RT)  $\geq 39.5^{\circ}\text{C}$  and a reddish-brown fetid vaginal discharge within the first 10 DIM. Cows meeting the inclusion criteria were randomly allocated to treatment with ketoprofen (3 mg/kg of BW,  $n = 300$ ) or treatment with ceftiofur (1 mg/kg of BW,  $n = 310$ ), both on 3 consecutive days. Rectal temperature was recorded daily for a period of 7 d after enrollment. Cows that showed RT  $\geq 39.5^{\circ}\text{C}$  on d 4 to 7 after inclusion received an extended treatment with ceftiofur for 3 (ketoprofen group) or 2 (ceftiofur group) more days. Between 21 and 34 DIM, cows were examined with the Metrichheck device and vaginal discharge was categorized on a 5-point scale according to the presence of pus. A total of 51 cows (34 from ketoprofen group, 17 from ceftiofur group) were excluded from analysis due to concurrent disease ( $n = 15$ ), additional medication of APM ( $n = 11$ ), and due to missing protocol compliance ( $n = 25$ ). Cows of the ketoprofen group (54%) received an extended treatment more often than cows of the ceftiofur group (30%;  $P < 0.01$ ). Occurrence of purulent vaginal discharge was similar for both treatment groups (ketoprofen: 57.8%, ceftiofur: 55.3%,  $P = 0.62$ ). More than half of the cows initially treated with ketoprofen needed ceftiofur treatment. However, there is potential of reducing antibiotic use by utilizing ketoprofen for the treatment of APM as up 46% of cows treated with ketoprofen did not need extended treatment. Animal welfare and economic aspect need to be considered.

**Key Words:** acute metritis, treatment, antibiotic

**21 Intrauterine cephalosporin infusion is associated with better reproduction performance in cows with purulent vaginal discharge and cytological endometritis.** José Denis-Robichaud\*<sup>1</sup> and Jocelyn Dubuc<sup>2</sup>, <sup>1</sup>*Department of Population Medicine, University of Guelph, Guelph, Ontario, Canada,* <sup>2</sup>*Faculté de Médecine Vétérinaire, Université de Montréal, Saint-Hyacinthe, Québec, Canada.*

The objectives of this study were to quantify the effect of an intrauterine infusion of cephalosporin on the reproductive performance at first service of postpartum dairy cows affected by purulent vaginal discharge (PVD) or cytological endometritis (ENDO) using different diagnostic strategies and to determine if the presence of prolonged anovulation would influence the magnitude of treatment benefit. A total of 2,259 Holstein cows in 28 herds were enrolled in a randomized clinical trial. At 35 ( $\pm 7$ ) DIM, cows were diagnosed for PVD (purulent vaginal discharge or worse using the metrichheck device) and ENDO ( $\geq 6\%$  polymorphonuclear cells using the cytobrush technique or at least small amounts of leukocytes using the leukocyte esterase colorimetric test). Regardless of reproductive tract disease status, cows were randomly assigned to receive an intrauterine cephalosporin infusion or no treatment. Serum progesterone

was measured at 35 and 49 ( $\pm 7$ ) DIM (14 d apart); cows were considered to have prolonged anovulation if progesterone was  $< 1$  ng/mL at both times. Statistical analyses were conducted using multivariable mixed logistic regression models adjusted for confounders and herd clustering effect. Intrauterine cephalosporin treatment was associated with an increased first service pregnancy risk in cows diagnosed with PVD (no treatment: 15.4%; treatment: 31.4%;  $P < 0.05$ ) and ENDO (cytobrush: no treatment: 16.2%, treatment: 24.4%,  $P < 0.05$ ; leukocyte esterase: no treatment: 15.8%; treatment: 25.1%,  $P < 0.05$ ), but not in cows unaffected by any form of reproductive tract disease (no treatment: 34.8%; treatment: 32.6%;  $P = 0.5$ ). The effect of cephalosporin treatment in anovular cows (no treatment: 21.0%; treatment: 26.4%;  $P = 0.26$ ) was numerically lower than in cyclic cows (no treatment: 22.7%; treatment: 34.1%;  $P < 0.05$ ). Overall, an intrauterine infusion of cephalosporin improved first service pregnancy risk in cows with postpartum reproductive tract disease and this effect was influenced by postpartum anovulation status.

**Key Words:** dairy cows, endometritis, treatment

**22 Potential role of lactic acid bacteria in the regulation of *Escherichia coli* infection and inflammation of the bovine endometrium.** Sandra Genís\*<sup>1</sup>, Àlex Bach<sup>1,2</sup>, Francesc Fàbregas<sup>1</sup>, Marta Terré<sup>1</sup>, and Anna Arís<sup>1</sup>, <sup>1</sup>*Department of Ruminant Production, Institut de Recerca i Tecnologia Agroalimentàries (IRTA), Torre Marimon, Caldes de Montbui, Barcelona, Spain,* <sup>2</sup>*Institució Catalana de Recerca i Estudis Avançats (ICREA), Barcelona, Spain.*

About 40% of dairy cattle develop uterine disease at postpartum, causing infertility. Some studies indicate that uterine infection, predominantly by *Escherichia coli* in the first week postpartum, is associated with metritis. Metritis is an inflammation of the uterus in which the cow fails to completely clear bacterial contaminants and reducing postpartum inflammatory processes. The aim of this study was to evaluate the potential role of 4 lactic acid bacteria (LAB; *Lactobacillus rhamnosus*, *Pediococcus acidilactici*, *Lactobacillus reuteri*, and *Lactobacillus sakei*) in the modulation of *Escherichia coli* infection and its respective inflammation response by endometrial cells. Primary endometrial epithelial cells were isolated from a fresh endometrium of a healthy cow and cultured at  $8 \times 10^4$  cells/well in 24-well plates to evaluate the effects of LAB at 3 different doses. Cell extracts were obtained with TriZol. Pro-inflammatory status was assessed by qPCR quantification of IL8, IL1 $\beta$ , TNF $\alpha$ , and IL6 gene expression. Internalization of *Escherichia coli* was determined by direct enumeration on MacConkey agar plates. Data were normalized and analyzed by an ANOVA. *Lactobacillus sakei* and *Lactobacillus reuteri* had a positive effect preventing *E. coli* infection (87% and 78% respectively,  $P < 0.001$ ) but were associated with a dose-variable effect on tissular inflammation that could further exacerbate the pro-inflammatory status of the endometrium. *Pediococcus acidilactici* clearly decreased ( $P < 0.001$ ) *E. coli* internalization up to an 83% reduction and slightly reduced the inflammation (up to 3.2-fold in IL1 $\beta$  expression,  $P < 0.001$ ). However, the pro-inflammatory cytokines IL8 and IL1 $\beta$  decreased significantly ( $P < 0.001$ ) up to 85.1 and 5.2 folds, respectively, in the presence of *L. rhamnosus*. In conclusion, these results demonstrate a clear beneficial effect of *P. acidilactici* and *L. rhamnosus* in the modulation of endometrial infection and inflammation in cattle.

**Key Words:** *Escherichia coli*, LAB bacteria, metritis

**23 Uterine microbiota from calving until establishment of metritis in dairy cows.** Soo Jin Jeon<sup>\*2</sup>, Achilles Vieira-Neto<sup>1</sup>, Mohanathas Gobikrushanth<sup>2,6</sup>, Rodolfo Daetz<sup>2</sup>, Rodolfo Mingoti<sup>1</sup>, Ana Carolina Parize<sup>2</sup>, Sabrina Freitas<sup>2</sup>, Antonio Nelson da Costa<sup>5</sup>, Rodrigo Bicalho<sup>3</sup>, Svetlana Lima<sup>3</sup>, Kwang C. Jeong<sup>1,4</sup>, and Klíbs N. Galvão<sup>2</sup>, <sup>1</sup>Department of Animal Sciences, University of Florida, Gainesville, FL, <sup>2</sup>Department of Large Animal Clinical Sciences, University of Florida, Gainesville, FL, <sup>3</sup>Department of Population Medicine and Diagnostic Sciences, Cornell University, Ithaca, NY, <sup>4</sup>Emerging Pathogens Institute, University of Florida, Gainesville, FL, <sup>5</sup>Departamento de Zootecnia, Universidade Federal do Ceará, Fortaleza, Ceara, Brazil, <sup>6</sup>Department of Agricultural, Food and Nutritional Science, University of Alberta, Alberta, Canada.

Objective was to characterize the progression of uterine microbiota from calving until establishment of metritis. Uterine swabs were collected at 0 (20 min from calving), 2, 4, 6, and 8 d postpartum (DPP) from 92 cows. Twelve cows were diagnosed with metritis at 4, 6, or 8 DPP (6 ± 2 DPP), and 12 healthy cows were selected for comparison. Swabs (n = 72) collected at 0, 2, and 6 ± 2 DPP were used for metagenomic sequencing of 16S rRNA gene on the Illumina MiSeq platform. A heat map showed that uterine microbiota was established in cows shortly after calving. The microbiota changed rapidly from 0 to 6 ± 2 DPP by decreasing the abundance of Proteobacteria and increasing the abundance of Bacteroidetes and Fusobacteria. At 6 ± 2 DPP, abundance of Bacteroidetes was significantly higher in metritic cows than healthy cows ( $P < 0.01$ ). Although most genera were shared, healthy and metritic cows could be discriminated based on relative abundance at 0, 2, and 6 ± 2 DPP using discriminant analysis ( $P < 0.01$ ). Also, discriminant analysis showed that *Bacteroides*, *Filifactor*, *Porphyromonas*, *Fusobacterium*, and *Arcanobacterium/Trueperella* were important predictors of metritis. Furthermore, *Bacteroides* and *Fusobacterium* were significantly correlated with uterine discharge score ( $r_s = 0.51$  and  $r_s = 0.49$ , respectively;  $P = 0.02$ ). Likewise, at species level, *Bacteroides heparinolyticus* and *Fusobacterium necrophorum* were the main bacteria for the development of metritis because they were both prevalent (16.8% and 20.2% in metritic cows; 7.0% and 15.8% in healthy cows) and correlated with uterine discharge score ( $r_s = 0.42$  and  $r_s = 0.42$ , respectively;  $P \leq 0.05$ ). In addition, there was a second (*Fusobacterium gonidiaformans*, *Helicoccus ovis*, and *Filifactor villosus*) and third (*Bacteroides pyogenes*, *Porphyromonas levii* and others) line of bacteria that acted synergistically with the main bacteria causing metritis.

**Key Words:** dairy cow, metritis, 16S metagenomics

**25 Association among health and fertility and survival of high-producing dairy cows in three geographic regions of Chile.** Pablo Pinedo<sup>\*1,2</sup>, Pedro Melendez<sup>3</sup>, Sushil Paudyal<sup>4</sup>, Felipe Arias<sup>5</sup>, Ricardo Krauss<sup>5</sup>, Hernando Lopez<sup>6</sup>, Alejandro Luco<sup>5</sup>, and Cristian Vegara<sup>5,6</sup>, <sup>1</sup>Texas A&M AgriLife Research, Amarillo, TX, <sup>2</sup>Department of Veterinary Pathobiology, College of Veterinary Medicine & Biomedical Sciences, Texas A&M University System, College Station, TX, <sup>3</sup>Department of Veterinary Medicine and Surgery, College of Veterinary Medicine, University of Missouri-Columbia, Columbia, MO, <sup>4</sup>West Texas A&M University, Canyon, TX, <sup>5</sup>ABS Chile Ltda., Santiago, Chile, <sup>6</sup>ABS Global Inc., DeForest, WI.

The objective was to analyze the association between disease occurrence during early lactation and reproductive performance and survival of dairy cows in high-producing herds, under different management practices in 3 geographic regions of Chile. Data included 30,157 lactation records of

cows calving between January, 2013 and June 2014, maintained under different production systems: Dry lot (DL; n = 3,881 cows in 6 herds); freestall (FS; n = 21,421 cows in 13 herds); grazing (G; n = 1,211 cows in 3 herds); and freestall/grazing (FG; n = 4,244 cows in 5 herds). Cows were also classified according to geographic location: central area (C, n = 6,375 cows); south-central area (SC, n = 17,073 cows); and south area (S, n = 7,309 cows). Logistic regression and ANOVA were used for the analyses (PROC GLIMMIX and PROC GLM, SAS), considering cow as the experimental unit. Covariables offered to the models included lactation number, cow, and herd milk yield. Average milk yield (305 ME) per cow was 11,793, 12,632, and 6,876 kg for C, CS, and S regions, respectively and 10,603, 12,270, 6,899, and 9,261 kg for DL, FS, G, and FG systems. The odds (95% CI) of pregnancy at 150 DIM (P150) and the odds of survival until 150 DIM (S150) for cows that had at least 1 disease event within 50 DIM were 0.82 (0.77–0.88) times the odds of pregnancy and 0.21 (0.17–0.26) times the odds of survival for healthy cows. The odds (95% CI) of P150 for cows under grazing systems were 1.48 (1.24–1.77), 1.07 (0.9–1.28), and 1.29 (1.09–1.52) times the odds of P150 for cows under DL, FG, and F systems, respectively. The odds (95% CI) of S150 for cows under grazing were 5.15 (3.0–8.8), 3.18 (1.88–5.40), and 2.0 (1.19–3.33) times the odds of S150 for cows under DL, FG, and F systems. The odds (95% CI) of P150 and S150 for cows located in the S area were 1.03 (0.95–1.11), and 1.11 (1.03–1.19) times the odds of P150 for cows in C, and SC areas and 1.42 (1.23–1.66), and 1.37 (1.20–1.60) times the odds of S150 for cows in C, and SC areas. Data suggested that cow's health status, geographic location, and production system had a significant association with reproductive performance and survival in this population of Chilean high-producing cows.

**Key Words:** dairy, health, fertility

**26 Bovine viral diarrhea diagnostic testing results in the Intermountain West—Comparison between test methods, age, sex and beef versus dairy breeds.** David J. Wilson<sup>\*</sup>, Thomas J. Baldwin, E. Jane Kelly, Arnaud VanWettere, and Gordon Hullinger, *Utah State University, Logan, UT.*

Bovine viral diarrhea (BVD) is an important cause of respiratory, gastrointestinal, and reproductive disease in cattle. The objectives were to calculate prevalence of BVD (“detected” test results) among all bovine samples tested for BVD at the Utah Veterinary Diagnostic Laboratory from 2009 to 2013, and to compare results by sex, age, dairy vs. beef breeds (if provided) of the cattle, and BVD test methods. For necropsied animals, most were tested by Ag capture ELISA, but fetuses were usually tested by PCR. Cattle were mainly from Utah, but also from surrounding states in the Intermountain West of the United States. This was not a planned experiment, but Chi-squared was used to test for significant differences in BVD prevalence between age, sex, breed and test methods. BVD was detected in 105/8,975 samples (1.2%), including 22/180 necropsies (12.2%). Test methods and results are given in Table 1. There were no significant differences in BVD detection by age or sex. Dairy breeds ( $P = 0.07$ ), all necropsied animals, and those tested with PCR were significantly more likely to test as “detected” with BVD. The overall prevalence of >1% and the 8% prevalence in aborted fetuses demonstrates that despite the low reported prevalence of persistently infected cattle, BVD remains an important bovine disease.



**Table 1 (Abstr. 26).** Results by BVD test method, by age, sex, or breed, and by dairy versus beef breeds

Item	Tested, no.	BVD detected, no. (%)
BVD test method		
Ag Capture ELISA	7,692	79 (1.0)
Serum ELISA	1,195	19 (1.6)
PCR	88	7 (8.0)*
Necropsy (all methods) <sup>1</sup>	180	22 (12.2)*
Age, sex or breed		
Male	215	5 (2.3) <sup>2</sup>
Female	382	9 (2.4) <sup>2</sup>
Fetus	36	3 (8.3) <sup>2,3</sup>
Calf	579	29 (5.0) <sup>2,4</sup>
Immature	183	4 (2.2) <sup>2,5</sup>
Adult	75	4 (5.3) <sup>2,6</sup>
Dairy <sup>7</sup>	754	25 (3.3) <sup>†</sup>
Beef <sup>8</sup>	1,600	26 (1.6) <sup>†</sup>

<sup>1</sup>Subset of above 3 categories.

<sup>2</sup>NSD among sex, age, or by test method within sex, age or breed.

<sup>3</sup>All 3 by PCR at necropsy.

<sup>4</sup>Twenty-seven by Ag Capture ELISA, including 16 of 114 necropsies (14.0%).

<sup>5</sup>Three by serum ELISA, 1 by Ag Capture ELISA, of 6 necropsies (16.7%).

<sup>6</sup>Two by serum ELISA, 2 by Ag Capture ELISA, of 24 necropsies (8.3%).

<sup>7</sup>95.0% Holsteins, 3.8% Jerseys.

<sup>8</sup>60.0% Angus, 22.8% "Beef."

\*Higher prevalence of "BVD detected" results,  $P < 0.0001$ .

<sup>†</sup> $P = 0.07$ .

**Key Words:** BVD, bovine, prevalence

**27 Prewaning plane of nutrition and *Mannheimia haemolytica* dose influence inflammatory responses to a combined bovine herpesvirus-1 and *Mannheimia haemolytica* challenge in postweaned Holstein calves.** K. P. Sharon<sup>\*1,2</sup>, Y. L. Liang<sup>1</sup>, N. C. Burdick Sanchez<sup>2</sup>, J. A. Carroll<sup>2</sup>, P. R. Broadway<sup>2</sup>, and M. A. Ballou<sup>1</sup>, <sup>1</sup>Department of Animal and Food Sciences, Texas Tech University, Lubbock, TX, <sup>2</sup>USDA-ARS, Livestock Issues Research Unit, Lubbock, TX.

To determine whether previous plane of milk replacer nutrition (PON) and *M. haemolytica* (MH) dose influences inflammatory responses to a combined viral-bacterial respiratory challenge, Holstein calves (1d of age;  $n = 30$ ) were assigned to treatments in a  $2 \times 3$  factorial with preweaned PON and dose of MH as main effects ( $n = 5$ /treatment). Calves were fed either a low (LPN;  $n = 15$ ) or a high PON (HPN;  $n = 15$ ) from birth through weaning. Calves fed the LPN were fed 445 g DM/d of milk replacer until weaning, and HPN calves were fed 830g DM/d of milk replacer from 1 to 10d and 1080g from 11d until weaning. Calf starter and water were offered ad libitum. Calves were step-down weaned beginning at 54d and moved into an enclosed barn at 70d. Indwelling rectal temperature (RT) devices and jugular catheters were inserted at 80d. Calves were challenged with  $1.5 \times 10^8$  PFU/mL/nostril of bovine herpesvirus-1 (BHV-1) at 81d and with  $10^6$ ,  $10^7$ , or  $10^8$  cfu of MH at 84d. Blood samples were collected at varying intervals respective to BHV-1 and MH challenges. There was a PON  $\times$  MH interaction ( $P < 0.01$ ) on RT, where calves receiving  $10^8$  MH had the greatest increase among HPN calves, but the least among LPN calves. Haptoglobin (Hp) concentrations were greater ( $P = 0.04$ ) in LPN vs HPN calves after BHV-1 challenge. There was a time  $\times$  MH interaction ( $P < 0.01$ ) for Hp concentrations after the MH challenge, where calves receiving  $10^7$  MH had the greatest Hp concentrations at 24 h, and calves receiving  $10^8$  MH the greatest at 144h. During the BHV-1 challenge, neutrophil concentrations (PMN) and change in total leukocytes were greater ( $P <$

0.01) among LPN vs HPN calves. After the MH challenge PMN were greatest ( $P \leq 0.05$ ) in calves receiving  $10^8$  MH at 5 and 24h. During the MH challenge calves receiving  $10^8$  MH had the greatest ( $P < 0.05$ ) IL-6 and TNF $\alpha$  concentrations. These data demonstrate that LPN calves responded more severely to a combined viral-bacterial respiratory challenge and greater doses of MH increase the acute inflammatory response and prolong inflammation.

**Key Words:** health, nutrition, respiratory

**28 Plasma leptin concentrations are increased during a vaccine-induced acute-phase response in beef cattle.** Rodrigo Marques<sup>\*1</sup>, Reinaldo Cooke<sup>1</sup>, Murilo Rodrigues<sup>1</sup>, Bruno Cappellozza<sup>1</sup>, Sergio Arispe<sup>2</sup>, and David Bohnert<sup>1</sup>, <sup>1</sup>EOARC, Oregon State University, Burns, OR, <sup>2</sup>Malheur County Extension, Oregon State University, Ontario, OR.

The objective of this experiment was to evaluate if a vaccine-induced acute-phase reaction also results in increased plasma leptin concentration, which would explain a potential DMI decrease in vaccinated beef cattle. Eighteen yearling Angus  $\times$  Hereford heifers were ranked by BW and allocated to 2 groups, which were randomly assigned to a crossover design containing 2 periods of 7 d and the following treatments on d 0 of each period: (1) vaccination against *Mannheimia haemolytica*, bovine rhinotracheitis virus, and bovine viral diarrhea Types 1 and 2 viruses (VAC; BoviShield One Shot; Zoetis, Florham Park, NJ), or (2) saline-injected control (CON). Heifers were maintained in individual pens, offered grass hay for ad libitum consumption, in addition to 3.5 kg/d (DM basis) of a corn-based supplement. During Period 1, hay and concentrate intake were evaluated daily. During Period 2, blood samples were collected before (-2 and 0 h) and at 2, 4, 6, 8, 12, 16, 24, 36, 48, 60, 72, 96, 120, 144, and 168 h after treatment administration. All samples were analyzed for plasma haptoglobin concentration. Samples collected from 0 to 48 h were also analyzed for plasma cortisol, insulin and leptin concentrations. Plasma variables were analyzed using the mean result from samples collected at -2 and 0 h as covariate. Treatment  $\times$  day interactions were detected ( $P < 0.01$ ) for hay and total DMI, given that these parameters were reduced ( $P \leq 0.05$ ) in VAC compared with CON on d 0 and 1. Treatment  $\times$  hour interactions were detected ( $P \leq 0.02$ ) for all plasma variables. Plasma cortisol concentrations were greater ( $P \leq 0.05$ ) in VAC compared with CON from 2 to 16 h. Plasma insulin concentrations were greater ( $P \leq 0.05$ ) in VAC compared with CON from 4 and 16 h. Plasma leptin concentrations were greater ( $P \leq 0.03$ ) in VAC compared with CON from 6 to 16 h. Plasma haptoglobin concentrations were greater ( $P \leq 0.02$ ) in VAC compared with CON from 16 to 120 h. In conclusion, plasma leptin concentration was increased during a vaccine-induced acute-phase reaction, which may explain the decrease in DMI observed herein in vaccinated cattle.

**Key Words:** cattle, acute-phase reaction, leptin

**29 Effect of an early-life LPS challenge on a subsequent LPS challenge in Holstein bull calves.** Aimee L. Benjamin<sup>\*1</sup>, Filiz T. Korkmaz<sup>1</sup>, Theodore Elsasser<sup>2</sup>, and David E. Kerr<sup>1</sup>, <sup>1</sup>University of Vermont, Burlington, VT, <sup>2</sup>USDA-ARS, Beltsville, MD.

The variable innate immune response between animals can be due to a variety of causes. Infections during the neonatal period could affect an animal's innate response phenotype. Ten Holstein calves were used to investigate the sustained effects of neonatal LPS exposure on the innate immune system. At 8d of age, 5 calves received an IV infusion of LPS (0.5  $\mu$ g/kg), while the remaining 5 received saline. A subsequent dose of

LPS (0.25 µg/kg) was given to all 10 calves at 30 d of age to determine if there was any lasting effects from the first LPS challenge. Skin biopsies were collected from all calves at 22 d of age to isolate dermal fibroblasts for a cell model to explore genetic and/or epigenetic changes that may have resulted from the early life LPS treatment. Two hours after the first challenge, LPS-treated calves had greater ( $P < 0.05$ ) plasma IL-6 (14.4 ± 2.8 vs. 0.6 ± 0.3 ng/mL) and TNF-α (3.2 ± 1.0 vs. 0.2 ± 0.1 ng/mL) compared to calves that had received saline, respectively. Response curves of these plasma cytokines from measurements at 0, 1, 2, 3, 5, and 7 h following the second LPS challenge were similar ( $P > 0.05$ ) between groups. Rectal temperatures were not significantly different between the treatment groups in either challenge. Dermal fibroblasts isolated from LPS- or saline-treated calves produced similar ( $P > 0.05$ ) levels of IL-6 (318.4 ± 96.8 vs. 359.3 ± 54.6 pg/mL) and IL-8 (183.6 ± 33.8 vs. 157.1 ± 11.7 pg/mL), respectively, in response to a 24h LPS challenge (500 ng/mL). Although the fibroblasts produced much greater levels of IL-6 (5178.6 ± 637.9 vs. 5353.4 ± 853.1 pg/mL) and IL-8 (1554.2 ± 96.8 vs. 1596.9 ± 93.4 pg/mL), respectively, in response to a 24h challenge with IL-1b (10 ng/mL), there was no early-life treatment effect. There was substantial inter-animal variation in clinical symptoms following the LPS challenges, but the early-life exposure to LPS at 8d of age did not clearly influence the response to a subsequent LPS challenge in 30-d-old calves, or affect the ability of fibroblasts to respond in vitro to LPS or IL-1b. More work is needed to determine how early-life exposure to infection may influence an animal's innate immune response.

**Key Words:** neonatal, variation

**30 ACTH-test reactivity affect disposition for storage of fat depots in dairy cows during the transition period.** Lena Ruda<sup>1</sup>, Claudia Raschka<sup>1</sup>, Lea Fieguth<sup>1</sup>, Asako Kinoshita<sup>1</sup>, Anja Schacht<sup>1</sup>, Marion Piechotta<sup>1</sup>, Korinna Huber<sup>2</sup>, Akos Kenez<sup>2</sup>, Ulrich Meyer<sup>3</sup>, Sven Dänicke<sup>3</sup>, and Juergen Rehage<sup>\*1</sup>, <sup>1</sup>*Clinic for Cattle, University of Veterinary Medicine Hannover, Hannover, Germany*, <sup>2</sup>*Department of Physiology, University of Veterinary Medicine Hannover, Hannover, Germany*, <sup>3</sup>*Institute of Animal Nutrition, Friedrich-Loeffler-Institute, Braunschweig, Germany*.

It is generally recommended to adjust feeding requirements to avoid overconditioning of dairy cows at calving because high body condition is a major risk factor for ketosis in early lactation. However, besides feeding and environmental factors individual disposition for high body condition appears to have also a genetic and epigenetic background. The individual reactivity of the hypothalamic-pituitary-adrenal axis (HPA-axis) and the release of cortisol as tested in the ACTH-test varies considerably between cows. Thus, the aim of the study was to test the relationship between HPA-axis reactivity and body condition in dairy cows during the transition period. In 22 pluriparous German HF dairy cows, kept in freestalls with cubicles and fed a TMR based on grass and corn silage and concentrate, an ACTH challenge was performed on d 100 postpartum (pp). Before and after injection of ACTH (80µg) cortisol concentrations were measured in blood samples taken in short-term intervals from indwelling jugular vein catheters from which baseline and peak concentrations and AUC were assessed. Results were used in a regression analysis (SAS statistical package) and correlated with sonographically estimated subcutaneous fat mass (SC fat) assessed at d -42 before parturition, and at d 1, d 21, d 100 pp. Cows were also classified

according the AUC cortisol results into high (HR; n = 7), intermediate (IR; n = 8), and low (LR; n = 7) responder and fat mass was tested for differences by ANOVA. Significant linear negative correlations were found between AUC and peak cortisol concentrations from ACTH challenge and SC fat at d -42, d 1, d 21 and d 100. In HR compared with LR cows in average SC fat was significantly less at d -42, d 1, d 21 and d 100 and gain before and loss of SC fat after parturition was lower. Cows with high reactivity of the HPA-axis are leaner during the whole transition period, gain less before and lose less SC fat after parturition. Dynamics in body condition appears to be closely correlated with HPA axis reactivity during the transition period in dairy cows. This work was funded by the German Research Foundation.

**Key Words:** ACTH-test, BCS, dairy cows

**31 Use of an ex vivo/in vitro laminitis model to elucidate the role of endotoxins during equine and bovine laminitis.** Nicole Reisinger<sup>\*1</sup>, Simone Schaumberger<sup>2</sup>, and Gerd Schatzmayr<sup>1</sup>, <sup>1</sup>*Biomim Research Center, Tulln, Austria*, <sup>2</sup>*Biomim Holding GmbH, Herzogenburg, Austria*.

Laminitis is one of the most common causes for lameness in horses and ruminants. The pathology of laminitis is still not fully understood. As it is a multifactorial disease, several substances and toxins such as endotoxins are discussed as possible trigger factors. The aim of our study was to test the influence of endotoxins on the lamellar integrity of hoof explants. Furthermore, the potential difference of effects of endotoxins on the lamellar integrity of hooves from horses and ruminants was investigated. Explants from hooves (n = 3) and claws (n = 3) were cultivated at 37°C and 5% CO<sub>2</sub> with D-MEM as culture medium. Lipopolysaccharides (LPS) of *Escherichia coli* O55:B5 were added to the equine [0, 2.5, 10, 100 µg/mL] and bovine [0, 1, 10, 100 µg/mL] explants for 24 h. After incubation, explants were tested for their integrity by measuring the force [Newton], which is needed to separate the explants (= separation force). Therefore, the explants were connected to a force transducer with clamps. Viability of explants was tested with the water-soluble tetrazolium (WST-1) assay. There was no effect on separation force when equine explants were incubated with 2.5 µg/mL LPS. Separation force of equine explants incubated with 10 and 100 µg/mL LPS was significantly decreased ( $P < 0.005$ ) by 45% and 49%, respectively, compared with control explants. Similar to equine explants, there was no effect on separation force when bovine explants were incubated with 1 µg/mL LPS. Separation force of bovine explants incubated with 10 and 100 µg/mL LPS was significantly decreased ( $P < 0.005$ ) compared with control explants. A reduction by 50% and 65% for LPS was observed, respectively. All explants were viable after 24 h incubation. In our study a concentration dependent reduction of separation force of explants incubated with LPS was observed. Similar effects of LPS were observed in explants from both species. Although laminitis has never been induced by endotoxins alone in animal experiments, our data suggest that endotoxins might play an important role during the onset of laminitis. The presented model could be used to test other potential trigger factors of laminitis and the interaction between these factors.

**Key Words:** laminitis, endotoxins, in vitro

# Bioethics Symposium: Effects of science, government, and the public in directing the future of animal agriculture

**32 Role of science in the future of animal agriculture.** Paul H. Hemsworth\*, *University of Melbourne, Parkville, Victoria, Australia.*

Animal welfare is a state within the animal. Scientists use 2 main conceptual frameworks for understanding animal welfare: biological functioning and affective state. These 2 frameworks were initially seen as competing, but more recently biological functioning is recognized to include affective experiences and affective experiences are recognized as products of biological functioning. While science provides us with the ability to understand how the body responds to physical challenges and affective states, our ability to monitor some affective states is under-developed. Thus most studies have used the biological functioning framework to infer compromised animal welfare, on the basis that suboptimal biological functioning accompanies negative affective states. There is increasing societal interest in providing domesticated animals with the opportunity for positive affective experiences and this will also be a major focus for animal welfare science in the early 21st century. Furthermore, animal welfare science will continue to identify conditions and strategies to prevent and ameliorate negative states in farm animals, and extend the range of animal welfare indicators for use on-farm in risk assessment and management, welfare benchmarking and welfare auditing. Science thus should provide the facts, but what society, groups or individuals do with these facts is a philosophical decision. Exclusion of science can result in emotive or self-interested arguments from sectional groups dominating community debate. This is not to say that such arguments should be ruled out; quite the reverse, as they reflect, in part, current community values. However, they should contribute to, not pre-empt, the debate. Furthermore, these ethical questions should include other considerations, such as our duties toward animals and human health, economic, social and environmental consequences. Thus, in resolving the question of whether or not a particular animal use is acceptable, science provides the means to understand the impact of each animal use on the animal and has a prominent role in underpinning decisions on animal use and the attendant conditions and compromises.

**Key Words:** animal welfare, science, role

**33 Update on the US Technical Advisory Group to the International Organization for Standardization (ISO) Working Group 16—Welfare of Food-Producing Animals.** Craig A. Morris\*, *United States Department of Agriculture, Agricultural Marketing Service, Washington, DC.*

World Organization for Animal Health (OIE) Member Countries mandated the organization to take the lead internationally on animal welfare and, as the international reference organization for animal health, to elaborate recommendations and guidelines covering animal welfare practices, reaffirming that animal health is a key component of animal welfare. However, there can be impediments to the ability of the OIE to assure worldwide adherence to the codes they develop; especially in the developing world. In 2011 to respond to this issue, the International Organization for Standardization (ISO) and the OIE established a Memorandum of Understanding to strengthen cooperation in all fields of mutual interest, including animal welfare. The ISO is a worldwide federation of national standards bodies and the work of preparing ISO standards is usually carried out through ISO Technical Committees. The ISO took on this work to help facilitate the international adoption of OIE codes, such as those dealing with animal welfare, through the private

sector implementing seller or buyer requirements. The ISO committee structure is broken down into Technical Committees (TCs) and the ISO assigned this work on animal welfare to TC34 (Food Products) with the creation of a Working Group (WG) 16. To participate in WG16, experts from the United States formed a Technical Advisory Group that is chaired by Dr. Morris of the United States Department of Agriculture. WG16 is currently working toward the development of the ISO's first Technical Specification (TS) in this area of animal welfare and plans for it to be complete by the end of 2015. The objective of this work is to have the private sector use this TS to not only further the international adoption of the OIE codes, but serve to better the living conditions of animals raised for food around the world while not impeding the international trade of animal products. This presentation will update attendees on the progress being made by the ISO in this important area.

**Key Words:** bioethics, animal welfare, ISO

**34 Effect of consumer choices on food animal production practices in the future.** Charlie Arnot and J. J. Jones\*, *Center for Food Integrity, Gladstone, MO.*

The application of technology in food and agriculture has provided countless benefits to society. Innovation and technology help us meet one of humanity's most basic needs—the need to provide safe, nutritious food for our children and our children's children. Today, our challenge is not just better technology, but finding better ways to support the informed public evaluation of those technologies and our food production system. Consumer choice is playing an increasing role in the food system. No matter what science says, many issues remain contentious because the social decision-making process is complex. The ability to break down the communication barriers is critical to fostering informed decision making that encourages technology and innovation in society's best interest. The Center for Food Integrity's 2014 research, combined with previous research provides great insight into how consumer choice—consumer decision-making—will have many effects on animal agriculture.

**Key Words:** consumers, choice, food

**35 US consumer perceptions of animal use for food, recreation, and more: Are feelings about Bambi the deer, Bessie the cow, and Buster the dog more related than we thought?** Elizabeth Byrd\* and Nicole Widmar, *Purdue University, W. Lafayette, IN.*

This research seeks to link sentiments about various animal uses, such as pets, circus animal, and racing dogs, to sentiments and perceptions of livestock animal welfare. A national-scale survey was used to explore linkages between animal uses and species. Researchers, marketers and livestock industries alike have also sought to uncover relationships between observable demographics (sex, age, education, pet ownership) and views of livestock practices. If owning a pet and/or interacting with animals in general is related to increased concern for livestock welfare, what else may be affecting consumers' perceptions of livestock rearing? Is the approval of using animals for working dogs, such as service/therapy or police/military, related to sentiments toward farm animals? When asked, 92% of US consumers agree it is acceptable to have animals as pets or for service/therapy animals. A total of 93% of US consumers agree with using animals to produce eggs, but



only 67% agree with keeping zoo animals. Is the approval of hunting and various hunting practices related to concern for food/farm animal welfare? More broadly, are sentiments toward animal species related and does it depend whether or not they are produced and consumed for food or even the perceived average age of those animals? Do those respondents who believe dairy cows have a long life expectancy on a farm also report increased concern for their welfare? Respondents reported the average life expectancy of a dairy cow on a dairy farm to be 9.71 years and the average life expectancy of an egg laying hen to be 5.29 years. Understanding how consumers' sentiments toward animal uses are related to each other and to key socio-demographic factors will provide valuable insight into the minds of consumers and provide guidance for the agricultural industry in understanding, communicating with, and meeting the demands of consumers.

**Key Words:** animal use, consumer demand, preferences

**36 Willingness to pay for pork chops and chicken breasts: Are hunters (and those who approve of hunting) different (from the average US consumer)?** Elizabeth Byrd\*, Nicole Widmar, and John Lee, *Purdue University, W. Lafayette, IN.*

Consumers, in general, are concerned about how their food, especially meat, is raised. This concern extends to animal welfare practices and the social and environmental impacts of production. Recently, studies have focused on consumer willingness to pay (WTP) for various production process attributes, such as gestation crate-free hog production, cage-free egg production, and rBST-free milk production. However, most studies have been limited to general samples of US residents or consumers. Other research has linked demographics, including pet ownership, to increased concern for pig welfare (McKendree, Croney, and Widmar, 2014). Relationships with animals, even those that are not consumed for food, are related to consumers' level of concern for farm animal welfare. Studies have explored the public's acceptance of lethal management of wildlife and feral animals. We seek to determine how views of wild animals, both consumptive and non-consumptive, may affect the level of concern for livestock animal welfare. Concern for animal welfare can extend to include WTP for attributes of the production process that affect animal welfare. The next step is to determine how interactions with wild animals, in terms of hunting (or approval of hunting), are related to concern for farm animal welfare. Preliminary results indicate those who regularly hunt are willing to pay less for animal welfare attributes such as cage or crate free production and antibiotic free production. For example, non-hunters have a mean WTP of \$3.13/lb for USDA-verified crate-free pork chops, but those who regularly hunt are not WTP anything. Likewise, non-hunters are willing to pay more than twice what hunters are willing to pay for USDA-verified antibiotic-free production. The goal of this analysis is to determine how consumers' outdoor activities and key demographic factors (sex, pet ownership, and opinions on

hunting) are related to the relative importance of food values for meat purchases and WTP for animal welfare production process attributes.

**Key Words:** animal welfare, consumer demand, preferences

**37 Willingness to pay for pork chops and bacon: Effects of perceived farm sizes and information shocks.** Ann Cummins\*, Nicole Widmar, Joan Fulton, and Candace Croney, *Purdue University.*

This research utilizes a willingness to pay (WTP) model for a variety of pork, specifically pork chop and bacon, attributes. There has been an identified gap between consumer's perceptions and the reality of even something as simple as the size of pig farm which pork originated from. This presentation explores the effect of the perceived size of pig farm on consumers' WTP for verified pork attributes. The data for this analysis is from a nationally representative survey (in terms of age, sex, income, and region of residency). A total of 1,004 respondents and 10,040 choice situations were obtained. Along with a collection of demographic, educational, perceptions about farming, and other information is used as part of the analysis, participants experienced a simulated shopping experience (designed choice experiment) where they made purchasing decisions about certified pork products with different attributes. The attributes included price per pound, permitted use of individual crates or stalls, farm size, antibiotic use, and certification entity. The choice experiment methodology is used for this analysis and the random parameter logit model is used to estimate the consumers' WTP for the different pork product attributes. The first set of analyses look at the average WTP for these two pork products in relation to individual consumer's self-reported perception of the farm size on which they believe most pigs raised for pork are raised (which they provided before entering the simulated shopping experience). The second set of analyses uses the choice experiment data, but in addition uses an information shock, in which a subset of approximately half of the respondents were randomly selected and given an information shock with included NASS statistics on the true pig farm size in the U.S. These responses are then compared between perceived size of pig farms in the US, the information shock, and consumers' WTP for pork verified attributes including farm size. We find that certain consumer segments are willing to pay statistically significant and positive amounts for the verified attributes studied. Further, there are differences in those WTP values amongst pork products, verifying parties, and the specific attributes in question.

**Key Words:** bacon, pork chops, willingness to pay

# Breeding and Genetics Symposium: Relevance of modeling in the genomics era

## 38 Is complex modeling important in the age of genomic selection? Guilherme J. M. Rosa\*, *University of Wisconsin, Madison, WI.*

Statistical methodology has always played a fundamental role in modern animal breeding and genetics. For example, regression and ANOVA techniques have been developed and applied extensively in the context of estimation of genetic parameters and prediction of genetic merit for complex traits. Later, linear mixed model approaches such as best linear unbiased prediction (BLUP) and residual maximum likelihood (REML) estimation of (co-)variance components became prevailing in the analysis of pedigreed data, given their flexibility to accommodate unbalanced data, complex genetic relationships and overlapping generations. Several extensions of mixed models techniques have also been applied in animal breeding, such as the analysis of binary and count data, growth curves, survival models, and gene mapping in outbred populations. These complex models have been frequently implemented using Bayesian and MCMC techniques, facilitated by recent advances in computing technology. More recently, accessibility to genomic technologies has allowed fine mapping of causative loci, high throughput functional genomics studies, and whole-genome prediction of complex traits in livestock species. However, advancements in genomic technologies have also brought several new challenges from data-storage and data-mining standpoints, given the dimensionality of current data sets. Nowadays, not only efficient computer algorithms are required for data storage and data management, but also carefully tailored data mining tools are essential to deal with issues of multiple testing, potential of over fitting, spurious associations, and nonlinearities and complex interactions inherent to genomic data. In this presentation I will review some of the contemporary statistical and data mining methods currently used in animal breeding and genetics, for both prediction and causal inference, with especial emphasis on mixture regression models and graphical models, and the incorporation of biological knowledge into the analyses. Through some examples, I will illustrate the importance of complex modeling in the age of genomic selection.

**Key Words:** statistical models, genomic data, animal breeding

## 39 BLUP, REML, and other tools in the age of genomic selection. Esa A. Mäntysaari\* and Martin Lidauer, *Natural Resources Institute Finland, Green Technology, Jokioinen, Finland.*

Onset of genomic selection changed the focus of animal evaluation experts into estimation of genomic breeding values (GEBV). This was because of enormous potential of genomic information, but also because of similar intellectual challenges in methodologies. Still, also GEBV rely on phenotypes as a source of information. The GEBVs and the ordinary estimated breeding values (EBV) have the same need of well-defined models to attain accurate and unbiased results. Milk and component yield EBVs can illustrate the value of accuracy. Although  $EBV_{protein}$  or  $EBV_{fat}$  can be used as indirect estimates of  $EBV_{milk}$  (correlations to milk EBV 0.91 and 0.79), the  $GEBV_{milk}$  trained on protein (fat) gave validation reliability of 0.27 (0.10), while the training on milk gave  $R^2 = 0.45$ . In this presentation we discuss particulars of breeding value estimation models and approaches for estimation of variance components (VC). The examples used are the joint Nordic test day model and the multiple trait-across country (MACE) model. The Nordic test day model is used to evaluate bulls and cows in Finland, Sweden and Denmark for 4 breeds: Holstein, Red Dairy Cattle, Jersey and FinnCattle. The challenges are the

varying production conditions and admixed populations. The model is a multilactation, multitrait (milk, protein, fat) random regression. In every evaluation run the heterogeneity of variance is estimated for each trait and herd-year. During the implementation, VC for 1,827 parameters were estimated for each country and breed combination. Estimation was done using Monte Carlo REML and EM-algorithm. Most genomic evaluations rely on genotype exchange and MACE results for training. Accuracy of MACE depends on the assumed correlations across countries. Currently Interbull estimates correlations among breeding values of bulls from 31 countries, 6 breeds and 40 traits. The largest single VC estimation is for the Holstein production traits involving all countries. The challenges are computing, and the lack of genetic ties among smaller countries. Current estimation is by subsets of countries, but another alternative is to use MC REML and all countries simultaneously.

**Key Words:** breeding value estimation, variance components

## 40 Practical implications for genetic modeling in the genomics era for the dairy industry. Paul M. VanRaden\*, *Animal Genomics and Improvement Lab, Agriculture Research Service, USDA, Beltsville, MD.*

Genetic models convert data into estimated breeding values and other information useful to breeders. The goal is to provide accurate and timely predictions of the future performance for each animal (or embryo). Modeling involves defining traits, editing raw data, removing environmental effects, including genetic-by-environmental interactions and correlations among traits, and accounting for nonadditive inheritance or nonnormal distributions. Data included phenotypes and pedigrees during the last century and genotypes within the last decade. Genomic data can include markers, haplotypes, and causative effects such as insertions, deletions, or point mutations; most models also include polygenic effects because the markers do not track causative variants perfectly. Total numbers of known variants have increased rapidly from thousands to hundreds of thousands to millions. Nonlinear models add precision for traits influenced by major genes, but linear models work well for traits with more normally distributed genomic effects. Numbers of genotyped animals in US dairy evaluations increased rapidly from a few thousand in 2009 to about 1 million in 2015. Most are young females that will contribute to estimating allele effects in the future, but only about 100,000 have phenotypes so far. Traditional animal models may become biased by genomic preselection because Mendelian sampling of phenotyped progeny and mates is no longer expected to average 0. Single-step models that combine pedigree and genomic relationships can account for such selection, but approximations and new algorithms are needed to avoid excessive computation. Traditional animal models may include all breeds and crossbreds, but most genomic evaluations are still computed within breed. Inclusion of inbreeding, heterosis, dominance, and interactions can improve precision. Multitrait genomic models may be preferred for traits with many missing records or when foreign records are included as pseudo-observations, but most countries use multitrait traditional evaluations followed by single-trait genomic evaluations. A final goal is to explain how the models work so that breeders can more confidently apply the predictions in their selection programs.

**Key Words:** genetic evaluation, genomic selection, mixed models

**41 Experiences in bioinformatics.** Luc L. Janss\*, *Aarhus University, Tjele, Denmark.*

Knowledge from bioinformatics research can in principle be used to improve genomic predictions. Examples are use of the QTLdb database with collected QTL mapping results, the use of genomic feature annotations, and pathway or Gene Ontology (GO) data. There are, however, several hurdles to appropriately use this information and include it in genomic models for analysis of livestock data. A limitation in the use of QTL mapping results is that results from classical linkage analysis studies have wide confidence intervals such that for any trait a large part of the genome will be tagged as "QTL region". Despite this limitation, SNPs in QTL regions can be found to explain more variance than those outside QTL regions, which is for instance shown for milk and fat yield in dairy cattle, but not for protein yield. A limitation of genomic feature annotations is that this information is not covering the whole genome and is not directly related to traits, or, for pathway or GO data, is mostly categorized in fundamental biological processes. This makes it difficult to attach genuine prior information to this data. The common approach to use this kind of bioinformatics data is to simply try which features or GO groups explain more variance or predict better. This leads to results such as that SNPs in/near genes (but not necessarily from the coding parts of genes) explain more variance in phenotypes. A final significant hurdle to use this information is that our animal populations are highly structured and include relatives. This leads to long-range LD and LD between chromosomes, which effectively spreads QTL effects over the whole genome. This creates a polygenic image of the trait architecture, which matches the assumptions of GBLUP that all SNPs contribute equally, and makes genomic relationships the main driver for genomic prediction within animal populations. Better modeling, notably a separation of effects of linkage and LD in genomic prediction, allows more meaningful inferences from bioinformatics data, and potentially allows to improve genomic predictions where relationships are weak; for example, across breed.

**Key Words:** bioinformatics, genomic feature, genomic prediction

**42 Practical implications for genetic modeling in the genomics era for the beef industry.** Andy D. Herring\*, *Texas A&M University, College Station, TX.*

The beef cattle industry is based on valuation of phenotypes, and its supply chain components vary across global region. In many areas, producers maintain ownership of animals until sale to an abattoir, yet in many other regions distinct industry segments (cow-calf, grower/stocker, finisher/feedlot, packer) exist where animal ownership changes across segment. Commodity cattle (of unknown genetic and/or management background) have different value potential as compared with cattle with known background. The utility of genomic data and analyses are, and will remain, different regarding these 2 types of cattle. Many genomic approaches have calculated molecular breeding values of animals; however, most beef industry managers other than seedstock producers would much rather have predicted phenotypes (predictions that could be collectively based on breeding value, non-additive genetic value, and environmental value) for improved management and marketing decisions. Current US beef industry trends show increasing carcass (and mature cow) weight and carcass quality grade, but static incidence of respiratory disease in feedlots, and no improvement in percent calf crop weaned in beef herds. Trends for more prevalent and less costly genomic data will also continue. There appears to be large potential to genomically characterize beef cattle for production-related physiological systems (health, growth, body composition, fertility, nutrient utilization) as well as potential interactions for optimal economic management and production system assessment. Better understanding of these systems and their components will require knowledge beyond the DNA sequence including RNA regulatory elements and products and protein function and structure; the roles of fetal programming and epigenetics on economically important traits in beef production remain largely unknown and need investigation. Resource cattle populations with detailed phenotypes and banked biological samples, and that evaluate multiple components of beef cattle production systems remain critically important; partnerships of industry groups and research institutions can assemble large, informative data sets. The incorporation of genomic data into economic assessments is also encouraged.

**Key Words:** genetic modeling, beef cattle production systems



# Comparative Gut Physiology and Nonruminant Nutrition Symposium: The gut–brain axis—Sensing and signaling

**43 Nutrient and non-nutrient sensing in the gastrointestinal tract.** Soraya P. Shirazi-Beechey\*, *University of Liverpool, Institute of Integrative Biology, Liverpool, UK.*

The intestinal epithelium is a major interface with the outside world. This interface is separated from the body's internal milieu by a single layer of epithelial cells consisting of absorptive enterocytes, goblet, enteroendocrine and Paneth cells. These cells are exposed, at the luminal domain, to an external environment that is continuously changing by types and amounts of nutrients, microorganisms, microbial products, gastrointestinal secretions and potentially toxic chemicals. The intestinal epithelium constantly monitors the composition of its contents to optimize nutrient absorption, as well as defending threats to its integrity. In recent years significant advances have been made in the understanding of the molecular recognition events involved in sensing the luminal contents of the gastrointestinal tract. The sensing of various nutrients in the gastrointestinal tract is accomplished by several G-protein coupled receptors, expressed on the luminal membrane of enteroendocrine cells. Sensing of nutrients by these receptors leads to secretion of hormones that control vital physiological functions such as food intake, nutrient digestion and absorption, intestinal barrier function and insulin secretion. The intestine also contains approximately 1000 different species of bacteria and has to discriminate between pathogenic and commensal bacteria to maintain a balance between immune protection and inflammatory over-reactions. A class of proteins known as pattern recognition receptors, in particular toll-like receptors (TLRs 1–10) play a key role in the recognition of microbes via detection of conserved molecular features. The sensory receptors that face the lumen of the intestine and are responsive to luminal contents provide a unique therapeutic opportunity. In my talk I will present data on the role of the gut expressed taste 1 receptor (T1R) family in intestinal nutrient sensing and the contribution of TLR9-recognition of bacteria in control of gut hormone release. The impact of these findings to animal nutrition will be discussed.

**Key Words:** intestine, taste 1 receptor (T1R), toll-like receptor 9 (TLR9)

**44 Effects of supplemental amino acids in low-protein diets on intestinal tight junction and amino acid transporters in growing pigs.** S. J. Zhang\*<sup>1,2</sup>, W. Parnsen<sup>1</sup>, and S. W. Kim<sup>1</sup>, <sup>1</sup>*Department of Animal Science, North Carolina State University, Raleigh, NC,* <sup>2</sup>*College of Animal Science and Technology, China Agricultural University, Beijing, China.*

This study was to determine the effects of supplemental AA in low-protein diets on tight junction and AA transporters in the small intestine of growing pigs. 72 pigs ( $19.7 \pm 1.1$  kg, 36 barrows and 36 gilts) were allotted to 3 treatments with 8 pens per treatment (3 pigs per pen, 4 barrow pens and 4 gilt pens) using sex and initial BW as blocks: NC (supplemental Lys, Met, and Thr at 18% CP), PC (supplemental Lys, Met, and Thr at 16% CP), and PCT (PC + 0.05% Trp). The NC and PC diets had AA to meet the NRC 2012 requirements (0.98% Lys, 0.55% Met + Cys and 0.59% Thr) whereas PCT diet had additional 0.05% Trp exceeding the requirement. After 4 wk feeding, 24 pigs (1 pig per pen with median BW) were euthanized to obtain duodenum and jejunum to quantify tight junction proteins (claudin-1, occludin-1, and ZO-1) and jejunum to quantify mRNA of AA transporters (CAT-1, b<sup>0,+</sup>AT, rBAT, y<sup>+</sup>LAT, 4F2hc, and B<sup>0</sup>AT). Tight junction proteins were separated by

Western blot and bands densities were measured. The mRNA of AA transporters was quantified by a real-time PCR. Data were analyzed using the Mixed procedure of SAS. Treatment and sex were the fixed effects and initial BW was a random effect. Pigs in PCT had increased ( $P < 0.05$ ) claudin-1 in duodenum and jejunum compared with pigs in NC and PC. No difference in ZO-1 in duodenum and jejunum was detected among treatments. Pigs in PC had decreased ( $P < 0.05$ , 39.3%) occludin-1 in duodenum compared with pigs in NC. There was no difference in occludin-1 between PC and PCT. Pigs in PC and PCT had increased ( $P < 0.05$ ) mRNA concentrations of CAT-1 (2.29 and 1.92 fold), 4F2hc (2.76 and 2.45 fold), and B<sup>0</sup>AT (2.12 and 2.26 fold, respectively) compared with pigs in NC. Collectively, use of supplemental amino acids (Lys, Met, Thr, and Trp) in low protein diet to meet the NRC 2012 requirements could increase AA transporters in jejunum and additional 0.05% Trp exceeding the NRC 2012 requirements enhanced intestinal tight junction.

**Key Words:** amino acid transporters, low-protein diet, tight junction

**45 The emerging role of bile acids as nutrient-sensing signals.** Ignacio R. Ipharraguerre\*<sup>1,2</sup>, <sup>1</sup>*Institute of Human Nutrition and Food Science, University of Kiel, Kiel, Germany,* <sup>2</sup>*Lucta S.A., Montornes del Valles, Spain.*

Traditionally, bile acids (BA) have been viewed as detergent molecules involved in the intestinal digestion and absorption of lipids and hepatic maintenance of cholesterol homeostasis. During the last 15 years, however, it has become clear that BA are important regulatory molecules with systemic endocrine functions that signal changes in luminal nutrients and microbial activity during the fed-fast cycle. The regulatory actions of BA are primarily mediated by the nuclear receptor FXR (farnesoid X receptor), the G-protein coupled receptor TGR5 and cellular signaling pathways (AKT and ERK1/2). These proteins are highly expressed in the liver as well as enterocytes, endocrine cells and enteric neurons in the small intestine. Surprisingly, BA receptors are also present in several tissues outside the biliary tree including adipose tissue, pancreas, and immune cells. In mice, via interaction with these sensors and pathways, BA control the expression of genes and concentration of circulating hormones involved in the regulation of lipid and glucose metabolism, energy expenditure, intestinal integrity, motility, and immune homeostasis, gut microbial growth, and inflammation. In pigs, evidence from seminal studies indicates that BA signaling is implicated in the intestinal secretion of glucagon-like peptides, gut mucosal growth and protection, nutrition-related liver disease, and changes in host weight and metabolism resulting from variations in gut microbiota. It is important to note that BA differ in their ability to activate signaling pathways and that the composition of the BA pool varies remarkably among non-ruminant animals. In fact, all non-ruminant species of interest (e.g., pig, chicken, rabbit, horse) have unique BA signatures. In view of these differences and the emerging role of BA as regulatory molecules, the study of their involvement in signaling outside the enterohepatic circulation and in mediating reciprocal communication between the host and its microbiota will become a relevant and expanding field of research in animal nutrition and physiology.

**Key Words:** bile acid signaling, FXR, TGR5

#### 46 The role of gut peptides in the gut-brain-axis of livestock.

Andrew P. Foote\*, *USDA-ARS, US Meat Animal Research Center, Clay Center, NE.*

Gut peptides are small hormones produced within the gut that are involved in many biological processes including, but not limited to, appetite regulation, mucosal growth, and metabolism regulation. Some peptides, such as cholecystokinin (CCK) and xenin-25 may affect appetite by altering gut motility through cholinergic pathways, but most of the hunger/satiety signals are processed through the brain. Ghrelin is a peptide produced mostly in the gastric stomach or abomasum and increases before a meal. The ghrelin receptor is expressed in neurons in the arcuate nucleus of the hypothalamus and binding leads to the release of neuropeptide Y and agouti-related peptide, thereby stimulating appetite. While ghrelin is thought to serve as a hunger signal in meal fed animals, it may also be involved in the variability of DMI in ad libitum fed animals. Other gut peptides, including peptide YY, oxyntomodulin, and glucagon-like peptide-1 (GLP-1), act as satiety signals and are inhibitory to ghrelin. These peptides stimulate neurons in the arcuate nucleus to release  $\alpha$ -melanocyte-stimulating hormone ( $\alpha$ -MSH) and cocaine and amphetamine regulated transcript (CART), thereby decreasing appetite. The actions of gut peptides are not limited to appetite regulation. Glucagon-like peptide-2 (GLP-2) is a potent stimulator of intestinal mucosa growth and gut blood flow, and could be important for gut health of livestock. Glucose-dependent insulinotropic polypeptide (GIP) increases insulin secretion and regulates lipid metabolism. Because the complete functions of many gut peptides in livestock species are not known, studying their regulatory roles is critically important in nutritional physiology and animal health.

**Key Words:** appetite regulation, metabolism regulation, gut function

#### 47 Nutrient sensing by glucagon-like peptide-1 secreting cells.

Frank Reimann\*, *Institute of Metabolic Science & MRC Metabolic Diseases Unit, University of Cambridge, Cambridge, UK.*

Glucagon-like peptide-1 (GLP-1) is an enteroendocrine hormone secreted by L-cells found throughout the intestinal epithelium, but with increased frequency in the ileum and colon. GLP-1 acts as an incretin, boosting postprandial insulin secretion, inhibits glucagon secretion and is anorexigenic. Using transgenic mouse models allowing labeling and manipulation of GLP-1 and GLP1R expressing cells we investigated molecular mechanisms underlying hormone secretion and responses in the GLP-1 axis. L-cells are electrically excitable cells, displaying an increased action potential firing rate when nutrients are available. This can be achieved by electrogenic nutrient uptake, for example by the sodium coupled glucose transporter (SGLT-1) or the proton coupled dipeptide transporter (PepT1). L-cells isolated from knockout mice for these transporters have attenuated responses to glucose and dipeptide, respectively. Lipid derived molecules and bile acids, by contrast were shown to be detected by the G-protein coupled receptors FFAR1 and TGR5 (GPBAR), predominantly coupling to increases in cytosolic  $Ca^{2+}$  and cAMP, respectively. In the distal intestine, where most nutrients are likely to be processed by the intestinal microbiota, L-cells can be shown to respond to short chain fatty acids (via FFAR2/3) and indole, with the latter inhibiting voltage gated potassium channels and ATP-production, resulting in stimulatory and inhibitory signals respectively. Distal L-cells also differ from small intestinal L-cells in that they co-secrete peptide YY (PYY) and the orexigenic peptide Insulin-like peptide-5 (InsI5), whereas duodenal/jejunal L-cells co-express glucose-dependent insulinotropic polypeptide (GIP, the other incretin) and cholecystokinin (CCK). It is hoped that an improved understanding of the gut hormone signaling will

facilitate the development of new therapies that harness endogenous gut hormone reserves for the treatment of metabolic disease.

**Key Words:** glucagon-like peptide-1 (GLP-1), hormone secretion, nutrient sensing

#### 48 Effect of feeding rate and glucose provision on plasma

**glucagon-like peptide 2 concentration in dairy calves.** Sarah Y. Morrison\*<sup>1</sup>, Juan. J. Castro<sup>1</sup>, Kristen M. Glosson<sup>1</sup>, Jens. J. Holst<sup>2</sup>, James K. Drackley<sup>1</sup>, and Ignacio R. Ipharraguerre<sup>3,4</sup>, <sup>1</sup>*University of Illinois, Urbana, IL,* <sup>2</sup>*Department of Biomedical Sciences, University of Copenhagen, Denmark,* <sup>3</sup>*Institute of Human Nutrition and Food Science, University of Kiel, Germany,* <sup>4</sup>*Lucta SA, Barcelona, Spain.*

Glucagon-like peptide 2 (GLP-2) may have therapeutic potential in young calves encountering stressors, such as weaning and diarrheal disease, because of its pleiotropic actions on intestinal mucosal and barrier function. During diarrhea, feed intake decreases, which is a major negative effector of GLP-2 secretion and increases intestinal atrophy. We determined the effects of feeding rate and supplementation with potential GLP-2 secretagogues in milk replacer (MR) on GLP-2 concentration [GLP-2] in calf plasma. Three days after birth, male Holstein calves (n = 45) were randomly assigned to 1 of 12 treatments arising from factorial combination of feeding rate (FR) and supplement type. Feeding rates were 25, 50, 75, and 100% of standard feeding level on d 5 (1.5% of BW as DM). Supplement treatments were Control: MR, no supplement; GLC: MR plus glucose (220 mg/kg BW per day); and 3OMeGLC, MR plus 3-O-Methyl glucose (6 mg/kg of BW per day). A commercial MR (12.5% solids) was fed twice daily at 10% of BW for d 1–2 and 12% of BW for d 3–5. On d 6–7 calves were fed the FR plus supplement treatments. On d 8, calves were fed 0, 25, 50, and 75% of respective MR allowance on d 5 plus supplement treatments, and plasma was obtained at –15, 15, 30, 60, 90, and 240 min relative to feeding for determination of [GLP-2]. No starter was fed but water was offered ad libitum. Generalized linear models with normal and Poisson distributions were used to analyze [GLP-2] and fecal score data, respectively. As designed, MR intake differed ( $P < 0.0001$ ) among treatments. Occurrence of diarrhea did not differ among treatments. For log [GLP-2], there was a quadratic effect of FR ( $P = 0.0003$ ) and effect of supplement, with 3OMeGLC greater than GLC but not control ( $P = 0.002$ ). Increasing FR increased ( $P < 0.0001$ ) area under the curve for [GLP-2] after feeding; calves fed the 25% rate had the lowest area under the curve and calves fed 100% had the greatest (10,188 vs. 22,023 pg·h/mL). Feeding rate had a significant effect on GLP-2 with calves allotted greater MR intakes having greater plasma [GLP-2].

**Key Words:** glucagon-like peptide 2, feeding rate, glucose

#### 49 The brain within the gut—Activation of enteric cells and sensory neurons.

John B. Furness\*<sup>1</sup>, David M. Bravo<sup>2</sup>, Jeremy J. Cottrell<sup>1</sup>, and Frank R. Dunshea<sup>1</sup>, <sup>1</sup>*University of Melbourne, Parkville, Australia,* <sup>2</sup>*InVivo Animal Nutrition & Health, Talhouët, Saint-Nolff, France.*

For optimal digestive efficiency, the contents of the gastrointestinal tract need to be detected and the information needs to be conveyed to control systems: the gut endocrine system, the nervous system and the immune and tissue defense systems. The contents include nutrients, products of digestion, bacteria, viruses, fungi and potentially injurious substances in foods. Sensory neurons that innervate the gut lining detect hormones released from enteroendocrine cells (EEC) and also detect other signals, for example cytokines and other substances released when

the gut is inflamed, and contractile activity of the gut. There are 4 classes of enteric sensory neurons, intrinsic primary afferent neurons (IPANs), with cell bodies in the gut wall, vagal primary afferent neurons, spinal primary afferent neurons and intestinofugal neurons, and within each class there are subclasses that detect different sensory signals. From a comparative point of view, the functioning of the enteric nervous system (ENS) is similar between species, although there are some differences in its organization, as will be discussed. The ENS works in concert with CNS reflex and command centers to control digestive function, so it cannot be considered in isolation. There is bidirectional information flow between the ENS and CNS. The major type of sensory ENS neuron is the IPAN. These neurons have distinctive shapes and electrophysiological characteristics, similar to primary afferent neurons of dorsal root ganglia. Their axons in the mucosa sense the chemical environment of the lumen and mucosal distortion. They also detect toxins (they are nociceptive neurons). Other processes of the neurons are sensitive to muscle movement. Their outputs are to other enteric neurons (interneurons and motor neurons) and to intestinofugal neurons. The major functions they control are gut movements, water and electrolyte secretion and blood flow.

**Key Words:** enteric nervous system, sensory neurons

**50 Xylanase supplementation in feed reduces incretin and PYY levels in piglets.** Katherine May\*<sup>1</sup>, Saoirse E. O'Sullivan<sup>2</sup>, John M. Brameld<sup>1</sup>, Helen V. Masey O'Neill<sup>3</sup>, Tim Parr<sup>1</sup>, and Julian Wiseman<sup>1</sup>, <sup>1</sup>*School of Biosciences, University of Nottingham, Loughborough, Leicestershire, UK*, <sup>2</sup>*School of Medicine, University of Nottingham, Derby, Derbyshire, UK*, <sup>3</sup>*AB Vista Feed Ingredients, Marlborough, Wiltshire, UK*.

The objective of this study was to investigate the effects of xylanase supplementation on gut hormone production in newly weaned piglets. In experiment 1, 32 female Camb12 weaned piglets ( $8.8 \pm 1.38$  kg, mean  $\pm$  SD) were randomly assigned to 1 of 2 diet groups in period 1 (0–2 wk postweaning; P1), a control diet (Co) or the same diet supplemented with xylanase (XS). During period 2 (2–6 wk postweaning; P2) half the pigs were kept on their original diet while the rest were swapped onto the other diet, resulting in 4 groups. At the end of P2 the pigs were culled by electrical stun and exsanguination at which point blood samples were collected using EDTA coated tubes. The samples were centrifuged at 3000 g for 10 min at 4°C and the plasma was aliquoted and stored at –80°C for further analysis. A Human Metabolic Hormone Milliplex HMHMAG-34K kit (Merck Millipore) was used to assess the concentrations of PYY, PP, Insulin, C-Peptide and GIP (total) in the plasma. In experiment 2, 16 female Camb12 weaned piglets ( $9.2 \pm 0.95$  kg) were assigned to the same 2 diet groups as in experiment 1 (Co and XS). After P1, the pigs were culled and blood samples taken as in experiment 1; however, protease inhibitors were added to the collection tubes so extra gut hormones could be analyzed [amylin (active), ghrelin (active) and GLP-1 (active)]. The data were tested for normality and analyzed with the appropriate parametric or non-parametric test (one/two-way ANOVA or Kruskal-Wallis), significance was accepted at  $P < 0.05$ . In experiment 1, XS in P2 significantly decreased plasma PYY concentrations ( $P = 0.008$ ). In experiment 2, XS significantly decreased plasma GIP ( $P = 0.027$ ) and GLP-1 ( $P = 0.002$ ) concentrations in P1. GIP and GLP-1 are known as incretin hormones that affect pancreas function. In conclusion, xylanase supplementation to newly weaned piglets affects the production of certain gut hormones. The effect was dependent upon the timing of supplementation as different hormones were affected between P1 and P2. Further trials are needed to investigate

whether there are longer-term effects when the piglets are grown to a commercial slaughter weight.

**Key Words:** gut hormone, piglet, xylanase

**51 Perinatal nutrition and the gut–brain axis.** Ryan N. Dilger\*, *University of Illinois, Urbana, IL.*

There exists a need to better understand complex interrelationships between dietary intake and cognitive function, and animal models are critically important in this endeavor. The field of nutritional neuroscience has, as a primary goal, the application of findings to the human clinical setting, and therefore, models that closely mimic the human condition are highly valued. Thus, based on similarities in patterns of brain development and structure, the pig has emerged as a biomedical and preclinical model for studying human brain development and cognitive function. Building upon the storied history of using the neonatal pig as a research model for studying pediatric nutrition and metabolism, recent emphasis has been placed on understanding how signaling mechanisms in the gut, being largely synonymous with microbial interactions, influence neurodevelopment and brain function. As a precocial species, the young pig can be artificially reared with relative ease, thus providing strict control over dietary intake. Moreover, validated methods to assess learning and memory using behavioral assays now exist, and these outcomes provide a powerful and integrated view of how nutrition influences brain development. Sensitive neuroimaging sequences are also available, and along with cellular and molecular techniques optimized for the pig, there exists a fruitful area to generate new knowledge of how early-life nutrition influences neurodevelopmental processes. As such, nutritional strategies to alter the delivery of specific components to the brain, or influence the microbial composition or production of bioactive compounds in the gastrointestinal tract, are currently being tested. There are many advantages to extending the field of pediatric nutrition research by integrating outcomes related to the microbiome, routes of information transfer between the gut and brain, and processes associated with cognitive function and brain development, and the young pig will play a pivotal role in these investigations.

**Key Words:** nutrition, neuroscience, pig

**52 Effects of increasing standardized ileal digestible tryptophan:lysine ratio on performance and ileal expression of cytokine mRNA in weaned pigs challenged with *Escherichia coli* K88.** B. Jayaraman\*<sup>1</sup>, A. Regassa<sup>1</sup>, W. K. Kim<sup>1</sup>, J. K. Htoo<sup>2</sup>, and C. M. Nyachoti<sup>1</sup>, <sup>1</sup>*University of Manitoba, Winnipeg, Manitoba, Canada*, <sup>2</sup>*Evonik Industries AG, Nutrition Research, Hanau-Wolfgang, Germany.*

A study was conducted to determine the optimal standardized ileal digestible (SID) tryptophan:lysine (Trp:Lys) ratio in piglets challenged with *Escherichia coli* K88 (*E. coli* K88) and fed antibiotic-free diets. Thirty individually housed mixed-sex pigs (Duroc  $\times$  [Yorkshire  $\times$  Landrace]) with an initial BW of  $6.41 \pm 0.4$  kg and weaned at  $21 \pm 1$  d were randomly assigned to 5 dietary treatments with 6 replicates per treatment. Dietary treatments consisted of increasing levels of SID Trp:Lys ratios (16, 18, 20, 22 and 24%). Diets were corn-wheat-soybean meal-based with a constant SID Lys of 1.18% that was set to be the second limiting AA but adequate in other AA. Pigs had ad libitum access to feed and water for 13 d. Piglets were fed the experimental diets for 6 d and on d 7, they were orally challenged with 6 mL of *E. coli* K88 ( $2 \times 10^9$  cfu/mL). Body weights and pen feed disappearance were recorded weekly to determine ADG, ADFI and G:F. On d 13, all pigs were euthanized



to obtain ileal tissue samples to measure mRNA expression of tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) and interleukin-10 (IL-10) using qRT-PCR. During the pre-challenge period, increasing dietary SID Trp:Lys increased (linear,  $P < 0.05$ ) ADG (157, 162, 173, 179 and 201 g) and G:F (0.71, 0.73, 0.74, 0.81 and 0.84). During the post-challenge period, there was an increasing trend (linear,  $P = 0.076$ ) in ADG (177, 180, 208, 210 and 213 g), whereas there was no effect on G:F ( $P > 0.10$ ). The optimal SID Trp:Lys determined using the broken-line regression analysis for ADG and G:F in piglets subjected to *E. coli* K88 challenge was 21 and 24.2%, respectively. The expression of pro-inflammatory cytokine, TNF- $\alpha$  mRNA, linearly decreased ( $P = 0.10$ ) with increasing SID Trp:Lys ratio. The expression of anti-inflammatory cytokine, IL-10 mRNA, increased (linear and quadratic,  $P < 0.01$ ) with increasing SID Trp:Lys ratio. In conclusion, in antibiotic-free starter diets, an average optimal SID Trp:Lys of 22.6% optimized performance and immune status of piglets under *E. coli* challenge.

**Key Words:** tryptophan:lysine, *Escherichia coli* K88, piglet

**53 Effect of dietary intervention of probiotic *Lactobacillus helveticus* MTCC 5463 on fecal beta-glucuronidase activity in geriatric volunteers.** Suja Senan<sup>\*1</sup>, Jashbhai Prajapati<sup>2</sup>, Chaitanya Joshi<sup>2</sup>, Sreeja V<sup>2</sup>, Manisha Gohel<sup>3</sup>, Sunil Trivedi<sup>3</sup>, Rupal Patel<sup>3</sup>, Himanshu Pandya<sup>3</sup>, Ajay Phatak<sup>3</sup>, Uday Shankar<sup>3</sup>, and Has Mukh Patel<sup>1</sup>, <sup>1</sup>South Dakota State University, Brookings, SD, <sup>2</sup>Anand Agricultural University, Anand, Gujarat, India, <sup>3</sup>H. M. Patel Center for Medical Care & Education, Karamsad, Gujarat, India.

Probiotics may reduce the risk for the onset of colorectal cancer by modulating the activity of fecal bacterial enzymes that catalyze the liberation of procarcinogenic substances in the intestine. The aim of this study was to investigate the influence of probiotic *Lactobacillus helveticus* MTCC 5463 (5463) on fecal microbial-related functions in geriatric volunteers. The subjects were randomized into 2 groups, fed either fermented drink containing 5463 (test group) or without 5463 (placebo group). The volunteers consumed 200 mL of the product once a day for 4 weeks. Fecal samples were collected in sterile plastic containers at the beginning and end of the intervention period.  $\beta$ -glucuronidase was considered as the marker enzyme for anti-colonic carcinogenesis activity, and measured in terms of unit activity per mg of protein in fecal samples. Total fecal protein was determined by Folin-Lowry method with bovine serum albumin as a standard. The data were analyzed with Student's *t*-test, and the levels of significance were expressed as *p* values. The mean  $\beta$ -glucuronidase activity was reduced in test group from 1.40 to 0.73 ( $\mu\text{g}/\text{min}/\text{mg}$  of protein) while in case of placebo group, no effect on enzyme activity was observed. Enzyme  $\beta$ -glucuronidase activity in the feces of all subjects in the probiotic group, were highly significant ( $P < 0.001$ ), whereas the placebo group showed nonsignificant differ-

ences ( $P = 0.40$ ). We were able to demonstrate consistent reduction in the fecal  $\beta$ -glucuronidase activity in the probiotic group. Results suggested that the metabolic activity of the fecal flora was altered by 5463 therapy, which further decreased procarcinogenic enzyme levels in the large intestine. Probiotic MTCC 5463 interaction with the geriatric host microbiome, affects  $\beta$ -glucuronidase activity, which can be exploited as an alternate therapy against colonic carcinogenesis.

**Key Words:** probiotics, geriatrics,  $\beta$ -glucuronidase

**54 Brain-gut interactions in stress.** Jackie D. Wood\*, *The Ohio State University, Columbus, OH.*

Stress can be viewed as exposure of an animal to a hostile environment that compromises bodily homeostasis. Stress of this nature can occur in many forms, examples of which are extremes in environmental temperature, fear, predator attack and crowding. Elevation of heart rate, blood pressure, respiration rate and emotional agitation are among the many bodily manifestations of stress. In the gut, they can include diarrhea, constipation, incontinence, gastric regurgitation and gastrointestinal mucosal inflammation. Corticotropin-releasing factor (CRF) is a neuropeptide that plays a major role in the body's overall responses to stress, including the gut. The physiology of central CRF signaling pathways in stress-induced changes in gastrointestinal motility and gastric acid secretion has been well characterized. Recent work elucidates how the physiology of peripheral CRF-related mechanisms contribute to stress-induced changes in gut motility and intestinal mucosal function. Acute stress, intramural CRF release and experimental application of selective CRF<sub>1</sub> receptor agonists evoke excitatory responses in neurons in the enteric nervous system (i.e., the brain-in-the-gut). Enteric neurons express CRF and its receptors. Exposure to acute and chronic stresses increases intestinal ion secretion and mucosal permeability to macromolecules and evokes diarrhea. Effects of stress on intestinal mucosal function are mimicked by peripheral injection of CRF and blocked by peripheral injection of non-selective peptide CRF receptor antagonists. CRF mRNA knock down in the enteric nervous system prevents intestinal responses to environmental stress in rodents. These findings constitute strong evidence that activation of peripheral CRF receptors in the enteric nervous system is a key mechanism involved in stress-related alterations of gut motility and mucosal function. In the cotton-top tamarin (*Saguinus oedipus*) model for ulcerative colitis and associated colon cancer, evidence suggests that environmental stress initiates the inflammatory response, which is then sustained by factors in the feces.

**Key Words:** stress, CRF, enteric nervous system

# Extension Education Symposium: Extension and industry outreach for tomorrow's producers

## 55 Developing partnerships between academia and industry.

Todd R. Bilby\*, *Merck Animal Health, Fort Worth, TX.*

Partnerships between academia and industry continue to be an important aspect of aiding in providing educational and research opportunities to advance the agriculture industry. Funding for applied research and extension continues to be difficult to both secure funds and develop long-term strategic partners. Understanding what industry partners are seeking in terms of strategic initiatives and corporate missions will be important in securing a long-term partner. In addition, extension needs to develop programs and other opportunities that will target a broad geography, deliver science-based outcomes, and aid in the implementation of these outcomes utilizing the latest technologies. Industry partners are actively seeking academic partners with a strong reputation in their field of interest who can deliver within a timely manner on tasks set forth by both parties. Academia would benefit by using more power of persuasion, salesmanship, and clearly illustrating to an industry partner the ROI for both parties. Once the right partner is chosen, articulation and documentation of terms, executing a contract that respects the integrity and commitment of all parties as well as timely delivery of outcomes is imperative to continued collaboration. Some significant roadblocks in negotiations between academia and industry occur during the processes of valuation, IP ownership, indemnification, royalties and conflict of interest. Often, academia representatives do not have enough information to understand the market factors during negotiations, however, industry has proprietary issues that restrict corporations from sharing information with regard to risk mitigation and value assessment for given products and markets. Lack of knowledge surrounding industry constraints coupled with the inability to share knowledge from industry can make negotiations and partnerships difficult. Enhanced appreciation for constraints, instead of focus on incentives, is required to share ideas and develop long-term partnerships.

**Key Words:** academia, industry, partnership

## 56 What I've seen and done on both sides: Academia and Industry.

Larry Corah\*, *Certified Angus Beef, Wooster, OH.*

There has been a progressive change in the evolution of information transfer in animal agriculture over the past 70 years. During the era of 1930s to 1960s, information flow centered on university research >> > transferred to the local county agent >> > and then shared with the producer. Starting in the 1960s to 1970s, that process started to change. Numerous commercial companies started adding expertise to support product research, the advent of consultants occurred, universities started filling extension positions with greater research expertise and focus and a new era of information flow started as these industry specialist and consultants played a much more active role. Today we find an even greater role of industry in technology transfer as they explain new R&D findings which coupled with reduced funding and staff downsizing at many universities which has resulted in a more educated clientele looking for information in new ways. Where are we heading? There are numerous opportunities for both industry and academia which likely will lead to greater partnership opportunities driven by varying needs. From a university perspective funding needs will likely drive partnerships. From an industry perspective, many companies are starting to recognize the important and needed role universities play in educating undergraduates and graduate students who fill vital staffing industry needs. These

collaborations will also expand in areas of mutual research interests and how that research is transferred to clientele groups.

**Key Words:** technology transfer, academia, industry

## 57 Training of students: Real-world experience on farm, in extension, and industry.

Robert E. James\*, *Virginia Tech, Blacksburg, VA.*

Significant challenges exist in training students to enter employment in the animal industry. Farm numbers continue to shrink while farm size and complexity has increased. Excellent opportunities exist for graduates with proficiency in communication and problem solving and are able to integrate sophisticated technical knowledge to practical problems on the farm. The challenges are many. Reductions in faculty, particularly in extension and more applied research and teaching positions, are common in our universities. Students are increasingly from urban and suburban backgrounds with little previous agriculture experience but considerable interest in food production. Declining state and federal support for undergraduate education and more applied graduate research are additional challenges. However, many new opportunities are available to provide needed training experiences. Summer internships are common for students at dairy farms and in industry positions in sales, market research and technical support. The North American Intercollegiate Dairy Challenge Contest and Academy has been successful in fostering better communication of students with dairy industry partners. Over 4,500 students have participated since its inception in 2002. Employment on campus dairy farms provides opportunities for non-traditional students to obtain farm experience. On farm research at the university and on commercial dairies expands student perspectives. Training graduate students presents similar challenges. In the past, formula funding permitted research to address timely research needs of our animal industries. However, these funds have been replaced with competitive grants directed toward specific narrow problems. Students begin their graduate studies with less input into research project design and have less time for exposure to our farms. Industry sponsored management research enables students to conduct on farm research on commercial farms. Farm sponsored check off programs such as the Virginia Set-aside program direct funds toward more applied research.

**Key Words:** students, extension, industry

## 58 Retaining qualified extension faculty: An administrator's perspective.

Gregory P. Lardy\*, *North Dakota State University, Fargo, ND.*

Retaining qualified faculty for extension positions is an important factor in the success of animal, dairy, and poultry science departments. To be successful in an extension appointment requires a unique combination of skills. I believe the following skills are critical: (1) sound technical training, (2) excellent written and oral communication skills, (3) a thorough understanding of the industry one is hired to serve and a desire to serve that industry, (4) excellent time management skills, (5) ability to translate scientific knowledge to practical applications in a systems approach, and (6) the ability to develop and maintain relationships with clientele. Candidates with these skills have an improved probability of being successful. Retaining highly qualified faculty requires support

from multiple levels of the institution. Factors affecting retention include (1) a positive departmental culture, (2) a competitive salary and benefits package, (3) clear job expectations and provision of resources required to effectively carry out the job duties including an institutional culture that supports and values extension programming, (4) a stimulating and challenging work environment, and (5) a departmental, college, and institutional culture that helps the faculty member balance the demands of work and family life and fosters professional development. In my experience, having a positive departmental culture is the most important. Department heads and chairs have a very direct effect on departmental culture (both positive and negative). Administrators and promotion and tenure committees at the departmental and college level must place a high value on extension programming in order for faculty with extension appointments to feel valued. Promotion and tenure documents at the department, college, and institutional level should reflect the scholarship of extension programming. Recruitment and retention of extension faculty is a multi-faceted challenge and one that requires commitments at the departmental, college, and institutional level to be successful. In my experience, hiring candidates with the right combination of skills and developing a positive departmental culture are the most important means to improving success.

**Key Words:** extension, faculty, recruitment

**59 Development of mobile applications in extension.** R. Lawton Stewart, Jr.\*<sup>1</sup> and G. Cliff Lamb<sup>2</sup>, <sup>1</sup>*The University of Georgia, Athens, GA*, <sup>2</sup>*The University of Florida, Marianna, FL*.

Mobile computing devices such as smartphones and tablets are quickly becoming the principal means of communication in the United States. The adoption of smartphone and tablet technology among adults over the age of 18 has increased from 35 and 8%, respectively, in 2011 to 55 and 42%, respectively, in 2014. This growth is visible in the agricultural field as well. A search of currently available mobile computing applications (apps) for agriculture revealed 42, 28, 19, and 11 apps are available for crops, livestock, spraying, and pests, respectively. Given that the role of extension is to extend lifelong learning opportunities through unbiased, research-based education, the utilization of apps provides an excellent opportunity to serve clientele. This technology can improve the power to disseminate information, make informed science-based decisions, and collect data. However, when considering the use of this technology for an Extension program, several aspects of implementing this technology should be addressed. Most attempts at Extension-based apps are based on currently available informative publications for dissemination of information, or spreadsheet-based decision aids for decision-aid tools. This development is initiated by the need of information in the field, however, the challenge often becomes getting information transferred to an app-accessible format. The most common form is an app available through application distribution platforms specific to a smartphone mobile operating system (e.g., Apple iTunes and Google Play). These require app-specific programming and approval through the distributor; however, once downloaded to a mobile device,

are accessible independent of a data connection. Another avenue is to develop a mobile-friendly website. This makes the information or decision aid available on any operating platform, however requires a data connection by the device. Finally, the impact of this technology should be quantifiable to allow extension personnel to measure the effect of their program. This can be performed through download statistics for mobile apps, or through website analytics for mobile-friendly websites. With the increased availability of information through mobile devices, mobile device applications are an excellent tool for extension personnel.

**Key Words:** mobile app, extension, decision-aid tools

**60 Social media to deliver extension.** Alison Van Eenennaam\*, *University of California, Davis, CA*.

The use of social media in public engagement and extension is a relatively new phenomenon, and many scientists are cautious about using it professionally. A recent survey of 4,000 researchers carried out by the Pew Research Center found less than half of the researchers ever used social media, 24% use it to blog about science, and only 12% said they currently use it to follow experts in their field. Depending upon the target audience, social media may or may not be a useful tool to deliver extension information. Social media gives opportunities to reach a much wider public audience than traditional extension meetings, but it may not always be the best approach to reach more traditional agricultural clientele who typically are an older demographic. One issue with the professional use of social media is the difficulty of objectively documenting impact. Although the number of followers or page views is an easy metric to report, it does not really evaluate impact. Merit and promotion evaluation systems will need to evolve to reflect the increased use of social media in extension programs, and appropriately reward academics for time spent effectively using these forms of communication. The UC Davis Animal Biotechnology and Genomics Extension program incorporates the use of social media including websites, YouTube educational videos, and the use of Twitter. For extension work in controversial areas, educators need to be prepared for the unpredictable nature of social media audiences. Several high-profile “public shamings” following an ill-advised tweet provide cautionary case studies of one of the risks of using social media, especially when discussing controversial topics. Although Twitter is useful for directing interested followers to a website or alerting them to an upcoming meeting, it is difficult to explain the nuances of complex topics in 140 characters, and maintaining a civil and respectful dialog can sometimes be challenging. Extension educators can certainly benefit from the expansive reach of social media and magnify the reach of their programs, and the next generation of educators will likely increasingly do so in the future. As with all extension communication, great care should be given to wording to ensure the professional dissemination of science-based information using social media.

**Key Words:** social media, Twitter, YouTube



# Forages and Pastures: Forages for livestock systems

**61 Forage-finished steer performance and carcass characteristics from grazing high-energy forages during the finishing period.** Rachel M. Martin<sup>\*1</sup>, Jason E. Rowntree<sup>1</sup>, Kim A. Cassida<sup>1</sup>, Joseph Paling<sup>1</sup>, and Douglas Carmichael<sup>2</sup>, <sup>1</sup>Michigan State University, East Lansing, MI, <sup>2</sup>Michigan State University AgBio Lake City Research Center, Lake City, MI.

The research objective was to compare high-energy forage options during the finishing period for Upper Midwestern forage-finished beef production systems. Twelve 0.80-ha pastures were randomly assigned to 1 of 3 forage treatments including: mixed pasture (MIX); simple cereal grain/brassica mixture (SIMP); and complex cereal grain/brassica mixture (COMP). Red Angus-influenced steers (BW = 439 kg ± 15.6, n = 24) were stratified by BW and randomly assigned to 1 of 12 paddocks and were grazed for a 64 d finishing period. Steers had *ad libitum* access to water and free choice mineral, and were given access to strips in each grazing treatment. Fasted BW was measured on d 0, 34, and 64. At the end of the finishing period, steers were slaughtered under Federal Inspection and carcass data were collected 48 h post-mortem. Data were analyzed using Proc Mixed (SAS v 9.4) where paddock was the experimental unit. There was a treatment by period interaction for d 64 BW ( $P < 0.01$ ) where steers on MIX and COMP had greater BW (517 kg ± 8.1 and 514 kg ± 8.1, respectively) than SIMP (490 kg ± 8.1). Steers in MIX and COMP had similar d 0 to 64 BW gains (78 ± 4.3 kg, and 74 ± 4.3 kg), however steers in COMP had greater ( $P < 0.01$ ) d 34 to 64 BW gains than MIX and SIMP (48 ± 1.6 kg versus 34 ± 1.6 kg and 31 ± 1.6 kg, respectively). Although d 64 BW were similar for COMP and MIX, there was a decided advantage ( $P < 0.01$ ) for steers in COMP for HCW and dressing percent (295 kg ± 5.6 and 57.29% ± 0.55). Although there were no differences for LM area, marbling score or USDA yield grade, carcasses from steers in COMP had numerically greater means (69.7 ± 2.8 cm<sup>2</sup>, 483 ± 16.9, and 2.5 ± 0.2, respectively) for each carcass trait when compared with MIX and SIMP. These data indicate that steers grazing MIX and COMP had reasonable gains and carcass merit and these systems can be a viable component of forage-finishing systems in the Upper Midwest.

**Key Words:** beef, forage-finished, grass-finished

**62 Using weekly pasture growth and utilization measurements to make management decisions on Oregon dairies.** Troy W. Downing<sup>\*</sup>, Oregon State University, Corvallis, OR.

The use of weekly pasture cover measurements have been shown useful in New Zealand dairies to estimate daily growth rates, determine grazing rotation, calculate feed inventories and they have been used for feed budgeting. The objectives of this study were to take weekly pasture growth measurements to estimate growth and utilization and to learn how to use this information to make management decisions on Oregon dairies. Additionally, all conserved feed fed was recorded daily and milk solids shipped were recorded and these data were used to estimate the percentage of milk produced from pasture. This study was done on one dairy for a 3-year period. Pastures were measured and mapped and total standing DM was estimated weekly in 22 pastures using a calibrated rising plate meter. Paddocks grazed and residual pasture covers were recorded daily and forage cover measurements were entered into management software. Paddock grazing and residual heights were also included in the electronic recordkeeping. Measurements started in March and continued until the end of November for all 3 years. Weekly grazing wedges were developed and were used to make grazing decisions that

week. Estimated dry matter yields for all paddocks each year averaged 18695 ± 3451, 17848 ± 3966 and 17306 ± 5058 kg/ha for each consecutive year. Daily pasture DM growth averaged 47, 49 and 56 kg/ha per day for each of the growing seasons. During year 1 the dairy averaged 1219 kg of milk solids per hectare from grass, 888 kg milk solids for year 2 and 962 kg of milk solids per hectare in year 3. Pasture accounted for 85, 72, and 80% of the total DM consumed on the dairy during the 9 mo grazing season for year 1, 2, and 3, respectively.

**Key Words:** rotational grazing, rising plate meter, pasture management

**63 Evaluation of sound-based detection of rumination in grazing dairy cows.** Santiago A. Utsumi<sup>\*1</sup>, Julio R. Galli<sup>2</sup>, Diego Milone<sup>3,4</sup>, Hugo L. Rufiner<sup>3,4</sup>, and Leonardo Giovanini<sup>3,4</sup>, <sup>1</sup>Michigan State University, East Lansing, MI, <sup>2</sup>Universidad Nacional de Rosario, Santa Fe, Argentina, <sup>3</sup>Universidad Nacional del Litoral, Entre Rios, Argentina, <sup>4</sup>CONICET, Argentina.

Individualized monitoring of rumination time (RT) could be used to guide nutritional recommendations for improved production, health and well-being of dairy cows. One commercial application is the QWES-HR Tag system (HR Tags), which uses a microphone and microprocessor in neck collars for sound-based detection of RT; filtered sounds in 12 × 2 h intervals are automatically integrated for estimation of 24 h moving RT, but inaccurate detection or processing of sounds could significantly affect estimations. A study was conducted to compare the automatic detection of RT by HR Tags against observed values obtained by supervised inspection of 24-h soundtracks. The RT of 5 lactating Holstein cows on ryegrass/white clover or orchardgrass/white clover pastures was monitored for 6 non-consecutive days, using 2 sound-based methods (1) HR Tags and, (2) Acoustic Halters, which included a digital recorder and one directional microphone pressed to the animal's forehead (Control). Five halters were used with halters rotated across cows according to a completely randomized design. Cross-validation of HR Tags (24-h and 2-h periods) was conducted by inspection of the root of the mean squared prediction error (RMSPE) and concordance correlation coefficient (CCC) between methods. The HR Tags consistently underestimated RT by 18%, regardless of the period of measurement (Table 1). Similarly, high RMSPE and low CCC indicated limited accuracy and low precision by HR Tags, which were slightly improved for 24 h evaluations (Table 1). Further research is needed to improve the automatic sound-based detection of rumination by commercial HR Tags.

**Table 1 (Abstr. 63).** Estimation of 24-h and 2-h rumination time by 2 acoustic methods

Rumination time/ measurement period		Control	HR Tags
24 hour (n = 29)	Mean (min)	534	437
	SE (min)	19.2	25.4
	RMSPE (min)		90.7
	CCC (%)		36.6
2 hour (n = 349)	Mean (min)	44.3	36.3
	SE (min)	1.7	1.2
	RMSPE (min)		30.6
	CCC (%)		23.8

**Key Words:** acoustics, grazing behavior, rumination

#### 64 Evaluation of forage quality of five grains for use in sprouted fodder production systems for organic dairy cattle.

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<sup>1</sup>University of Minnesota, West Central Research and Outreach Center, Morris, MN, <sup>2</sup>University of Minnesota Extension, Rochester, MN, <sup>3</sup>University of Minnesota, Southern Research and Outreach Center, Waseca, MN.

The objective of the study was to evaluate 5 grains for use in sprouted fodder production systems at the University of Minnesota's West Central Research and Outreach Center, Morris, MN. Forage mass, mold score, dry matter, and forage quality were evaluated for varieties of sprouted organic barley, oats, wheat, rye, and triticale harvested at 7 d after the start of sprouting. During September 2014, on every Monday for 6 weeks, 28 fodder trays (0.6 m x 1.8 m) from a FarmTek Fodder Pro system were filled with 4.1 kg of pre-soaked grain, which was soaked for 24 h. Each tray was automatically watered 3 times a day for 4 min each time. On the seventh day, each tray was harvested, weighed, and visually scored on a 1 to 5 scale for mold by one observer. Ten random samples from each sprouted grain each week were saved for dry matter and forage quality analysis. Sprouted forage samples were sent to Rock River Laboratory, Inc., Watertown, Wisconsin, and were analyzed by wet chemistry for DM, CP, NDF, and TDN. Data were analyzed using the MIXED procedure of SAS. Independent variables for analyses were the fixed effects sprouted grain, and date of harvest and replicate were random effects. Sprouted barley (9.3 kg), oats (9.0 kg), and wheat (8.8 kg) had greater ( $P < 0.05$ ) forage mass per tray than sprouted rye (7.8 kg) and triticale (6.3 kg). Mold scores were lower ( $P < 0.05$ ) for sprouted barley (0.04) and oats (0.03) compared with sprouted rye (2.8) and triticale (4.8). Sprouted barley DM (15.4%) was lower ( $P < 0.05$ ) than sprouted oat (19.1%), rye (19.8%), triticale (24.2%), and wheat (18.9%) DM. Concentrations of CP averaged 15.6%, 13.1%, 12.8%, 17.0%, and 17.9% for sprouted barley, oats, rye, triticale, and wheat, respectively, and they were different ( $P < 0.05$ ) from each other. The NDF was greater ( $P < 0.05$ ) for sprouted barley and oats (34.4% and 44.8%, respectively) compared with sprouted rye, triticale, and wheat (23.6%, 20.4%, and 26.7%, respectively). Sprouted triticale (79.7%) had higher TDN than sprouted oats (71.8%), which was the lowest for TDN. In summary, the results show that sprouted barley has the highest forage quality for fodder production systems.

**Key Words:** sprouted fodder, NDF, forage quality

#### 65 Protein quality of grass silage as affected by silage additives and its effects on dairy cow performance.

Elisabet Nadeau<sup>\*1</sup>, Björn Johansson<sup>2</sup>, Wolfram Richardt<sup>3</sup>, and Michael Murphy<sup>2</sup>, <sup>1</sup>Swedish University of Agricultural Sciences, Skara, Sweden, <sup>2</sup>Lantmännen Feed Division, Malmö, Sweden, <sup>3</sup>LKS mbH, Lichtenwalde, Germany.

This experiment was conducted to evaluate the effects of improved protein quality of grass silage by additive use on dairy cow intake and performance. Grass from the first regrowth was wilted to 33% DM before being precision chopped and treated with the inoculant Kofasil Duo (*Lactobacillus plantarum*/*Lactobacillus buchneri*, 200,000 cfu/g) or with the chemical additive Kofasil Ultra K (sodium nitrite, hexamine, potassium sorbate, sodium benzoate and sodium propionate, 2 L/T, Addcon Europe GmbH), which were compared with a control without additive. The forage was ensiled in hard-pressed round bales before being fed to 48 dairy cows (150 DIM) in a continuous trial for 9 wk using 8 cows per treatment. Cows were blocked according to breed, parity, DIM and milk yield. The silage contained 15% CP, 47% NDF, 3.3% WSC, 8.2% lactic acid, 2.1% acetic acid and 0.25% NH<sub>3</sub>-N of DM with minor differences between treatments. Rumen undegradable

protein (RUP) of the silage at 5% passage rate was 22, 23 and 24% of CP for control (C), inoculant (I) and salt-based (S) additive, respectively. Diets were isonitrogenous (15.1% of DM) and isoenergetic (11.4 MJ/kg DM) varying in RUP (4.7% (high) and 2.5% (low) of DM). Concentrate UDP was 2.9 and 8.6% of DM. Dietary forage proportion of the TMR was 58% of DM. Data were analyzed using PROC MIXED of SAS with RUP, silage and week as fixed factors and block as a random factor. High RUP diet had higher milk yield than low RUP diet (29.4 vs. 27.9 kg;  $P < 0.05$ ). The DM intake was not affected by RUP and silage treatment. Yields of milk and ECM were higher for the I and S diets than for the C diet at low RUP (28.9 vs. 26.0 kg milk,  $P < 0.01$ ; 30.6 vs. 27.1 kg ECM,  $P < 0.001$ ) whereas there was no effect of additive treatment in the high RUP diet. Milk fat and protein did not differ between silage treatments. Feed efficiency was higher for the I and S diets than for the C diet at the low RUP (1.6 vs. 1.3 kg of ECM/kg DM intake,  $P < 0.001$ ) but not at the high RUP. The increased milk yield and feed efficiency when fed a diet with low RUP can partly be explained by increased RUP of the I and S silages.

**Key Words:** additive, protein quality, silage

#### 66 Effects of type and level of energy supplementation on stocker cattle performance from annual ryegrass.

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Supplementation of high-quality grazed forage with high-energy feedstuffs can improve animal performance and enable increased stocking rates. However, the extent of performance improvement and direction of forage utilization response may be dependent upon supplementation level and whether the supplement is a high-starch or highly-digestible-fiber feedstuff. For these reasons, a grazing experiment was conducted to determine the type and level of supplementation with select high-energy feedstuffs that yield optimum animal performance and forage utilization from annual ryegrass (*Lolium multiflorum*). Twenty 0.81-ha pastures were each grazed by 4 crossbred steers (235 kg  $\pm$  15 kg initial BW) between Feb. 6 and May 15, 2014. Cracked corn (CC), pelleted citrus pulp (CP) or pelleted soybean hulls (SH) were fed at rates of 0.25, 0.50 and 0.75% of steer BW daily (2 rate  $\times$  supplement replicates per treatment, including replicate pastures in which steers received no supplement). Steers were weighed every 28 d following an overnight shrink. Forage mass was measured every 28 d using the destructive harvest/disk meter dual-sampling method. Data were analyzed as a completely randomized design by the PROC GLM procedure of SAS 9.2 using pasture as the experimental unit. There were no supplement type  $\times$  level interactions ( $P > 0.05$ ) observed for either ADG or net forage mass change (FMC). Steers receiving CC and SH supplementation had greater ( $P < 0.05$ ) ADG (1.30 and 1.23 kg/d, respectively), and steers receiving CP tended ( $P = 0.07$ ) to have greater ADG (1.20 kg/d) than steers receiving no supplement (0.95 kg/d). Steers supplemented at 0.25 and 0.75% BW had greater ( $P < 0.05$ ) ADG (1.23 and 1.30 kg, respectively) than steers receiving no supplement (0.95 kg/d). Net FMC over the entire grazing season increased ( $P < 0.05$ ) in pastures grazed by steers receiving CC (+38 kg DM/ha) compared with those grazed by steers receiving SH (-277 kg/ha). There were no differences in net FMC between any of the supplementation types or levels and the unsupplemented control. Patterns of net FMC response indicate relative underutilization of forage by steers receiving CC (i.e., substitution effect of supplementation) compared with steers receiving SH or CP.

**Key Words:** beef cattle, ryegrass, supplementation

**67 Performance of *Bos indicus* versus *Bos taurus* stocker cattle grazing 'Coastal' bermudagrass supplemented with distillers dried grains.** W. Brandon Smith<sup>\*1</sup>, F. M. "Monte" Rouquette<sup>1</sup>, Joel L. Kerby<sup>1</sup>, Luis O. Tedeschi<sup>2</sup>, Jamie L. Foster<sup>3</sup>, Jason P. Banta<sup>4</sup>, Kimberly C. McCuiston<sup>5</sup>, and Tanner J. Machado<sup>5</sup>, <sup>1</sup>Texas A&M AgriLife Research, Overton, TX, <sup>2</sup>Texas A&M University, College Station, TX, <sup>3</sup>Texas A&M AgriLife Research, Beeville, TX, <sup>4</sup>Texas A&M AgriLife Extension, Overton, TX, <sup>5</sup>Texas A&M University-Kingsville, Kingsville, TX.

The objective of this study was to evaluate performance of "long yearling" stocker steers grazing 'Coastal' bermudagrass (*Cynodon dactylon* [L.] Pers.) and supplemented daily with titrated levels of DDG. Steers (n = 63 [7 testers per pasture; 4 Brahman, 3 other], 352 ± 7.6 kg initial BW, approx. Fifteen mo of age) were stratified by BW within source (Overton or McGregor) and known sire breed type (Angus or Charolais, both with Brahman crossbred dams; or purebred Brahman) and allocated randomly to 1 of 9 paddocks (1.3 ± 0.17 ha), and paddocks were allocated randomly to 1 of 3 levels of DDG supplementation for the 96-d study: 0.00, 0.25 or 1.00% BW hd<sup>-1</sup> d<sup>-1</sup>. Animals were group-fed daily at 0800 h, and bunk space was allowed at a minimum of 61 cm hd<sup>-1</sup>. Forage mass was assessed by destructive harvest of four 0.09-m<sup>2</sup> quadrats throughout each pasture on 21-d intervals. Steers were weighed every 21 d, and grazers were added to each pasture based on visual and numerical assessments to maintain vegetative growth while preserving ad libitum intake and selection. Data were analyzed using SAS PROC MIXED. Average daily gain was greater ( $P < 0.05$ ) from steers offered 1.00% BW daily (0.97 kg d<sup>-1</sup>) compared with those offered 0.00 or 0.25% BW daily (0.71 and 0.69 kg d<sup>-1</sup>, respectively), and *Bos indicus* steers gained more (0.93 kg d<sup>-1</sup>;  $P < 0.05$ ) than *B. taurus* steers (0.72 kg d<sup>-1</sup>). Supplement to gain ratio was not different among treatments ( $P = 0.29$ ) or species ( $P = 0.76$ ). Pastures in which supplement was offered at 1.00% BW tended to sustain more ( $P = 0.06$ ) AU (363-kg steer) per ha (8.8 AU ha<sup>-1</sup>) compared with 0.00% pastures (6.3 AU ha<sup>-1</sup>), with 0.25% pastures intermediate (8.2 AU ha<sup>-1</sup>). Additionally, pastures supplemented at 1.00% BW supported more ( $P < 0.05$ ) gain per unit area (851 kg ha<sup>-1</sup>) than pastures supplemented at 0.00 or 0.25% (431 and 575 kg ha<sup>-1</sup>, respectively). Thus, supplementation of stocker calves with DDG at the 1% level, especially *B. indicus*, may be a viable option for increased performance from stocker cattle, as well as increased stocking density, on bermudagrass pastures, depending on the economic factors of the current market.

**Key Words:** Coastal bermudagrass, DDG, stocker

**68 Interaction between a tannin-containing legume and endophyte-infected tall fescue seed on lamb feeding behavior and physiology.** Juan J. Villalba<sup>\*1</sup>, Casey Spackman<sup>1</sup>, Ben Goff<sup>2</sup>, James L. Klotz<sup>3</sup>, and Jennifer W. MacAdam<sup>1</sup>, <sup>1</sup>Utah State University, Logan, UT, <sup>2</sup>University of Kentucky, Lexington, KY, <sup>3</sup>USDA-ARS, Lexington, KY.

It was hypothesized that a tannin-rich legume like sainfoin reduces the negative postingestive effects of ergot alkaloids in tall fescue. Thirty-two 3-mo-old lambs were individually penned and randomly assigned to a 2 × 2 factorial experimental design with 2 legume species (sainfoin [SF; tannins] or cicer milkvetch [CIC; no tannins]) and a ration (seed:beet pulp:alfalfa, 50:30:20) containing 2 types of tall fescue seed (endophyte-infected [E+; 1,745 ppb ergovaline] or endophyte-free [E-]). For a 10-d baseline period, all groups were fed their respective supplemental legumes (17 to 25 g DM/kg BW) and ad libitum amounts of E-. In an ensuing 10-d testing period, the protocol was the same but

half of the lambs received E+ instead of E-. Daily feed intake and rectal temperatures were measured for both periods and jugular blood was extracted at the beginning and end of the testing period. Subsequently, all lambs had choices between their respective legume (SF; CIC) and seed-containing rations (E+; E-). Response variables were analyzed using a mixed effects model, which included legume, ration and day as fixed effects and lamb as a random factor. All groups consumed similar amounts of E- during baseline ( $P > 0.10$ ). However, lambs ate more E- than E+ during testing ( $P < 0.05$ ) and lambs offered SF ate more E+ than lambs offered CIC ( $P < 0.05$ ). Groups fed E- during baseline and testing periods had similar rectal temperatures ( $P > 0.10$ ). However, lambs fed E+ had lower rectal temperatures when supplemented with SF than with CIC ( $P < 0.05$ ). Lambs fed E+ had a greater concentration of hemoglobin and number of red blood cells than lambs fed E- ( $P < 0.05$ ). Plasmatic concentrations of cortisol and prolactin did not differ between treatments ( $P > 0.10$ ). All lambs preferred their respective rations over their respective legumes and they preferred E- over E+ ( $P < 0.05$ ). In summary, SF increased intake of E+ and reduced rectal temperatures relative to CIC supplementation. However, SF did not lead to changes in preference for E+ or improvements in other physiological parameters assessed.

**Key Words:** ergot alkaloids, ingestive behavior, prolactin

**69 Effect of a combination of lactic acid producing bacteria and fibrolytic enzymes on the ensiling characteristics of cool season grasses: A farm-scale application.** Jeffrey M. Chilson<sup>\*</sup>, Chia-Yu Tsai, Kirk C. Ramsey, Richard Scuderi, and Pedram Reza-mand, University of Idaho, Moscow, ID.

In prior research, using a combination inoculant containing homofermentative lactic acid bacteria and fibrolytic enzymes we showed an improvement in rumen DM degradation, lactic acid production and rate of pH decline, for alfalfa using small scale setting. The objective of the present study was to test the effectiveness of the inoculant on the quality of ensiled mixed cool season grasses (orchard, oat, mountain brome and Italian rye grasses) in a farm scale setting. Grasses were harvested and the windrows randomly assigned to either the control (Con) or inoculant (Trt). The Con was treated with water only and Trt was treated with an equal amount of water plus the inoculant. Control was baled, and wrapped first, followed by Trt. On d 86, 88, 90, 92, 94, 3 bales from each group were removed, weighed, core samples taken and composite samples prepared for analysis. Composite samples were analyzed for pH, lactic acid, DM, and NDF. Statistical analysis was conducted using a T-Test, on SAS v. 9.4. Initial DM did differ between Con and Trt ( $P = 0.03$ ; 52.3 vs. 58.4 ± 1.9%) possibly related to temperature change (start 16°C, finish 32°C) during baling. Initial pH was not different between Con and Trt. Initial NDF was not different between groups ( $P = 0.15$ ; 60.02 vs. 58.29 ± 0.72%). Final pH was not different between Con and Trt, either ( $P = 0.45$ ; 5.24 vs. 5.36 ± 0.12). DM loss was numerically greater for Con, but not significant, either on kg/bale basis ( $P = 0.39$ ; 91.7 v. 85.9 ± 12.5kg) or as a percent/bale basis ( $P = 0.32$ ; 24.9 vs. 20.9 ± 3.0%). Lactic acid production was not different between groups ( $P = 0.14$ ; 40.89 vs. 75.24 ± 19.88 μmol/g DM). There was, however, a significant difference in final NDF between Con and Trt ( $P = 0.04$ , 55.25 vs. 57.84 ± 0.81%). Whereas Trt showed greater NDF, Con had numerically greater DM loss, possibly indicating that Con lost more soluble nutrients during ensiling. Furthermore, final pH for both Con and Trt were above desired pH of 4.00–4.20, which could indicate that fermentation was in part inhibited by the high initial DM.

**Key Words:** inoculant, silage, grass



# Graduate Student Competition: ADSA Dairy Foods

## Graduate Student Oral Competition

**70 Structural changes and texture development in milk protein concentrates induced by high hydrostatic pressure.** Lee Cadesky\*<sup>1</sup>, Markus W. Ribeiro<sup>1</sup>, Mukund V. Karwe<sup>2</sup>, and Carmen I. Moraru<sup>1</sup>, <sup>1</sup>*Cornell University, Ithaca, NY*, <sup>2</sup>*Rutgers University, New Brunswick, NJ*.

Milk protein concentrate (MPC) and micellar casein concentrate (MCC), obtained by membrane filtration, are increasingly available. This research focuses on utilizing high hydrostatic pressure (HHP) to induce structural modifications of milk proteins in MPC and MCC and enable the creation of new textures that may find application in milk protein based products. MPC and MCC powders were reconstituted by dispersing the powders in water to 2.5 and 10% casein (w/v), and allowed to fully hydrate. The concentrates were packaged in flexible pouches and subjected to 15 min HHP treatments at 150, 250, 350, and 450 MPa, at temperatures below 25°C, in a 10 L HHP unit. Pressure induced structural changes to casein micelles in HHP treated samples were assessed by (a) evaluating the renneting behavior of the protein concentrates using dynamic rheology; (b) mineral and proteomic profiling of ultracentrifugation supernatants. The study was replicated, and significant differences among treatments were assessed by statistical analyses. In all samples, HHP treatment increased calcium levels in the serum as compared with untreated controls ( $P < 0.05$ ). HHP treatments at 250 and 350 MPa resulted in levels of serum calcium over 3.5 times higher than in untreated samples. HHP above 250 MPa induced significant ( $P < 0.05$ ) concentration and pressure dependent changes in MCC and MPC. In both 10% MCC and 10% MPC, HHP above 350 MPa led to the formation of weak gel structures. HHP treatment also increased  $G'$  values of rennet gels and reduced coagulation times, which indicates a destabilization of casein micelles following pressure treatment. In 10% MCC, maximum gel strength was observed after 150 MPa treatment, with  $G'$  of 140 Pa as compared with 45 Pa in untreated samples. For 10% MPC, HHP treatment enabled rennet coagulation, which did not occur in untreated samples. MPC renneted gels were weak, with 450 MPa treatment resulting in the strongest gels ( $G'$  of 36 Pa). These observations suggest that HHP treatment of milk protein concentrates can be used for developing new types of dairy-based, protein rich foods with unique structure and texture.

**Key Words:** high-pressure processing, proteins, rheology

**71 Solubilization of rehydrated frozen highly concentrated micellar casein concentrate for use in liquid food applications.** Ying Lu\*<sup>1</sup>, Donald McMahon<sup>1</sup>, Lloyd Metzger<sup>2</sup>, Anil Kommineni<sup>2</sup>, and Almut Vollmer<sup>1</sup>, <sup>1</sup>*Western Dairy Center, Utah State University, Logan, UT*, <sup>2</sup>*Midwest Dairy Foods Research Center, South Dakota State University, Brookings, SD*.

Highly concentrated micellar casein concentrate (HC-MCC), a potential ingredient for protein fortification, containing ~20% casein with most whey proteins removed by microfiltration, diafiltration, and vacuum evaporation of skim milk. Our objective was to determine the conditions needed to obtain complete solubility of thawed frozen HC-MCC in water, and to understand its gelation upon cooling. Dispersibility (ability to pass through a 250- $\mu$ m mesh sieve), suspendability (percentage of protein not sedimented at 80  $\times$  g within 5 min), and solubility (percentage of protein not sedimented at 20,000  $\times$  g within 5 min) were measured at

various mixing conditions. Gelation upon cooling from 50°C to 5°C was monitored based on storage ( $G'$ ) and loss ( $G''$ ) modulus and gelled HC-MCC examined using transmission electron microscopy. Thawed HC-MCC was added to water (3% of protein) using high shear (7,500 rpm) for 1 min or low shear (800 rpm) for 30 min at 4, 12, 20, or 50°C and at pH 6.4 to 7.2. The HC-MCC completely dispersed at 50°C, or at  $\leq 20^\circ\text{C}$  followed by overnight hydration at 4°C. Suspendability at 50°C was ~90%, while mixing at  $\leq 20^\circ\text{C}$  followed by overnight hydration yielded only ~57%. Solubility followed a similar trend with ~83% at 50°C and only ~29% at  $\leq 20^\circ\text{C}$ . Mixing HC-MCC in 60 mM trisodium citrate increased dispersibility to 99%, and both suspendability and solubility to 81% at 20°C. Cold gelling temperature, defined as temperature at which  $G' = G''$ , was positively correlated ( $R^2 = 0.97$ ) with protein level in HC-MCC. Gelation occurred at 38, 28 and 7°C with 23, 20, and 17% of protein, respectively. Gelation was reversible upon heating. In micrographs of gelled HC-MCC, casein micelles were observed to be within the normal range and close packed together with only ~20 to 50 nm space between them. We propose that cold gelation of HC-MCC occurs when the kinetic energy of the casein micelles is sufficiently reduced to inhibit their mobility in relation to adjacent casein micelles. Understanding solubilization of rehydrated frozen HC-MCC and its rheological property can help designing process systems for using it as a potential ingredient in liquid food.

**Key Words:** casein micelle, microfiltration, solubility

**72 Development of a method for characterizing high-protein dairy powders with an ultrasonic flaw detector.** Mary Hauser\* and Jayendra Amamcharla, *Kansas State University, Manhattan, KS*.

When choosing a high-protein dairy powder such as milk protein concentrate (MPC), dissolution behavior is an important property to consider. Current methods for testing powder dissolution are time consuming, difficult to reproduce, and subjective. Ultrasound spectroscopy is a rapid and precise method, but expensive equipment and skilled technicians are needed. An economical alternative is to use an ultrasonic flaw detector (UFD). The objective of study was to develop a method to characterize the dissolution behavior of MPC with an UFD. The experimental setup included an UFD (Epoch LTC) in pulse-echo mode and a 1MHz immersion transducer that was kept a constant distance from the reflector plate. Powder concentration, stirring speed, UFD settings, and path length were optimized. To validate the method, 2 batches of MPC80 from a commercial manufacturer were procured and stored at 25°C and 40°C for 4 weeks. MPC was slowly added to water at a 5% concentration. During the dissolution, A-Scan data from UFD was acquired to calculate velocity and attenuation. Focus beam reflectance measurement (FBRM) and solubility index were used as reference methods. Velocity had a trend of fluctuating and then stabilizing with fluctuation time increasing with storage time. Due to the variation in fluctuation, the standard deviation from 900 to 1800s was set as a parameter. Day 0s standard deviation was 0.01 m/s and increased to 0.1 m/s after 4 wk at 40°C. During dissolution, attenuation increased and then gradually decreased. From the attenuation, 3 parameters were extracted. The area under the curve from 0 to 1800s showed the most changes. From Day 0 to wk 4 at 40°C, the area decreased from 100 to 110 Np·s/m to below 30 Np·s/m. After comparing all the data, it was observed that fresh powders dispersed quickly, had a low standard deviation and a larger area. As the MPC aged at 40°C, the

particle dispersion rate decreased and an increase in standard deviation and reduction in area were observed. Overall, the UFD can be a low cost method to characterize the dissolution behavior of dairy powders.

**Key Words:** ultrasonic flow detector, milk protein concentrate, solubility

**73 The effect of spray-drying parameters on the flavor of skim milk powder.** C. W. Park\*, M. A. Stout, and M. A. Drake, *South-east Dairy Foods Research Center, North Carolina State University, Raleigh, NC.*

Unit operations during production influence the sensory properties of skim milk powder (SMP). Off-flavors associated with SMP decrease consumer acceptance of ingredient applications. Previous work has shown that spray drying parameters such as inlet temperature and feed solids concentration affect physical and sensory properties of whole milk powder and whey protein concentrate. The objective of this study was to determine the effect of inlet temperature and feed solids concentration on the flavor of SMP. Pasteurized skim milk was evaporated to 50% total solids by a falling film evaporator. The condensed milk was then spray dried at either 160, 210, or 260°C inlet temperature and either 30, 40, or 50% total solids in a randomized order. Warm deionized water was added to achieve 30 and 40% solids condensed milk. The entire experiment was replicated 3 times. Flavor of the SMP was evaluated by sensory and instrumental analyses, which included descriptive analysis and solvent extraction with solvent-assisted flavor evaporation followed by gas chromatography mass spectrometry. Surface free fat and particle size were also analyzed. Both main effects (30, 40, 50% solids and 160, 210, and 260°C inlet temperature) and interactions between solids concentration and inlet temperature were investigated. Interaction effects were not significant ( $P > 0.05$ ). Decreasing inlet temperature decreased overall aroma, sweet aromatic and cooked flavors, and 2-acetyl-1-pyrroline and furaneol concentrations and increased cardboard flavor and hexanal, heptanal, nonanal, furfuryl alcohol, and 2,4-decadienal concentrations ( $P < 0.05$ ). Increasing solids concentration increased sweet aromatic flavor and 2-acetyl-1-pyrroline concentration, while 30% solids concentration increased cardboard flavor and hexanal, nonanal, DMTS, and furfuryl alcohol concentrations compared with 40 or 50% solids ( $P < 0.05$ ). Particle size increased and surface free fat decreased with increasing inlet temperature and solids concentration ( $P < 0.05$ ). These results demonstrate that increasing inlet temperatures and solids concentration during spray drying decrease off-flavor intensities in SMP.

**Key Words:** skim milk powder, spray drying, flavor

**74 Using membrane filtration to fractionate acid whey into value-added ingredients.** Bang Chen\*<sup>1</sup>, Karen E. Smith<sup>2</sup>, John A. Lucey<sup>2,1</sup>, Rebecca Kalscheuer<sup>2</sup>, and Michael Molitor<sup>2</sup>, <sup>1</sup>*University of Wisconsin-Madison, Madison, WI*, <sup>2</sup>*The Wisconsin Center for Dairy Research, Madison, WI.*

There has been a huge expansion in acid whey production due to the rapid growth in Greek yogurt manufacture, therefore it is critical to find an economically feasible way to process acid whey. The objective of this study was to determine the suitability of novel experimental nanofiltration (NF) membranes to reduce the calcium content of acid whey, so that value added streams (e.g., lactose) could be produced from this whey. Approximately 1,000 L of acid whey was obtained from a local Greek yogurt manufacturer for each trial. A 10,000 Da UF membrane was used to produce a permeate stream (i.e., remove any residual proteins). Acid whey was then processed by one of 4 different NF membranes. Three

novel NF membranes were evaluated for divalent ion permeation and compared with a control NF membrane. Permeates were concentrated to 1× and 2×, and were sampled at processing pressures of 1380, 2760 and 4140 kPa, and at operating temperatures of 4, 21, 43 and 54°C. Flux also was recorded. Total solids, lactose, galactose, lactic acid and calcium contents of the NF permeates/ retentates were determined. The calcium/lactose ratio in the permeate was determined. We wanted a high ratio of calcium to lactose in the permeate so that we could successfully reduce the ash content of acid whey while retaining lactose. Higher temperatures and higher pressures yielded higher rates of component permeation for each type of membrane. The novel NF membranes had higher permeation of all components, compared with the control NF membrane. Lactose and calcium permeated the novel NF membranes to differing degrees, in contrast to the control NF membrane. Permeates were further concentrated (6×) by 1 of 2 novel membranes, or a control membrane. The composition of final concentrates and samples taken during the process of concentration were analyzed. The results indicated that the novel membranes had good retention of lactose while calcium permeation increased as the permeate concentration increased. Lactose hydrolysis tests were performed on NF concentrates of acid whey by utilizing a commercially available lactase enzyme with an acidic pH optimum. The end product of this hydrolysis was a dairy syrup with enhanced sweetness.

**Key Words:** acid whey, membrane processing, nanofiltration

**75 Hydrodynamic cavitation as a tool to improve texture, mouthfeel, and creaminess in formulated and high-protein, low-fat Greek yogurts.** Gopinathan H. Meletharayil\* and Hasmukh A. Patel, *South Dakota State University, Brookings, SD.*

Greek yogurt (GY) manufacturing involves a straining step that generates acid whey, which has led to processing and environmental concerns. Non-strained yogurt (GSY) can be the solution to this problem. However, GSY is unacceptable to consumers because of its tart astringent taste, gritty and chalky texture and lack of creamy mouthfeel, taste, and flavor. We explored the use of hydrodynamic cavitation in conjunction with CO<sub>2</sub> treated functional milk proteins to develop a low fat GSY having rheological and organoleptic profiles similar to GY. CO<sub>2</sub> treated milk retentate was added to skim milk and NFDM base to yield 9% (w/w) protein and 17% (w/w) total solids. The mixture was heated to 90°C for 10 min followed by cooling to 42°C. The milk was fermented with yogurt culture to pH 4.6, followed by rapid cooling to less than 10°C. The yogurt was then subjected to hydrodynamic cavitation using an APV cavitator (SPX Flow Technology, Denmark). Rheology, large deformation studies, % lactic acid (LA) organic acid profiles using HPLC, graininess using microscopy and organoleptic profile (mouthfeel and creaminess) using sensory panel were investigated. Experimental data were tested for ANOVA and statistical significance ( $P < 0.05$ ) was determined, using statistical software SAS. Hydrodynamic cavitation reduced the consistency coefficient of GSY to values similar to those obtained in commercial GY. Cavitation of GSY reduced the number of grains from 2389 to 35 grains/g compared with 293 grains/g in commercial GY. There was no significant difference in the LA content between the cavitated GSY and commercial GY. Organic acid profiles of the strained and non-strained cavitated yogurts were superimposable. GSY subjected to hydrodynamic cavitation had better mouthfeel and creaminess compared with commercial GY. Based on these results, it can be concluded that the combination of CO<sub>2</sub> treated proteins and hydrodynamic cavitation can be used as a promising solution to manufacture GSY with rheological and organoleptic profiles better or similar to GY. This processing innovation will help to reduce processing times,

capital investments and more importantly the vexing problem of acid whey disposal.

**Key Words:** hydrodynamic cavitation, Greek yogurt, texture

**76 Engineering of infant formula emulsions to enhance protein thermal stability through Maillard conjugation.** Kamil P. Drapala\*, Daniel M. Mulvihill, and James A. O'Mahony, *School of Food and Nutritional Sciences, University College Cork, Cork, Ireland.*

Comfort-type infant formula (IF) emulsions are frequently manufactured using whey protein hydrolysate (WPH) ingredients; however, stability of these emulsions to heating is often poor. The objective of this study was to improve heat stability of such emulsions by conjugation of WPH with maltodextrin (MD) through wet heating. Model IF emulsions (1.55% protein, 3.50% oil, 7.00% carbohydrate) were prepared using whey protein isolate (WPI), WPH, heated WPH or WPH-MD conjugate. The conjugate was prepared by heating a WPH/MD solution (5.00% protein and 5.00% maltodextrin, pH 8.2) at 90°C for 8 h; heated WPH was prepared in a similar manner but in the absence of MD. Emulsions were heated at 75°C or 95°C for 15 min using a rheometer, with viscosity data recorded throughout the heat treatments, or at 100°C for 15 min using an oil bath. Emulsions were recovered after all heat treatments and changes in viscosity, fat globule size distribution (FGSD) and microstructure, determined using confocal laser scanning microscopy (CLSM), were used to monitor the effects of heating on the structure/stability of the emulsions. Emulsions with similar, monomodal size distributions (mean oil droplet diameter  $\leq 1.0 \mu\text{m}$ ) were formed with all protein ingredients (no significant differences,  $P < 0.05$ ). Heat stability of emulsions increased in the order WPH < WPI  $\ll$  heated WPH  $\ll\ll$  WPH-MD conjugate. A sharp increase in viscosity during heating at 75°C (WPH) or 95°C (WPI) and significantly higher viscosity ( $P < 0.05$ ) post-heat treatment (as compared with initial viscosity) indicated structural rearrangement/coagulation in the WPH and WPI stabilized emulsions; no changes in viscosity on heating were observed for emulsions stabilized with heated WPH or WPH-MD conjugate. After heat treatment at 100°C, flocculation and coalescence of oil droplets in emulsions stabilized by heated WPH were mediated by protein aggregation (as evidenced by CLSM analysis) while no changes in FGSD or microstructure were observed in emulsions stabilized by WPH-MD conjugate. Modification of WPH through conjugation with MD yielded a protein ingredient with superior thermal stability in oil-in-water IF emulsions.

**Key Words:** protein-carbohydrate conjugation, emulsion thermal stability, infant formula

**77 Improvement of the physicochemical and functional properties of whey protein hydrolysates by conjugation.** Eve M. Mulcahy\*, Daniel M. Mulvihill, and James A. O'Mahony, *School of Food and Nutritional Sciences, University College Cork, Cork, Ireland.*

Proteins can be modified by enzymatic hydrolysis to alter their functionality. The objective of this study was to determine the effect of conjugation on selected physicochemical functionalities (i.e., solubility and solution clarity/heat stability) of either intact whey protein isolate (WPI) or hydrolysed whey protein isolate (WPH). Conjugation of WPI or WPH (degree of hydrolysis of 8%) with maltodextrin (dextrose equivalent value of 6; MD6) was achieved by heating solutions of 5% WPI or WPH and 5% MD6, initial pH of 8.2, at 90°C for up to 24 h. Samples were taken after 3, 5, 8 and 24 h of heating; the greatest reduction ( $P < 0.05$ ) in available amino groups (21.0% reduction in AAG, measured

by the o-phthalaldehyde assay) occurred during the first 8 h of heating for WPH-MD6, with a considerably smaller decrease in AAG between 8 and 24 h of heating, which was consistent with the limited (0.8%) reduction of AAG in WPI-MD6 after 8 h. The number of amino groups available to react with the carbonyl groups of MD6 were 55% higher in WPH than in WPI which contributed to a greater extent of conjugation with the former. Unheated WPI had a protein solubility of 87.3% at pH 4.5; WPI-MD6 conjugate solutions (8 h of heating) had a protein solubility of 40.0% at pH 4.5 while WPH-MD6 conjugate solutions (8 h of heating) had enhanced protein solubility of 78.6% at pH 4.5. Unheated WPI increased in turbidity after thermal treatment for 3 min at 85°C, with 40 mM NaCl (48.5% decrease in transmission compared with control) while WPI-MD6 and WPH-MD6 conjugates retained, to a greater extent, solution clarity after heating (3.5 and 0.2% decrease in transmission compared with respective controls). Conjugation of WPH with MD6 resulted in higher levels of conjugation than for WPI and resulted in a conjugate with enhanced functional properties including improved solubility and lower levels of turbidity development in a heated high ionic strength environment compared with unconjugated WPI and conjugated WPI-MD6.

**Key Words:** conjugation, whey protein hydrolysate, physicochemical functionality

**78 Novel application of a fungal catalase preparation to control spore-forming bacteria in the dairy industry.** Nuria Garcia-Fernandez\*<sup>1,2</sup> and Ashraf Hassan<sup>1</sup>, <sup>1</sup>*Dairy Science Department, South Dakota State University, Brookings, SD,* <sup>2</sup>*Midwest Dairy Foods Research Center, South Dakota State University, Brookings, SD.*

Spores can resist pasteurization, germinate, and grow in the dairy products during storage, causing spoilage. The aim of this work was to investigate the antimicrobial properties of several catalase preparations (Cat) against spore-forming bacteria isolated from dairy sources and their possible applications in milk and cleaning of separation membranes. The antimicrobial activity of a food-grade (FG) commercially available Cat produced by *Aspergillus niger*, a non-food-grade Cat (NFG), and a Cat from bovine liver (BL) against *Bacillus sporothermodurans* (Bs), *Geobacillus stearothermophilus* (Gs), *Bacillus mojavensis* (Bc), *Bacillus licheniformis* (K1), and *Bacillus* spp. (10/1) was assessed by the agar diffusion assay and broth microdilution. The effect of FG Cat on biofilms formed by 2 single strains of slime-producing *Bacillus* (Bc and K1) and a cocktail of 4 strains (Bc, Bs, Gs and 10/1) was studied alone (150 mg/mL) and after a pretreatment with 0.1% Tween 20-PBS buffer. Last, a cocktail of  $10^3$  spores/mL of 4 strains of *Bacillus* (Bc, Gs, Bs, and 10/1) inoculated in UHT milk was challenged with 0, 6.25 and 12.5 mg/mL of FG Cat for 30 min and the number of survivors was determined. The FG and NFG Cat inhibited the growth of vegetative cells of all tested species of *Bacillus* at 12.5 mg/mL with inhibition zones of up to 25 mm, while BL did not show any inhibition. The FG Cat reduced the number of viable cells in one-day-old Bc and K1 biofilms by 2 and 4.87 log cfu/cm<sup>2</sup> respectively ( $P < 0.05$ ). While the application to a 3-d-old multispecies biofilm reduced the viable counts by only 0.73 log cfu/cm<sup>2</sup>, a pretreatment with Tween 20 followed by FG, resulted in a 1.18 log cfu/cm<sup>2</sup> reduction ( $P < 0.05$ ). The FG Cat at 12.5 mg/mL reduced the number of germinating spores in milk by 36.7%. The FG Cat lost antimicrobial activity after heating at 100°C for 10 min. This work describes for the first time the antimicrobial activity of Cat preparations against bacterial spores, which would create new opportunities for the dairy industry to control germination and outgrowth of spore-forming bacteria (patent pending).

**Key Words:** catalase, antimicrobial, *Bacillus*



**79 Impact of buttermilk serum fractions on the rennet coagulation properties of bovine milk.** M.-P. Gauvin\*<sup>1</sup>, M. Britten<sup>1,2</sup>, and Y. Pouliot<sup>1</sup>, <sup>1</sup>STELA Dairy Research Center, Institute on Nutrition and Functional Foods (INAF), Université Laval, Québec, Québec, Canada, <sup>2</sup>Food Research and Development Center (FRDC), Agriculture and Agri-Food Canada, St-Hyacinthe, Québec, Canada.

Buttermilk is the aqueous phase obtained after churning of cream into butter. Its composition is similar to that of skim milk except for higher proportion of milk fat globule membrane (MFGM) components. These components seem to interfere with rennet coagulation of milk. The purpose of the present study was to separate buttermilk serum into 3 fractions using centrifugation and determine the effect of each fraction on milk rennet-induced coagulation. Buttermilk and pasteurized skim milk (control) were centrifuged (31,000 g, 34°C, 1 h). Three distinct layers of the supernatants were collected: low-density opalescent (LDO), clear (CL) and high-density opalescent (HDO) layers. Protein and fat in these fractions were characterized by gel electrophoresis (SDS-PAGE) and estimation of phospholipid content by colorimetric phosphorus assay after fat extraction by the Mojonnier procedure. Casein micelles from skim milk were collected by centrifugation (31,000 g, 20°C, 1 h), redis-

persed in the different fractions and used to monitor rennet aggregation and coagulation kinetics using dynamic light scattering and rheology. MFGM protein and residual casein micelles were mainly concentrated in the HDO fraction. This fraction from buttermilk contained 3 times more MFGM protein than the same fraction from skim milk, but 35% less casein. Phospholipids were also concentrated in this fraction, 200 mg/100 g and 17 mg/100 g, representing 43 and 39% total fat of this fraction in buttermilk and skim milk, respectively. All buttermilk serum fractions, especially HDO, showed negative effect on rennet coagulation kinetics. Compared with the results obtained with their respective CL fraction (which is the LDO and HDO-depleted fraction), buttermilk HDO slowed down the aggregation rate (-62%) while skim milk HDOL increased it (+42%). In addition, the growth of casein aggregates was rapidly stopped in presence of buttermilk HDO fraction. This suggests that components in this fraction from buttermilk interfere with the aggregation of renneted casein micelles and more investigation is underway to determine the role of MFGM components.

**Key Words:** buttermilk, MFGM, rennet-induced coagulation

# Graduate Student Competition: ADSA Production Division

## Graduate Student Oral Competition, MS

### 80 Automated detection of estrus using multiple commercial precision dairy farming technologies in synchronized dairy cows.

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Detecting dairy cattle estrus helps determine optimal breeding time. Dairy cattle estrus varies in length and intensity, increasing the need for accurate and continuous estrous detection. The objective of this study was to evaluate precision dairy farming technologies (PDFT) for estrous detection. The estrous detection efficiency of PDFT is predicted to be greater than traditional visual observation of estrus. Estrus was synchronized in 24 lactating Holstein dairy cows using a modified G7G-Ovsynch protocol (last GnRH injection withheld to permit expression of estrus) beginning 45 to 85 DIM. Resumption of ovarian cyclicity at enrollment, presence of a corpus luteum (CL) on the day of the final injection (designated experimental day 0), regression of the CL by d 5, and presence of a new CL on d 11 were verified by transrectal ultrasonography. Cows were observed for estrous behaviors for 30 min, 4X per day, on days 2 to 5. Blood samples were collected on d -2, -1, 0, 1, 2, 5, 7, 9, and 11 to quantify progesterone to verify luteal regression and ovulation. Potential periods of estrus (gold standard) were defined by the temporal pattern of progesterone (>1.0 ng/mL on d -2, -1 and 0, <1.0 ng/mL on d 2, and >1.0 ng/mL on d 9 and 11). Detection of estrus by PDFTs, an estrous behavioral scoring system, and by visual observation of standing estrus were compared to the gold standard (Table 1). Sensitivity and specificity for detection of estrus was similar among all PDFT. Only 56% of cows that ovulated were observed standing by visual estrous detection. All systems tested are capable of detecting estrus at least as effectively as visual observation.

**Table 1 (Abstr. 80).** Detection of estrus using alerts generated by PDFT<sup>1</sup> and visual observation (n=24)

Detection method	TP	FP	TN	FN	Sensitivity (%)	Specificity (%)
Afimilk AfiAct Pedometer Plus	16	0	6	2	89	100
GEA CowScout (leg version)	15	0	6	3	83	100
ENGs Track A Cow	14	0	6	4	78	100
Agis SensoOr	14	0	6	4	78	100
Estrus behavioral score <sup>2</sup>	12	1	5	6	67	83
Standing behavior	10	0	6	8	56	100

<sup>1</sup>Sensitivity = TP/(TP + FN) and specificity = TN/(TN + FP); where TP = true positive, TN = true negative, FP = false positive, and FN = false negative.

<sup>2</sup>Scoring system as defined by Van Eerdenburg et al. (1996).

**Key Words:** automated estrus detection, precision dairy technology, estrous behavior

**81 Effect of nutrition and management practices on de novo fatty acid synthesis in northeastern US dairy herds.** Melissa E. Woolpert\*<sup>1,2</sup>, Catarina Melilli<sup>3</sup>, Kurt W. Cotanch<sup>1</sup>, Heather M. Dann<sup>1</sup>, Rick J. Grant<sup>1</sup>, Larry E. Chase<sup>3</sup>, and David M. Barbano<sup>3</sup>, <sup>1</sup>William

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This study investigated the effects of management practices and dietary chemical characteristics on de novo fatty acid (FA) concentrations in bulk tank milk from commercial dairy herds. De novo FA are synthesized primarily from rumen fermentation products acetate and butyrate. It was hypothesized that farms with higher de novo FA concentrations will prioritize management and nutrition that promote rumen health. Herds (n = 44) located in Vermont and northern New York State were selected based on high (HDN; 25.8 ± 1.0; mean ± SD) or low (LDN; 23.6 ± 0.9) de novo FA expressed as a proportion of total FA (relative %) in bulk tank milk. Management practices were assessed during one visit to each farm in March or April, 2014. Total mixed ration samples were collected and analyzed by NIR for chemical composition. Data were analyzed using the MIXED procedure of SAS with de novo group as the fixed effect, farm as the random effect, and breed as the covariate in the model. Milk (26.3 vs. 22.7 kg/d, SE = 1.3, P = 0.06), fat (1.1 vs. 0.9 kg/d, SE = 0.1, P < 0.01), true protein (0.9 vs. 0.7 kg/d, SE < 0.1, P < 0.01), and de novo FA (25.6 vs. 23.7 relative %, SE = 0.2, P < 0.01) were higher for HDN vs. LDN farms, respectively. Days in milk (170 ± 7 d; least squares means ± SE), freestall bunk space (41 ± 3 cm), manure score (2.8 ± 0.0), and freestall feeding frequency (1.5 ± 0.4 times/d) were not different between groups. However, tiestall feeding frequency (4.8 vs. 2.8 times/d, SE = 0.7, P < 0.1), freestall stocking density (1.05 vs. 1.20 cows/stall, SE = 0.06, P = 0.07), and body condition score (3.07 vs. 2.95, SE = 0.04, P < 0.01) were different between HDN vs. LDN herds, respectively. Dietary DM (40.7 ± 2.0), CP (16.0 ± 0.6%), ADF (22.9 ± 1.0%), NDF (37.5 ± 1.3%), starch (21.7 ± 1.5%), ash (8.5 ± 0.4%), and forage to concentrate ratio (1.47 ± 0.14) were not different (P > 0.12) between groups. However, dietary ether extract was lower in HDN herds (3.5 vs. 4.5%, SE = 0.2, P < 0.01). Overcrowded freestalls, reduced feeding frequency, and greater dietary ether extract content compromised de novo FA synthesis thereby decreasing milk fat and protein yields.

**Key Words:** stocking density, milk protein, milk fat

**82 Pregnancy outcomes based on milk pregnancy-associated glycoprotein levels.** Erin M. Wynands\*, Stephen J. LeBlanc, and David F. Kelton, *University of Guelph, Guelph, Ontario, Canada.*

Timely diagnosis of pregnancy and pregnancy losses is economically important. A commercially available pregnancy-associated glycoprotein (PAG) milk assay (Idexx Laboratories Inc.) is offered through routine Dairy Herd Improvement (DHI) testing for diagnosis of pregnancy. The objective of this observational study was to describe the relationship between PAG at various stages of gestation and the likelihood of successful calving. The hypothesis was that higher PAG levels would be associated with successful calving. Data were collected from CanWest DHI for all cows with a milk PAG assay between January 1 and May 31, 2013. The PAG milk assay result is reported as a relative PAG level (S-N value). Cows that tested pregnant (PAG >0.25 according to the manufacturer's cut-point; 6196 cows in 967 herds) were included in this analysis. A dichotomous outcome of calving between 270 and 290 d after the relevant insemination was determined for each cow with 5139 successful outcomes. There was a significant interaction between days in gestation (DIG) at time of PAG assay and PAG, so the data were

stratified by DIG. Milk PAG levels increased after breeding, decreased at 45–75 DIG, then increased through the remainder of gestation. For cows  $\leq 45$  DIG ( $n = 793$ ) increasing PAG level was associated with an increased likelihood of calving ( $P = 0.01$ ). For cows  $> 45$  and  $\leq 75$  DIG ( $n = 1653$ ) relative PAG levels ( $P = 0.001$ ) and linear score ( $P = 0.01$ ) were negatively associated with the outcome, while test-day milk yield was positively associated with the outcome ( $P = 0.01$ ). For cows  $> 75$  DIG ( $n = 3750$ ) relative PAG level ( $P = 0.001$ ), and milk yield ( $P = 0.005$ ) were positively associated with full-term pregnancy; linear score was negatively associated with the outcome ( $P = 0.05$ ). These results indicate that while higher PAG levels are positively associated with a positive calving outcome in general, a decrease in PAG level around 45–75 d in gestation was associated with a successful pregnancy outcome.

**Key Words:** pregnancy-associated glycoprotein, pregnancy diagnosis

### 83 Prepartum dietary cation-anion difference (DCAD) and 25-hydroxyvitamin D<sub>3</sub> supplementation modulate $\beta$ -defensin responses in postpartum dairy cattle.

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Postpartum hypocalcemia has been associated with suppressed neutrophil and lymphocyte function in dairy cattle. It was hypothesized that mitigation of postpartum hypocalcemia would enhance activation of host-defense mechanisms during the postpartum period. The objective of this study was to determine the effects of pre-partum DCAD in combination with vitamin D supplementation on expression of a cluster of  $\beta$ -defensin (DEFB) host-defense genes. Pregnant dry Holstein cows ( $n = 51$ , multiparous) of 255 d gestation were blocked by parity and randomly assigned to 4 prepartum diets of positive (+130 mEq/kg) or negative (-130 mEq/kg) DCAD and either 3 mg vitamin D<sub>3</sub> or 3 mg of 25-hydroxyvitamin D<sub>3</sub> (25D) per 11 kg of DM/d. Treatment diets were fed for the last 21 d of gestation, and, upon calving, cows were fed the same lactation ration for the first 49 DIM. Peripheral blood neutrophils and monocyte cultures were collected at d 0 and d 3 of calving and stimulated with 75 ng/mL of 25D and a combination of 100ng/mL of lipopolysaccharide (LPS) and 75 ng/mL of 25D. The mRNA transcripts of DEFB genes were quantified by real-time PCR, and data were analyzed with a general linear mixed model to test for fixed effects and interactions of experimental variables. Neutrophil gene expression of DEFB4 ( $P \leq 0.05$ ), DEFB5 ( $P \leq 0.05$ ), DEFB7 ( $P = 0.1$ ), and DEFB10 ( $P = 0.1$ ) was greater in cows fed the negative DCAD. Neutrophils had lower expression of DEFB3, DEFB4, DEFB7, DEFB10, and lingual antimicrobial peptide in response to LPS at d 3 of calving compared with d 0, ( $P \leq 0.05$ ). Cows supplemented with vitamin D<sub>3</sub> showed greater DEFB3, DEFB4, DEFB5, DEFB7, and DEFB10 gene expression in monocytes ( $P \leq 0.05$ ) than 25D supplemented cows. In conclusion, pre-partum DCAD and 25D affect neutrophil and monocyte  $\beta$ -defensin responses and have implications for improving transition cow immunity.

**Key Words:** defensin, DCAD, vitamin D

### 84 Effects of elevated subcutaneous adipose stores on fractionated peripheral blood mononuclear cells and polymorphonucleocytes fatty acid profile and polymorphonucleocytes gene expression in periparturient dairy cow.

Cynthia M. Scholte<sup>\*</sup>, Zahra Mohammadi-Amiri, Bahman Shafii, and Pedram Rezamand, University of Idaho, Moscow, ID.

During the periparturient period, elevated circulating nonesterified fatty acids (NEFA) affect not only the circulating fatty acid (FA) composition, but also that of the peripheral blood mononuclear cells (PBMC) and polymorphonucleocytes (PMN). The changes to specific lipid fractions, however, remain unknown. We hypothesize elevated lipid mobilization will alter FA profiles and therefore gene expression of pro-inflammatory mediators. Starting -28 d before calving, treatment cows ( $n = 18$ ) received a dry cow ration with an additional 10 kg of corn / head per day, while the control cows ( $n = 16$ ) received the dry cow ration (no additional corn) with an additional 400 mg of monensin / head per day to minimize lipid mobilization. Postpartum, treatment cows were fasted for 8 h on d +3. Plasma samples were collected on d -28, +3, +12, and +21 relative to parturition to isolate PBMC and PMN for analysis of fatty acid methyl ester (FAME) of cells' NEFA and phospholipids (PL) fractions. Isolated PMN were analyzed using real-time q-PCR for caspase-1 (CASP), interleukin-8 receptor (IL-8R), and L-selectin (SELL). Data were analyzed as repeated measures ANOVA using mixed model procedures in SAS (9.3) with significance declared at  $P \leq 0.05$ . Several FA concentrations of NEFA and PL fractions in PBMC and PMN changed over time. Eicosanoid precursor C20:4 in the NEFA fraction of PBMC were greater in treatment than control cows (0.51 vs.  $1.29 \pm 0.27$  g/100g FAME;  $P = 0.02$ ). In contrast, C20:4 of PBMC in PL fraction were greater in control animals ( $10.06$  vs.  $8.18 \pm 0.56$  g/100g FAME;  $P = 0.02$ ). Anti-inflammatory C20:5 concentrations in the PL fraction of PMN were greater in the treatment group and multiparous animals in a treatment across parity effect ( $P = 0.05$ ). Although gene expression of CASP, IL-8R and SELL in PMN changed over time ( $P < 0.0001$ ), no treatment effect was detected. In summary, dietary manipulation altered FA profiles within PBMC and PMN fractions, but did not alter gene expression in PMN.

**Key Words:** lipid mobilization, immune cell, fatty acid

### 85 Cows at a high risk for subacute rumen acidosis exhibit different feeding behavior.

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Previous studies reported that cows fed the same diet have a large variation in rumen pH; cows at a high-risk of sub-acute rumen acidosis (SARA) have a lower pH for a longer period of time than low-risk cows. The objective of this study was to determine if feeding behavior is different between cows with a high or low risk for SARA. We hypothesized that high-risk cows would eat longer during each meal, contributing to a lower pH. Sixteen ruminally cannulated lactating cows (DIM =  $265 \pm 36$ ; BW =  $632 \pm 80$  kg) were fed a diet consisting of 35% forage and 65% concentrate once daily to increase the risk of SARA. After a 17-d diet adaptation period, ruminal pH was measured every 30 s over a 24-h period. Cows were classified as high or low risk to SARA based on an acidosis index (area of pH  $< 5.8$ /DMI). Feeding behavior of animals was observed for the same 24-h period; each animal was checked every 5 min and each behavior seen was assumed to last for 5 min. The 24-h observation period was split into 3 8-h time periods following feeding, and eating behavior was summarized for each period. All response variables were evaluated for the group effect using the PROC TTEST procedure of SAS. Minimum pH ranged from 5.13 to 6.10 among the 16 cows fed the same diet. The 7 high-risk cows had a higher acidosis index than the 9 low-risk cows (8.03 vs. 0.11,  $P = 0.002$ ). Despite no significant difference in DMI ( $P = 0.87$ ), high-risk cows spent more time eating in the first 8-h period after feeding (186 vs. 153 min,  $P = 0.014$ ) and less in the third 8-h period (19 vs. 43 min,  $P = 0.012$ ) than low-risk cows. In the third period, high-risk cows spent more time ruminating (243 vs. 197 min,  $P = 0.012$ ). Average meal duration was longer for



high-risk cows (92 vs. 59 min,  $P = 0.011$ ). Our results suggest that feeding behavior may be a contributing factor to SARA, where cows with a high-risk for low rumen pH eat for longer periods of time during the first 8-h after feeding. Increasing feeding frequency to encourage distribution of eating throughout the day may be an effective approach to mitigate SARA, which warrants further investigation.

**Key Words:** subacute rumen acidosis, feeding behavior

### 86 Factors associated with aerobic plate count, coliform count, and log reduction of bacteria in automated calf feeders.

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As automated calf feeders become more widely adopted, there is a need for research on sanitation management. The objective was to identify factors associated with bacteria counts and log reduction (LR) as a result of circuit cleaning (CC) in milk/replacer samples collected from automated calf feeders (Förster Technik, Engen, Germany). It was hypothesized that increased cleaning is associated with low bacteria counts and increased LR. Six VA and 4 MN dairy farms (14 feeders) were visited weekly for 7 mo. Samples of milk/replacer were collected aseptically from each feeder, iced, and frozen at  $-5^{\circ}\text{C}$ . For a 4-wk period, VA farms were instructed to collect and freeze samples immediately before and after CC. Samples were thawed and plated on Petrifilms to find  $\log_{10}$  aerobic plate count (APC) and  $\log_{10}$  coliform count (CCP). From a model of state, farm(state), LSM  $\pm$  SE APC was  $3.84 \pm 0.26$  and  $5.02 \pm 0.21 \log_{10}$  cfu/mL ( $P < 0.01$ ), and CCP was  $0.62 \pm 0.47$  and  $2.89 \pm 0.39 \log_{10}$  cfu/mL ( $P < 0.01$ ) for MN and VA. The LSM by farm ranged from 3.30 to  $5.51 \log_{10}$  cfu/mL APC and 0.07 to  $3.82 \log_{10}$  cfu/mL CCP. The LR of APC (LRAPC) and CCP (LRCCP) after CC was  $0.78 \pm 0.82$  and  $0.67 \pm 1.17 \log_{10}$  cfu/mL (mean  $\pm$  SD). By regression, each  $\log_{10}$  increase in pre-CC APC and CCP increased LRAPC and LRCCP by  $0.45 \log_{10}$  cfu/mL ( $P < 0.01$ ). Farms with higher pre-CC APC and CCP cfu/mL performed more CC/wk and used more mL detergent/L water ( $P < 0.05$ ). More mixer cleanings (MCHE)/d lowered pre-CC APC cfu/mL ( $P < 0.05$ ), whereas use of silicon hoses increased pre-CC CCP cfu/mL ( $P = 0.04$ ), and use of bleach decreased pre-CC CCP cfu/mL ( $P < 0.01$ ). By stepwise regression with farm in the model, more days since last MCHE increased LRCCP ( $P < 0.01$ ) within farm. Number of calves, milk L/d, and feeder visit frequency were not associated with LR of bacteria within farm. LR was more dramatic at high pre-CC bacteria, which was reduced by more MCHE/d and use of bleach.

**Key Words:** bacterial count, automated calf feeders

### 87 Effects of supplementing slow-release urea in combination with steam-flaked corn or high-moisture corn on ruminal fermentation and lactational performance of dairy cows.

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Associative effects between slow-release urea (SRU) and different types of corn grains [i.e., steam-flaked corn (SFC) vs. high-moisture corn (HMC)] need to be investigated to improve N utilization and lactational performance of dairy cows. This experiment was performed to test a hypothesis that supplementing SRU (Optigen, Alltech, Nicholasville, KY) in HMC-based diet would improve microbial fermentation and nutrient utilization by lactating dairy cows due to a better synchronous supply of dietary N and ruminally fermentable carbohydrates. Eight

multiparous Holstein cows ( $32 \pm 8.2$  DIM) were used in a duplicated  $4 \times 4$  Latin square design, and within each square, cows were randomly assigned to a sequence of 4 diets during each of the 4 21-d periods (14 d of treatment adaptation and 7 d of data collection). The 4 dietary treatments included: SFC without SRU, SFC with SRU, HMC without SRU, and HMC with SRU. Data were analyzed with a model that included the fixed effect of type of corn grain, SRU supplementation, and their interaction with cow and period as the random effects. Supplementing SRU with SFC but not with HMC increased intake of DM and CP, resulting in tendencies for an interaction between corn grain (CG) and SRU supplementation (CG  $\times$  SRU;  $P = 0.06$ ). Milk yield did not differ among treatments ( $39.0 \pm 0.37$  kg/d). Milk true protein concentration increased with SFC with SRU, whereas it decreased with HMC with SRU ( $P = 0.01$ ). Dietary treatments did not affect ruminal concentration of volatile fatty acids and ammonia-N. Feeding HMC tended to increase feed efficiency for milk production ( $P = 0.09$ ) and N utilization efficiency for milk N ( $P = 0.10$ ), but supplementing SRU did not influence the efficiency parameters. Supplementing SRU with either SFC or HMC resulted in minor effects on ruminal fermentation and lactational performance possibly due to relatively less dietary concentrations of the corn grains (13.6% DM on average) in high-forage diets (60% of the diet DM). However, feeding HMC improved feed and N utilization efficiency for milk production.

**Key Words:** steam-flaked corn, high-moisture corn, slow-release urea

### 88 Regulation of microRNA-221 gene expression in ovarian theca cells of cattle: A possible role in follicular development.

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Cystic ovarian follicles result in reproductively inefficient dairy cows. Fibroblast growth factor-9 (FGF9) gene expression in granulosa cells (GC) is downregulated in cystic vs. non-cystic follicles in cattle. FGF9 is thought to act as an intraovarian regulator by stimulating proliferation and reducing steroid production by GC and theca cells (TC). Analysis using Affymetrix GeneChip bovine Genome Arrays ( $n = 8$ ) revealed that microRNA-221 (*MIR221*) in TC was upregulated by FGF9. We selected this gene for further study due to recent evidence indicating *MIR221* is involved in cell proliferation, angiogenesis and cancer. We hypothesized that *MIR221* is regulated during follicle growth and by steroids and hormones that induce cAMP. Three experiments were conducted to evaluate hormonal regulation of *MIR221* mRNA in TC and its possible role in follicular development using sensitive real-time PCR (*U6* as housekeeping gene). TC were isolated from large ( $>8$  mm) follicles ( $n = 6$  to 8 per pool) collected from an abattoir, cultured for 48 h in medium with 10% fetal calf serum, and then treated in serum-free medium for an additional 24 h. Data were analyzed via GLM procedure of SAS. Exp. 1 tested effects of cAMP pharmacologic agents (dbcAMP at 0.1 mg/mL; forskolin at 4.1  $\mu\text{g/mL}$ ) on FGF9-induced *MIR221* mRNA abundance; neither affected ( $P > 0.10$ ) *MIR221* expression whereas FGF9 increased ( $P < 0.05$ ) *MIR221* abundance 1.94-fold. Exp. 2 tested the effect of 300 ng/mL of steroids (estradiol; androstenedione) and phytoestrogens (genestein;  $\alpha$ - and  $\beta$ -zearanol) on *MIR221*; none of the treatments influenced ( $P > 0.10$ ) *MIR221* mRNA levels ( $3.45 \pm 0.62$  relative abundance). In Exp. 3, follicular fluid (FF) was collected from ovaries obtained at an abattoir, and *MIR221* mRNA abundance measured; *MIR221* mRNA was 3-fold greater ( $P < 0.05$ ) in FF aspirated from small (1 to 5 mm) follicles compared with FF from large follicles. These novel results indicate *MIR221* is increased by FGF9 but not regulated by steroids or cAMP. Greater levels of *MIR221* in FF

of small vs. large follicles suggest a role for FGF9-induced *MIR221* in follicular development.

**Key Words:** theca cells, cattle, microRNA-221

**89 Feeding a rumen-degradable amino acid can enhance milk production.** Kayla M. Hultquist\* and David P. Casper, *South Dakota State University, Brookings, SD.*

The study objective was to determine if feeding a rumen degradable AA can increase milk production comparable to growth hormone (GH). Eight multiparous late-lactating ( $255 \pm 26.4$  d in milk) Holstein dairy cows were blocked by milk yield ( $34.1 \pm 8.25$  kg/d) and randomly assigned to 1 of 4 treatments in a replicated  $4 \times 4$  Latin square design having 21 d periods with 7 d for dietary adaptation and 14 d for data collection. Treatments were control (C), Posilac (rBST), and valine fed at 40 (V40) and 80 g/d (V80). Cows were fed a total mixed ration (TMR) with a distillers grains carrier at 113.4 g/d containing none or added AA. Rumen and blood samples were collected during wk 2 and 3 for volatile fatty acids and endocrine assays. Dry matter intake was similar ( $P > 0.10$ ) (21.3, 22.0, 22.8, and 21.5 kg/d for C, rBST, V40, and V80, respectively) for cows receiving all treatments, but milk yield (22.0, 26.1, 25.2, and 24.9 kg/d), FCM (22.1, 25.4, 24.4, and 24.3 kg/d), and ECM (22.7, 26.1, 25.1, and 24.9 kg/d) were significantly ( $P < 0.05$ ) increased for cows receiving rBST, V40, and V80 compared with C cows. Milk yield improvements were similar ( $P > 0.10$ ) between rBST, V40, and V80. Milk fat percentages (3.51, 3.36, 3.32, and 3.38%) were highest ( $P < 0.05$ ) for cows receiving C compared with cows receiving V40, while cows receiving other treatments were intermediate and similar ( $P > 0.10$ ). Milk protein percentages (3.20, 3.12, 3.15, and 3.13%) were greatest ( $P < 0.05$ ) for cows receiving C compared with cows receiving rBST. Ruminal isobutyric acid (1.01, 1.05, 1.28, and 1.49 molar %) concentrations were increased ( $P < 0.05$ ) for cows receiving V40 and V80 compared with cows receiving C and rBST treatments. Plasma IGF-1 concentrations (60.4, 106.1, 65.9, and 58.3 ng/mL) were greater ( $P < 0.05$ ) for cows receiving rBST compared to cows receiving other treatments. Plasma GH concentrations (1.78, 1.99, 1.55, and 1.45 ng/mL) were greater ( $P < 0.05$ ) for cows receiving rBST compared with V40 and V80 cows, while C cows were intermediate and similar ( $P > 0.10$ ). This study suggests that feeding a rumen degradable AA can increase milk yield comparable to rBST.

**Key Words:** milk production, dairy cow, growth hormone

**90 Investigation of a new anti-slip flooring technology to address slips and falls by dairy cows.** Nancy Franco-Gendron\*<sup>1</sup>, Renée Bergeron<sup>1</sup>, Walt Curilla<sup>2</sup>, Sabine Conte<sup>3</sup>, Trevor J. DeVries<sup>4</sup>, and Elsa Vasseur<sup>1</sup>, <sup>1</sup>Organic Dairy Research Center, University of Guelph, Alfred Campus, Alfred, Ontario, Canada, <sup>2</sup>AB Silikal Hygienic Floors, Diamond Hard Surfaces Inc., Calgary, Alberta, Canada, <sup>3</sup>Centre de Recherche et de Développement sur le Bovin Laitier et le Porc, Agriculture and Agri-Food Canada, Lennoxville, Quebec, Canada, <sup>4</sup>Department of Animal and Poultry Science, University of Guelph, Kemptville Campus, Kemptville, Ontario, Canada.

Freestall dairy facilities are known to have issues with slips and falls by cows caused by the combination of smooth flooring and manure slurry. Eighteen Holstein dairy cows (parity 1 to 6) selected to maximize gait range were evaluated on various flooring types to test the hypothesis that flooring with greater anti-slip properties will improve cattle's ease of movement. Cows were trained and randomly assigned to walk on 6 flooring types covered with thin coat of manure 3x/wk for 6 wks; 2

controls: rubber mat (Animat), traction milled concrete (Agri-Trac), and 4 different coated treatments: Red (Silikal 0.7–1.2 mm quartz with 1 seal coat), Green (Silikal 0.7–1.2 mm, 2 seal coats), Yellow (Silikal 0.4–0.8 mm, 1 seal coat) and Orange (Silikal 0.4–0.8 mm, 2 seal coats). Cows were filmed walking a straight corridor and a corridor with a turn (5 passages/corridor). Cows had 2 accelerometers on their rear legs to measure the average acceleration of a passage. Reflective markers were placed on both right hooves before walking the straight corridor measuring stride length, foot height, swing and stance time via kinematics. Steps taken, passage time and time/steps were recorded for the curved corridor. Stride for cows on the straight corridor was longer for rubber ( $1.54 \pm 0.026$  m; mean  $\pm$  SE) compared with all other treatments:  $1.44 \pm 0.041$  m for Red (Wilcoxon sign rank test:  $S = 54$ ;  $P = 0.003$ ),  $1.38 \pm 0.027$  m for Green ( $S = 68$ ;  $P < 0.0001$ ),  $1.48 \pm 0.031$  m for Yellow ( $S = 43$ ;  $P = 0.02$ ), and  $1.46 \pm 0.033$  m for Orange ( $S = 47$ ;  $P = 0.01$ ). Stride of cows walking on the straight corridor was shorter on groove concrete ( $1.41 \pm 0.034$  m) compared with Yellow ( $S = -52$ ;  $P = 0.02$ ), but no differences were found with the other treatments. The time taken per step for cows on the curve corridor was not different between flooring (Kruskal-Wallis test:  $X^2 = 0.9$ ;  $P > 0.1$ ). In conclusion, longer strides or faster pace reflects cow ease of movement on a particular flooring type. Based on these 2 variables, cow ease of movement was greater on rubber versus coating treatments, but was better on the Yellow treatment compared to concrete. Other aspects of ease of movement like acceleration or gait need to corroborate these first findings.

**Key Words:** dairy cows, welfare, flooring

**91 2,4-Thiazolidinedione improves liver function but does not affect insulin sensitivity and expression of genes in adipose and mammary tissue of lactating dairy goats.** Fernanda T. da Rosa\*<sup>1</sup>, Johan Osorio<sup>1</sup>, Francisco Y. Rivera<sup>1</sup>, Erminio Trevisi<sup>2</sup>, Charles T. Estill<sup>1</sup>, and Massimo Bionaz<sup>1</sup>, <sup>1</sup>Oregon State University, Corvallis, OR, <sup>2</sup>Universita Cattolica del Sacro Cuore, Piacenza, PC, Italy.

Previous studies have shown that daily injection of 2,4-thiazolidinedione (TZD) in lactating dairy goats decreases milk somatic cell count but does not affect milk fat yield. The latter could be due to a lipogenic competition between adipose and mammary tissues, owing to an activation of peroxisome proliferator-activated receptor gamma (PPAR $\gamma$ ), a potential TZD target, in the adipose tissue. This study investigated the hypothesis that activation of PPAR $\gamma$  improves the response to mastitis and increase milk fat synthesis. To test this, 24 Saanen goats received either TZD or saline and intramammary infusion of either bacteria or saline (6 goats/group). A metabolic and inflammation profiling in blood (including insulin), adipocyte size by histology, and expression by RT-qPCR of PPAR $\gamma$  target genes in adipose tissue obtained through biopsy and in mammary epithelial cells (MEC) isolated from milk were assessed. In MEC, expression of *CCL2* and *IL8* was also measured. Data were analyzed by GLIMMIX of SAS with Mastitis, TZD, and Time and all interactions as main effects. Significance was determined by Tukey-adjusted  $P < 0.05$ . Mastitis induction decreased myeloperoxidase but TZD treatment increased haptoglobin, cholesterol, paraoxonase,  $\gamma$ GT, and myeloperoxidase in blood compared with saline, indicating a positive effect on liver and increased neutrophil host defense capability. Insulin sensitivity (estimated using QUICKI) was not affected by TZD but overall was lower in mastitic goats. Adipocyte size increased over time but was not affected by treatments. Sub-clinical mastitis increased expression of *CCL2* and decreased expression of *FASN* in MEC but increased expression of *FASN* and *SREBF1* in adipose. No overall effects of TZD on expression of PPAR $\gamma$  target genes were observed in MEC and adipose tissue. In summary, the data revealed limited effects

of TZD on the response of goats to mastitis. Furthermore, the data does not support TZD being a strong PPAR $\gamma$  agonist and insulin sensitizer in dairy goats but indicates that it might improve directly or indirectly the liver function and inflammatory response.

**Key Words:** goats, mastitis, 2-4-thiazolidinedione

**92 Inferring the causal effect of number of lambs born on milk yield in dairy sheep using propensity score methods.** Vera C. Ferreira\*, Bruno D. Valente, David L. Thomas, and Guilherme J. M. Rosa, *University of Wisconsin, Madison, WI*.

Assigning causal interpretation to associations obtained from observational data is challenging as they are prone to confounding. Number of lambs born (prolificacy) in dairy sheep may be considered a potential factor contributing to milk yield (MY). However, inferring this effect using traditional regression or ANOVA techniques can generate spurious results if confounder variables affect both the outcome (MY) and treatment (prolificacy). Propensity score (PS) methods tackle this issue by balancing baseline covariate distributions between treatment levels, allowing unbiased inference of marginal effects. This method belongs to the framework of causal models dealing with potential outcomes. It intends to mimic aspects of randomized trials, in which the difference

among treatment groups is causally meaningful. Under the assumption that ewe prolificacy affects MY, our objective was to estimate the magnitude of such a causal effect using PS based on Matched Samples. Data comprised 4,319 records from 1,534 crossbred dairy ewes. The set of potential confounders was composed by lactation number (1th, 2nd and 3th – 6th) and dairy breed composition (<.5, 0.5-.75 and > 0.75 of East Friesian or Lacaune). For the treatment variable, single lamb birth was assigned to Group 0, while multiple birth (2, 3 or 4 lambs) was assigned to Group 1. MY represented the volume of milk produced for the whole lactation (mean = 268.5 L and SD = 116.4 L). The R package “nonrandom” was used. A total of 1,166 pairs of treated/nontreated individuals with similar PS values were formed. The criterion for similarity was defined by a caliper size equal to 20% of the sd in the PS logit (0.13) and a ratio of treated/untreated = 1. All covariates were deemed balanced after matching (cutoff for standardized bias = 0.2). The estimated causal effect of prolificacy on MY was 20.52 L, se = 3.77 L, 95% CI = 13.13–27.91 L. This means that ewes that gave birth to a single lamb would be expected to have MY increased by 20.52 L if they had given birth to multiple lambs and all other variables were held constant. This implies that any management practice that increases (decreases) prolificacy would affect MY positively (negatively).

**Key Words:** causal inference, milk yield, prolificacy



# Graduate Student Competition: ADSA Southern Section Graduate Student Oral Competition

**93 The effect of somatic cell score on milk yield of dairy cattle in the southeastern United States.** Derek T. Nolan\* and Jeffrey M. Bewley, *University of Kentucky, Lexington, KY.*

The objective of this study was to quantify the effect of somatic cell score (SCS) on milk yield of dairy cattle. Collection of Dairy Herd Information cow records included: herd, days in milk (DIM), parity, birth date, calving date for each parity, and SCS. Records were collected from 6 states in the Southeast (Tennessee, Kentucky, Virginia, Georgia, Florida, and Mississippi). To be included in the data set, cows must have been within one standard deviation of the mean age of the parity group and have DIM between 240 and 305 d. Herds containing less than 5 cows that met the criteria were not included in the analysis. As a result, 10,333, 5,917, 3,213, and 3,266 cows were included from 468, 357, 238, and 244 herds in the analysis of parities 1, 2, 3, and 4 respectively. The GLM procedure of SAS version 9.3 (SAS Institute Inc. Cary, NC) was used to analyze the following model:  $Y_{ijklmn} = S_i + H_j + M_k + D_l + A_m + I_n + E_{ijklmn}$ , where  $Y_{ijklmn}$  = lactation milk yield,  $S_i$  = effect of the  $i$ th state,  $H_j$  = effect of the  $j$ th herd,  $M_k$  = effect of  $k$ th calving season,  $D_l$  = effect of the  $l$ th days the cow was in lactation,  $A_m$  = effect of the  $m$ th age at calving,  $I_n$  = effect of  $n$ th SCS, and  $E_{ijklmn}$  = residual error. Analysis of parities 1 to 4 occurred separately. Herd was a random effect in the model; all other variables were considered fixed. For every 1-unit increase in SCS, a milk yield loss of -113.58, -209.89, -214.42, and -252.09 kg/lactation occurred for parities 1, 2, 3, and 4 respectively. Overall, first-parity cows have half of the milk production loss from a unit increase in SCS than older parity cows. The Southeast Quality Milk Initiative project is supported by Agriculture and Food Research Initiative Competitive Grant no. 2013-68004-20424 from the USDA National Institute of Food and Agriculture.

**Key Words:** somatic cell score, milk yield, Southeast Quality Milk Initiative

**94 Effect of early lactation increased milking frequency on milk yield and local regulators of mammary cell activity.** Diana K. Hardin\*<sup>1</sup>, Andrea J. Lengi<sup>1</sup>, Hollie H. Schramm<sup>2</sup>, and Benjamin A. Corl<sup>1</sup>, <sup>1</sup>*Department of Dairy Science, Virginia Tech, Blacksburg, VA,* <sup>2</sup>*Department of Large Animal Clinical Sciences, Virginia-Maryland College of Veterinary Medicine, Blacksburg, VA.*

Increased milking frequency (IMF) during early lactation increases milk yield not only during the IMF period, but also during continuing lactation after a return to twice daily milking (2×). The increase in milk yield is locally regulated within the mammary gland; however the mechanism for the increase in milk yield is unknown. The objective of this study was to demonstrate a difference in milk and component yield, both during the IMF period and throughout remaining lactation, and examine potential local regulation mechanisms that increase production due to early lactation IMF. Eight multiparous dairy cows were assigned to unilateral frequent milking [2× left udder half and 4-times-daily milking (4×) right udder half] for the first 21d of lactation. Both udder halves were milked 2× for the remainder of lactation. Milk weights from each udder half were recorded at 7, 14, 21, 60, 120 and 180 DIM. Mammary biopsies were obtained from each rear quarter at 21 DIM, and tissue was used to measure protein and mRNA expression. During the IMF period, between 0 and 21 DIM, 4× milking increased milk yield  $5.5 \pm 0.5$  kg/d more than 2× ( $P < 0.05$ ). Between 60 and 180 DIM, the 4× half tended to produce  $4.4 \pm 0.9$  kg/d more than the 2× half ( $P < 0.1$ ). Overall, between d 0 and d 180, IMF increased milk yield by  $4.6 \pm 0.5$  kg/d more than 2× ( $P < 0.05$ ). The 4× gland had reduced phosphorylated and activated Akt (phospho:total Akt) compared with the 2× gland ( $P < 0.05$ ). There was no significant difference between the mRNA expression of IGF-1 or IGFBP-5 between 4× and 2× milking. There was no significant difference between total and phosphorylated STAT5, however the 4× gland tended to have increased activation of STAT5 (phospho:total STAT5) compared with the 2× gland ( $P < 0.06$ ). There was no significant difference between the expression of total, phospho, or activated STAT3. We conclude that early lactation IMF significantly increased milk yield throughout lactation, and this increase in milk yield may be regulated by changes in cell activity resulting from activation of STAT5.

**Key Words:** milking frequency, lactation, STAT5

# Lactation Biology Symposium: Mammary gland biology revisited

**95 Mammary gland growth—It's not just estrogen.** Russell C. Hovey\*<sup>1</sup>, Grace E. Berryhill<sup>1</sup>, Josephine F. Trott<sup>1</sup>, and Adam L. Lock<sup>2</sup>, <sup>1</sup>University of California, Davis, Davis, CA, <sup>2</sup>Michigan State University, East Lansing, MI.

The mammary gland goes through a coordinated series of developmental states during postnatal life in preparation for lactation. These changes ultimately reflect a changing endocrine environment alongside local alterations in the microenvironment. A longstanding assumption has been that estrogens serve as the first essential component for any of these developmental changes to be realized in the mammary glands. Indeed, estrogens can initiate the onset of allometric growth during puberty, and they subsequently potentiate the effects of other endocrine cues on the mammary epithelium during lobulo-alveolar development throughout gestation. More recent evidence from our laboratory indicates that alternative pathways are equally effective in promoting growth of the mammary glands independent of a primary role for estrogens. One fascinating example is the ability of dietary *trans*-10,*cis*-12 conjugated linoleic acid (10,12 CLA) to promote allometric growth of the mammary ducts of ovariectomized mice. In subsequent studies we showed that the specific effects of this dietary fat are independent of estrogenic action, and are mediated through IGF-I receptor signaling. The relevance of these findings is emphasized by the fact that this dietary intervention mimics several aspects of the metabolic syndrome manifest in obese humans. At the same time, additional findings from our laboratory indicate that prolactin and progesterone have synergistic effects on epithelial growth and morphogenesis independent of any role for estrogens, which are partly mediated by changes in the local microenvironment. These data collectively support the notion that while estrogens are clearly mitogenic for the growing and developing mammary glands, they are not necessarily requisite. These findings have potential implications for our understanding of mammary growth in all mammals including livestock, as well as direct relevance to the regulation of mammary cancers.

**Key Words:** prolactin, progesterone, lipid

**96 Body condition of gilts at the end of gestation affects their mammary development.** Chantal Farmer\*<sup>1</sup>, Marie-France Palin<sup>1</sup>, and Michel Vignola<sup>2</sup>, <sup>1</sup>Agriculture and Agri-Food Canada, Dairy and Swine R&D Centre, Sherbrooke, QC, Canada, <sup>2</sup>Nutreco Canada, St-Elzéar, QC, Canada.

The goal of this project was to determine if changes in body condition that incur during gestation affect mammary development of gilts on d 110 of gestation. Gilts of a similar BW (138.1 ± 8.2 kg) and backfat thickness (BF, 16.4 ± 1.0 mm) at mating were fed different amounts of a commercial gestation diet to create 3 groups of animals based on body condition at the end of gestation. These were low (LO; 12–15 mm, n = 13), medium (ME; 17–19 mm, n = 13), and high (HI; 21–26 mm, n = 13) BF. All gilts were weighed and had their BF measured ultrasonically at P2 the day of mating and on d 30, 50, 100 and 110 of gestation. Blood samples were obtained on d 109 of gestation to measure IGF-I and adiponectin concentrations. Gilts were then slaughtered on d 110 to collect mammary glands for compositional analyses. The MIXED procedure of SAS using a univariate model (3 levels) was used for statistical analyses and means were compared using the Tukey's test. As expected, BW and BF were similar across all groups at mating ( $P > 0.10$ ). Treatment differences were present for both variables as of 30 d

of gestation onward ( $P < 0.01$ ). Neither IGF-I nor adiponectin concentrations were affected by treatment ( $P > 0.10$ ). Mammary extraparenchymal tissue weight was lesser in LO than in ME or HI gilts (1074.7, 1360.2 and 1578.4 ± 64.3 g, respectively,  $P < 0.01$ ) and tended to be lesser in ME than in HI gilts ( $P = 0.06$ ). Weight of parenchymal tissue was also affected by treatment ( $P < 0.05$ ), being lesser in LO than HI gilts ( $P < 0.05$ ), and tending to be lower in LO than ME gilts ( $P = 0.12$ ). Values for LO, ME and HI gilts were 1058.6, 1369.6 and 1443.9 ± 198.7 g, respectively. Mammary cell number (DNA) in parenchyma was not affected by treatment, whereas metabolic activity (RNA) was greater in LO than HI gilts ( $P < 0.05$ ). Total parenchymal fat was also lesser in LO than ME or HI gilts ( $P < 0.001$ ). Body condition of gilts therefore has an impact on mammary development at the end of gestation. Thanks to Swine Innovation Pork for partial funding.

**Key Words:** backfat thickness, gilt, mammary development

**97 Autocrine-paracrine regulation of the mammary gland.**

Samantha R. Weaver and Laura L. Hernandez\*, University of Wisconsin-Madison, Madison, WI.

The mammary gland has a remarkable capacity for regulation at a local level, particularly with respect to its main function: milk secretion. Regulation of milk synthesis has significant effects on animal and human health, at the level of both the mother and the neonate. Control by the mammary gland of its essential function, milk synthesis, is an evolutionary necessity and is therefore tightly controlled at a local level. For at least the last 60 years, researchers have been interested in elucidating the mechanisms underpinning the mammary gland's ability to self-regulate, largely without the influence from systemic hormones or signals. By the 1960s, researchers realized the importance of milk removal in the capacity of the gland to produce milk and that the dynamics of this removal, including emptying of the alveolar spaces and frequency of milking, were controlled locally as opposed to through systemic hormonal regulation. Using both in vitro systems and various mammalian species, including goats, marsupials, humans, and dairy cows, it has been demonstrated that the mammary gland is largely self-regulating in its capacity to support the young, which is the evolutionary basis for milk production. Local control occurs at the level of the mammary epithelial cell through pressure and stretching negative-feedback mechanisms and also in an autocrine fashion through bioactive factors within the milk which act as inhibitors, regulating milk secretion within the alveoli themselves. It is only within the last 20 to 30 years that potential candidates for these bioactive factors have been examined at a molecular level. Several factors, including parathyroid hormone related protein (PTHrP), growth factors (transforming growth factor, insulin-like growth factor, epidermal growth factor) and serotonin (5-HT), are both synthesized within and act upon the gland, and possess dynamic receptor activity resulting in diverse effects on growth, calcium homeostasis, and milk composition. This review will focus on the autocrine-paracrine regulation of the mammary gland, with an examination of both foundational work and the progress made within the last 10 to 20 years of research.

**Key Words:** lactation, milk, secretion

**98 New insights in the importance of prolactin in dairy ruminants.** Pierre Lacasse<sup>\*1</sup>, Séverine Ollier<sup>1</sup>, Vanessa Lollivier<sup>2</sup>, and Marion Boutinaud<sup>2</sup>, <sup>1</sup>*Dairy and Swine R&D Centre, Sherbrooke, QC, Canada*, <sup>2</sup>*INRA, Agrocampus Ouest, UMR1348 PEGASE, Saint Gilles, France*.

In most mammals, prolactin (PRL) is essential for maintaining lactation and its suppression inhibits lactation. However, the involvement of PRL in the control of ruminant lactation is less clear because inconsistent effects on milk yield have been observed with short-term suppression of PRL by bromocriptine. Therefore, several experiments were conducted to assess the galactopoietic role of PRL. In an initial experiment, cows in early lactation received daily injections of the dopamine agonist quina-golide (QUIN) for 9 weeks. QUIN reduced milking-induced PRL release and caused a faster decline in milk production. Milk production was correlated with the amount of PRL released at milking. QUIN reduced mammary epithelial cell activity, survival and proliferation. In goats, QUIN did not affect either basal or milking induced PRL release and milk production, whereas injection of cabergoline, another dopamine agonist, caused a decrease of 28% of milk yield the day after the injection. In another experiment, cows were injected for 5 d with QUIN; QUIN + injection of bPRL at milking time; or vehicles. Again, milk, protein and lactose yield were decreased by QUIN. Although PRL injections were not sufficient to restore milk yield, they tended to increase milk protein and lactose yields and increased the viability of milk purified mammary epithelial cells. In late lactation cows, QUIN decreased milk production within the first day of treatment and induced a more rapid changes in several markers of mammary gland involution after drying-off. Similarly, injection at drying-off of cabergoline hastened mammary involution and enhanced mammary gland remodeling. Recently, we stimulated PRL secretion with daily injection of the dopamine antagonist domperidone for 5 weeks. Milk production increased gradually and was greater in domperidone-treated cows during the last 4 weeks of the treatment period. Milk production of both groups became similar again 5d after the last injection. In conclusion, these data, combined with those from other studies, provide a good body of evidence that PRL is galactopoietic in dairy ruminants.

**Key Words:** prolactin, milk production, cows

**99 Regulation of cell number in the mammary gland via the control of the exfoliation process in milk in ruminants.** Lucile Hervé<sup>1,2</sup>, Vanessa Lollivier<sup>1,2</sup>, Hélène Quesnel<sup>1,2</sup>, and Marion Boutinaud<sup>\*1,2</sup>, <sup>1</sup>*INRA UMR1348, Saint Gilles, France*, <sup>2</sup>*Agrocampus Ouest UMR1348, Rennes, France*.

Milk yield is partly influenced by the number of mammary epithelial cells (MEC) in the mammary gland. It is well known that MEC number varies due to cell proliferation and apoptosis. The exfoliation of MEC from the mammary epithelium into milk is another process which might influence MEC number in the mammary tissue. Yet, little is known about the control of MEC exfoliation process. The rate of MEC exfoliation can be assessed by measuring the milk MEC content through flow cytometry analysis or through an immuno-magnetic method for MEC purification. Various experimental models were used to affect milk yield and study the rate of MEC exfoliation. Reducing milking frequency from twice to once daily increased MEC loss per day in goat but not in cow milk. An increased daily rate of MEC exfoliation was also observed during short days as compared with long days or in response to an endotoxin-induced mastitis in cows. Other animal models were designed to investigate the endocrine control of the exfoliation process and its link with milk production. Suppression of ovarian steroids by ovariectomy resulted in a greater persistency of lactation and a decrease in MEC exfoliation.

Administering prolactin inhibitors enhanced MEC loss while exogenous prolactin tended to prevent this negative effect of prolactin inhibitors. These findings suggest that prolactin could regulate MEC exfoliation. In most of these studies, variations of MEC exfoliation were associated with variations in milk yield and changes in mammary epithelium integrity. Exfoliation of MEC could be a process that regulates MEC number in the mammary tissue, and thereby could influence milk yield and lactation persistency.

**Key Words:** cow, lactation, mammary epithelial cell

**100 Mammary response to infection: A critical balance between pathogen elimination and collateral damage.** David E. Kerr<sup>\*</sup>, *University of Vermont, Burlington, VT.*

Mastitis is an inflammatory disease of the mammary gland. The disease is generally classified as sub-clinical (no obvious signs) or 3 levels of clinical disease including: mild (abnormal milk); moderate (abnormal milk with swelling or redness of the gland); severe (abnormal milk, gland inflammation, with systemic signs of illness). Recent large-scale studies indicate that mild, moderate, and severe forms make up approximately 60%, 30%, and 10% of clinical cases of mastitis, with severe mastitis predominately caused by gram-negative bacteria. Efforts to reduce severe mastitis are of utmost importance to dairy animal welfare, and these cases are associated with greatest milk production losses. Experimental challenge studies under controlled conditions reveal animal-to-animal variation in the severity of the resulting mastitis. This suggests a genetic basis to disease severity and the potential for finding genetic markers for use in breeding programs to produce animals with a reduced tendency to develop severe mastitis. However, the evolving field of epigenetics suggests that in utero and early life environments can modify gene expression and thus modify an animal's phenotype. Our approach is to develop a cell culture challenge model predictive of an animal's innate response phenotype. Such a model could potentially be used with cells from young animals to determine their response phenotype and thus facilitate selection of herd replacements. In our dermal fibroblast model, the cells are cultured under controlled conditions and then challenged with LPS to determine innate response magnitude. This model has revealed breed differences (Angus vs. Holstein) and epigenetic differences in samples from the same animals (i.e., same genotype) collected at 5 and 16 mo of age. Further, animals with low vs. high fibroblast response phenotype produce less BSA in milk following experimentally induced mastitis. Future studies employing this and other model systems, combined with well-controlled disease challenges of extreme phenotypes will lead to a greater understanding of factors contributing to animal variation in the severity of response to mammary infection.

**101 Blood-derived proteins in milk during the colostrum period: Active or passive transfer?** Samantha K. Wall<sup>\*1</sup>, Josef J. Gross<sup>1</sup>, Evelyne C. Kessler<sup>1</sup>, Kris Villez<sup>2</sup>, and Rupert M. Bruckmaier<sup>1</sup>, <sup>1</sup>*Veterinary Physiology, Vetsuisse Faculty University of Bern, Bern, Switzerland*, <sup>2</sup>*Swiss Federal Institute of Aquatic Science and Technology, Dübendorf, Switzerland*.

Colostrum has a different composition than milk in established lactation. This difference is in part due to the partially open blood-milk barrier, which prevents the interdiffusion of blood and milk components. In the first days of lactation,  $\alpha$ -lactalbumin (LALBA), a milk protein, is typically present in blood and several blood-derived proteins are present in milk such as IgG<sub>1</sub> (very high concentration), IgG<sub>2</sub>, serum albumin (ALB), and lactate dehydrogenase (LDH). With the exception of IgG<sub>1</sub>, which is transferred by active transcellular transport, other proteins are



thought to pass paracellularly through the temporarily open barrier. This study aimed to examine the decline patterns of each protein relative to IgG<sub>1</sub>, to distinguish between paracellular and transcellular transport through the blood-milk barrier during the first days of lactation. Ten Holstein cows were milked at 4 h after parturition, the next 5 consecutive milkings, and the afternoon milking on d 5, 8, 10, and 14 of lactation for a total of 10 milking time points and blood samples were taken in parallel. Blood and milk samples were analyzed for the concentrations of LDH, ALB, IgG<sub>1</sub>, IgG<sub>2</sub> and LALBA. Protein concentration curves were generated from all 10 time points and were evaluated using the tau time constant model to determine the rate of decline of the slope of each protein. When examining blood-derived proteins in milk, the

concentration of IgG<sub>1</sub> declined significantly faster than the proteins IgG<sub>2</sub> and LDH. Interestingly, the decline of ALB was not statistically different from IgG<sub>1</sub> nor IgG<sub>2</sub> and LDH. IgG<sub>1</sub> concentration in milk far exceeded levels in plasma, and this protein exhibited a recovery increase in plasma during the experimental period. IgG<sub>2</sub>, ALB, and LDH concentrations in milk did not reach plasma levels. Plasma LALBA followed a different pattern, declining significantly slower than all blood-derived proteins in milk. These results indicate that there is active transport of only IgG<sub>1</sub>, with a sharp decline at parturition, compared with IgG<sub>2</sub>, ALB, LDH, and LALBA which are following the closure of the blood-milk barrier.

**Key Words:** colostrum, blood-milk barrier, blood-derived protein

# Meat Science and Muscle Biology

## 102 Timing of exposure to high-concentrate diets vs. pasture on lipogenic enzyme gene expression of steers at slaughter.

Brandon M. Koch<sup>\*1</sup>, Emanuel A. Oliveira<sup>2</sup>, and Susan K. Duckett<sup>1</sup>,  
<sup>1</sup>Clemson University, Clemson, SC, <sup>2</sup>Universidade Estadual Paulista, Jaboticabal, SP, Brazil.

Forty Angus steers (278 ± 21.4 kg) were used to evaluate the effect of feeding strategy during stocker (P1) and finishing (P3) phases on the relative mRNA expression of lipogenic enzymes. Steers were randomly assigned to 2 feeding treatments during P1 (111 d): high-concentrate diet (cracked corn, corn silage, and soybean meal) or high-quality pasture (winter annuals, alfalfa, and non-toxic fescue). An intermediate phase (P2) consisted of 98 d where all steers grazed high-quality pastures. At the start of P3 (until 568 kg BW), each group from P1 was randomly divided into 2 groups that received either a high-concentrate diet or grazed high-quality pastures resulting in 4 treatments (FPF, FPP, PPF, PPP). At slaughter, s.c. adipose tissue samples were collected from each steer and flash frozen for later analysis. No differences were observed for the relative expression of Acetyl CoA carboxylase, carnitine palmitoyltransferase 1A, or glucose transporter type 4 mRNA ( $P > 0.162$ ). An interaction between P1 and P3 ( $P = 0.048$ ) was observed for stearoyl CoA desaturase (SCD) mRNA with 107-, 140-, 10-fold increases for FPF, PPF, FPP, respectively, compared with PPP. Steers on a high-concentrate diet during P3 had greater relative expression of fatty acid synthase (FASN) mRNA ( $P < 0.001$ ) with 59- and 21-fold increases for FPF and PPF treatments, respectively compared with PPP. Similarly, elongase-5 (ELOVL5) and elongase-6 (ELOVL6) mRNA expression increased ( $P < 0.001$ ) when finished on a high-concentrate diet, with FPF and PPF treatments having 18- and 10-fold increases, respectively, for ELOVL5, and 8 and 5-fold increases, respectively, for ELOVL6, when compared with PPP. Adiponectin receptor 1 adipocyte protein 2, and lipoprotein lipase expression were all downregulated when cattle received a high-concentrate diet during P1 ( $P < 0.018$ ). Glycerol-3-phosphate acyltransferase was highly upregulated when steers were finishing on a high-concentrate diet ( $P < 0.001$ ; 56- and 33-fold increase for FPF and PPF, respectively, when compared with PPP). Feeding high-concentrate diets upregulates key lipogenic genes that enhance MUFA content of beef.

**Key Words:** gene expression, lipogenic

## 103 Effects of high-concentrate diets during stocker and finishing phase on lipid fractions in longissimus muscle.

Adriana Pordomingo, Gabriela Volpi Lagreca, Brandon M. Koch, and Susan K. Duckett<sup>\*</sup>, Clemson University, Clemson, SC.

Twenty Angus steers (278 ± 21.4 kg BW) were used to evaluate the effect of high concentrate diets during stocker and finishing phases on lipid fractions [neutral lipid (NL), phospholipid (PhL) and free fatty acid (FFA)] in longissimus muscle. Steers were randomly assigned to 2 treatments: (1) high quality forage grazed during all phases (PAST) or (2) high concentrate diets fed during early stocker (111 d) phase and finishing phase (77 d) with an intermediate period on high quality forage (98 d) between stocker and finishing periods (FPF). Steers were slaughtered at a similar live weight endpoint (568 kg BW). Longissimus muscle samples were obtained from each animal. Lipids were extracted from the LM, separated into lipid fractions (NL, PhL, and FFA) and fatty acid composition of each fraction analyzed by GLC. Feeding of high concentrates during the stocker phase and finishing phase increased ( $P$

$< 0.05$ ) total fatty acid content of the LM compared with PAST (4.96% vs. 3.38%, respectively). On a gravimetric basis, FPF had greater NL and FFA content than PAST. However on a percentage basis, the lipid fractions were similar ( $P > 0.05$ ) between PAST and FPF. Overall, the LM contained 93.7% NL, 5.0% PhL, and 1.6% FFA. In the NL, FPF had greater ( $P < 0.05$ ) concentrations of MUFA, n-6 PUFA, and n-6 to n-3 ratio of PUFA compared with PAST. The PAST had greater ( $P < 0.05$ ) concentrations of n-3 fatty acids in NL than FPF. In the PhL, PAST had greater ( $P < 0.05$ ) MUFA and n-3 PUFA compared with FPF. The PhL fraction of FPF had greater ( $P < 0.05$ ) concentrations of n-6 PUFA and higher ratio of n-6 to n-3 PUFA. Fatty acid composition of FFA fraction did not differ ( $P > 0.05$ ) between treatments. These results indicate that PAST finished beef has increased n-3 PUFA concentrations in the LM due to greater accumulation of n-3 PUFA in both NL and PhL fractions and not because of a greater PhL contribution to the total lipid fraction.

**Key Words:** beef, high concentrate diet, fatty acid

## 104 Feeding microalgae meal (*Schizochytrium limacinum* CCAP 4087/2) to finishing cattle I: Effects on visceral and subcutaneous adipocyte size and *Longissimus lumborum* muscle fiber characteristics.

Kelsey J. Phelps<sup>\*1</sup>, John M. Gonzalez<sup>1</sup>, Derris D. Burnett<sup>1</sup>, Christian A. Alvarado-Gilis<sup>1</sup>, Mathew A. Vaughn<sup>1</sup>, Caleb P. Weiss<sup>1</sup>, Cadra L. Van Bibber-Krueger<sup>1</sup>, Justin E. Axman<sup>1</sup>, Jake D. Thieszen<sup>1</sup>, Kate A. Jacques<sup>2</sup>, and James S. Drouillard<sup>1</sup>, <sup>1</sup>Kansas State University, Manhattan, KS, <sup>2</sup>Alltech Inc., Nicholasville, KY.

The objective of this study was to examine effects of feeding microalgae meal (MA; *Schizochytrium limacinum* CCAP 4087/2) on visceral adipocyte, subcutaneous adipocyte, and *Longissimus lumborum* (LL) muscle fiber characteristics. Heifers (36 pens; 8 heifers/pen) were blocked by initial pen BW (4014 ± 223 kg) and assigned within strata to 1 of 4 treatments. Treatments consisted of 0, 50, 100, and 150 g·heifer<sup>-1</sup>·d<sup>-1</sup> of MA (Alltech Inc., Nicholasville, KY) added to a basal diet consisting of steam-flaked corn, wet corn gluten feed, alfalfa hay, glycerin, and supplement. Heifers were harvested on d 89 of the study and visceral and subcutaneous adipose tissue were collected from approximately the 13th rib of 3 heifers selected at random from each pen. The strip loin from the left side of the same randomly selected heifers was removed and transported to Kansas State University for analysis. One 1.27-cm steak was removed from the 13th-rib end of each loin for measurement of muscle fiber characteristics of the LL. There were no treatment × depot interactions for cross-sectional area or diameter of subcutaneous and visceral adipocytes ( $P > 0.33$ ). Increasing MA in the diet did not affect cross-sectional area or diameter of visceral or subcutaneous adipocytes ( $P > 0.56$ ) and did not affect distribution of LL myosin heavy chain type I, IIA, and IIX muscle fibers or cross-sectional area of fibers ( $P > 0.16$ ; Table 1). Supplementing microalgae meal does not affect visceral and subcutaneous adipocyte size or muscle fiber characteristics of the LL.

Contd.

**Table 1 (Abstr. 104).** Cross-sectional area ( $\mu\text{m}^2$ ) of adipocytes and muscle fibers in heifers fed microalgae meal

Item	Algae, g·heifer <sup>-1</sup> ·d <sup>-1</sup> DM				SEM
	0	50	100	150	
Subcutaneous adipocyte	9,962	9,395	9,632	9,899	523
Muscle fiber					
Type I	2,601	2,321	2,289	2,521	119
Type IIA	2,794	2,692	2,710	2,676	139
Type IIX	4,171	3,809	3,954	4,020	181

**Key Words:** omega-3, adipocyte, muscle fiber type

### 105 Feeding microalgae meal (*Schizochytrium limacinum* CCAP 4087/2) to finishing cattle II: Effects on *Longissimus*

*lumborum* fatty acid profile and meat quality. Kelsey J. Phelps<sup>\*1</sup>, John M. Gonzalez<sup>1</sup>, Christian A. Alvarado-Gilis<sup>1</sup>, Derris D. Burnett<sup>1</sup>, Mathew A. Vaughn<sup>1</sup>, Sara M. Ebarb<sup>1</sup>, Caleb P. Weiss<sup>1</sup>, Cadra L. Van-Bibber Krueger<sup>1</sup>, Justin E. Axman<sup>1</sup>, Kate A. Jacques<sup>2</sup>, and James S. Drouillard<sup>1</sup>, <sup>1</sup>Kansas State University, Manhattan, KS, <sup>2</sup>Alltech Inc., Nicholasville, KY.

Effects of feeding microalgae meal (MA; *Schizochytrium limacinum* CCAP 4087/2) on fresh meat quality were examined. Heifers (36 pens; 8 heifers/pen) were blocked by initial pen BW (4014 ± 223 kg) and assigned within strata to 1 of 4 treatments. Heifers were fed diets containing steam-flaked corn, wet corn gluten feed, alfalfa hay, glycerin, supplement, and 0, 50, 100 or 150 g·d<sup>-1</sup> MA (Alltech, Inc., Nicholasville, KY). Heifers were harvested on d 89 of the study and strip loins were collected from 3 randomly selected heifers per pen. One 1.27-cm steak was removed from the 13th-rib end of each loin for fatty acid analysis. Loins were weighed, vacuum packaged, and aged for 14 d. Loins were reweighed and fabricated into 2.54-cm steaks for analysis of lipid oxidation and color stability during retail display, Warner-Bratzler shear force, and sensory attributes. Feeding MA did not affect concentrations of C16:0, C18:0, C18:3n-3, or total fatty acids within loins ( $P > 0.16$ ), but increased concentrations of C18:2n-6c and C20:5n-3 (linear,  $P < 0.01$ ) and C22:5n-3 and C22:6n-3 (quadratic,  $P < 0.02$ ). There were treatment × day interactions for all color attributes and TBARS during display ( $P < 0.01$ ). From d 0 to 2 of display, increasing MA decreased L\* (linear,  $P < 0.03$ ). For the remainder of display, increasing MA tended to decrease L\* (quadratic,  $P < 0.07$ ). From d 2 to 4 of display, increasing MA decreased a\* (linear,  $P < 0.04$ ). For the remainder of display, increasing MA decreased a\* (quadratic,  $P < 0.02$ ). Surface oxymyoglobin decreased and surface metmyoglobin increased with increased MA (d0–4, linear,  $P < 0.05$ ; d5–7 quadratic,  $P < 0.04$ ), and lipid oxidation was elevated on d 0 and 7 of display with increased MA (quadratic,  $P < 0.01$ ). Treatments did not affect loin purge loss, loss of weight during cooking, or shear force ( $P > 0.15$ ). For sensory panel, off-flavors increased (quadratic,  $P < 0.01$ ) with increased MA. Increases in measures of oxidation in response to feeding microalgae meal suggest that it may be prudent to include antioxidants in diets of microalgae-fed cattle to preserve oxidative stability of meat.

**Key Words:** omega-3, color, sensory

**106 Intratesticular injection of zinc solution effectively castrates male pigs without affecting pork quality.** Jason K. Apple<sup>\*1</sup>, Tsung-Cheng Tsai<sup>1</sup>, Hae-Jin Kim<sup>1</sup>, Min Wang<sup>2</sup>, Brian P. Corbett<sup>2</sup>, Tim M. Johnson<sup>1</sup>, and Charles V. Maxwell<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Arkansas Division of Agriculture, Fayetteville, AR, <sup>2</sup>Ark Science Inc., Irvington, NY.

Zinc gluconate neutralized by Arg (Zeuterin; Ark Science Inc., Irvington, NY) is directly injected into the testes of dogs and has a 99.6% sterility rate; thus, a study was designed to test the effects of intratesticular injections of a Zn solution (Testrin (T); Ark Science Inc.) as a method of castration in swine. Within 10 litters, 7-d-old male pigs (birth weights of 1 kg, or greater) were assigned randomly to 1 of 3 intratesticular T dosages: 0.15 (n = 1/litter), 0.20 (n = 2/litter), or 0.30 mL/testicle (n = 2/litter). Scrotal area was thoroughly cleansed before T was administered by deep intratesticular injection using a sterile syringe and a sterile 28-gauge (1.26-cm long) needle. A surgically-castrated male (B) and an intact female (G) from each litter were designated as industry controls. Pigs were weaned at 21 d, mixed, and subsequently moved first to an off-site nursery then to a grower-finisher unit. Live weights were recorded at birth, weaning, and at the end of the study to calculate ADG, and scrotums were palpated monthly to monitor testicle growth. One month after injection, the proportion of pigs receiving 0.15, 0.20, and 0.30 mL/testicle with at least 1 testicle was 88.9, 95.0, and 70.0%, respectively, and declined to 55.6 (end of trial), 40.0 (end of trial), and 0% (4 mon after injection), respectively. Testrin-injected pigs were heavier ( $P < 0.05$ ) at weaning than B, but neither pre-weaning ( $P = 0.549$ ), post-weaning ( $P \geq 0.185$ ), nor overall ADG ( $P = 0.262$ ) differed among treatments. Carcasses of T-pigs were heavier ( $P = 0.015$ ) than B, but fat depths, LM area, and calculated fat-free lean yield were similar ( $P \geq 0.333$ ) among treatments. Although the LM from G and males treated with 0.20 mL T/testicle were darker (lesser L\* value;  $P < 0.05$ ) than the LM from B, LM color ( $P \geq 0.071$ ), drip and cooking losses ( $P \geq 0.370$ ), marbling ( $P = 0.164$ ), firmness ( $P = 0.185$ ), and shear force values ( $P = 0.378$ ) did not differ among T-treated males, B, and G. Results indicated that male pigs can be effectively castrated by intratesticular injections of 0.30 mL T/testicle, and produce carcasses of equal composition and quality to B and G.

**Key Words:** castration, intratesticular injection, pork quality

### 107 Prediction of red meat yield and trimmable fat yield from beef carcasses utilizing bioelectrical impedance analysis.

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An experiment was performed utilizing bioelectrical impedance technology (BIA) to predict red meat and trimmable fat yields for over-finished beef carcasses. Fifty-six single-sired steers were finished to above typical slaughter weights (603.5 kg ± 48.1 kg; 2.0 ± 0.7 cm 12th rib back fat) before harvest. After a 24-h chill period, standard grading procedures were used to derive a calculated yield grade for each animal (3.7 ± 0.9). Measures of BIA [resistance (Rs, 160.2 ± 16.5 ohms), reactance (Xc, 44.6 ± 4.8 ohms)] were quantified by introducing an alternating electrical current between positive (detector) and negative (source) electrodes placed at opposite ends of the right side of each carcass. Source electrodes introduced current through the carcass and detector electrodes detected any decrease in voltage caused by resistance to electrical current. Other measured variables included temperature (Tp, 3.1 ± 0.7°C), length between electrodes (L, 118.7 ± 5.1 cm), and hot carcass weight (HCW, 381.0 ± 32.8 kg). Impedance (I; (Rs<sup>2</sup> + Xc<sup>2</sup>)<sup>0.5</sup>; 166.3 ± 17.0) electrical volume (EVOL; L<sup>2</sup>/Rs; 89.4 ± 16.8), resistive density (RsD; RSW<sup>2</sup>/(L<sup>2</sup>/Rs); 392.4 ± 63.0), and reactive density (XcD; RSW<sup>2</sup>/(L<sup>2</sup>/Xc); 108.9 ± 16.3) were derived from measured variables. Correlations were calculated between dependent and independent variables. Stepwise regression procedures were used to develop models for prediction of percentage red meat yield (RMY%) and trimmable fat yield (TFY%). Pearson correlation coefficients indicate that RMY was



highly correlated ( $P < 0.05$ ) to RsD ( $r = -0.65$ ), XcD ( $r = -0.51$ ), Tp ( $r = -0.46$ ), HCW ( $r = -0.39$ ), EVOL ( $r = 0.30$ ), Rs ( $r = -0.30$ ), and I ( $r = -0.28$ ) whereas FY was correlated ( $P < 0.05$ ) with RsD ( $r = 0.75$ ), XcD ( $r = 0.60$ ), HCW ( $r = 0.57$ ) and Tp ( $r = 0.50$ ). Regression models indicate that 65% and 72% of the variation in RMY% and TFY% may be attributed to BIA measures. By comparison, the calculated USDA yield grade accounted for 50.0% and 61.0% of the variation in RMY% and TFY%, respectively. These results suggest that BIA technology can be utilized as a predictor of beef carcass composition.

**Key Words:** beef, bioelectrical impedance technology (BIA), zilpaterol

**108 Dietary lysine affected the expression of genes related to lipid metabolism in skeletal muscle of finishing pigs.** Taiji Wang\*, Naresh Regmi, Jean M. Feugang, Mark A. Crenshaw, John R. Blanton Jr., and Shengfa F. Liao, *Department of Animal and Dairy Sciences, Mississippi State University, Mississippi State, MS.*

It has been reported that some amino acids can function as signaling molecules to regulate skeletal muscle growth in mammals. This study was conducted to identify those genes that may be regulated by amino acid lysine and responsible for muscle growth and meat quality of pigs. Nine crossbred barrows ( $94.4 \pm 6.7$  kg BW) were randomly allotted to 3 dietary treatments (3 pigs/treatment). Three corn and soybean-meal based diets were formulated to meet the NRC (2012) requirements for nutrients except for lysine, whose concentrations were 0.43, 0.71, and 0.98% for Diets 1 (lysine deficient), 2 (lysine adequate), and 3 (lysine excess), respectively. After 5 weeks on trial, pigs were killed and muscle samples collected from *longissimus dorsi* (between the 10th and 12th ribs). Total RNA was extracted from 50 mg of each sample using a TRIzol reagent. Porcine Gene 1.0 ST Array (Affymetrix, Inc.) was used to quantify the expression levels of 19,211 genes. Raw microarray data were normalized with gcRMA algorithm and analyzed with ANOVA using Partek Genomics Suite (Partek Inc.). A total of 674 transcripts were differentially expressed ( $P < 0.05$ ); 60 out of 131 transcripts ( $P < 0.01$ ) belong to 59 genes and 71 were unannotated. GO Enrichment analysis of this 59-gene set identified 11 genes in 5 categories of molecular functions: binding, catalytic activity, transcription regulator activity, transporter activity, and molecular transducer activity. Interestingly, 4 genes are associated with lipid metabolism: PSPH: lipid binding and key enzyme for serine (precursor of phospholipids and glycolipids) synthesis; CFD: stimulating glucose transport for triglyceride accumulation and inhibiting lipolysis; ME1: associated with backfat thickness and meat quality; SCD: playing a key role in intramuscular fat formation. It appears that lysine can regulate the expression of multiple genes, and at least 4 genes are related to lipid metabolism. Further studies are needed to elucidate the association of dietary lysine level with the expression levels of these genes and the gene network for lipid metabolism. (Supported by USDA Hatch/Multistate Project 233803)

**Key Words:** lysine, muscle, gene expression

**109 Transcriptomic and metabolomic assessment of growth promoter effects on porcine muscle growth.** John Brameld\*<sup>1</sup>, Kevin Ryan<sup>1</sup>, Hannah Williams<sup>1</sup>, Doug Harris<sup>2</sup>, David Brown<sup>1</sup>, Richard Emes<sup>1</sup>, Tom Giles<sup>1</sup>, Chungui Lu<sup>1</sup>, Charlie Hodgman<sup>1</sup>, and Tim Parr<sup>1</sup>, <sup>1</sup>University of Nottingham, Nottingham, UK, <sup>2</sup>Zoetis, Kalamazoo, MI.

This study compared the effects of growth hormone (GH) and  $\beta$ -adrenergic agonist (BA) on porcine muscle transcriptome and blood

metabolome. Duroc  $\times$  (Landrace  $\times$  Large White) gilts ( $77 \pm 7.1$  kg,  $n = 165$ ) were all fed a high protein/energy diet ad libitum, with the GH group receiving an intramuscular injection, 10mg once every 2d of porcine GH (Reporcin, Zamira), the BA group receiving Ractopamine at 20mg/kg feed, whereas the control group just had feed. Pigs were treated for 1, 3, 7, 13d ( $n = 10$  per treatment for each period) and 27d ( $n = 15$  per treatment for each period). After each treatment period muscles were harvested and blood collected, then plasma immediately prepared. The remaining carcass was incinerated. Plasma was analyzed by Metabolon's biochemical platform technology. Total RNA from LD was extracted and subjected to transcriptome analysis (Agilent pig microarray) followed by gene cluster analysis using MaSigPro. Gene expression was verified by quantitative RT-PCR. Protein expression was determined by Western blot. Treatment groups were compared by 2-way ANOVA (Genstat). The BA treatment increased Vastus Lateralis weight ( $P < 0.001$ ), and induced a switch to faster muscle fiber type in Longissimus dorsi (LD), as myosin heavy chain isoform IIB gene expression was increased ( $P < 0.001$ ). Within 1d of treatment plasma fatty acids were increased in BA, but not GH ( $P < 0.05$ ). Both GH and BA decreased certain plasma amino acids, such as lysine ( $P < 0.05$ ), but only GH decreased the concentration of others, such as serine ( $P < 0.05$ ) and glycine ( $P < 0.05$ ). Only GH increased glucose ( $P < 0.05$ ) but there was no effect of either GH or BA on lactate ( $P > 0.1$ ). Predominant effect of treatment was a BA coordinate increase in LD serine synthesis pathway gene expression (PHGDH,  $P < 0.001$ ; PSAT,  $P < 0.001$ ; PSPH,  $P < 0.001$ ) by 3d, which was confirmed at the protein level, as PHGDH was increased with BA at 7d ( $P < 0.001$ ). The effect of GH and BA treatment on metabolism appears to lead to differential effects on muscle mass, with BA potentially elevating serine synthesis, which could lead to the generation of metabolites required for growth.

**Key Words:** growth promoters, pig, transcriptomics

**110 Molecular factors underlying the discrepancy of marbling between Nellore and Angus beef.** Taiane Martins<sup>1</sup>, Walmir Silva<sup>1</sup>, Letícia Sanglard<sup>1</sup>, Ivan Carvalho Filho<sup>1</sup>, Ygor Cassani<sup>1</sup>, Nick Serão<sup>2</sup>, Mario Chizzotti<sup>1</sup>, Marcio Ladeira<sup>3</sup>, and Marcio Duarte\*<sup>1</sup>, <sup>1</sup>Federal University of Vicosa, Vicosa, MG, Brazil, <sup>2</sup>Iowa State University, Ames, IA, <sup>3</sup>Federal University of Lavras, Lavras, MG, Brazil.

Studies have reported that intramuscular adipogenesis and fibrogenesis may concomitantly occur in skeletal muscle of beef cattle. Thus, we hypothesized that the discrepancy of intramuscular fat content in beef from Nellore and Angus bulls was associated with differences in intramuscular adipogenesis and fibrogenesis during the finishing phase. To test our hypothesis, longissimus muscle samples of Nellore ( $n = 6$ ; BW =  $372.5 \pm 37.3$  kg) and Angus ( $n = 6$ ; BW =  $382.8 \pm 23.9$  kg) bulls with 20 mo of age were collected for analysis of gene and protein expression, and chemical quantification of intramuscular fat and collagen. Least squares means were estimated for the effect of breed and differences were considered at  $P \leq 0.05$ . A greater intramuscular fat content was observed in skeletal muscle of Angus compared with Nellore cattle ( $P < 0.01$ ). Despite the lack of differences in gene expression of adipogenic markers *Zfp423* ( $P = 0.62$ ), *PPAR $\gamma$*  ( $P = 0.42$ ), and *C/EBP $\alpha$*  ( $P = 0.14$ ), a greater protein expression of PPAR $\gamma$  was observed in skeletal muscle of Angus compared with Nellore cattle ( $P = 0.05$ ). A greater abundance of adipo/fibrogenic cells, evaluated by the PDGFR $\alpha$  content, was observed in skeletal muscle of Angus than Nellore cattle ( $P = 0.05$ ). No differences in fibrogenesis were observed in skeletal muscle of Angus and Nellore cattle, which is in accordance with the lack of differences in intramuscular collagen content in beef from both breeds ( $P = 0.16$ ). These findings demonstrate that difference in intramuscular fat content

is associated with a slightly enhanced adipogenesis in skeletal muscle of Angus compared with Nellore cattle, but no difference in fibrogenesis.

**Key Words:** adipocyte, collagen, intramuscular fat

### 111 Effect of rearing system on meat quality, lipid, and amino acid profiles of lambs.

Jian-min Chai<sup>1</sup>, Hai-chao Wang<sup>1</sup>, Qi-yu Diao<sup>1</sup>, Kai-dong Deng<sup>2</sup>, Yan Tu<sup>1</sup>, Min-li Qi<sup>1</sup>, and Nai-feng Zhang\*<sup>1</sup>, <sup>1</sup>Feed Research Institute of Chinese Academy of Agricultural Sciences, Key Laboratory of Feed Biotechnology of the Ministry of Agriculture, Beijing, China, <sup>2</sup>College of Animal Science, Jinling Institute of Technology, Nanjing, Jiangsu, China.

To determine the effect of rearing system on meat quality, lipid and amino acid profiles of lambs, 24 Hu lambs (12 rams and 12 ewes) were randomly divided into 2 treatments: ewe-reared (ER) or weaned at d 10 and fed milk replacer (MR). Ewe milk or MR was available from 0 to 60 d and a creep feed was offered ad libitum to all lambs from d 15 to 90. The fatty acid and amino acid composition of the diet and meat samples were determined by gas chromatography and automated dedicated amino acid analyzer, respectively. All data were analyzed using *t*-test procedure of SAS. Lambs fed MR had a greater growth rate ( $P = 0.03$ ) and creep feed intake ( $P < 0.001$ ) than those in ER treatment. Lambs fed MR had a greater harvest weight ( $P = 0.003$ ), HCW ( $P = 0.004$ ) and fat thickness over *L. dorsi* ( $P = 0.05$ ) compared with those of ER treatment. The meat of lambs in MR treatment had lesser L\* ( $P = 0.04$ ) and b\* ( $P = 0.02$ ). There was no difference ( $P = 0.82$ ) in a\* values between the 2 treatments. Lambs fed MR had greater ether extract content ( $P = 0.003$ ) than that in ER treatment. No differences were found in crude protein ( $P = 0.58$ ) and ash ( $P = 0.11$ ) between 2 treatments. The content of unsaturated fatty acids and monounsaturated fatty acids in meat were greater ( $P = 0.02$ ) for MR treatment compared with ER treatment. On the contrary, the proportion of saturated fatty acids in ER treatment was greater ( $P = 0.03$ ) than that in MR treatment. The proportion of C14:0 of ER lambs was greater ( $P = 0.01$ ) than that in MR treatment, while the proportion of C18:0 ( $P = 0.003$ ), C18:2 ( $P = 0.04$ ), C18:3 ( $P = 0.008$ ) and the ratio of polyunsaturated fatty acids ( $P = 0.002$ ) and saturated fatty acids (P/S) ( $P = 0.03$ ) were lesser than those of MR treatment. The proportion of leucine ( $P = 0.003$ ), alanine ( $P < 0.001$ ), tyrosine ( $P = 0.002$ ), and proline ( $P < 0.001$ ) were greater, while histidine ( $P = 0.014$ ) was lesser for lambs of MR treatment compared with those of ER treatment. In conclusion, the MR rearing system could increase meat production and improve the proportion of lipid and amino acid profiles of lambs.

**Key Words:** meat quality, lipid, amino acid profile

**112 The effects of growth-promoting agents on ovine metabolism and growth.** Shaker Al-Doski\*<sup>1</sup>, Tim Parr<sup>1</sup>, Krystal Hemmings<sup>3</sup>, Zoe Daniel<sup>1</sup>, David Brown<sup>1</sup>, Doug Harris<sup>2</sup>, Chungui Lu<sup>1</sup>, Charlie Hodgman<sup>1</sup>, Sean May<sup>1</sup>, and John Brameld<sup>1</sup>, <sup>1</sup>University of Nottingham, Nottingham, UK, <sup>2</sup>Zoetis, Kalamazoo, MI, <sup>3</sup>University of Derby, Derby, UK.

This study sought to investigate the short-term effects of bovine growth hormone (GH) and  $\beta$ -adrenergic agonist ( $\beta$ A), on lamb liver and muscle, particularly protein and energy metabolism. Wether lambs (120 d old) were all fed a high protein/energy diet ad libitum, with the GH group ( $n = 10$ ) receiving a single subcutaneous injection of bovine GH (Posilac, Monsanto, 3.75mg/kg BW) on d 1; the  $\beta$ A group ( $n = 10$ ) receiving  $\beta$ A (cimaterol) at 10mg/kg in the feed, whereas the control group (C,  $n = 11$ ) only had ad libitum feed. After 6 d sheep were slaughtered blood was collected, plasma immediately prepared, and subsequently analyzed using Metabolon's biochemical platform technology. Samples of Longissimus dorsi (LD) and Supraspinatus (SS) muscles were snap frozen in liquid nitrogen and stored at  $-80^{\circ}\text{C}$  until analysis, the remaining carcass was incinerated. From extracted total RNA first strand cDNA was generated using random primers. Gene expression was determined by quantitative RT-PCR analysis relative to total cDNA, as measured using oligreen. Protein expression was determined by Western blot. Treatment groups were compared by one-way ANOVA (Genstat) and post hoc Dunnett's test. Although there were no significant effects of  $\beta$ A and GH in body weight of lambs ( $P = 0.122$ ),  $\beta$ A, but not GH, significantly increased the weights of both SS ( $P < 0.01$ ) and ST ( $P < 0.05$ ). In the blood, GH significantly increased the concentration of more fatty acids than  $\beta$ A ( $P < 0.05$ ), both GH and  $\beta$ A significantly decreased certain plasma amino acids ( $P < 0.05$ ). Treatment with  $\beta$ A, but not GH, increased mRNA expression of genes involved in glycolysis ( $P < 0.05$ ) and the serine synthesis pathway ( $P < 0.05$ ) in both SS and ST, but decreased expression of genes in the TCA cycle ( $P < 0.05$ ). Effects on the serine pathway were confirmed as protein levels for PHGDH were significantly increased with  $\beta$ A ( $P < 0.001$ ). It appears that GH and  $\beta$ A have differential effects on metabolism that lead to differential effects on muscle mass over this short time frame. Treatment with GH has wider effects on whole body metabolism, while  $\beta$ A appears to have more specific effect on both muscle metabolism and its growth.

**Key Words:** growth promoter, sheep, transcriptomics

## Nonruminant Nutrition: Enzymes and processing

**113 Comparative efficacy of xylanases on energy and nutrient digestibility in growing pigs fed corn- or wheat-based diets.** Elijah Kiarie<sup>\*1,2</sup>, Luis F. Romero<sup>1</sup>, Susan Arent<sup>3</sup>, Rikke Lorentsen<sup>3</sup>, and Hans H. Stein<sup>4</sup>, <sup>1</sup>DuPont Industrial Biosciences-Danisco Animal Nutrition, Marlborough, Wiltshire, UK, <sup>2</sup>Department of Animal Science, University of Manitoba, Winnipeg, MB, Canada, <sup>3</sup>DuPont Nutrition Biosciences-Enzyme Research & Development, Brabrand, Arhus, Denmark, <sup>4</sup>Department of Animal Sciences, University of Illinois, Urbana, IL.

Microbial xylanases differ with respect to source, substrate specificities, and biochemical characteristics, but these differences are rarely considered in their evaluation for animal feed applications. Therefore, an experiment was conducted to determine effects of 5 xylanases on apparent ileal (AID) and apparent total-tract digestibility (ATTD) of energy and nutrients in growing pigs fed diets based on corn plus 40% corn distillers dried grains with solubles (DDGS) or wheat plus 25% wheat co-products. The 5 xylanases (XA, XB, XC, XD and XE) were mono-component preparations of endo-1,4- $\beta$ -xylanase produced by different fungal organisms except XC, which was of bacterial origin. Xylanases were fed at 75 mg of xylanase protein/kg of feed. All diets also contained microbial phytase (500 units/kg). All xylanases and phytase were provided by Danisco Animal Nutrition, Marlborough, Wiltshire, UK. Twelve ileal cannulated barrows (29.3 kg BW) were allotted to two 6  $\times$  6 Latin squares with 6 corn-based and 6 wheat-based diets in each square. Corn and wheat diets had similar levels of insoluble (11.6 vs. 11.0%) non-starch polysaccharides (NSP), but corn diets contained less soluble NSP (1.3 vs. 1.9%) and starch (31.1 vs. 39.0%) than wheat diets. Corn diets containing XD or XE had greater ( $P < 0.05$ ) AID of GE (68.5%) than the control diet (64.6%) or diets containing XB (64.3%) or XC (64.3%), whereas in wheat diets, the diet containing XB had lower ( $P < 0.05$ ) AID of GE (67.9%) compared with diets containing XA, XC, and XD that all had an AID of 72.8% (interaction,  $P < 0.05$ ). Wheat based diets had greater ( $P < 0.05$ ) AID of GE and nutrients compared with corn diets. There was no main effect of diet and no interactions between xylanase and diet for ATTD of GE, but diets containing XE or XD had greater ( $P < 0.05$ ) ATTD of GE compared with the control diets and diets containing XC, whereas results for diets containing XA and XB were intermediate. These data indicate that not all xylanase sources are similar in efficacy at the pig gut level, which may be a result of the unique characteristics of each xylanase.

**Key Words:** digestibility, xylanase, pig

**114 Effects of supplemental xylanase on digesta viscosity, gut health, and growth performance of nursery pigs.** Hongyu Chen<sup>\*1</sup>, Rafael Cabrera<sup>2</sup>, and Sung Woo Kim<sup>1</sup>, <sup>1</sup>North Carolina State University, Raleigh, NC, <sup>2</sup>Huvepharma Inc., Peachtree City, GA.

A total of 40 pigs (10.7  $\pm$  1.2 kg initial BW at 6 wk of age) were used in a 21 d trial to evaluate the effects of supplemental xylanase (Huvepharma Inc., Peachtree City, GA) in nursery diets on digesta viscosity, gut health, and growth performance. Pigs were individually housed and randomly allotted to 4 dietary treatments in a 2  $\times$  2 factorial arrangement (n = 10 per treatment, 0 or 1,500 EPU/kg xylanase and 0 or 30% DDGS as 2 factors). Body weight and feed intake were recorded weekly. Plasma samples were collected on d 19 to measure tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) and malondialdehyde (MDA). On d 21, all pigs were euthanized to collect tissues from duodenum, jejunum, and colon for morphology

and to measure TNF- $\alpha$ , and MDA concentrations. Distal jejunal digesta were collected to measure viscosity. During the entire period, supplementation of xylanase increased ( $P < 0.05$ ) ADG (616 to 660 g/d) of nursery pigs whereas DDGS (0 or 30%) did not affect ADG (628 vs. 648 g/d, respectively). There was no interaction between the 2 factors indicating that the effect of xylanase on ADG was independent to the use of DDGS in the nursery feed. On wk 3, ADFI was increased ( $P < 0.05$ ) when fed DDGS (1,141 to 1,267 g/d) and there was an interaction ( $P < 0.05$ ) between 2 factors indicating that xylanase decreased ADFI when DDGS was used in the feed. Use of DDGS increased ( $P < 0.05$ ) viscosity (1.86 to 2.38 cP), whereas supplemental xylanase reduced ( $P < 0.05$ ) viscosity (2.27 to 1.96 cP) of jejunal digesta. Plasma TNF- $\alpha$  was decreased ( $P < 0.05$ , 108.45 to 69.87 ng/mL) by supplemental xylanase. Use of DDGS reduced ( $P < 0.05$ ) villus height: crypt depth ratio (1.46 to 1.27) in duodenum. Supplemental xylanase increased ( $P < 0.05$ ) the crypt depth (360 to 404  $\mu$ m) in duodenum. Collectively, this study indicates that supplemental xylanase (1,500 EPU/kg) improved growth performance and inflammatory status of nursery pigs by reducing digesta viscosity and plasma TNF- $\alpha$  regardless of the use of DDGS in nursery diets.

**Key Words:** digesta viscosity, pig, xylanase

**115 Effects of supplemental xylanase on growth, gut health, and ileal nutrient digestibility in nursery pigs fed corn-soybean meal-based diets with two dietary energy levels.** I. Park<sup>\*1</sup>, H. Chen<sup>1</sup>, J. Tyus<sup>2</sup>, J. J. Wang<sup>2</sup>, and S. W. Kim<sup>1</sup>, <sup>1</sup>North Carolina State University, Raleigh, NC, <sup>2</sup>BioResource International Inc., Durham, NC.

This study was conducted to determine the effects of supplemental xylanase (Xylamax, BioResource International, Inc., Durham, NC) on growth performance, gut health, and ileal nutrient digestibility in nursery pigs fed corn-soybean meal based diets with 2 dietary energy levels. Pigs (60 barrows and 60 gilts weaned at 21 d of age) had a 4-d adaptation period after weaning. On 25 d of age (7.77  $\pm$  0.63 kg BW), pigs were allotted to 4 treatments (2  $\times$  2 factorials arrangement: 3,427 or 3,323 kcal ME/kg of feed and 0 or 1,400 U of enzyme/kg feed) with 10 pens (5 barrow and 5 gilt pens, and 3 pigs per pen) per treatment in a randomized complete block design, and the feeding period lasted for 3 wks. Growth performance was measured weekly. Titanium dioxide (0.3%) was added to diets from d17 to 21 as an indigestible marker. Blood samples were taken on d 20 to measure tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) and malondialdehyde (MDA). On d 21, 24 pigs (1 from each pen and 6 pens per treatment) were euthanized to obtain the duodenal and jejunal mucosa to measure TNF- $\alpha$  and MDA. Jejunal and ileal digesta were collected to measure viscosity and ileal nutrient digestibility. Data were analyzed using the Mixed procedure in SAS with pen as the experimental unit using treatments and sex as fixed effects and initial BW as a random effect. The G:F was improved ( $P < 0.05$ ) by increasing dietary ME (0.774 to 0.800) and xylanase supplementation (0.775 to 0.799), respectively. Dietary energy levels and xylanase supplementation did not affect viscosity and ileal nutrient digestibility. High ME diet increased ( $P < 0.05$ ) the concentrations of MDA in jejunal mucosa (0.74 to 1.25 nmol/mg protein) and plasma (16.89 to 23.79  $\mu$ M). Supplemental xylanase decreased ( $P < 0.05$ ) MDA (1.02 to 0.88 nmol/mg protein) and TNF- $\alpha$  (3.03 to 2.50 pg/mg protein) concentrations in duodenal mucosa. Collectively, supplemental xylanase (1,400 U of enzyme/kg feed) in corn-soybean meal based pig diets enhances growth



performance and gut health of nursery pigs as indicated by increased feed efficiency and reduced MDA and TNF- $\alpha$  concentrations in duodenal mucosa, respectively.

**Key Words:** growth performance, gut health, xylanase

**116 Porcine in vitro digestion and fermentation characteristics of wheat and wheat millrun without or with xylanase and  $\beta$ -glucanase blend.** Tofuko A. Woyengo<sup>\*1,2</sup>, Elijah Kiarie<sup>3,4</sup>, and Ruurd T. Zijlstra<sup>1</sup>, <sup>1</sup>University of Alberta, Edmonton, AB, Canada, <sup>2</sup>South Dakota State University, Brookings, SD, <sup>3</sup>Danisco Animal Nutrition-DuPont Industrial Biosciences, Waukesha, WI, <sup>4</sup>University of Manitoba, Winnipeg, MB, Canada.

A study was conducted to determine porcine in vitro digestion and fermentation characteristics of the wheat and wheat millrun without or with a with xylanase and  $\beta$ -glucanase blend (XB, Danisco UK Ltd., Marlborough, Wiltshire, UK) that supplied 4,880 units of xylanase and 608 units of  $\beta$ -glucanase per kilogram of treated feedstuff in a completely randomized design. Samples were hydrolyzed in 2 steps using pepsin and pancreatin. Subsequently, residues were incubated in a buffer solution with minerals and fresh pig feces as inoculum. Gas production was measured for 72 h, and modeled to estimate kinetics of gas production. Concentration of VFA per unit weight of residue incubated or feedstuff was measured in fermented solutions. In vitro digestibility of DM (IVDDM) for wheat (77.7%) was greater ( $P < 0.05$ ) than that for wheat millrun (63.2%). Feedstuff and XB interacted ( $P < 0.05$ ) on IVDDM such that XB supplementation increased ( $P < 0.05$ ) IVDDM for wheat, but not for wheat millrun. Total gas production of residue incubated for wheat (176.6 mL/g DM) was greater ( $P < 0.05$ ) than that for wheat millrun (117.4 mL/g DM). Also, total VFA production per unit weight of residue incubated for wheat (3.12 mmol/g DM) was greater ( $P < 0.05$ ) than that for wheat millrun (2.16 mmol/g DM). However, XB did not affect total gas production and total VFA production per unit weight of residue incubated. Total VFA production per unit weight of feedstuff for wheat (0.79 mmol/g DM) was lower ( $P < 0.05$ ) than that for wheat millrun (0.93 mmol/g DM). Feedstuff and XB tended to interact ( $P = 0.096$ ) on total VFA production per unit weight of feedstuff such the XB decreased ( $P < 0.05$ ) total VFA production per unit weight of feedstuff for wheat but not for wheat millrun. In conclusion, wheat was more digestible by porcine pepsin and panceatin than wheat millrun, whereas wheat millrun was more fermentable than wheat. The XB supplementation increased the digestion of wheat, but had limited effect on the digestion and fermentation of wheat millrun.

**Key Words:** wheat and wheat millrun, in vitro fermentation, pig

**117 Supplemental effects of  $\beta$ -mannanase on growth performance, ileal nutrient digestibility, and gut health of nursery pigs.** Hongyu Chen\*, Inkyung Park, Shihai Zhang, and Sung Woo Kim, North Carolina State University, Raleigh, NC.

A total of 84 pigs ( $17.6 \pm 2.8$  kg initial BW at 6 wk of age) were used in a 40-d trial to evaluate effects of dietary supplemental  $\beta$ -mannanase (400 U/kg feed, CTCBIO Inc., Seoul, Korea) on growth performance, ileal nutrient digestibility and gut health of nursery pigs. Pigs were allotted to 2 treatments (14 pens per treatment, 3 pigs per pen) based on a randomized complete block design with sex and initial BW as blocks. Experimental diets were corn (phase 3: 50%, phase 4: 57%), soybean meal (phase 3: 27%, phase 4: 20%), and 20% DDGS based with or without 400 U  $\beta$ -mannanase/kg feed. Growth performance (ADG, ADFI, and G:F) was recorded weekly. Plasma was collected on d 35 to

quantify tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) and malondealdehyde (MDA). On d 35, 0.3% titanium oxide was added as an indigestible marker to the diets for an additional 4 d feeding. On d 40, 16 pigs (1 pig per pen, 8 pens per treatment) were euthanized to collect digesta from jejunum, ileum, and colon (to measure viscosity and pH value) and to collect tissues from duodenum, jejunum and ileum (for morphology, TNF- $\alpha$ , and MDA evaluation). Supplementation of  $\beta$ -mannanase reduced ( $P < 0.05$ ) viscosity of jejunal digesta (2.52 to 1.97 cP, respectively). Supplementation of  $\beta$ -mannanase improved ( $P < 0.05$ ) G:F during phase 3 (0.593 to 0.617) and overall period (0.572 to 0.589), pH of colon digesta (5.99 to 6.33), and ileal digestibility of NDF (31.3 to 41.1%) and ADF (26.8 to 38.7%). Supplementation of  $\beta$ -mannanase increased ( $P < 0.05$ ) ileal villus height (377 to 432  $\mu$ m), decreased ( $P < 0.05$ ) jejunal crypt depth (249 to 212  $\mu$ m), and improved ( $P < 0.05$ ) villus height: crypt depth ratios in jejunum (1.58 to 2.10) and ileum (1.65 to 2.02). Supplementation of  $\beta$ -mannanase tended to reduce ( $P = 0.078$ ) TNF- $\alpha$  concentration (7.94 to 6.46 pg/mg) in jejunal mucosa. Collectively, dietary  $\beta$ -mannanase (400 U/kg) could improve feed efficiency by decreasing digesta viscosity and increasing nutrient digestibility, and could also maintain gut health by improving gut morphology and reducing inflammatory response.

**Key Words:** digesta viscosity, nutrient digestibility,  $\beta$ -mannanase

**118 Effect of superdosing two sources and two levels of phytases on pig performance when supplemented to commercial nursery diets.** Chris Sparks<sup>\*1</sup>, Rafael A. Cabrera<sup>\*1</sup>, Gene Gourley<sup>2</sup>, and Emily Weber<sup>2</sup>, <sup>1</sup>Huvepharma US Inc., Peachtree City, GA, <sup>2</sup>Gourley Research Group LLC, Webster City, IA.

The objective of this study was to determine the effect of superdosing 2 sources and levels of phytases on nursery pig performance in a commercial setting. Phase 1 (crumble/meal mix), phase 2 (common meal) and phase 3 diets were corn-SBM with 4, 10, and 20% DDGS respectively and contained 0.47, 0.40, and 0.35% av. P respectively. They were budgeted at 5.7 and 6.4 kg/pig for phase 1 and 2 and until they moved out of the nursery for phase 3. Six experimental diets were fed: Positive Control (PC), Negative Control (NC, PC minus 0.12% av. P), 500 and 1000 FTU/kg of Optiphos 2000 (Huvepharma USA, Inc.); and 500 and 1000 Quantum Blue 5G (AB Vista) were added to the NC. We had 10 pens per treatment and 21 pigs/pen. Pigs (Geneticpork Fertilis 25 female x PIC Line 359 Sire) were sourced out of a sow farm negative to PPRS, PED virus and *M. hyopneumonea*. Pens were weighed at 7 d interval. Pens were the experimental units and we found no gender by treatment interactions therefore gender was removed from the model. The ANOVA was performed using SAS statistical software (SAS Institute, Cary, NC). From d 0 to 44 (Overall), all phytase treatments had higher ( $P < 0.01$ ) final pig BW, increased ( $P < 0.01$ ) ADG, and higher ( $P < 0.01$ ) ADFI when compared with either PC or NC. Positive Control and NC treatments were not different for those traits. All phytase treatments had decreased ( $P = 0.03$ ) FCR when compared with the PC but were not different than the NC. There were not significant differences in any performance traits between Optiphos and Quantum Blue. Increasing the levels of phytases from 500 FTU/kg of complete feed to 1000 FTU/kg of complete feed with either source did not result in improved performance. In this trial, 500 FTU/kg of complete feed performed as well as 1000 FTU/kg of complete feed of either Optiphos or Quantum Blue for commercially raised nursery pigs.

**Key Words:** nursery pig, superdosing, phytase level

**119 Effects of superdosing of microbial phytase in diets for weanling pigs.** T. D. Crenshaw\*<sup>1</sup>, O. Adeola<sup>2</sup>, M. J. Azain<sup>3</sup>, S. K. Baidoo<sup>4</sup>, S. D. Carter<sup>5</sup>, G. M. Hill<sup>6</sup>, S. W. Kim<sup>7</sup>, P. S. Miller<sup>8</sup>, M. C. Shannon<sup>9</sup>, and H. H. Stein<sup>10</sup>, <sup>1</sup>University of Wisconsin, Madison, WI, <sup>2</sup>Purdue University, Lafayette, IN, <sup>3</sup>University of Georgia, Athens, GA, <sup>4</sup>University of Minnesota, Minneapolis, MN, <sup>5</sup>Oklahoma State University, Stillwater, OK, <sup>6</sup>Michigan State University, East Lansing, MI, <sup>7</sup>North Carolina State University, Raleigh, NC, <sup>8</sup>University of Nebraska, Lincoln, NE, <sup>9</sup>University of Missouri, Columbia, MO, <sup>10</sup>University of Illinois, Urbana-Champaign, IL.

An experiment using 390 crossbred pigs (20 replications of 3 or 4 pigs/pen) was conducted at 10 research stations to assess the effects of superdosing phytase on nursery pig growth performance. Corn-soybean meal phase 2 nursery diets were fed for 21-d from 8.5 to 17.6 kg BW. A common premix was formulated using the NCCC42 formulations, and mixed at a central location. Diets were formulated to contain 1.35% standardized ileal digestible (SID) Lys. The positive control diet contained 0.8% Ca, 0.69% P and 0.40% standardized total-tract digestible (STTD) P. The negative control diet contained 0.50% Ca, 0.42% P and 0.18% STTD P. Dietary phytase (Ronozyme HiPhos) was added to the negative control diet premix to provide 0, 500, 1,500 or 3,000 phytase units (FTU)/kg complete diet. On d21, one pig per pen was killed and the 3rd and 4th metacarpals from the right front foot were analyzed for total bone ash. Data were analyzed using the Glimmix procedure of SAS. Pigs were randomly selected based on weight, age, sex, and ancestry. Station by treatment effects were observed for ADG in wk 2 and for the entire period, and for ADFI in wk 2, wk 3, and for the entire period. Body weight gain linearly increased as increased concentrations of phytase were added to the negative control diet (360, 440, 450, and 470 g/d;  $P < 0.01$ ), but ADFI (640, 710, 700, 720 g/d) was not affected during the 21-d study. A linear increase ( $P < 0.05$ ) in G:F was observed as phytase additions increased (0.56, 0.62, 0.64, 0.65 g/g for pigs fed diets containing 0, 500, 1,500, or 3,000 FTU of phytase). Percent bone ash also increased (linear,  $P < 0.01$ ) as addition of phytase increased (44.6, 48.4, 49.2, and 50.6% for pigs fed diets containing 0, 500, 1,500 or 3,000 FTU of phytase, respectively). Percent bone ash was not different between pigs fed the positive control diet and the negative control diet containing 3,000 FTU/kg of phytase (51.1 vs. 50.5%, respectively). In this 21-d study, feeding phase 2 nursery diet containing up to 3,000 FTU/kg resulted in similar growth performance and bone ash percentage as pigs fed a positive control diet containing 0.8% Ca, 0.69% P and 0.40% STTD P.

**Key Words:** phosphorus, pig, phytase

**120 Effect of water-to-feed ratio on the digestibility of diets fed to growing-finishing pigs.** Cinta Sol\*, Lorena Castillejos, Ramon Muns, and Josep Gasa, *Animal Nutrition and Welfare Service, Department of Animal and Food Sciences, Universitat Autònoma de Barcelona, Bellaterra, Spain.*

Dilution rates water to feed may affect the apparent total-tract digestibility (ATTD) of barley-wheat diets fed to growing-finishing pigs. The experiment lasted 52 d and it was divided in 2 studies of 26 d each. In the first study 16 female pigs (46.7 ± 1.98 kg of body weight, BW) were individually allotted and assigned to 1 of 4 treatments (n = 4). Treatments consisted in 4 water-to-feed ratios: in dry form (CON), with no added water, and ratios of 0.6:1, 2.1:1 and 2.7:1. In the second study the same animals (65.4 ± 3.14 Kg of BW) were again assigned to 4 new treatments: CON and 1.35:1, 2.7:1 and 3.5:1 ratios. Each pen was provided with a free-choice nipple drinker. Diet was fed close to “ad

libitum” trying to avoid refusals, a 0.3% of TiO<sub>2</sub> was added to the feed as an indigestible marker and fecal grab samples were collected from the rectum twice a day the 2 last days of each study. Data of ATTD of OM, GE and nutrients were analyzed by one-way ANOVA using the GLM procedure of SAS and differences among mean values were assessed by the Tukey *t*-test. In the first study, pigs fed diets with ratios 2.1:1 and 2.7:1 had higher organic matter digestibility (OMd) and gross energy digestibility (GEd) than CON ( $P < 0.05$ ). Pigs fed diets with a ratio of dilution 2.7:1 also had higher ( $P < 0.05$ ) intake of digestible organic matter (DOMi) and digestible energy (DEi) than pigs fed CON. In the second study, pigs fed water diluted diets (the 3 ratios) had better OMd and GEd than CON ( $P < 0.05$ ) but no differences were found in DOMi. Compared with CON, the dilution of 3.5:1 also showed a higher ( $P < 0.05$ ) digestibility coefficients for crude protein (CP), ether extract (EE) and crude fiber (CF) and the dilution 1.35:1 and 2.7:1 also for CF. Using a quadratic regressions approach, the maximum feed OM and GE digestibility coefficient was found with dilution rates of 1.83:1 and 2.70:1 for the first and second study, respectively. In conclusion, the water-to-feed ratio which optimized OM and GE digestibility varies depending on the age of the animals; being lower ratios better for young pigs and higher for finishing pigs. Despite the fact that dilution rates below 2.5–2.7:1 are not usually achieved in liquid feeding commercial farms.

**Key Words:** water-to-feed ratio, growing-finishing pig, nutrient digestibility

**121 Effects of pelleting and extrusion on energy digestibility in pig diets containing different levels of fiber.** Oscar J. Rojas\*<sup>1</sup>, Ester Vinyeta<sup>2</sup>, and Hans H. Stein<sup>1</sup>, <sup>1</sup>University of Illinois, Urbana, IL, <sup>2</sup>Bühler AG, Uzwil, Switzerland.

An experiment was conducted to determine effects of pelleting, extrusion, and extrusion plus pelleting on energy and nutrient digestibility in diets containing low, medium, or high levels of fiber. Three diets were formulated: (1) a low fiber diet contained corn and soybean meal; (2) a medium fiber diet contained corn, soybean meal, and 25% distillers dried grains with solubles (DDGS); and (3) a high fiber diet contained corn, soybean meal, 25% DDGS, and 20% soybean hulls. Each diet was divided into 4 batches after mixing and the treatments included the following: (1) no further processing; (2) pelleted at 85°C; (3) extruded at 115°C using a single screw extruder; and (4) extruded at 115°C and then pelleted at 85°C. Thus, 12 different diets were produced. A total of 24 growing pigs (initial BW: 26.5 ± 1.5 kg) with a T-cannula fitted in the distal ileum were allotted to the diets in a split-plot design with 8 pigs allotted to the low fiber diets; 8 pigs were allotted to the medium fiber diets; and 8 pigs were allotted to the high fiber diets. Diets were fed to the pigs during 4 14-d periods. Within each type of diet, the 8 pigs were fed the diets produced using the 4 processing technologies. Thus, there were 8 replicate pigs per diet. Each of the 4 14-d periods consisted of 5 d for adaptation, 5 d of fecal collection, and ileal digesta were collected on d 13 and 14 of each period. Results indicated that pelleting, extrusion, or extrusion and pelleting improved ( $P < 0.05$ ) the apparent ileal digestibility (AID) of starch and most indispensable AA. In most cases, there were no differences between the pelleted, the extruded, and the extruded plus pelleted diets. The apparent total-tract digestibility (ATTD) of energy and the ME of the diets were also improved ( $P < 0.05$ ) by pelleting and by the combination of extrusion plus pelleting, and extrusion alone improved ATTD of energy and ME in the low fiber and the high fiber diets. Results of this experiment indicate that energy utilization may be improved by pelleting or extrusion or by the

combination of the 2 technologies, but the response to extrusion seems to be greater in diets that are relatively high in fiber.

**Key Words:** extrusion, fiber, pelleting

**122 Mix time does not affect growth performance in finishing pigs fed meal and pelleted diets with high inclusion (32% DDGS and 32% wheat midds) of alternative ingredients.** Megan E.

Morts\*, Joe D. Hancock, C. Greg Aldrich, Charles R. Stark, Joel D. McAtee, and Kayla L. Kohake, *Kansas State University, Manhattan, KS.*

A total of 200 finishing pigs (average initial BW of 72 kg) were used in a 51-d growth assay to determine the effects of mix time in meal and pelleted diets with high inclusion of alternative ingredients (32% DDGS and 32% wheat middlings) on growth performance and carcass measurements. The pigs were sorted by gender and ancestry and assigned to pens (5 pigs/pen and 10 pens/treatment) in a completely randomized design. The diets (907 kg batches) were blended in a Hayes & Stolz twin-shaft double-ribbon mixer. All ingredients (corn, soybean meal, DDGS, wheat middlings, limestone, salt, vitamin mix, mineral mix, crystalline amino acids, and tylosin) were added to the stopped mixer. Mix times were 0 and 180 s before transfer to sack-off or being pelleted before transfer to sack-off. Treatments were arranged as a 2 × 2 factorial with main effects of diet form (meal vs. pellets) and mix time (0 vs. 180 s). Diets were formulated to at least 120, 120, and 110% of requirements for essential amino acids, vitamins, and minerals, respectively, as suggested in the NRC (2012) Nutrient Requirements of Swine. Feed and water were consumed on an ad libitum basis until the pigs were harvested (avg BW of 119 kg) at a commercial abattoir for collection of carcass data. There were no interactions among diet form and mix time ( $P = 0.45$  or greater). Pelleting improved ( $P < 0.01$ ) ADG and G:F, but did not affect ( $P = 0.49$  or greater) hot carcass weight (HCW), last rib backfat thickness (BF), or dressing percentage (dress %). Mix time did not affect ( $P = 0.17$  or greater) ADG, G:F, HCW, BF, or dress %. In conclusion, increasing mix time from 0 to 180 s in meal or pelleted diets with high inclusion of DDGS and wheat middlings did not improve growth performance or carcass characteristics.

**Table 1 (Abstr. 122).** Effects of diet form (meal vs. pellet) and mix time (0 vs. 180 s) on performance and carcass traits

Item	Mash		Pellet		SE	P-value		
	0 s	180 s	0 s	180 s		Form	Mix	Form x Mix
ADG, g	909	893	969	944	28	0.01	0.17	0.76
G:F, g/kg	294	298	312	319	5	0.01	0.31	0.78
HCW, kg	87.4	86.9	88.2	88.1	3.2	0.49	0.84	0.90
Dress %	73.3	72.9	72.8	73.1	0.5	0.71	0.98	0.45
BF, mm	23	22	23	22	2	0.76	0.54	0.66

**Key Words:** duration of mixing, DDGS, pigs

**123 Effect of sieving methodology on determining particle size of ground corn, sorghum, and wheat by sieving.** Julie R. Kalivoda\*, Cassandra K. Jones, and Charles R. Stark, *Kansas State University, Manhattan, KS.*

Particle size determination by sieving is a standard method used to estimate the geometric mean (dgw) and geometric standard deviation (Sgw) of feedstuffs. However, the method allows for variation in time of sieving, sieve agitator setup, and the use of dispersing agent (fumed silica). The objectives of this experiment were to determine which method of particle size analysis best estimates the particle size of cereal grain and assess analytical variation within each method. Treatments were arranged in a 4 × 3 × 3 × 2 factorial design with 4 sieving methods: (1) 10 min with sieve agitators (10A), (2) 15 min with no sieve agitators (15N), (3) 15 min with sieve agitators (15A), or (4) 15 min with sieve agitators and a dispersing agent (15AS), 3 cereal grains (corn, wheat, or sorghum), 3 grinds (coarse, medium, or fine), and 2 mills (hammermill or roller mill). Mill parameters were adjusted to achieve 3 grinds with coarse being the largest and fine being the smallest. Samples were analyzed for particle size according to ASAE S319.3. Data were analyzed using GLIMMIX procedure of SAS with 4 replicates per treatment. Interactions were removed from the model if  $P > 0.05$ . The main effects of method, grain, grind size, and grind method all differed ( $P < 0.01$ ) for both dgw and Sgw. All methods were significantly different from one another ( $P < 0.05$ ; 587, 615, 576, and 541 for dgw and 2.23, 2.09, 2.27, and 2.63 for Sgw for 10A, 15N, 15A, and 15AS, respectively). All grains differed from one another, with corn having the smallest dgw and Sgw ( $P < 0.05$ ; 537 and 2.28, 550 and 2.34, 652 and 2.31, respectively for corn, sorghum, and wheat dgw and Sgw). Hammermill ground grain had a smaller dgw and larger Sgw than roller mill ground grain ( $P < 0.05$ ; 415 and 2.62 vs. 744 and 1.99, respectively). As expected, varying grind size resulted in different dgw ( $P < 0.05$ ), but Sgw was also affected as the coarse and medium grinds had greater Sgw than the fine grind (2.36 and 2.35 vs. 2.23, respectively). In summary, the particle size analysis method of 15AS provided the lowest dgw and highest Sgw, suggesting that all particles had reached their appropriate spot within the sieve stack and thus was the most accurate of the tested methods.

**Key Words:** corn, grain, particle size analysis



# Physiology and Endocrinology: Reproduction and estrous synchronization

## 124 Characterization of endometrial immune cells adjustments along pregnancy in the cow.

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Successful of embryo implantation requires synchronization between maternal and fetal factors to promote endometrial receptivity. In this scenario, the maternal immune system may play an important role in the establishment as well in the development of conceptus until onset of parturition in eutherian mammals. Our study aimed to identify and quantify the subpopulations of immune cells in the bovine pregnant endometrium. We hypothesized that the endometrial immune cells pool change as pregnancy progresses. Samples from early ( $33 \pm 1.5$  d), mid ( $170 \pm 16.9$  d) and late pregnancy ( $227 \pm 10.8$  d) were obtained from a local abattoir ( $n = 5$ , per group). Cryostat sections were labeled for CD4 (T helper), CD8 (T cytotoxic), CD14 (macrophage), CD25 (activation T cells) and WC1 ( $\gamma$ d). The samples were analyzed under epifluorescence microscope. The number of positive cells was counted and normalized to the total number of cells per  $\text{mm}^2$  in 5 random fields for each group/ marker. Data were analyzed separately for each marker by least square ANOVA using the GLM procedure of SAS. The WC1<sup>+</sup> cells were most prominent in the endometrial stroma, with greater cell number in mid than in early and late stages of pregnancy ( $P < 0.01$ ). Moreover, CD4<sup>+</sup> ( $P = 0.09$ ) and CD8<sup>+</sup> ( $P < 0.01$ ) decreased in number in late pregnancy, whereas CD25<sup>+</sup> cells were low only in mid-pregnancy ( $P = 0.02$ ). CD14<sup>+</sup> cells increased, respectively, 3- and 4-fold in mid and late stages of pregnancy ( $P < 0.01$ ). Our results show that there is a deviation of endometrial immune cells subpopulations during pregnancy in the cow. In early pregnancy, there is a predominance of immune cells involved in regulation of immune response (CD3, CD4 and CD8) to allow embryo attachment and survival; whereas in mid-pregnancy, number of cells with trophic function are increased (CD14 and WC1) likely to allow placental development. In late pregnancy, there is a recruitment of cells involved in inflammatory response to induce placental detachment and parturition (CD14 and CD25).

**Key Words:** endometrial immune cell, pregnancy, bovine

## 125 Expression of nonclassical MHC-I isoforms is deregulated in cloned placenta.

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Trophoblast cell evades the maternal immune recognition by expressing non-classical (NC) MHC-I isoforms, in humans and mice. In the cow, MHC-I proteins are expressed from 120 d of pregnancy, however in clones MHC-I expression can begin as early as d 35. This abnormal expression of MHC-I in clones may play a role on high rates of embryo/fetal losses in this model. Our study aimed to determine expression of MHC-I and its NC isoforms in normal and cloned bovine pregnancy. Samples of normal and cloned bovine placenta in early ( $n = 5$ ) and term ( $n = 6$ ) pregnancy were used for immunolocalization of MHC-I by IHC, using antibodies for bovine MHC-I (IL-A88) and murine NC isoform

(Qa-2). Also, the expression of classical MHC-I (JSP-1) and NC isoforms (NC1–4) was analyzed by qRT-PCR. Data were analyzed by least square ANOVA using the GLM procedure of SAS. The model included main effects (group and animal) and standard errors. In normal placenta, NC1 was equally expressed in early and late pregnancy, whereas in clones, NC1 tended to increase expression in late pregnancy ( $P = 0.07$ ). NC2 was higher expressed during early pregnancy in clones than early normal pregnancy ( $P = 0.03$ ) and both normal and clone late pregnancies ( $P = 0.04$ ). For NC3 expression showed interaction between type and stage of pregnancy ( $P = 0.01$ ). The expression of NC3 was higher in later in normal pregnancy ( $P < 0.01$ ), while in clones the NC3 expression was higher in early stages ( $P = 0.04$ ). Neither NC4 and JSP1 showed changes in expression among groups. Immunohistochemistry analysis showed that IL-A88 stained the maternal epithelium and non-invading trophoblast but not trophoblast giant cells. In early pregnancy, IL-A88 staining was weak in normal and almost absent in cloned placenta. At term, IL-A88 staining increased in normal placenta. For Qa-2, no staining was detected in early pregnancy in normal whereas the cloned placenta showed strong staining. Furthermore, in the term pregnancy the Qa-2 staining did not differ between normal and cloned placenta. Altogether, our data suggest that the MHC-I expression is dysregulated in clone pregnancy, which may contribute to their low pregnancy rates and success.

**Key Words:** nonclassical MHC, bovine, cloned placenta

## 126 Behavioral and hormonal pattern around estrus and the characteristics of preovulatory follicles of repeat breeder dairy cows.

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Repeat breeding in dairy cows results in large economic loss; however, the etiology of this phenomenon remains elusive. The objectives were to examine the behavioral and endocrine patterns around estrus, and the characteristics of preovulatory follicles in repeat breeder (RB) vs. normal (CTL) cows. The CTL cows were  $>60$  d in lactation, cycling and not inseminated, and a cow was considered as RB if it did not become pregnant after at least 4 successive inseminations (average  $7.0 \pm 2.0$ ), with normal intervals. A total of 27 and 31 cycles in 12 RB and 18 CTL cows, respectively, were synchronized. Data were analyzed as repeated measurements by the MIXED procedure of SAS, and the model included the effects of treatment, cow and cluster. Behavioral estrus and ovulation were observed in 81.5 and 83.8% of the synchronized cycles in the RB and CTL cows, respectively. The RB and CTL cows had similar estrus durations of 21.4 and 19.6 h, respectively, but estrus was more intense in the RB, as indicated by numerically higher overall activity indexes and higher peak neck activity. The interval from PG injection to estrus onset (i.e., proestrus) was 8.2 h shorter in RB than in CTL cows (47.9 and 56.1 h, respectively;  $P < 0.007$ ), but the average preovulatory follicle size was similar. Plasma estradiol ( $E_2$ ) concentration at peak was numerically higher (21%) and the AUC tended to be higher in the RB than in the CTL cows. Also, LH secretion during the period from 18 to 3 h before the LH peak was lower in RB than in CTL: 2.5 and 4.6 ng/mL, respectively ( $P < 0.01$ ). In another study, 16 preovulatory follicles (8 CTL and 8 RB) were aspirated 48 h after PG injection. No differences were observed in androstenedione and progesterone ( $P_4$ ) in follicular fluid, but concentrations were 1.7 fold higher ( $P < 0.005$ )

and  $E_2/P_4$  ratio tended to be higher ( $P < 0.07$ ) in RB than in CTL cows. In conclusion, better estrus expression, similar follicular diameter and higher  $E_2$  in RB indicate that the etiology of repeat breeding lies beyond these parameters. However, short proestrus and subdued LH concentrations before the LH peak, which could impair oocyte competence and development, are first reported in RB cows.

**Key Words:** repeat breeders, estrus intensity, LH secretion

**127 Effects of dry period length on onset of ovarian activity and ovarian cyclicity in the subsequent lactation.** Juncai Chen<sup>\*1</sup>, Nicoline M. Soede<sup>1</sup>, Gerrit J. Remmelink<sup>2</sup>, Bas Kemp<sup>1</sup>, and Ariette T. M. Van Knegsel<sup>1</sup>, <sup>1</sup>*Adaptation Physiology Group, Wageningen University, Wageningen, the Netherlands*, <sup>2</sup>*Livestock Research, Wageningen University and Research Centre, Wageningen, the Netherlands*.

Negative energy balance (NEB) caused by high milk yield and insufficient feed intake in early lactation has been related to compromised cow health and fertility. Recent studies show that the NEB in early lactation could be alleviated by omitting or shortening dry period in dairy cows. It can therefore be hypothesized that omitting or shortening dry period improves fertility in dairy cows. The objective of this study was to evaluate the effects of dry period length on onset of ovarian activity and ovarian cyclicity in Holstein-Friesian dairy cows within 100 d in milk (DIM). The cows (60 primiparous and 108 multiparous) were randomly assigned to one of 3 dry period lengths (0, 30, and 60 d). Milk samples were collected 3 times a week for progesterone concentration analysis until 100 DIM after calving. Onset of luteal activity (OLA) was defined as the occurrence of at least 2 succeeding milk samples with progesterone concentrations  $\geq 2$  ng/mL. Normal resumption of ovarian cyclicity was defined as onset of first luteal activity occurring within 45 DIM, and followed by regular ovarian cycles of 18 to 24 d in length. Data are expressed as percentage or LSMEANS  $\pm$  SEM. Within 100 DIM postpartum, cows with a 0-d dry period had greater incidence of normal resumption of ovarian cyclicity compared with cows with a 60-d dry period (53.2% vs. 26.0%;  $P = 0.01$ ). Cows with a 30-d dry period tended to have greater incidence of normal resumption of ovarian cyclicity compared with cows with a 60-d dry period (47.7% vs. 26.0%;  $P = 0.09$ ). In addition, cows with a 0-d dry period tended to have shorter interval from calving to first commencement of luteal activity (23.1 vs. 28.9  $\pm$  2.0 d;  $P = 0.07$ ) compared with cows with a 60-d dry period. Overall, our results demonstrate that omitting dry period improves resumption of ovarian cyclicity within 100 DIM in the subsequent lactation.

**Key Words:** continuous milking, progesterone, ovulation

**128 Effect of extending the duration of the postpartum voluntary waiting period on reproductive performance of lactating dairy cows.** Matias L. Stangaferro<sup>\*1</sup>, Robert Wijma<sup>1</sup>, Magdalena Masello<sup>1</sup>, Mark J. Thomas<sup>2</sup>, and Julio O. Giordano<sup>1</sup>, <sup>1</sup>*Department of Animal Science, Cornell University, Ithaca, NY*, <sup>2</sup>*Dairy Health & Management Services, Lowville, NY*.

Our objective was to investigate the effect of extending the duration of the voluntary waiting period (VWP) on reproductive performance of lactating dairy cows. Holstein cows [ $n = 1,082$ ; 434 primiparous (PP) and 648 multiparous (MP)] from 3 commercial dairy farms in New York were blocked by parity and total milk production in their previous lactation (MP only) and were randomly assigned to receive first timed AI (TAI) service at 60  $\pm$  3 (SVWP) or 88  $\pm$  3 DIM (LVWP) after synchronization of ovulation with the Double-Ovsynch protocol

(GnRH-7 d-PGF-3 d-GnRH-7 d- GnRH-7 d-PGF-56 h-GnRH-16 h-TAI). Pregnancy diagnosis was conducted by rectal palpation or transrectal ultrasound 39 and 90 d after AI. Data for pregnancies per AI (P/AI) at 39 d after TAI, pregnancy loss (PL) from 39 to 90 d after AI, and percentage of cows pregnant up to 150 DIM were analyzed by logistical regression using PROC GLIMMIX of SAS. Models included the effect of treatment, parity, and their interaction. Farm was included as a random effect in all models. At 39 d after TAI, P/AI were greater ( $P < 0.01$ ) for cows in the LVWP (46.0%; 228/496) than cows in the SVWP (38.4%; 225/586) group. Pregnancies per AI were greater ( $P < 0.01$ ) for PP (49.5%; 215/434) than for MP (36.7; 238/648) cows. Pregnancy loss, was similar ( $P = 0.88$ ) for cows in the LVWP (4.5%) and the SVWP (4.7%) group, and for PP (4.6%) and MP (4.6%) cows ( $P = 0.97$ ). The percentage of cows pregnant by 150 DIM was similar ( $P = 0.23$ ) for cows in the LVWP (62.6%; 278/444) and the SVWP (69.6%; 261/375) group but were greater ( $P < 0.01$ ) for PP (74.5%; 222/298) than for MP (62.6%; 278/444) cows. The interaction between treatment and parity group did not affect ( $P > 0.05$ ) P/AI, PL, or percent pregnant by 150 DIM. We conclude that extending the duration of the VWP from 60 to 88 DIM for cows receiving TAI after synchronization with the Double-Ovsynch protocol increased overall P/AI. Also, the initial differences in P/AI after first service were compensated by earlier re-insemination of cows in the SVWP group. Supported by New York Farm Viability Institute project AOR13006.

**Key Words:** timed AI, first service, dairy cow

**129 Cows under heat stress have increased uterine size, reduced circulating progesterone, and decreased fertility compared with cows in cooler conditions.** Giovanni M. Baez<sup>\*1,2</sup>, Rafael V. Barletta<sup>1</sup>, Eduardo Trevisol<sup>1</sup>, Jerry N. Guenther<sup>1</sup>, João P. Ferreira<sup>3</sup>, and Milo C. Wiltbank<sup>1</sup>, <sup>1</sup>*University of Wisconsin-Madison, Madison, WI*, <sup>2</sup>*Universidad Francisco de Paula Santander, Cucuta, NS, Colombia*, <sup>3</sup>*São Paulo State University, Botucatu, SP, Brazil*.

Seasonal differences in progesterone (P<sub>4</sub>) concentrations and pregnancies per AI (P/AI) have been demonstrated in lactating dairy cows. Moreover, an association between uterine size and P/AI has been recently reported. Our objective was to evaluate seasonal differences in uterine size, circulating P<sub>4</sub>, and fertility in primiparous and multiparous dairy cows. Lactating Holstein cows ( $n = 704$ ) were synchronized to receive timed AI (TAI) on d 81  $\pm$  3 of lactation by using the Double-Ovsynch protocol (GnRH-7d-PGF-3d-GnRH-7d-GnRH-7d-PGF-56h-GnRH-16h-TAI). At the time of the last injection of PGF, uterine diameter was determined at the greater curvature using ultrasound, uterine length was determined by rectal palpation, and uterine volume was calculated. Circulating P<sub>4</sub> at final PGF and GnRH were evaluated to assure synchronization of all cows used in the final analysis ( $n = 616$ ; primiparous (P),  $n = 289$ ; multiparous (M),  $n = 327$ ). At the same time, respiratory rate (number of breaths per minute, BPM) and body temperature was determined in all cows. Fischer's exact test and *t*-test were used to analyze categorical and continuous variables respectively, and logistic regression analysis was used to calculate probabilities of P/AI related to uterine volume. There was a clear delineation in average respiratory rate (40 to 28 BPM) indicating a change of season (hot (H)  $n = 226$  (112 P, 114 M), cool (C)  $n = 390$  (177 P, 213 M)). Uterine volume was greater in H than C season for all cows (150.3  $\pm$  2.9 vs. 131.2  $\pm$  1.9 mm<sup>3</sup>, H vs. C,  $P < 0.0001$ ), and for P (134.6  $\pm$  4.1 vs. 122.0  $\pm$  2.8,  $P = 0.009$ ) or M (165.7  $\pm$  4.2 vs. 138.8  $\pm$  2.6,  $P < 0.0001$ ) cows. Circulating P<sub>4</sub> was lower during H than C season in P (7.5  $\pm$  0.3 vs. 8.1  $\pm$  0.2,  $P = 0.08$ ) and M (6.0  $\pm$  0.3 vs. 6.8  $\pm$  0.2,  $P = 0.02$ ) cows. The P/AI was reduced in H vs. C for all cows (38.5% vs. 47.7%,  $P = 0.02$ ), and for P (41.1%

vs. 56.5%,  $P = 0.03$ ) but not for M (36.0% vs. 40.4%,  $P = 0.21$ ) cows. Logistic regression analyses indicated a relationship between uterine volume and P/AI for M cows in H ( $P = 0.03$ ) and C ( $P = 0.03$ ) seasons but for P cows only in C season ( $P = 0.02$ ) with greater uterine volume associated with reduced P/AI. Thus, heat stress is associated with an increase in uterine size. Greater uterine size partially explains differences in fertility within parities and seasons.

**Key Words:** fertility, uterine size, heat stress

**130 Effect of heat stress during pregnancy on intact and adrenal de-medullated fetuses: Placental, fetal, and mammary development in ewes.** Antoni Macko\*, Sean Limesand, and Robert Collier, *University of Arizona, Tucson, AZ.*

It is well established that heat stress during mid- and late-gestation induces intrauterine growth retardation (IUGR) and decreased milk yield in the subsequent lactation in cattle and sheep. We hypothesized that elevated fetal adrenal norepinephrine (NE) secretion contributes to impaired mammary development in heat stressed pregnant ewes. Pregnant ewes were assigned to one of 4 treatment groups that were a combination of control (C) or hyperthermia-induced IUGR (I) and surgical sham (S) or bilateral fetal adrenal demedullation (D) at 98 d gestational age (dGA; term = 148 dGA)(n = 4 CS, 4 CD, 6 IS, and 4 ID fetuses). At 134 dGA, fetal plasma NE was measured, animals were euthanized, and fetal and placental weights obtained. Ewes' mammary glands were collected, weighed and prepared for analyses of DNA content, and histological evaluation to quantify the numbers of alveolar units per microscopic field and mammary epithelial cells per alveolus. Data were analyzed by Proc Mixed ANOVA, SAS 9.3. In the IS group, fetal plasma NE was 4-fold higher ( $P < 0.05$ ) and placental, fetal and maternal mammary weights and mammary DNA were lower ( $P < 0.05$ ) compared with the CS group and these parameters were partially reversed in the ID group. Placental weight: CS 337 ± 34, CD 271 ± 39, IS 160 ± 36, ID 215 ± 50 g; Fetal weight: CS 3495 ± 214, CD 3343 ± 233, IS 1746 ± 330, ID 2574 ± 296 g; Mammary wet weight: CS 3043 ± 445, CD 2496 ± 345, IS 785 ± 300, ID 1326 ± 316 g; Mammary dry weight: CS 343 ± 34, CD 353 ± 59, IS 192 ± 44, ID 278 ± 58 g; Mammary DNA content: CS 3043 ± 456, CD 2946 ± 353, IS 868 ± 353, ID 1327 ± 322 ng/μl. There were no treatment effects on the number of alveolar units per field: CS 33.6 ± 9.6, CD 17.3 ± 4.0, IS 16.5 ± 5.0, ID 28.5 ± 3.9; or number of cells per alveolus: CS 6.5 ± 1.0, CD 7.5 ± 2.0, IS 8.2 ± 0.3, ID 6.4 ± 1.0. We conclude that heat stress during gestation reduces placental, fetal and mammary development and fetal adrenal de-medullation partially reverses these effects. We also propose that an endocrine signal from the placenta regulates maternal mammary growth during gestation and this signal is responsive to fetal catecholamines.

**Key Words:** heat stress, placenta, mammary gland

**131 Timing of GnRH administration based on estrous response in beef heifers following administration of the 14-d CIDR-PG protocol with split-time AI.** Brianne E. Bishop\*, Jordan M. Thomas, Jillian M. Abel, Mark R. Ellersieck, Scott E. Pooch, Michael F. Smith, and David J. Patterson, *University of Missouri, Columbia, MO.*

The experiment was designed to evaluate timing of GnRH administration in beef heifers based on estrous status with split-time AI. Estrus was synchronized in 816 heifers across 4 locations by using the 14-d CIDR-PG protocol (CIDR insert [1.38 g progesterone] on d 0 with removal on d 14; 25 mg PGF<sub>2α</sub> [PG] 16 d after CIDR removal on d 30; and 100 μg

GnRH depending on treatment). Estrous detection aids (Estroject) were applied at PG on d 30, with estrus recorded at 66 and 90 h after PG on d 33 and 34, respectively. Treatments were balanced across locations using tract score and weight. Timing of insemination was based on expression of estrus 66 h after PG. Heifers in each treatment that exhibited estrus by 66 h were inseminated; whereas, AI was delayed 24 h until 90 h after PG for heifers failing to exhibit estrus by 66 h. Heifers in treatment 1 were administered GnRH 66 h after PG irrespective of estrus expression; whereas, in treatment 2, heifers were administered GnRH coincident with delayed insemination only if not detected in estrus at 66 h after PG. Data were analyzed using PROC FREQ in SAS. There was no effect of treatment on overall estrous response (1 = 85%; 2 = 87%;  $P = 0.49$ ) or AI pregnancy rate (1 = 55%; 2 = 58%;  $P = 0.54$ ). There were no differences between treatments in estrous response at 66 h (1 = 70%; 2 = 69%;  $P = 0.64$ ); and pregnancy rate resulting from AI for heifers inseminated at 66 h was not influenced by GnRH (1 = 62%; 2 = 64%;  $P = 0.65$ ). Furthermore, there were no differences between treatments in estrous response during the 24 h delay period (1 = 50%; 2 = 58%  $P = 0.22$ ), or pregnancy rate resulting from AI (1 = 40%; 2 = 44%;  $P = 0.55$ ). In summary, when split-time AI is used in conjunction with the 14 d CIDR-PG protocol, administration of GnRH at AI to heifers that exhibit estrus by 66 h after PG is not warranted. These data suggest, however, that among heifers for which AI is delayed based on failure to exhibit estrus by 66 h after PG, timing of GnRH administration (66 vs. 90 h after PG) may be more flexible.

**Key Words:** estrous synchronization, split-time artificial insemination, beef heifer

**132 Timing of GnRH administration based on estrous response in beef cows following administration of the 7-d CO-Synch + CIDR protocol with split-time AI.** Brianne E. Bishop\*, Jordan M. Thomas, Jillian M. Abel, Mark R. Ellersieck, Scott E. Pooch, Michael F. Smith, and David J. Patterson, *University of Missouri, Columbia, MO.*

The experiment was designed to evaluate timing of GnRH administration in beef cows based on estrous status with split-time AI. Estrus was synchronized in 622 cows across 6 locations by using the 7-d CO-Synch + CIDR protocol (100 μg GnRH + CIDR insert [1.38 g progesterone] on d 0; 25 mg PGF<sub>2α</sub> [PG] at CIDR removal on d 7; and 100 μg GnRH depending on treatment). Estrous detection aids (Estroject) were applied at CIDR removal and PG on d 7, with estrus recorded at 66 and 90 h after PG on d 10 and 11, respectively. Treatments were balanced across locations; cows within location were randomly assigned to one of 2 treatments based on age, BCS, and days postpartum. Timing of AI was based on expression of estrus 66 h after PG. Cows in each treatment that exhibited estrus by 66 h were inseminated; whereas AI was delayed 24 h until 90 h after PG for cows failing to exhibit estrus by 66 h. Cows in treatment 1 were administered GnRH 66 h after PG irrespective of estrus expression; whereas in treatment 2, cows were administered GnRH coincident with delayed AI only if not detected in estrus at 66 h after PG. Data were analyzed using PROC FREQ in SAS. Treatment affected overall estrous response (1 = 85%; 2 = 90%;  $P = 0.04$ ) but did not affect total AI pregnancy rate (1 = 58%; 2 = 57%;  $P = 0.89$ ). There were no differences between treatments in estrous response at 66 h (1 = 73%; 2 = 75%;  $P = 0.47$ ); and pregnancy rate resulting from AI for cows inseminated at 66 h was not influenced by GnRH (1 = 63%; 2 = 59%;  $P = 0.50$ ). Estrous response during the 24 h delay period differed between treatments (1 = 45%; 2 = 61%  $P = 0.04$ ), although AI pregnancy rate for cows inseminated at 90 h did not differ (1 = 44%; 2 = 49%;  $P = 0.51$ ). In summary, when split-time AI is used in conjunction with



the 7 d CO-Synch + CIDR protocol, administration of GnRH at AI to cows that exhibit estrus by 66 h after PG is not warranted. These data indicate that delayed administration of GnRH to 90 h coincident with AI among cows failing to exhibit estrus by 66 h after PG results in a greater overall estrous response.

**Key Words:** estrous synchronization, split-time artificial insemination, beef cow

### 133 Influence of estrus expression prior to fixed-time AI on embryo survival to maternal recognition of pregnancy.

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Estradiol has been reported to play a critical role in pregnancy establishment and embryonic survival. Our objective was to focus on the role of preovulatory estradiol in embryo survival from fertilization to maternal recognition of pregnancy. Estrus was synchronized in beef cows (n = 29) with the CO-Synch protocol and artificially inseminated (d 0). Blood was collected to determine estradiol (d -2 to 0) and progesterone (d 0 to 16) concentrations. Cows were then divided into 2 groups based on expression of estrus (estrus and no estrus). On d 16 uteri were flushed to collect embryos. Total cellular RNA was extracted from blood leukocytes (d 16) to measure the expression of interferon-stimulated genes (ISG): ISG-15, OAS-1, and MX2. Flush media was analyzed for protein and glucose concentrations. Data were analyzed by PROC MIXED. There was an effect of estrus, time, and estrus by time ( $P < 0.01$ ) on circulating concentrations of estradiol, but there was no effect of estrus ( $P = 0.85$ ) or estrus by time ( $P = 0.26$ ) on circulating concentrations of progesterone. There was no difference in embryo recovery rate ( $P = 0.97$ ; 45% vs. 44%). When corrected for volume of flush media, there was no difference between estrus and no estrus for uterine flush protein ( $P = 0.51$ ;  $218 \pm 94$  vs.  $124 \pm 104$  mg/mL); or glucose ( $1960 \pm 274$  vs.  $2003 \pm 304$  mg/dL) content between estrus and no estrus. There was no difference ( $P > 0.20$ ) in d 16 expression of ISG-15, OAS-1, or MX2 between estrus and no estrus animals, nor a difference between cows an embryo was or was not recovered from. However, there was a tendency for cows in which an embryo was recovered from to have greater concentrations of protein ( $P = 0.056$ ;  $314 \pm 104$  mg/mL) and glucose ( $P = 0.09$ ;  $2347 \pm 303$  mg/dL) in uterine flushes compared with cows in which an embryo was not recovered ( $28 \pm 94$  mg/mL and  $1616 \pm 274$  mg/dL). In summary, there were no differences between cows that did or did not express estrus in ISG expression, or in protein or glucose concentration of uterine flushes. Therefore, the increased embryo survival to d 30 of gestation among cows that express estrus is not associated with embryo survival until maternal recognition.

**Key Words:** embryo survival, estradiol, fixed-time AI

### 134 Post insemination interventions: Effect of human chorionic gonadotropin, gonadotropin-releasing hormone, and progesterone on ovulation and conception rates in Nili-Ravi buffaloes.

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We hypothesized that enhancing luteal tissue (or progesterone) would improve conception rate in Nili-Ravi buffaloes. The objective of the present studies were to standardized the dose of human chorionic gonadotropin (hCG) for ovulation induction on 7 d post insemination (experiment 1) and to see the effect of standardized dose of hCG, GnRH and exogenous progesterone (CIDR insert) on ovulation and conception rates in Nili Ravi buffaloes (experiment 2). In experiment 1, ovaries of the adult Nili Ravi buffaloes were scanned and mapped 7 d post synchronized estrus to confirm the presence of corpus luteum (CL) and dominant follicle (>8 mm) and assigned randomly to treatments of saline, or 500, 1000, 1500, 2000, 2500, or 3000 IU hCG (IVF-C, LG, Korea; i.m., n = 47) and ovulation induction or accessory CL, was confirmed a week later with ultrasound. Ovulation incidence in response to  $\geq 2500$  IU hCG was more (100%) than that of other doses or saline. The minimum effective dose was determined through dose response curve. In experiment 2, ovarian responses and conception rates were assessed by minimal effective dose of hCG (2500 IU, n = 58), GnRH (n = 28), CIDR insert for a week (1.38 g, n = 26) and control (n = 44) on 7 d post AI in previously CIDR synchronized in Nili-Ravi buffaloes. Overall significance was determined with Chi-squared and association between groups was checked using Z test in SAS software. More buffaloes were induced to ovulate with hCG (95%) and GnRH (80%) when compared with control and CIDR insert ( $P < 0.05$ ). Conception rates were significantly higher ( $P < 0.05$ ) in hCG (62%; 36/58) treated buffaloes compared with control (32%; 14/44). In contrast, these were non-significant in GnRH (50%; 14/28) and CIDR (42%; 12/26) treated buffaloes when compared with control. In conclusion 2500 IU of hCG is sufficient to induce the accessory CL and this treatment enhance the conception rate when given 7 d post insemination in Nili-Ravi buffaloes.

**Key Words:** progesterone, conception rate, Nili-Ravi buffaloes

### 135 Relationships between hair coat shedding and hair cortisol concentrations and age at first calving in crossbred beef heifers.

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The objective was to evaluate the relationships among hair coat shedding, hair length and hair cortisol concentrations to age at first calving. Spring born (n = 101) crossbred beef heifers (7 to 8 mo. of age) were evaluated for hair length (HL; 1 = short and 5 = long), hair luster (HST; 1 = glossy, healthy appearance and 5 = dull and unthrifty), hair shedding (HS; 1 = short, slick hair, sheds early and 5 = long, late shedding, or full winter coat) at weaning. Calves were weighed, blood samples collected for evaluation of serum cortisol concentrations pre-weaning and at weaning by RIA. At weaning, hair samples were collected over the shoulder, rib and hip area (7.6 cm x 12.7 cm clipped area) for evaluation of hair cortisol concentrations. Hair samples were washed and dried overnight, samples ground using a Retsch mixer miller (200 mm for 10 min), sonicated (at 25 Hz) and incubated 8 h at room temperature. The supernatant was pipetted off and dried under a stream of atmospheric air. Samples were reconstituted before quantification of cortisol using Salimetrics cortisol EIA assay. Data were analyzed using PROC CORR and PROC Mixed of SAS. Hair cortisol concentrations at the rib, hip and shoulder were ( $P < 0.001$ ) greater in cattle with a red or brown coat color compared with cattle with black coat color. Hair length, HST or HS were not influenced by hair coat color ( $P > 0.10$ ). Hair length, HST

and HS were negatively correlated with adjusted 205-d weaning weight ( $P < 0.006$ ) and serum cortisol ( $P < 0.001$ ). Hair length, HST, HS or hair cortisol concentrations were not correlated with age at first calving ( $P > 0.10$ ). Calving day of the year was negatively correlated with hair coat luster ( $P < 0.04$ ) and tended to be negatively correlated with hair length ( $P < 0.07$ ). Pregnancy status was not influenced by hair cortisol concentrations or HL, HST or HS ( $P > 0.10$ ). In summary, hair cortisol

concentrations were influenced by hair coat color. However, hair cortisol concentrations and hair length, luster or hair coat shedding were not adversely associated with age at first calving.

**Key Words:** hair coat shedding, hair cortisol concentrations, beef heifers

# Production, Management, and the Environment I

## 136 Changes in milking procedures on US dairy operations:

**1996–2014.** Jason E. Lombard<sup>\*1</sup>, Charles P. Fossler<sup>1</sup>, Ashley E. Adams<sup>1,2</sup>, Chelsey B. Shivley<sup>1,2</sup>, Natalie J. Urie<sup>1,2</sup>, Christine A. Kopral<sup>1</sup>, and Lindsey P. Garber<sup>1</sup>, <sup>1</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO, <sup>2</sup>Colorado State University, Fort Collins, CO.

The National Animal Health Monitoring System conducted national dairy studies in 1996, 2002, 2007, and 2014. In all 4 studies, information regarding milking procedures was collected via face-to-face interviews. Although the states participating in each study varied slightly, the goal for all the studies was to account for close to 80% of dairy operations and 80% of dairy cows in the US. An objective of this study was to describe how milking facilities and practices have changed over the last 18 years. In 1996, the average herd size on US dairies was 70 cows, and by 2014 herd size had more than doubled to about 160 cows. During this time, the average bulk-tank somatic cell count decreased from 300,000 to approximately 200,000 cells/mL. Although the majority of operations (52.8%) still milk cows in a tie stall or stanchion barn, parlor use increased from 28.8% of operations in 1996 to 45.7% in 2014. In 1996, 54.9% of cows were milked in a parlor compared with 86.0% in 2014. Although the percentage of operations that milked cows twice daily has remained around 90% since 1996, the percentage of operations that milked 3 times per day nearly doubled from 5.8% in 1996 to 10.2% in 2014. Glove use during milking has almost doubled at both the operation level (32.9% of operations in 2002 and 59.1% in 2014) and at the cow level (48.7% of cows in 2002 and 87.9% in 2014). Forestripping all cows has increased from 44.5% of operations in 2002 to 71.5% in 2014. From 1996 to 2014, compounds containing iodine were the most frequently used pre-milking and post-milking teat disinfectants. Automatic take-off use increased from 36.0% of operations in 2002 to 49.1% in 2014, while operations using a backflush system has remained between 6 and 7% since 2002. The US has seen an increase in dairy herd size over the past 18 years, and this increase is associated with more cows being milked in parlors, more operations using gloves and checking for mastitis via forestripping, and an improvement in milk quality, as evidenced by lower somatic cell counts.

**Key Words:** milking practice, parlor, milk quality

**137 Assessing the farm-level cost of mastitis.** Jacqueline Holland<sup>\*1</sup>, Jason Lombard<sup>2</sup>, Joleen Hadrich<sup>1</sup>, and Christopher Wolf<sup>3</sup>, <sup>1</sup>Dept. of Agricultural and Resource Economics, Colorado State University, Fort Collins, CO, <sup>2</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO, <sup>3</sup>Department of Agricultural, Food, and Resource Economics, Michigan State University, East Lansing, MI.

Although mastitis is the most common disease of dairy cattle, estimates of financial losses due to mastitis vary within the literature. The objective of this research is to create a cost-calculation tool that allows dairy producers to estimate their farm's losses due to mastitis, and to provide dairy managers with more accurate information for making decisions about mastitis control and prevention. The tool takes into account direct and indirect loss components, including fatalities; early culling; milk-yield loss; and costs for treatment, labor, and replacement animals. The tool allows dairy producers to input their herd information (e.g., mastitis prevalence, treatment costs) into a cost worksheet and obtain an estimate

of the cost of mastitis on their operation. Information from the National Animal Health Monitoring System's Dairy 2014 study was used to estimate mastitis prevalence, outcome percentages, as well as treatment and labor costs at the farm level. All operations in the study had at least one case of clinical mastitis; overall, 24.1% of cows were affected with mastitis during 2013. Of affected cows, 72.9% recovered, 24.0% were sold, and 3.1% died. The operation average cost of treating a single case of mastitis in 2013 was \$42.05. Additionally, cow-level records from DHIA Dairy Records Processing Centers (DRPCs) throughout the US were used to determine milk-yield losses due to mastitis, as milk-yield reduction accounts for the largest loss associated with mastitis. A hierarchical model using cow-level lactation information from the DRPCs was used to construct milk-yield curves of varying production levels, taking into account herd effects. The construction of yield curves through the hierarchical model allows for the comparison of cows based on lactation average somatic cell counts, thus establishing a loss value. The resulting mastitis-cost estimation tool focuses on determining an accurate cost of mastitis, which justifies investment in mastitis prevention measures.

**Key Words:** mastitis losses, mastitis management, animal health economics

**138 Associations between housing and management practices on the incidence of lameness, hock lesions, and thin cows on US dairy operations.** Ashley E. Adams<sup>\*1,2</sup>, Jason E. Lombard<sup>2</sup>, Ivette N. Roman-Muniz<sup>1</sup>, Charles P. Fossler<sup>2</sup>, and Christine A. Kopral<sup>2</sup>, <sup>1</sup>Colorado State University, Fort Collins, CO, <sup>2</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO.

The objective of this study was to determine the association among different on the incidence of lameness, hock lesions, and thin cows on US dairy operations. This study was conducted as part of the National Animal Health Monitoring System's Dairy 2014 study, which included dairy operations in 17 states. Size categories were assigned as follows: small (30–99 cows), medium (100–499 cows), and large ( $\geq 500$  cows). Trained assessors visited 192 dairy operations between March and July 2014, and recorded locomotion and hock scores (on a 3-point scale), and the number of thin cows (BCS  $< 2.25$ ) on a total of 22,773 cows (average 119 cows per farm). The majority of dairy cows (90.3%) were considered to be sound (LS = 1), 6.9% were mild/moderately lame (LS = 2), and 2.8% were severely lame (LS = 3). Similarly, most dairy cows (87.3%) had no hock lesions (HS = 1), 10.1% had mild lesions (HS = 2), and 2.6% had severe hock lesions (HS = 3). A limited percentage of cows (4.2%) were thin. Univariate comparisons were performed using PROC LOGLINK, which accounts for study design and weighting. Statistical significance was declared at  $P < 0.05$ . Herd size had an effect on all parameters examined, with the percentage of cows scored as LS2, LS3, HS2, HS3, and thin increasing as herd size decreased. Additionally, the percentage of cows scored as LS2, HS2, and HS3 differed by housing type, primary flooring type, and primary bedding material used, and cows scored LS3 differed by housing type and primary bedding used. Operations that housed cows on pasture had fewer LS2 and LS3 cows than those that housed cows in tie-stalls or freestalls, whereas operations that housed cows in open/dry lots had fewer HS2 and HS3 cows than those that housed cows in tie-stalls, freestalls, or on pasture. Feeding a TMR and having a nutritionist balance rations for dairy cows was associated with a lower percentage of thin cows. Although herd size



was found to be an important indicator of the percentage of LS2, LS3, HS2, HS3, and thin cows on dairy operations in the US, other housing and management practices were also found to have an effect.

**Key Words:** dairy cow, lameness, hock lesions

**139 Management of nonambulatory dairy cows on US dairy operations.** Ashley E. Adams<sup>\*1,2</sup>, Jason E. Lombard<sup>2</sup>, Ivette N. Roman-Muniz<sup>1</sup>, Charles P. Fossler<sup>2</sup>, and Christine A. Kopral<sup>2</sup>, <sup>1</sup>Colorado State University, Fort Collins, CO, <sup>2</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO.

The objective of this study was to describe practices used on US dairy operations for the management and care of nonambulatory dairy cattle. This study was conducted as part of the National Animal Health Monitoring System's Dairy 2014 study, which included dairy operations in 17 states. Information on the management of nonambulatory dairy cows was collected on 1,261 dairy operations via in-person interviews. For the purpose of this study, nonambulatory cows were defined as those unable to stand at any time during a period of at least 24 h. The majority of operations (76.5%) had at least one nonambulatory cow in 2013, with an average of 2.6% of US dairy cows becoming nonambulatory during the year. Less than a quarter of all operations (22.6%) had written guidelines for handling nonambulatory cattle: 57.1% of large dairies ( $\geq 500$  cows) had written guidelines, compared with just 24.1% of medium dairies (100–499 cows), and 15.5% of small dairies (30–99 cows). After becoming nonambulatory, cows were offered water within 1 h on 62.6% of operations, within 4 h on 83.0%, and within 12 h on 92.6% of operations; 2.6% of operations did not offer nonambulatory cows water. Food was offered to nonambulatory cows within 1 h on 60.6% of operations, within 4 h on 81.3%, and within 12 h on 91.6% of operations; 3.4% of operations did not offer food to nonambulatory cows. Cows were offered shelter within 1 h after becoming nonambulatory on 65.5% of operations, within 4 h on 80.1%, and within 12 h on 84.9% of operations; 9.1% of operations did not offer shelter to nonambulatory cows. Of nonambulatory cows, 30.0% recovered, 49.7% were euthanized, and 17.7% died. Of those that were euthanized, 59.1% were euthanized within 2 d of being recognized as nonambulatory. With 17.7% of nonambulatory cattle dying, and 41.9% being nonambulatory for 2 d or more before being euthanized, an opportunity exists for dairy producers to improve in this area. Implementing written guidelines, including the identification of cows that qualify for humane euthanasia, may help ensure the welfare of nonambulatory dairy cattle.

**Key Words:** dairy cattle, nonambulatory, management practices

**140 Morbidity and mortality of preweaned dairy heifer calves.** Natalie J. Urie<sup>\*1,2</sup>, Jason E. Lombard<sup>1</sup>, and Chelsey B. Shivley<sup>1,2</sup>, <sup>1</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO, <sup>2</sup>Colorado State University, Fort Collins, CO.

During the calf component of the National Animal Health Monitoring System's Dairy 2014 study, information was collected on morbidity and mortality in preweaned dairy heifers. In addition, data were collected on birthing parameters, serum IgG, and treatments administered. Data have been collected from 851 calves on 89 farms in 12 of the nation's top dairy states. These results from the interim analysis are based on approximately 25% of the total number of calves expected to be enrolled in the study. The objective of the study was to assess morbidity and mortality of preweaned dairy heifers in the US to aid management decisions and increase heifer survival. Of the 851 calves, 33.4% experienced at least 1 disease event, and 6.1% had more than 1 disease event. The

maximum number of disease events reported was 6. Clinical signs of morbidity were classified as dull; dehydrated; gastrointestinal; respiratory; lameness; and neurological. Gastrointestinal signs were observed in 18.0% of calves, while dullness was reported in 13.3% of calves and respiratory signs in 10.6%. Of the 33.4% of calves with clinical signs, 93.3% received treatment. Medications, including antibiotics, were administered to 82.4% of calves and electrolytes were administered to 35.6% of calves. Interestingly, of the 5.4% of calves that died, 39.1% died without displaying clinical signs before dying. The average age at death was 22.8 d, with a range from 3 to 78 d. Birthing parameters, including calving-ease score, number of calves born, and disinfection of the navel were not associated with morbidity or mortality. Serum IgG was collected and calves were classified as excellent  $\geq 15.0$  g/L (71.2%); fair 10.0–14.9 g/L (11.4%); or failure  $< 10.0$  g/L (17.4%) of passive transfer. Calves with fair passive transfer had higher levels of morbidity (44.3%) compared with calves with excellent passive transfer (30.4%). Serum IgG categories were not associated with mortality. High serum IgG levels were associated with decreased morbidity. Digestive and respiratory diseases were the 2 most common causes of morbidity in preweaned heifer calves.

**Key Words:** preweaned dairy calf, morbidity, mortality

**141 Evaluation of colostrum quality and passive transfer status of dairy heifer calves on US dairy operations.** Chelsey B. Shivley<sup>\*1,2</sup>, Natalie J. Urie<sup>1,2</sup>, Deborah M. Haines<sup>3,4</sup>, Jason E. Lombard<sup>2</sup>, and Manuel F. Chamorro<sup>3</sup>, <sup>1</sup>Colorado State University, Fort Collins, Colorado, <sup>2</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, Colorado, <sup>3</sup>Research and Technical Services, The Saskatoon Colostrum Company, Saskatoon, SK, Canada, <sup>4</sup>Department of Veterinary Microbiology, Western College of Veterinary Medicine, Saskatoon, SK, Canada.

Passive transfer of immunity is essential for the short- and long-term health of dairy calves. The objective of this study was to evaluate colostrum quality and the passive-transfer status of US dairy heifer calves. This study was conducted as part of the calf component of the National Animal Health Monitoring System's Dairy 2014 study, which included 92 dairy operations in 13 states. Dairy 2014's calf component is a yearlong longitudinal study focused on dairy heifer calves from birth to weaning; data were collected on 851 calves. The results from the interim analysis are based on approximately 25% of the total number of calves expected to be enrolled in the study. The mean colostrum IgG level was 74.9 g/L (SE 1.2), with 66.3% of samples having IgG levels above 50 g/L. The mean serum IgG level was 22.2 g/L (SE 0.4), and 71.2% of calves had serum IgG levels above 15 g/L. The highest percentage of calves (60%) received colostrum from their dam. Pasteurized colostrum was fed to 9.6% of calves. On average, colostrum was fed within 3.4 h following birth. The mean volume of colostrum fed at first feeding was 3.0 L, and the mean volume of colostrum fed in the first 24 h of life was 4.5 L. The highest percentage of calves (63.7%) were fed colostrum by bottle, while 31.3% were fed colostrum by esophageal feeder. Backward elimination model selection in Proc Mixed was used to determine which colostrum management factors were most important for determining serum IgG levels. The model using initial results included grams of total colostrum fed at the first feeding, the number of hours following birth that calves received their first colostrum feeding, and the calves' birth weight. Serum IgG increased with more grams of total colostrum fed at first feeding; serum IgG decreased as the number of hours from birth to first feeding increased; serum IgG also decreased as birth weights increased. These results show that feeding appropriate

amounts of high-quality colostrum shortly following birth is crucial to the passive-transfer status of dairy calves.

**Key Words:** dairy heifer calf, colostrum quality, passive transfer

**142 Evaluation of the Brix refractometer for measuring colostrum and serum IgG concentrations.** Manuel F. Chamorro\*<sup>1</sup>, Ron Sargent<sup>1</sup>, Deborah M. Haines<sup>2,1</sup>, and Jason Lombard<sup>3</sup>, <sup>1</sup>The Saskatoon Colostrum Company Ltd., Saskatoon, Saskatchewan, Canada, <sup>2</sup>Western College of Veterinary Medicine, Saskatoon, Saskatchewan, Canada, <sup>3</sup>Center for Epidemiology and Animal Health USDA, Fort Collins, CO.

The objective of this study was to evaluate the use of a digital Brix refractometer to assess the levels of IgG in maternal colostrum and to measure passive transfer in newborn calves. Colostrum (n = 1,590) and serum (n = 1,756) samples from 1- to 5-d-old calves were collected from 130 dairy farms across the US from April through December 2014. Colostrum and serum IgG were determined by radial immunodiffusion (RID), and a digital refractometer was used to indirectly estimate IgG concentrations by %Brix. The mean IgG colostrum level was 76.7 g/L (SEM ± 0.84) with a range of 3.0 to 227.4 g/L. The mean %Brix in colostrum samples was 23.3 (SEM ± 0.12) with a range of 2.3% to 40%. The mean IgG serum level was 21.8 g/L (SEM ± 0.26) with a range of 0.1 to 79.3 g/L. The mean %Brix in serum samples was 9.1 (SEM ± 0.02) with a range of 6.4% to 18.7%. The %Brix in colostrum and serum samples was highly correlated with IgG levels in colostrum and serum (r = 0.76 and r = 0.85, respectively). A receiver-operating characteristic curve was created to plot the true positive rate against the false positive rate for consecutive %Brix values for colostrum and serum samples. Compared with a cut-point of 50 g/L IgG (consistent with good quality) as determined by RID in colostrum samples, the lowest false-positive rate occurred at 22% Brix, with a combined sensitivity and specificity of 78% and 84%, respectively. The positive predictive value (PPV) and negative predictive value (NPV) of 22% Brix were 94% and 53.5%, respectively. For serum samples, the optimal combination of sensitivity (88.8%) and specificity (90.6%) compared with a cut-point of 10 g/L (consistent with adequate passive transfer) occurred at 8.4% Brix. The PPV and NPV of 8.4% Brix were 98.3% and 56.9%, respectively. The digital brix refractometer is a convenient method to assess colostrum quality and adequacy of passive transfer in calves at the herd level. However, individual %Brix values below these threshold values underestimate the actual IgG level in about 45% of the samples.

**Key Words:** colostrum IgG, serum IgG, Brix

**143 Prevalence of *Cryptosporidium* and *Giardia* in pre-weaned dairy heifer calves.** Natalie J. Urie\*<sup>1,2</sup>, Jason E. Lombard<sup>1</sup>, Ronald Fayer<sup>3</sup>, Monica Santin-Duran<sup>3</sup>, and Chelsey B. Shivley<sup>1,2</sup>, <sup>1</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO, <sup>2</sup>Colorado State University, Fort Collins, CO, <sup>3</sup>USDA:ARS:NEA:BARC: Environmental Microbial and Food Safety Lab, Beltsville, MD.

As part of the calf component of the National Animal Health Monitoring System's Dairy 2014 study, fecal samples were collected from 782 calves on 87 farms in 11 of the top dairy states. These results from the interim analysis are based on approximately 25% of the total number of calves expected to be enrolled in the study. The samples were examined for *Cryptosporidium* (*Crypto*) and *Giardia* by immunofluorescence microscopy. Samples were collected from 612 calves in the East [IA, MI, MN, MO, NY, OH, PA, VT, VA, WI] and from 239 calves in the West [CA,

CO, WA]. Calves were from 3 to 54 d of age at the time of sampling, with a mean age of 22 d. Overall, 48.2% of calves were infected with *Crypto* and 36.5% were infected with *Giardia*. One fifth of calves (20.0%) were infected with both *Crypto* and *Giardia*; 28.3% were infected with *Crypto* only, and 16.5% were infected with *Giardia* only. Approximately one-third of calves (35.3%) were not infected with either organism. *Crypto* was detected in 54.1% of calves 21 d of age or younger (n = 442) and in 40.6% of calves older than 21 d of age (n = 340). *Giardia* infection also differed between age groups: *Giardia* was detected in 32.4% of calves 21 d of age or younger and in 41.8% of calves older than 21 d of age. The percentage of samples positive for *Crypto* was similar between the East and West (47.4 and 50.2%, respectively). A higher percentage of calves in the East were infected with *Giardia* compared with calves in the West (40.0 and 27.8%, respectively). Calves were sampled from March through December 2014; 29.5% were sampled in April–June (spring), and 58.6% were sampled in July–September (summer). Only 11.9% were sampled during October–December (fall). The prevalence of *Crypto* was not different between seasons. *Giardia* levels differed among the seasons with a prevalence of 36.8% in the spring, 33.8% in the summer and 48.4% in the fall. *Giardia* prevalence in calves was lower than *Crypto*, but *Giardia* was more prevalent in the East region and during the fall. Species and assemblage identification for *Crypto* and *Giardia*, respectively, for these and additional samples await molecular applications.

**Key Words:** calves, *Cryptosporidium*, *Giardia*

**144 Evaluation of average daily gain in preweaned dairy heifer calves based on different liquid diets and management practices.** Chelsey B. Shivley\*<sup>1,2</sup>, Natalie J. Urie<sup>1,2</sup>, and Jason E. Lombard<sup>2</sup>, <sup>1</sup>Colorado State University, Fort Collins, CO, <sup>2</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO.

The objective of this study was to evaluate average daily gain (ADG) in US dairy heifer calves based on different liquid diets and management practices. This study was conducted as part of the calf component of the National Animal Health Monitoring System's Dairy 2014 study, which included 123 dairy operations in 13 states. Dairy 2014's calf component is a yearlong longitudinal study focused on dairy heifer calves from birth to weaning; data were collected on 851 calves from 92 operations. The results from the interim analysis are based on approximately 25% of the total number of calves expected to be enrolled in the study. Liquid diets were categorized by type (i.e., milk replacer, waste/whole milk, or a combination of the 2 (combination)) and by volume (i.e., > 4.9 kg/d or ≤ 4.9 kg/d). The mean ADG was 0.7 kg per day, and calves were fed liquid diets an average of 58.8 d. Backward elimination model selection was used to determine which diet and management practices significantly affected ADG. Using initial results, the model included liquid diet ( $P = 0.004$ ), disease ( $P < 0.001$ ), housing ( $P = 0.0189$ ), and dehorning ( $P = 0.001$ ). After controlling for other independent variables in the model, calves fed a combination diet gained, on average, 0.14 kg/day more than calves fed any milk replacer or a low volume of milk. Calves fed a high volume of milk outgained calves fed milk replacer or a low volume of milk by 0.15 kg/day. Calves without any disease events gained 0.07 kg/day more than calves with one or more disease events. Calves housed in groups gained 0.15 kg/day more than calves housed individually. Preweaned heifer calves that were dehorned gained 0.11 kg/day more than calves that were not dehorned. These initial results indicate the type of liquid diet fed, the amount fed, as well as other management practices unrelated to feeding, affect ADG.

**Key Words:** dairy heifer calf, average daily gain, liquid diet

**145 Prevalence of *Campylobacter* spp. in bulk tank milk and filters from US dairies.** Laura P. Del Collo\*<sup>1,3</sup>, Jeffrey S. Karns<sup>1</sup>, Debabrata Biswas<sup>3</sup>, Jason E. Lombard<sup>2</sup>, R. Camilla Kristensen<sup>2</sup>, Charles P. Fossler<sup>2</sup>, and Jo Ann S. Van Kessel<sup>1</sup>, <sup>1</sup>USDA-ARS-NEA Environmental Microbial and Food Safety Laboratory, Beltsville, MD, <sup>2</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO, <sup>3</sup>Dept. of Animal and Avian Sciences, University of Maryland, College Park, MD.

*Campylobacter* spp. is an important zoonotic bacterial pathogen that caused the majority of US outbreaks associated with nonpasteurized milk from 2007 to 2012. Bulk tank milk and milk filter samples were collected from 234 dairy operations in 17 top dairy states from March through July 2014 as part of the National Animal Health Monitoring System's Dairy 2014 study. Overall, 234 bulk tank samples and 396 milk filter samples were collected. Bulk tank and milk filter pairs were collected from 231 operations, and only bulk tank samples were collected from 3 operations. Bulk tank milk and buffered peptone water extracts of milk filters were enriched in Bolton Broth and struck onto mCCDA plates. A real-time PCR method was used to determine the presence of pathogenic species (*C. jejuni*, *C. coli*, and *C. lari*) in enrichments and to confirm isolates. Analysis of prevalence (%) incorporated operation weights to reflect the population from which samples were collected. Positive operations were those with either a positive milk filter or a positive bulk tank sample (or both). PCR indicated the presence of pathogenic *Campylobacter* spp. in enrichments from 24.3% of operations, while isolates were obtained from 12.0% of operations. *Campylobacter* was more frequently identified in samples from operations in the west region than in the east region (46.5% and 21.8%, respectively) and was more common in medium and large operations than in small operations (42.8%, 47.3%, and 4.9%, respectively). Based on multiplex PCR analysis, 41 (91.3%) of the *Campylobacter* isolates were *C. jejuni*, 3 were *C. lari*, and 1 was identified as *C. coli*. This first national survey of *Campylobacter* spp. in bulk tank milk and milk filters suggests that pathogenic *Campylobacter* spp. are common on US dairies and, therefore, are a continued risk for foodborne disease outbreaks.

**Key Words:** *Campylobacter*, bulk tank milk, milk filter

**146 *Salmonella* Dublin antibodies in bulk-tank milk on U.S. dairy operations.** Jason E. Lombard\*<sup>1</sup>, Belinda S. Thompson<sup>2</sup>, Paul D. Virkler<sup>2</sup>, Bettina Wagner<sup>2</sup>, R. Camilla Kristensen<sup>1</sup>, and Charles P. Fossler<sup>1</sup>, <sup>1</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO, <sup>2</sup>Animal Health Diagnostic Center, College of Veterinary Medicine, Cornell University, Ithaca, NY.

Bulk-tank milk was collected and tested for antibodies to *Salmonella* Dublin during the National Animal Health Monitoring System's Dairy 2014 study. Information regarding management practices and dairy-cow health and productivity was also collected via 2 face-to-face interviews. One objective of the Dairy 2014 study was to estimate the herd-level prevalence of *Salmonella* Dublin antibodies and associated factors. A single bulk tank sample was collected from 230 operations from March through July 2014. After adjusting for study design and incorporating weighting procedures, estimates indicated that 8.0% of dairy operations had *Salmonella* Dublin antibodies present in bulk-tank milk. A higher percentage of operations with 500 or more cows (39.2%) had bulk-tank antibodies compared with operations with 100–499 cows (2.1%), and 30–99 cows (1.0%). In the West region (CA, CO, ID, TX, WA), 52.1% of bulk tanks were antibody positive, and in the East region [IA, IN,

KY, MI, MN, MO, NY, OH, PA, VT, VA, WI] 2.8% of operations were antibody positive. Although bulk tanks in closed herds were not at lower risk of being antibody positive, bulk tanks on a higher percentage of operations that reared heifers offsite (20.6%) were antibody positive compared with bulk tanks on operations that did not raise heifers offsite (6.0%). In addition, bulk tanks on operations that reared heifers offsite and commingled heifers with cattle from other operations were at higher risk of being antibody positive (28.0%) compared with bulk tanks on operations in which heifers did not have contact with other cattle at the offsite facility (3.2%). Operations with *Salmonella* Dublin antibodies detected in bulk-tank milk had rolling herd average milk production of 10,764 kg compared with 9,672 kg for operations in which antibodies were not detected. Operation-level mortality of pre-weaned and weaned heifers did not differ by Dublin result. Results suggest that large operations, operations in the West and operations that raise heifers offsite are more likely to have *Salmonella* Dublin antibodies detected in milk.

**Key Words:** *Salmonella* Dublin, bulk milk, prevalence

**147 Prevalence of *Salmonella* and *Listeria monocytogenes* in bulk tank milk and filters from US dairies.** Jo Ann S. Van Kessel\*<sup>1</sup>, Jeffrey S. Karns<sup>1</sup>, Jason E. Lombard<sup>2</sup>, R. Camilla Kristensen<sup>2</sup>, and Charles P. Fossler<sup>2</sup>, <sup>1</sup>USDA-ARS-NEA Environmental Microbial and Food Safety Laboratory, Beltsville, MD, <sup>2</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO.

Zoonotic bacterial pathogens are frequently isolated from the feces of dairy animals and their environments; therefore, bulk tank milk (BTM) and meat from culled dairy animals are at risk for contamination. As part of the National Animal Health Monitoring System's (NAHMS) Dairy 2014 study, 234 BTM and 396 milk filter (MF) samples were collected from 234 operations in 17 states from March through July 2014. BTM and MF pairs were collected from 231 operations, and only BTM samples were collected from 3 operations. Samples were enriched for *Salmonella* and *Listeria* spp. in selective broths. The presence of *Salmonella* was determined by real time PCR, and positive enrichment broths were struck onto selective agar for isolation. *L. monocytogenes* was distinguished from other *Listeria* species via the presence of phosphatidylinositol-specific phospholipase. Analysis of prevalence (%) incorporated operation weights to reflect the population from which samples were collected. Positive operations were those with either a positive MF or a positive BTM sample (or both). PCR analysis indicated that 18.6% of operations were positive for *Salmonella*. *Salmonella* isolates were obtained on 96.5% of these PCR-positive operations. Sixteen *Salmonella* serotypes were identified, with multiple serotypes detected on 9 operations. The most common serotypes were Cerro, Montevideo, Kentucky, and Newport, which were isolated from 33, 8, 5, and 5 operations, respectfully. *Listeria* spp. were isolated from 20.0% of operations and *L. monocytogenes* was isolated from 3.0% of operations. Prevalences of *Salmonella* and *L. monocytogenes* increased as herd size increased. The 2014 prevalences of *Salmonella* and *L. monocytogenes* are significantly lower than the prevalences from the NAHMS 2007 Dairy study (28.1% for *Salmonella*, 7.1% for *L. monocytogenes*). A significant presence of *Salmonella* and *L. monocytogenes* in BTM or MF samples indicates potential health risks to raw milk consumers.

**Key Words:** *Salmonella*, *Listeria*, bulk tank milk



**148 Dry-off procedures on US dairy operations.** Jason E. Lombard\*<sup>1</sup>, Gosia Zobel<sup>2</sup>, Ashley E. Adams<sup>1,3</sup>, Charles P. Fossler<sup>1</sup>, Chelsey B. Shivley<sup>1,3</sup>, Natalie J. Urie<sup>1,3</sup>, and Christine A. Kopral<sup>1</sup>, <sup>1</sup>USDA:APHIS:VS:Center for Epidemiology and Animal Health, Fort Collins, CO, <sup>2</sup>University of British Columbia Animal Welfare Program, Vancouver, BC, Canada, <sup>3</sup>Colorado State University, Fort Collins, CO.

The National Animal Health Monitoring System's Dairy 2014 study collected information via face-to-face interviews from 17 states. An objective of the study was to describe dry-off procedures on US dairy operations. Size categories were created based on number of lactating and dry cows: small (30–99 cows), medium (100–499 cows), and large (500 or more cows). During 2013, the operation average dry period length was 57.1 d. Almost all operations (98.8%) dried off at least some cows based on a set schedule, and 81.3% of operations dried off at least some cows based on a minimum milk-production level. Overall, 88.1% of cows were dried off on a set schedule and 11.9% were dried off based on milk production. Almost 3-fourths of operations (73.6%) abruptly stopped milking some cows at dry-off, while 47.5% of operations gradually stopped milking some cows at dry-off. However, only 10.2% of cows experienced gradual cessation of lactation, while 89.8%

experienced abrupt cessation. At dry-off, 15.0% of operations performed a California Mastitis Test; 65.7% reduced the quality/energy content of feed; 10.9% restricted access to feed; and water was restricted on 3.9% of operations. More than 3-fourths of operations (80.3%) treated all cows with dry-cow intramammary antibiotics and, overall, 93.0% of cows were treated. Cephapirin benzathine was the single most commonly administered antibiotic (31.6% of cows dry-treated). Alcohol pads were used to clean teat ends before administering dry-cow antibiotics on 90.3% of operations. Dry-cow intramammary antibiotics were not used on 9.2% of operations. Internal teat sealants were used on 37.3% of operations, while external teat sealants were used on 14.5%. The percentage of operations using sealants on all cows increased as herd size increased. The operation average cost of antibiotics and sealants used at dry-off was \$13.09 per cow. Gradual cessation of milking, which may reduce new intramammary infections during dry-off, is infrequently practiced in the US. Most cows are treated with antibiotics at dry-off, but this practice may be examined due to pressure to reduce antibiotic use in livestock and poultry.

**Key Words:** dry-off procedure, antibiotic, teat sealant

# Ruminant Nutrition: Dairy calves

**149 Effects of intensive whole-milk feeding in calves on subsequent growth of dairy heifers.** Camila Flávia de Assis Lage<sup>1</sup>, Mariana Magalhães Campos<sup>2</sup>, Fernanda Samarini Machado<sup>2</sup>, Paulo Campos Martins<sup>1</sup>, Luigi Francis Lima Cavalcanti<sup>3</sup>, Marcelo Neves Ribas<sup>3</sup>, Luiz Gustavo Ribeiro Pereira<sup>2</sup>, Thierry Ribeiro Tomich<sup>2</sup>, Rafael Alves de Azevedo<sup>1</sup>, and Sandra Gesteira Coelho<sup>1</sup>, <sup>1</sup>Federal University of Minas Gerais, Belo Horizonte, Minas Gerais, Brazil, <sup>2</sup>EMBRAPA Dairy Cattle, Coronel Pacheco, Minas Gerais, Brazil, <sup>3</sup>CNPq, RHAÉ – SEVA Engenharia, Projeto Intergado, Contagem, Minas Gerais, Brazil.

The effects of intensive whole milk feeding in calves on subsequent growth of Holstein-Gyr females was evaluated. Up to 56 d old, calves received 6 L/d of 4 different liquid diets consisting of whole milk with increasing addition of milk replacer (Sprayfo Violet SSP) to adjust the concentration of total solids (TS) to 13.5 (n = 15), 16.1 (n = 15), 18.2 (n = 13), 20.4% (n = 15). After weaning, animals were randomly housed in 4 paddocks, each one equipped with electronic feed and water bins (Intergado, Brazil) in Embrapa Dairy Cattle facilities, Brazil. Diet (70:30, corn silage:concentrate, 195 g CP/kg, DM basis) was fed ad libitum, twice a day, until 210 d old. The withers height (WH), hip height (HH), rump width (RW) and chest circumference (CC) measures were carried out fortnightly using a flexible tape measure and a teletape (Ketchum, Canada). WH and CC were analyzed as a completely randomized design with repeated measures using nonlinear mixed models approach. A regression model ( $Y(\text{age}) = A + (B - A) \exp[-\exp(-c \times \text{age})]$ ), where Y = response at a specific age, A = asymptote as age  $\rightarrow \infty$ , B =  $Y_{(0)}$ , and c = logarithm of the rate constant) was fit to the data, where age and function parameters were allocated as fixed effects while animal was considered as random. It was evaluated the necessity of adding random terms to model error dependence and heteroscedasticity by monitoring Schwarz criterion, estimates stability and correlation. HH and RW, due to their linear growth pattern, were analyzed by linear mixed models, allowing polynomial models of first and second degree orders to model variable change over time ( $\alpha = 0.05$ ). All variables were steadily influenced by age, but only WH and CC were influenced by TS. For both responses, the c parameter was linearly increased by TS, revealing that a more intensive feeding strategy increases the growth rate of these 2 traits. TS influenced all CC parameters, where increasing total solid level caused a linear decrease of A, while increased B. TS effects over parameters A and B should be interpreted with parsimony, because animals in the present study had not achieved their mature weight.

**Key Words:** milk replacer, intensive farming, heifer

**150 Effect of bacteria level in colostrum on dairy heifer serum IgG concentration.** Christine Cummins and Emer Kennedy\*, Teagasc, Ireland.

Storage of colostrum  $>4^{\circ}\text{C}$  increases total bacterial count (TBC) which may compromise passive transfer of immunity. This experiment investigated the effect of colostrum stored at varying temperatures, to induce a difference in bacteria levels, on the rate of passive transfer of immunoglobulin G (IgG) in dairy heifer calves. Colostrum was collected immediately postpartum from Holstein-Friesian (HF) cows, tested for IgG concentration, and assigned to 1 of 5 treatments: (1) pasteurized, (2) fed when freshly collected, (3) stored at  $4^{\circ}\text{C}$  for  $\geq 48$  h (fridge), (4) stored at  $13^{\circ}\text{C}$   $\geq 48$  h, and (5) stored at  $22^{\circ}\text{C}$   $\geq 48$  h. Colostrum fed to each calf was tested for TBC, using serial dilution and IgG concentration

using radial immunodiffusion (RID; Triple J Farms, WA). Seventy-five HF and HF  $\times$  Jersey (JEX) heifer calves were removed from their dam and assigned to a treatment immediately postpartum at Teagasc Moorepark Research Farm, from 3 Feb to 25 Mar 2014. A randomized block design accounting for breed, birth date and birth weight (BW) was used. Calves were fed 8.5% of their BW in colostrum via stomach tube within 2 h. Calf blood samples were collected at 0 and 24 h of age and analyzed for IgG concentration using RID. Data were checked for normality and the MIXED procedure in SAS was used to examine the effect of treatment on serum IgG concentration. Pasteurised colostrum had a TBC  $<9,000$  cfu/mL, fresh colostrum had 68,000 cfu/mL; both below the recommended level of 100,000 cfu/mL. Colostrum stored at  $4^{\circ}\text{C}$  had a TBC  $> 2$  million cfu/mL, currently not recommended for feeding. Colostrum stored at  $13^{\circ}\text{C}$  and  $22^{\circ}\text{C}$  had significantly higher ( $P < 0.01$ ) TBCs ( $>92$  and  $>1000$  million cfu/mL, respectively). Colostrum stored at  $22^{\circ}\text{C}$  had the lowest IgG concentration (62 g/L). The overall average colostrum IgG concentration across all treatments was 97 g/L. Zero-hour serum contained no IgG. Serum IgG of calves at 24 h from the pasteurised, fresh and  $4^{\circ}\text{C}$  treatments were similar, but were significantly higher ( $P < 0.05$ ) than colostrum from the  $13^{\circ}\text{C}$  and  $22^{\circ}\text{C}$  treatments. Colostrum with high levels of bacteria reduced IgG absorption in dairy calves. Colostrum should be stored  $\leq 4^{\circ}\text{C}$  to minimize bacterial growth and improve subsequent passive transfer of IgG.

**Key Words:** calf, IgG, colostrum

**151 Performance of calf reared on waste milk or nonmedicated milk replacer contained sodium butyrate and *Bacillus amyloliquefaciens*.** O. V. Vazquez-Mendoza<sup>1</sup>, A. E. Kholif<sup>2</sup>, M. M. Y. Elghandour<sup>3</sup>, A. Z. M. Salem<sup>3</sup>, V. L. Garcia-Flor<sup>4</sup>, and T. A. Morsy<sup>2</sup>, <sup>1</sup>Norel México S.A. de C.V., Parque Industrial El Marqués, Querétaro, México, <sup>2</sup>Dairy Science Department, National Research Centre, Giza, Egypt, <sup>3</sup>Facultad de Medicina Veterinaria y Zootecnia, Universidad Autónoma del Estado de México, Toluca, Estado de México, Mexico, <sup>4</sup>Centro de Ciencias Agropecuarias, Universidad Autónoma de Aguascalientes, Aguascalientes, México.

More interest is paid for the accelerated growth programs for dairy calves through enhancing early nutrition programs based on greater rates of liquid feeding for better mammary gland development and milk production. Appropriate supply of nutrients for calves through liquid feed including milk or milk replacer is essential for better performance and welfare. In a completely randomized design, the nutritional and economic efficiencies, and growth performance of 18 Holstein female dairy calves ( $41 \pm 3.7$  kg BW, 1 d old) fed either pasteurized waste milk (PWM) or calf milk replacer (CMR) were tested. Calves were fed colostrum (IgG; 70–100 mg/mL) within the first 2 h of life at the rate of 10% of their BW, and then offered 2 L every 12 h for 3 d, without access to solid feed. Calves were fed individually on PWM (n = 9) or CMR containing *Bacillus amyloliquefaciens* and sodium butyrate (n = 9) twice daily at 0900 and 1600 h for 60 d. From the fourth day, calves were offered pelleted starter feed (180 g CP and 338.2 g of NDF/kg DM) in the morning at 0900 h. Water was provided ad libitum. Health condition, body measurements, fecal bacteriological analysis and economic analysis were measured. No differences were observed for liquids and total starter intakes; however, calves fed on PWM consumed more ( $P < 0.05$ ) starter DM during the period from d-16 to d-45 with greater metabolizable energy intake. Greater ( $P < 0.05$ ) BW changes

for PWM-calves than CMR-calves during the period from d-30 to d-60; with higher ( $P < 0.05$ ) average daily gains during the period from d-16 to d-60. Health conditions did not differ between calves with greater faces score and lower cough score for CMR-calves. Calves fed CMR had greater fecal number from *Klebsiella oxytoca* and *Proteus vulgaris* with the same count of *E. coli*. Greater economic evaluation ( $P < 0.0001$ ) for CMR-calves than PWM-calves were obtained. It could be concluded that the CMR can be used efficiently rather than PWM without reducing feed efficiency and with more profits in feeding dairy calves during the pre-weaning period.

**Key Words:** calf, health condition, milk replacer

### 152 Effects of colostrum feeding programs on passive immunity, health, and performance of Holstein dairy calves. Weina Shi and Zhijun Cao\*, *China Agricultural University, Beijing, China.*

The objective of this study was to investigate effects of 4 colostrum feeding programs on passive transfer of immunity, health, and growth performance of Holstein dairy calves before 70 d age. Experiment 1, 76 newborn Holstein calves (24 bulls and 52 heifers) were blocked by colostrum quality and sex and assigned randomly to 4 treatments. Calves were housed in individual hutches and were moved to the heifer pens at the age of 9 week. Colostrum feeding programs were as follows: calves in T<sub>322</sub> were fed 3 L at 0 h (birth), 2 L at 6 h, and 2 L at 12 h; calves in T<sub>402</sub> were fed 4 L at 0 h and 2 L at 12 h; calves in T<sub>400</sub> were fed 4 L only at birth; calves in T<sub>220</sub> were fed 2 L at 0 h and 2 L at 6 h. Blood sample collected at 24 h after birth showed that serum total protein (TP, g/dL) and IgG (mg/mL) levels were significantly higher for calves in T<sub>322</sub> (TP = 6.11 g/dL, IgG = 22.49 mg/mL,  $P < 0.01$ ) and T<sub>402</sub> (TP = 5.89 g/dL, IgG = 19.99 mg/mL,  $P < 0.05$ ) compared with calves in T<sub>400</sub> (TP = 5.56 g/dL, IgG = 16.05 mg/mL) and T<sub>220</sub> (TP = 5.58 g/dL, IgG = 16.31 mg/mL). At 48 h after birth, serum TP and IgG levels were significantly higher for calves in T<sub>322</sub> (TP = 6.37 g/dL, IgG = 25.61 mg/mL,  $P < 0.01$ ) and T<sub>402</sub> (TP = 6.12 g/dL, IgG = 22.61 mg/mL,  $P < 0.01$ ) compared with calves in T<sub>400</sub> (TP = 5.58 g/dL, IgG = 16.30 mg/mL) and T<sub>220</sub> (TP = 5.66 g/dL, IgG = 17.24 mg/mL). Experiment 2, 40 heifers in experiment 1 were involved. Results showed that the incidence and frequency of diarrhea and the fecal index tended to be greater ( $P > 0.05$ ) in T<sub>400</sub> and T<sub>220</sub> than in the other 2 treatments. At the first week post-weaning, calves from T<sub>322</sub> (1536.54 g/d, DM,  $P < 0.01$ ) and T<sub>400</sub> (1321.23 g/d, DM,  $P < 0.01$ ) had significantly higher starter intake than that of calves from T<sub>220</sub> (1162.01 g/d, DM). These results suggest that colostrum feeding program 322 (3 L at 0 h, 2 L at 6 h, and 2 L at 12 h) and 402 (4 L at 0 h and 2 L at 12 h) can improve passive immunity, health, and growth performance of Holstein dairy calves compared with the other 2 feeding programs. Further research is needed to evaluate the influence of colostrum feeding programs on later growth, reproduction and lactation performance of dairy cattle.

**Key Words:** colostrum, passive immunity, growth performance

### 153 Influences of SmartCare in milk replacer and XPC in calf starter on the performance and health of preweaning Holstein calves challenged with *Salmonella enterica* serotype Typhimurium. Tyler L. Harris\*<sup>1</sup>, Yu Liang<sup>1</sup>, Matt D. Sellers<sup>1</sup>, Jeff A. Carroll<sup>2</sup>, Ilkyu Yoon<sup>3</sup>, Mark F. Scott<sup>3</sup>, and Michael A. Ballou<sup>1</sup>, <sup>1</sup>Department of Animal and Food Sciences, Texas Tech University, Lubbock, TX, <sup>2</sup>USDA-ARS, Lubbock, TX, <sup>3</sup>Diamond V, Cedar Rapids, IA.

This study investigated how supplementing milk replacer with SmartCare (SC) and calf starter with original XPC (XPC) would affect performance and health following an oral challenge with *Salmonella enterica* serotype Typhimurium. The study was performed in 2 35 d periods with 30 Holstein bull calves per period. In each period, calves were blocked by location in the barn and randomly assigned to treatments that included: CON: base milk replacer and calf starter with no added products, SC: milk replacer with 1 g SC/calf/d and base calf starter, and SC+XPC: milk replacer with 1 g SC/calf/d and calf starter with 0.5% XPC. Calves were fed 350 g of milk replacer at 0700 and 1700. Calf starter and water were offered ad libitum and recorded daily. Calves were challenged with 10<sup>8</sup> cfu of sulfamethazine-resistant *Salmonella enterica* orally on d 14 of the study. Fecal *Salmonella* shedding was determined on d 14 to 21, and d 24, 28, and 35 using selective media. Blood samples were collected on d 0, 7, 14, 16, 18, 21, 24, 28, and 35 and analyzed for hematology. Contrasts were analyzed to determine SC and XPC effects. All data were reported as CON, SC, and SC+XPC, respectively. The LSMeans with various superscripts differ ( $P \leq 0.05$ ). Calf starter intake (0.178<sup>a</sup>, 0.218<sup>ab</sup>, 0.254<sup>b</sup> ± 0.022 kg) was improved in both SC and SC+XPC (SC contrast  $P = 0.029$ ; XPC contrast  $P = 0.035$ ). Average daily gain (0.48<sup>a</sup>, 0.48<sup>a</sup>, 0.58<sup>b</sup> ± 0.028 kg) was greater among SC+XPC calves vs. the other treatments (XPC contrast,  $P = 0.001$ ). Post challenge, the SC+XPC calves had a lower neutrophil to lymphocyte ratio (0.81<sup>a</sup>, 0.83<sup>a</sup>, 0.69<sup>b</sup> ± 0.051) vs. CON and SC calves (XPC contrast,  $P = 0.005$ ). The SC+XPC calves also had lower hematocrits (35.1<sup>a</sup>, 35.3<sup>a</sup>, 33.4<sup>b</sup> ± 0.54%) and hemoglobin concentrations (10.8<sup>a</sup>, 10.6<sup>a</sup>, 10.1<sup>b</sup> ± 0.16 mg/dL) after the challenge. Fecal *Salmonella* shedding tended to be reduced by both SC and SC+XPC (3.21, 2.84, 2.42 ± 0.279 d shedding; SC and XPC contrasts  $P \leq 0.071$ ). Calves fed both SC in milk replacer and XPC in calf starter had improved performance and enteric health.

**Key Words:** calf, health, yeast culture

### 154 Effect of feed type and presentation on feeding behavior, intake, and growth of dairy calves fed a high level of milk. Morgan A. Overvest\*<sup>1</sup>, Renee Bergeron<sup>2</sup>, Derek B. Haley<sup>3</sup>, and Trevor J. DeVries<sup>1</sup>, <sup>1</sup>Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada, <sup>2</sup>Department of Animal and Poultry Science, University of Guelph, Campus d'Alfred, Alfred, ON, Canada, <sup>3</sup>Department of Population Medicine, Ontario Veterinary College, University of Guelph, Guelph, ON, Canada.

The objective of this study was to assess the effect of different feed types and method of feed presentation in the first 12 wk of life on the feeding behavior, intake and growth of calves fed a high milk level. Forty-eight neonatal Holstein calves (~24 h old) were individually housed and randomly assigned to 1 of 4 treatments: silage-based total mixed ration (TMR), concentrate (CON), and chopped hay and concentrate presented in 2 manners: mixed (MIX) or separate (SEP). All calves were offered 12L/d of acidified milk replacer until d 38 at which time step-down weaning by 1 L/d began. At d 50 calves no longer received milk and all calves on SEP and CON treatments were offered the MIX diet until the end of the trial while TMR and MIX calves did not change feeds. Feed intakes were recorded daily and calves were weighed 2x/wk. For the last 2 d during wk 3, 5, 7, 9, and 11 video recordings were analyzed for time spent feeding. Data were summarized by week and analyzed in a repeated measures general linear mixed model. In the preweaning stage (d 1–37) ADG was similar for all calves (1.1 kg/d; SE = 0.05;  $P = 0.16$ ). TMR calves had lower ADG than calves on the other 3 treatments during both the weaning (d 38–49; 0.2 vs. 0.7 kg/d; SE = 0.06;  $P < 0.01$ ) and post-weaning (d 50–84; 0.5 vs 1.2 kg/d; SE = 0.09;  $P <$



0.01) stages. This result is related to the lower DMI of calves fed TMR in comparison to MIX, SEP and CON calves in the weaning (0.2 vs. 0.5 kg/d; SE = 0.07;  $P < 0.01$ ) and post-weaning (1.8 vs. 2.8 kg/d; SE = 0.17;  $P < 0.01$ ) stages. It should be noted that, given DM content of the feeds (TMR = 52%, all other diets = 89%), the as-fed intake of the calves was actually similar ( $P \geq 0.40$ ) across treatments in all 3 stages. Interestingly, TMR calves spent more time feeding during the postweaning stage than MIX, SEP and CON calves (308 vs 194 min/d; SE = 16.0;  $P < 0.001$ ). It appears that during weaning and post-weaning, calves fed TMR were attempting to maximize their nutrient intake, but were unable to match that of those in other treatments due to the high moisture content of their feed, and thus were unable to perform at a similar level.

**Key Words:** dairy calf, weaning, feeding behavior

**155 Extensive, noninvasive measurements of body temperature and posture in neonatal Holstein dairy calves bedded with deep straw in response to changes in ambient temperature and amount of milk replacer fed.** T. Mark Hill\*, H. Gale Bateman, II, F. Xavier Suarez-Mena, James D. Quigley, and Rick L. Schlotterbeck, *Nurture Research Center, Provimi North America, Cargill Premix and Nutrition, Brookville, OH.*

Posture of calves can be influenced by nutrition and ambient temperature. Calf tail vein temperature (a proxy for body temperature) and standing time were extensively measured in 14 male calves ( $46 \pm 1.5$  kg initial BW) between 4 and 18 d of age. Thermocrons (Maxum Integrated Products Inc.) were taped to the tail vein to capture temperatures hourly. Accelerometers (Onset Computer Corp.) were attached to a plastic ankle bracelet and taped to the medial side of the right, rear leg to record posture every minute. Calves were fed free-choice dry feed and water and a 27% CP, 17% fat (DM) milk replacer fed at 2 rates (0.66, LOW, and 1.0 HIGH, kg DM daily) in 2 equal meals fed at 0600 and 1600 h. Calves were bedded with deep straw and maintained in an unheated, naturally ventilated, curtain-sided nursery with a well-drained rock floor. Ambient temperature averaged  $2.5^{\circ}\text{C}$  (daily averages ranged from  $-2.4$  to  $8.1^{\circ}\text{C}$ ). Data were analyzed as repeated measures in a randomized ANOVA to test MR rate and with linear regression analysis to test ambient temperature and time. Mean calf temperature was  $38.4 \pm 0.11^{\circ}\text{C}$  and it changed diurnally with a  $0.5^{\circ}\text{C}$  daily range being highest at night ( $P < 0.05$ ). Calf temperature was  $0.1^{\circ}\text{C}$  greater for calves fed a HIGH vs. LOW rate and it increased  $0.02^{\circ}\text{C}$  per  $1^{\circ}\text{C}$  ambient temperature ( $P < 0.05$ ). Standing time averaged  $299 \pm 17$  min/d and it increased 7.1 min daily with age and decreased 2.6 min daily per  $1^{\circ}\text{C}$  ambient temperature ( $P < 0.05$ ). Standing bouts (a standing plus lying event) averaged  $16.2 \pm 0.5$  daily and decreased 0.3 bouts daily per  $1^{\circ}\text{C}$  ambient temperature ( $P < 0.05$ ). Bouts were 2.8 more daily for calves fed at the high vs. low rate and these extra bouts occurred during the AM and PM meals ( $P < 0.05$ ). Standing time and bouts changed over the 24-h day and were greatest during the AM and PM meals, intermediate during the day between meals, and were the least overnight ( $P < 0.05$ ). Feeding more milk replacer slightly increased body temperature and increased standing bouts during meals. A greater body temperature and less standing time overnight suggests that calves bedded with deep straw during cold temperatures conserved body heat by lying.

**Key Words:** calves, posture, temperature

**156 Commercial dairy farm evaluation of highly digestible corn grain for calf starters when calves are fed pasteurized waste milk.** David P. Casper\*<sup>1</sup> and Mark Kirk<sup>2</sup>, <sup>1</sup>South Dakota State University, Brookings, SD, <sup>2</sup>Masters Choice, Anna, IL.

New corn hybrids have been developed by Masters Choice (MC) that vary in energy density and starch digestibility. These MC hybrids have lower starch densities due to an altered starch structure, which allows for greater ruminal and intestinal starch digestion. Our previous work (Casper et al. 2014) had demonstrated improved nutrient digestibilities, feed efficiency, and reduction in cost of gain when feeding a calf starter (CS) based on MC corn when calves were fed an accelerated milk replacer (28:18 CP:fat). Forty-five (1 to 3 d old) Holstein heifer calves raised on a large commercial Eastern South Dakota Dairy Operation were randomly assigned to 1 of 3 CS to evaluate growth performance of Holstein heifer calves through 7 wk of age from mid-June 15 through mid-August, 2013. Treatments were: 1) Control (C) CS: containing 40% (DM basis) conventional ground shelled corn 24% CP; 2) MC CS: containing (40% DM basis) MC corn and 24% CP; and 3) Standard commercially available CS (S) being fed at the dairy operation containing 16% conventional corn and 16.5% CP. All CS were fed as a pellet. Colostrum was fed the first 3 d of life and then pasteurized waste milk was fed twice daily at a rate of 3.78 L/d for 2–13 d, 5.68 L/d for 14–35 d, and once daily at 2.85 L/d for 35–42 d of age. Feedings were reduced to 1x/d at 35 d to facilitate weaning at 42 d and the study ended at 49 d. Calves were housed in individual poly hutches with ad libitum CS and water. Data were analyzed as a completely random design using the PROC MIXED procedure of SAS Version 9.4. Initial body weights were similar ( $P > 0.08$ ) for calves fed all CS (38.8, 37.5, and 34.9 kg for C, MC, and S, respectively). Body weight gains (18.0, 16.8 and 13.6 kg) and average daily gains (0.34, 0.32, and 0.28 kg/d) were greater ( $P < 0.03$ ) for calves fed C and MC compared with calves fed S CS. Calves fed MC CS gained greater ( $P < 0.03$ ) more body length (4.6, 12.9, and 6.4 cm) than calves fed C or S. Feeding a higher CP CS improved growth rates, but MC CS further increased body length when fed pasteurized waste milk during the summer heat stress.

**Key Words:** waste milk, corn hybrid, calf starter

**157 Vitamin D status of dairy calves fed pasteurized whole milk.** Jessica L. Powell\*<sup>1</sup>, Kathryn E. Merriman<sup>1</sup>, Mary E. Drewnoski<sup>2</sup>, and Corwin D. Nelson<sup>1</sup>, <sup>1</sup>University of Florida, Gainesville, FL, <sup>2</sup>University of Nebraska, Lincoln, NE.

Calves need vitamin D to support bone growth and immunity. Feeding dairy calves pasteurized whole milk is a common practice, but the vitamin D status of milk-fed calves has not been widely appreciated. The objectives of this study were to determine serum 25-hydroxyvitamin D (25D) concentrations of dairy calves fed pasteurized whole milk and the effects of subcutaneous vitamin D injections on serum 25D concentrations of milk-fed dairy calves. Two experiments were conducted on 2 separate farms and serum 25D concentrations were measured using a 25D ELISA. In the first experiment, 23 Holstein calves received a vitamin AD&E (1200 IU  $\alpha$ -tocopherol, 400,000 IU retinyl-palmitate, and 40,000 IU vitamin D<sub>3</sub>; n = 11) or saline injection (n = 12) at birth, and were fed pasteurized whole milk and housed indoors. Serum 25D concentrations of the control calves were  $8.1 \pm 1.0$ ,  $11.2 \pm 2.6$ ,  $13.0 \pm 3.2$ , and  $13.3 \pm 1.6$  ng/mL (mean  $\pm$  SE) at 0 d, 7 d, 22 d, and 37 d of age, respectively. In contrast, serum 25D concentrations of the vitamin AD&E treated calves were the same as control calves at birth ( $10.0 \pm 1.0$  ng/mL), greater at 7 d and 22 d ( $25.4 \pm 2.8$  and  $24.9 \pm 3.4$  ng/mL,

respectively;  $P < 0.05$ ), and not different from controls at 37 d ( $14.3 \pm 1.6$  ng/mL; treatment x time interaction,  $P < 0.05$ ). In the second experiment, 13 Holstein bull calves received either 80,000 IU of vitamin D<sub>3</sub> via subcutaneous injection at birth and once weekly for 3 weeks ( $n = 5$ ) or no injection (control,  $n = 8$ ), and were fed pasteurized whole milk and housed under shaded structures. Serum 25D concentrations of the non-treated calves were  $13.4 \pm 3.3$ ,  $5.1 \pm 3.4$ ,  $8.0 \pm 4.1$ , and  $8.7 \pm 4.9$  ng/mL (mean  $\pm$  SE) at 0 d, 7 d, 14 d, and 21 d of age, respectively. In the vitamin D-treated calves, serum 25D was the same as control calves at birth, but was increased to  $25.6 \pm 4.4$ ,  $38.1 \pm 5.2$ , and  $41.2 \pm 6.2$  ng/mL (mean  $\pm$  SE) at 7 d, 14 d and 21 d of age, respectively (treatment x time interaction,  $P < 0.01$ ). In conclusion, milk-fed dairy calves, particularly if housed indoors or under shade, are at risk for vitamin D deficiency (serum 25D  $< 10$  ng/mL) if they do not receive supplemental vitamin D, and continuous vitamin D supplementation is needed to maintain vitamin D status of calves.

**Key Words:** dairy calf, nutrition, vitamin D

**158 Gradual weaning affects pre- and postweaning feed intake, growth, and gastrointestinal development in Holstein calves fed an elevated plane of nutrition during the pre-weaning stage.** Michael A. Steele<sup>\*1,2</sup>, Leonel Leal<sup>3</sup>, Michelle Carson<sup>1</sup>, John H. Doelman<sup>1</sup>, and John A. Metcalf<sup>1</sup>, <sup>1</sup>Nutreco Canada AgResearch, Guelph, Ontario, Canada, <sup>2</sup>University of Alberta, Edmonton, Alberta, Canada, <sup>3</sup>Nutreco Research and Development, Boxmeer, the Netherlands.

The short and long-term benefits of feeding elevated quantities of milk have been recently been established and these feeding strategies have been implemented in many production systems worldwide. The objective of this study was to characterize the effect of abrupt and gradual weaning when calves are fed an elevated plane of nutrition (1.35 kg/d milk replacer) in a twice-daily feeding scheme. At total of 55 calves were randomly assigned to treatments of abrupt (0 d step-down) or gradual (12 d step-down) weaning at 48 d of life. Calves were housed and sampled in individual pens for the duration of the experiment. Milk, starter, straw and water intake was measured on a daily basis. Bodyweight was measured every 6 d until d 35 and every 3 d thereafter, while blood, rumen fluid and fecal matter was collected on d 35, 48 and 54 of the experiment. Although the growth rates of the step-down calves were lower from d 35 to weaning ( $0.48 \pm 0.08$  vs.  $1.00 \pm 0.10$  kg/d;  $P < 0.01$ ), the post-weaning average daily gain was greater compared with the abruptly weaned group ( $0.86 \pm 0.12$  vs.  $0.15 \pm 0.10$  kg/d;  $P < 0.01$ ). Total rumen volatile fatty acid concentration was greater in the step-down group compared with the abrupt group on the day of weaning on d 48 ( $76.84 \pm 1.28$  vs.  $46.72 \pm 0.78$  mmol;  $P < 0.01$ ). Fecal starch percentage was lower postweaning compared with the abruptly weaned calves on d 54 ( $2.97 \pm 1.28$  vs.  $6.67 \pm 1.22\%$ ;  $P < 0.05$ ), whereas serum amyloid A concentration was not affected by weaning strategy. These results showcase the benefits of a step-down feeding scheme from an overall energy balance standpoint in twice daily feeding schemes, presumably due to the increased opportunities for the gut to adapt before weaning.

**Key Words:** calf, weaning, development

**159 Effects of pre- and postweaning nutrition on growth, efficiency, and rumen fermentation of Holstein calves.** Tana S. Dennis<sup>\*1</sup>, Michael W. Grott<sup>1</sup>, Brad W. Shelton<sup>1</sup>, and Tamilee D. Nennich<sup>1,2</sup>, <sup>1</sup>Purdue University, West Lafayette, IN, <sup>2</sup>Famo Feeds, Freeport, MN.

The objective of this study was to evaluate the interaction of preweaning and postweaning nutrition on calf performance, blood metabolites, and rumen fermentation. Holstein calves ( $43.5 \pm 5.1$  kg BW at birth; 39 heifers and 18 bulls) were assigned at 1 d of age to 1 of 4 treatments in a randomized complete block design with a  $2 \times 2$  factorial arrangement of treatments. Preweaning milk replacer (MR) treatments were a 22% CP, 20% fat (as-fed basis) MR (C) or 28% CP, 20% fat MR (H), with weaning based on starter intake. Postweaning treatments were low NFC (27% NFC on DM basis; LC) or high NFC (42% NFC; HC) grower diets fed individually for ad libitum intake from 12 to 28 wk of age. Weights, skeletal measurements, and blood were taken every 2 wk during the preweaning period. Postweaning, BW were taken every 2 wk and skeletal measurements, blood, and rumen fluid were collected monthly. Pre- and postweaning periods were analyzed separately and overall from birth to 28 wk. Calves fed H were 15 d older, 18.0 kg heavier, and consumed 58% more DM through weaning compared with C ( $P < 0.01$ ); however, feed efficiency (FE) was similar between H and C from birth to weaning ( $P = 0.24$ ). From weaning to 11 wk, DMI was 53% greater for C ( $P < 0.01$ ); however, ADG from weaning to 11 wk was similar, resulting in greater ADG from birth to 11 wk for H ( $P < 0.01$ ). Hip height, hip width, and heart girth increased 2.7, 3.6, and 3.7%, respectively, for H over C at 8 wk ( $P < 0.01$ ). Postweaning, ADG was improved for HC ( $P = 0.01$ ), resulting in an 8.7 kg advantage in BW at 28 wk ( $P = 0.04$ ). Total DMI was similar between postweaning treatments, and FE was significantly improved for HC from 12 to 28 wk ( $P < 0.01$ ). Rumen fermentation and blood profiles were altered in favor of decreased acetate ( $P = 0.09$ ), increased butyrate ( $P = 0.01$ ), and reduced rumen NH<sub>3</sub> and plasma urea N ( $P < 0.01$ ) for HC. Overall, calves fed H+HC were 12.4 kg heavier at 28 wk compared with calves fed H+LC, but similar in BW to calves fed C+HC. These results suggest calves fed a high plane of nutrition preweaning should continue to receive high planes of nutrition postweaning to maintain growth advantages.

**Key Words:** dairy calf, weaning, growth

**160 Commercial dairy farm evaluation of milk replacers with different protein sources and concentrations.** K. A. Froehlich<sup>\*1</sup>, U. Salga Vegas<sup>1</sup>, C. Soderholm<sup>2</sup>, and D. P. Casper<sup>1</sup>, <sup>1</sup>South Dakota State University, Brookings, SD, <sup>2</sup>Milk Specialties Global, Eden Prairie, MN.

High protein (accelerated) milk replacers (MR) have increased skeletal frame growth and average daily gain (ADG) in dairy calves. However, high protein MR often cost more money than dairy producers are willing to spend for the improved performance. Our objectives were to determine the growth performance of calves fed a 24/20 (protein/fat) MR made from alternative protein sources that equal the cost of a 20/20 all milk protein MR. Eighty-eight Holstein calves raised on a South Dakota (SD) commercial dairy operation were blocked by birth date and randomly assigned to 1 of 3 treatments consisting of a control 20:20 all milk protein MR (C20), 24:18 all milk protein MR (C24), and a 24:18 alternative protein MR (A24) where the CP sources were based on 11% milk, 5% wheat, and 8% plasma. Colostrum was fed the first 3 d of life and then MR was fed at a rate of 0.68 kg/calf/d at twice daily for 35 d via bottle. Feedings were reduce to once daily at 36 d to facilitate weaning at 42 d. All MR had neomycin and oxytetracycline added at 1,330 g/ton each and supplemented with SAF Mannan. Calves were housed in individual poly-dome hutches bedded with straw with ad libitum access to a 20% CP calf starter (CS) and water. Calves fed all 3 MR were similar ( $P > 0.10$ ) in ADG (0.59, 0.63, 0.63 kg/d for C20, C24, and A24, respectively). Gains in body weight, hip width, wither

height, and body length during the 56 d study were similar ( $P > 0.10$ ) among calves fed all MR. Calves fed C24 MR had greater ( $P < 0.05$ ) gains in hip height than calves fed C MR (6.2, 10.6 and 8.5 cm) with calves fed A24 being intermediate. Calves fed A24 MR tended ( $P < 0.08$ ) to have greater gains in heart girth (15.8, 16.8 and 18.0 cm) than calves fed C20 with calves fed C24 being intermediate. Feeding a MR having more protein can support greater frame growth. The use of wheat and

plasma protein to replace portions of all milk protein in MR can result in better frame growth when calves are raised on a commercial dairy. Thus, feeding a 24/18 MR can be cost neutral to the feeding of a 20/20 MR, but potentially support improved frame growth.

**Key Words:** calf, wheat protein, plasma protein



## Ruminant Nutrition: Dairy rumen fermentation

**161 Methanogenesis reduction ability of monensin and essential oils from two Nigerian citrus species.** Musibau A. Bamikole<sup>1,2</sup>, Ibukun M. Ogunade<sup>\*1</sup>, Felipe Amaro<sup>1</sup>, Yun Jiang<sup>1</sup>, Thiago F. Bernardes<sup>1</sup>, Darren D. Henry<sup>3</sup>, Vania R. Vasconcelos<sup>1</sup>, F. O. Ugiagbe<sup>2</sup>, U. J. Ikhatua<sup>2</sup>, Nicolas DiLorenzo<sup>3</sup>, and Adegbola T. Adesogan<sup>1</sup>, <sup>1</sup>University of Florida, Gainesville, FL, <sup>2</sup>University of Benin, Benin City, Nigeria, <sup>3</sup>North Florida Research and Education Center, University of Florida, Marianna, FL.

The effects of essential oils from sweet orange (*Citrus sinensis*, EOS) and tangerine (*Citrus tangerina*, EOT) on rumen fermentation, methane production and digestibility were studied. A corn silage-based TMR (0.5 g; CP 16.6%; NDF 35.9%) was treated with EOT or EOS at rates of 0 (Control), 10 (Low), 20 (Med) and 30 (High)  $\mu\text{L}/50\text{ mL}$  of rumen fluid-buffer inoculum (ratio 1:2) and with monensin (0.6 mg/50 mL). Each suspension was incubated in a 120-mL gas-tight culture bottle in triplicate at 39°C for 24 h in each of 2 runs. Fermentation parameters, gas and methane production, in vitro DM digestibility (DMD), and fermentation efficiency (FE; DMD  $\text{g kg}^{-1}/\text{gas volume}$ ) were measured. Data for each EO were separately analyzed with the Glimmix procedure of SAS. Adding EOS or EOT did not depress DMD (g/kg; 541 and 548, respectively) but adding monensin did (555 vs. 526;  $P < 0.05$ ). Gas volume (mL/g DM) was increased by Low EOS or EOT (84.5 vs. 92.8 and 96.3) and decreased by High rates or monensin (75.3, 73.6 and 66.8), respectively. Hence, FE was reduced by Low EOS or EOT (6.58 vs. 5.81 and 5.70) and increased by High rates or monensin (7.52, 7.43 and 7.92), respectively. Methane production (mg/g DM digested) was increased by Low EOS and EOT (11.4 vs. 14.4 and 15.3) and reduced by High rates and monensin (7.10, 7.10 and 6.58; decreases of 37.7, 37.7 and 42.2%), respectively. Ammonia-N concentration (mg/dL) was increased by Med EOS (21.43) and EOT (20.6) versus monensin (4.63) or Control (11.3). Ruminant pH was increased by monensin (5.66 vs. 5.75) but not by EOT (5.63 – 5.72) or EOS (5.67 to 5.71). Total VFA concentrations (mmol) of EOS (54.7 to 69.2), EOT (68.24 to 121.8), monensin (71.0) and control (80.2) treatments did not differ ( $P > 0.05$ ). Monensin decreased acetate molar proportion and increased that of valerate, and Low EOT reduced the molar proportion of propionate. High doses of EO from *Citrus sinensis* and *Citrus tangerina* reduced in vitro methane production without adverse effects on feed digestibility and VFA production. By comparison, monensin reduced methane production and digestibility.

**Key Words:** essential oil, citrus, in vitro fermentation

**162 Changes in fermentation and biohydrogenation intermediates in continuous cultures fed corn grains differing in rates of starch degradability.** Kaylin Young<sup>1</sup>, Louisa Bowen<sup>1</sup>, Mariano Alende<sup>1</sup>, Gustavo Lascano<sup>1</sup>, Mark D. Holt<sup>2</sup>, and Thomas Jenkins<sup>\*1</sup>, <sup>1</sup>Clemson University, Clemson, SC, <sup>2</sup>Matrix Nutrition LLC, Chandler, AZ.

Excessive amounts of starch in diets for lactating dairy cattle is a known risk factor for milk fat depression but little is known about how these risks are altered by differences in rates of starch degradability ( $K_d$ ) in the rumen. The objective of this study was to compare accumulation of biohydrogenation intermediates causing milk fat depression, including conjugated linoleic acid (CLA), when corn with low or high  $K_d$  were fed to continuous cultures. Diets contained (DM basis) 50% forage (alfalfa pellets and grass hay) and 50% concentrate with either no added fat (LF) or 3.3% added soybean oil (HF). Three LF and 3 HF diets contained corn

sources with either low, medium, or high  $K_d$  (48.4, 66.2, or 84.0% in a 7 h in vitro test) giving 6 diet treatments with a  $2 \times 3$  factorial arrangement. Each diet was fed to dual-flow continuous fermenters 3 times daily at 0800, 1600, and 2400 h. Diets were fed for 4 10 d periods, with 7d for adaptation and 3 d for sample collection. No fat  $\times$  starch interactions occurred ( $P > 0.05$ ) so main effects are presented. Starch effects were tested by linear (L) or quadratic (Q) contrasts. The LF and HF treatments differed ( $P < 0.05$ ) in acetate (53.0 vs. 49.0 mol/100 mol), acetate/propionate (2.00 vs. 1.70), *trans*-10 18:1 (2.7 and 13.6% of total), and *trans*-10,*cis*-12 CLA (0.15 and 0.41% of total), respectively. Increasing starch  $K_d$  from low to high increased ( $P < 0.05$ ) culture pH (L, 6.22, 6.23, 6.38) and acetate (Q, 48.7, 50.6, 53.9 mol/100 mol) but decreased ( $P < 0.05$ ) butyrate (Q, 16.4, 15.0, 12.2) mol/100 mol. Changes in biohydrogenation intermediates (expressed as % of total fatty acids) from low to high  $K_d$  included decreases ( $P < 0.05$ ) in *trans*-11 18:1 (Q, 8.58, 4.78, 4.46) and *cis*-9,*trans*-11 CLA (Q, 0.40, 0.34, 0.20) but an increase ( $P = 0.07$ ) in *trans*-10,*cis*-12 CLA (L, 0.23, 0.24, 0.37). The results show that increasing the starch  $K_d$  in continuous culture while holding starch level constant causes elevation of some biohydrogenation intermediates linked previously to milk fat depression.

**Key Words:** lipid, biohydrogenation, starch

**163 Effects of functional oils and monensin supplementation on ruminal fermentation and milk production and composition in Holstein cows under heat stress.** Maurício F. Martins<sup>1</sup>, Arlindo S. Netto<sup>1</sup>, Paulo R. Leme<sup>1</sup>, Maria G. Pinheiro<sup>2</sup>, Joan Torrent<sup>\*3</sup>, Katiéli C. Welter<sup>1</sup>, and Isadora Arruda<sup>4</sup>, <sup>1</sup>Univ. São Paulo, Pirassununga, SP, Brazil, <sup>2</sup>Agência Paulista de Tecnologia dos Agronegócios, Ribeirão Preto, SP, Brazil, <sup>3</sup>Oligo Basics USA LLC, Cary, NC, <sup>4</sup>Univ. Estadual Paulista, Botucatu, SP, Brazil.

Eight Holstein lactating cows ( $68 \pm 3.46$  d in milk) were assigned to 2 contemporary Latin squares in a  $2 \times 2$  factorial design during the summer months. The 4 treatments were (1) no additive, (2) monensin supplementation at a rate of 30 mg/kg DM, (3) supplementation of 0.5 g/kg DM of a commercial blend of functional oils containing cashew nut shell liquid and castor oil as active ingredients (Essential, Oligo Basics Agroind. Ltda., Cascavel, Brazil), and (4) supplementation of both monensin and functional oils at the rates supplemented in treatments 2 and 3. Average temperature heat indexes ranged between 70 and 90. Whereas the inclusion of functional oils increased intake (15.30 vs. 16.67 kg;  $P < 0.001$ ), monensin decreased it (16.89 vs. 15.57 kg;  $P < 0.001$ ). Feeding functional oils increased 3.5% fat corrected milk ( $P < 0.001$ ) but only when cows were not supplemented with monensin (interaction monensin  $\times$  functional oils:  $P < 0.01$ ). Also, the supplementation of functional oils increased both the protein and fat yield ( $P < 0.01$ ) and the protein percentage in the milk ( $P < 0.01$ ). Monensin supplementation improved milk production efficiency ( $P < 0.01$ ) but decreased fat percentage ( $P < 0.05$ ) and total solids in milk ( $P < 0.01$ ). Only the supplementation of monensin changed the ruminal fermentation parameters, increasing propionate ( $P < 0.05$ ), and consequently decreasing the acetate to propionate ratio ( $P < 0.05$ ). Rectal and skin temperatures were not affected by any treatment. However, the respiratory frequency was increased by the supplementation of functional oils ( $P < 0.05$ ). In conclusion, whereas the inclusion of functional oils improved the cows' productive parameters without altering rumen fermentation, monensin supplementation decreased the amount of milk fat.

**Key Words:** functional oil, monensin, heat stress

**164 Shifts in methanogen archaea and anaerobic fungi in the rumen of dairy cows during the transition period.** Sanjay Kumar\*, Nagaraju Indugu, Bonnie Vecchiarelli, and Dipti Pitta, *Department of Clinical Studies, School of Veterinary Medicine, New Bolton Center, University of Pennsylvania, Kennett Square, PA.*

Anaerobic rumen fungi (ARF) play an important role in the degradation of dietary plant cellulosic material. Subsequent decomposition products (mainly H<sub>2</sub> and CO<sub>2</sub>) are utilized by other microbes, including methanogens. In the present study, we investigated the shift in ARF and methanogenic communities in dairy cows fed S1 diet (80% forage) 4 weeks before calving and moved to S2 diet (50% forage) after calving. Five cows from 2 study groups (SG: primiparous and multiparous) were sampled for ruminal contents before and after calving using a stomach tube. The genomic DNA from all rumen samples were amplified using archaeal and fungal specific primers, sequenced on a 454 Jr Roche platform and analyzed using QIIME pipeline. Approximately 18,317 and 35,582 reads were analyzed from 20 archaeal and ARF communities, respectively, resulting in 180 and 310 OTUs for archaea and ARF, respectively. Community comparisons (the Bray-Curtis distance matrix) revealed the effect of diet ( $P < 0.001$ ) on ARF composition, while archaea communities differed between primiparous and multiparous cows ( $P < 0.05$ ). Among ARF lineages, genus *Cyllamyces* was higher on S2 diet whereas, *Cecomycetes* and *Orpinomyces* were higher on S1 diet, irrespective of parity. *Methanobrevibacter* showed >95% abundance across all samples. A co-occurrence analysis using dice index was performed, incorporating taxa from bacteria (published recently) and archaea and fungi from this study to determine the effect of diet and parity on inter-microbial relationships within the rumen. The co-existence patterns both within and between bacteria, archaea and ARF were more influenced by SG than by diets. In conclusion, the findings presented here indicate the influence of dry matter intake, stage of production and parity on rumen microbial communities.

**Key Words:** anaerobic fungi, co-occurrence, diet

**165 Effects of stocking density and source of forage fiber on short-term responses in ruminal fermentation and behavior of Holstein dairy cows.** Mackenzie A. Campbell\*<sup>1,2</sup>, Kurt W. Cotanch<sup>1</sup>, Catherine S. Ballard<sup>1</sup>, Heather M. Dann<sup>1</sup>, and Richard J. Grant<sup>1</sup>, <sup>1</sup>The William H. Miner Agricultural Research Institute, Chazy, NY, <sup>2</sup>The University of Vermont, Department of Animal Science, Burlington, VT.

Understanding the interaction of stocking density and diet is vital for dairy cow well-being and ruminal health. Multiparous (n = 11) and primiparous (n = 5) ruminally cannulated Holstein cows (116 ± 84 d in milk and 46 ± 9 kg milk/d) were assigned to 1 of 4 pens to determine the short-term effects of stocking density and source of forage fiber on ruminal fermentation and behavior. Pens were assigned to treatments in a 4 × 4 Latin square with 14-d periods using a 2 × 2 factorial arrangement. Two stocking densities (STKD; 100 or 142% of stalls and headlocks) and 2 diets (straw; S and no straw; NS) resulted in 4 treatments: (1) 100NS, (2) 100S, (3) 142NS, and (4) 142S. Dietary forage content consisted of 39.7% corn silage and 6.9% haycrop silage versus 39.7% corn silage, 2.3% haycrop silage, and 3.5% chopped straw (dry matter; DM basis) for NS and S, respectively. Both diets were formulated for 16% crude protein, 28% neutral detergent fiber (NDF), and 28% starch (DM basis). Alterations in forage fiber source resulted in physically effective NDF values of 18.8% and 20.1% for NS and S, respectively. Ruminal pH measurements were recorded on d 12–14 of each period using indwelling pH loggers. Time spent feeding and ruminating were measured using 72-h direct observation on d 8–10 of each period. Data were analyzed

using the MIXED procedure in SAS with pen as the experiment unit. Eating time (248 min/d, SEM = 9) and rumination time (496 min/d, SEM = 23) did not differ ( $P > 0.05$ ) among treatments. Higher STKD tended ( $P < 0.07$ ) to have a lower mean and maximum pH and significantly reduced ( $P < 0.01$ ) time that pH < 5.8. Area under the curve (AUC) and time spent below pH 5.8 were significantly reduced with S. Higher STKD reduces ruminal pH and S tends to counteract this effect.

**Table 1 (Abstr. 165).** Ruminal pH in Holstein cows fed 2 stocking densities (STKD; 100 or 142% of stalls and headlocks) and 2 diets (straw; S and no straw; NS)

Item	100%		142%		SEM	STKD	P-value	
	NS	S	NS	S			Diet	STKD × Diet
Mean pH	6.17	6.13	6.09	6.10	0.03	0.07	0.62	0.39
Minimum pH	5.70	5.67	5.62	5.59	0.05	0.11	0.53	0.95
Maximum pH	6.63	6.58	6.56	6.53	0.04	0.07	0.22	0.68
ph < 5.8, h/d	2.29	1.90	4.12	2.77	0.41	<0.01	0.01	0.10
AUC < 5.8, pH units × min/d	0.38	0.19	0.58	0.34	0.10	0.06	0.03	0.75

**Key Words:** overcrowding, physically effective fiber, ruminal pH

**166 Rumen bacterial communities in three breeds of dairy cattle shift from early to peak lactation.** Melissa L. Bainbridge\*<sup>1</sup>, Laura M. Cersosimo<sup>1</sup>, André-Denis G. Wright<sup>2</sup>, and Jana Kraft<sup>1</sup>, <sup>1</sup>University of Vermont, Burlington, VT, <sup>2</sup>University of Arizona, Tucson, AZ.

Rumen bacteria form a dynamic, complex symbiotic relationship with their host, degrading fibrous forages to provide volatile fatty acids (VFA) as energy to the animal. The objective of this study was to characterize rumen bacteria and VFA in 3 breeds of primiparous dairy cattle, Holstein (HO, n = 7), Jersey (JE, n = 8), and HO × JE crossbred (CB, n = 7), at early (3 DIM) and peak lactation (93 DIM). All cows were fed a consistent TMR at a 70:30 forage to concentrate ratio. Rumen digesta were collected via esophageal intubation at 3 and 93 DIM. Microbial DNA was extracted and sequenced using Illumina MiSeq (v. 3) following PCR amplification of the V1-V3 region of the 16S rRNA gene. Sequences were analyzed using Mothur. The 16S copy numbers of rumen bacterial densities were quantified by real-time PCR. Data were analyzed using a repeated measures general linear mixed model in SAS. Breed (B) had no effect on rumen VFA, however, molar concentrations of acetate, butyrate, and propionate were lower at 93 DIM (74.7, 10.7, and 20.8 mM, respectively) than at 3 DIM (93.8, 14.7, and 26.9 mM, respectively;  $P < 0.01$ ). The quantity of bacteria in rumen digesta was unaffected by B or lactation stage (LS). Overall, Bacteroidetes (Bd) was the predominant phylum, accounting for 54–78% of total bacteria, followed by the phyla Firmicutes (Fc; 19–42%) and Proteobacteria (1–4%). *Prevotella* was the predominant genus of the Bd phylum and was unaffected by B, however, *Prevotella* species did increase with LS (49.8% vs. 67.3% for 3 DIM and 93 DIM, respectively;  $P < 0.01$ ). At 93 DIM there was a lower abundance of Fc (38.5%) than at 3 DIM (25.0%;  $P < 0.01$ ), with bacteria belonging to the genera *Butyrivibrio* and *Coprococcus* decreasing from 3 DIM (1.74% and 1.33%) to 93 DIM (0.26% and 0.30%;  $P < 0.01$ ). Bacteria belonging to the genera *Mogibacterium* and *Ruminococcus* within the Fc phylum were affected by B ( $P < 0.05$ ), HO had less *Mogibacterium* species than CB and JE (0.58% vs. 1.22% and 1.26%), and HO had higher abundance of *Ruminococcus* species than CB and JE (2.3% vs. 1.5% and 1.3%;  $P <$

0.05). In conclusion, LS had a greater effect on bacterial communities and VFA than host genetics.

**Key Words:** volatile fatty acid, bacterial diversity

**167 Effect of 3-nitrooxypropanol on ruminal fermentation, methane and hydrogen emissions, and methane isotopic composition in dairy cows.** Laiz F. de Matos<sup>1</sup>, Michael T. Harper<sup>1</sup>, Juliana Lopes\*<sup>1</sup>, Fabio Giallongo<sup>1</sup>, Joonpyo Oh<sup>1</sup>, Danielle Gruen<sup>2</sup>, Alexander N. Hristov<sup>1</sup>, Maik Kindermann<sup>3</sup>, and Stephane Duval<sup>4</sup>, <sup>1</sup>Department of Animal Science, The Pennsylvania State University, University Park, PA, <sup>2</sup>Department of Earth, Atmospheric and Planetary Sciences, Massachusetts Institute of Technology, Cambridge, MA, <sup>3</sup>DSM Nutritional Products, Animal Nutrition and Health, Basel, Switzerland, <sup>4</sup>DSM Nutritional Products France, Research Centre for Animal Nutrition and Health, Saint Louis Cedex, France.

The objective of this crossover experiment was to investigate the effect of a methane inhibitor, 3-nitrooxypropanol (3NOP), on rumen fermentation and enteric CH<sub>4</sub> emission in lactating dairy cows. Six ruminally-cannulated late-lactation (235 DIM; SD = 20 d) Holstein cows were assigned to 2 treatments: control and 3NOP (60 mg/kg DMI). Each experimental period consisted of 10 d for adaptation and 4 d for sample collection. Compared with the control, 3NOP decreased ( $P < 0.001$ ) CH<sub>4</sub> emission by 31% (487 vs. 335 g/d, respectively) and increased ( $P < 0.001$ ) that of H<sub>2</sub> from 0.005 to 1.33 g/d. CH<sub>4</sub> emissions per kg of DMI or milk yield were also decreased ( $P < 0.001$ ) 34 and 37%, respectively, by 3NOP. The isotopic composition of CH<sub>4</sub> was similar between treatments: control,  $\delta^{13}\text{C}_{\text{CH}_4} = -20.91 \pm 0.32\text{‰}$ ,  $\delta\text{D}_{\text{CH}_4} = -266.92 \pm 0.14\text{‰}$ , and  $\Delta^{13}\text{C}_3\text{D} = -1.96 \pm 1.78\text{‰}$ ; and 3NOP,  $\delta^{13}\text{C}_{\text{CH}_4} = -24.91 \pm 1.72\text{‰}$ ,  $\delta\text{D}_{\text{CH}_4} = -266.94 \pm 0.27\text{‰}$ , and  $\Delta^{13}\text{C}_3\text{D} = -1.72 \pm 2.97\text{‰}$ . Concentrations of total VFA and propionate in ruminal fluid were not affected by treatment. Acetate concentration tended to be lower ( $P = 0.08$ ) and acetate:propionate ratio was lower ( $P < 0.001$ ) for 3NOP compared with the control. Butyrate and isovalerate concentrations tended to be or were increased ( $P \leq 0.08$ ) by 3NOP. Methanogenic archaea (*Methanobrevibacter*, *Methanosphaera*, and *Methanomicrobium*) were not affected ( $P \geq 0.46$ ) by 3NOP. *Prevotella* spp., the predominant bacterial genus in ruminal contents (22 to 23% of the total isolates), was also not affected ( $P = 0.54$ ) by 3NOP. Compared with the control, *Ruminococcus* and *Clostridium* spp. were decreased ( $P \leq 0.03$ ) and *Butyrivibrio* spp. was increased by 3NOP: 8.2 vs. 6.5%, 6.2 vs. 4.1%, and 3.6 vs. 4.8%, respectively. This experiment demonstrated that a substantial inhibition of enteric CH<sub>4</sub> emission in dairy cows resulted in increased H<sub>2</sub> emission and decreased acetate concentration, but had no effect on rumen archaea. The isotopic composition of CH<sub>4</sub> was similar between the 2 treatments, supporting the conclusion that there was little to no change in the metabolic strategy of the rumen archaeal population.

**Key Words:** methane, 3-nitrooxypropanol, archaea

**168 Divergent fermentation patterns of grass fructan, inulin, and glucose.** Mary Beth Hall\*, US Dairy Forage Research Center, USDA-ARS, Madison, WI.

Fructans are an important nonfiber carbohydrate in cool season grasses. Their fermentation by rumen microbes is not well described, though such information is needed to understand their nutritional value to ruminants. Fermentation kinetics and product formation from orchardgrass fructan (phlein; PHL), chicory inulin (INU), and glucose (GLC) were compared when fermented in vitro with mixed rumen microbes. Studies were carried out as randomized complete block designs. All rates given are fractional exponential. Significance was declared at  $P < 0.05$ , and tendency at  $0.05 \leq P < 0.10$ . Rate of substrate disappearance tended to

be greater for GLC than for PHL and INU which tended to differ from each other (0.74, 0.62, and 0.33 h<sup>-1</sup>, respectively). Disappearance of GLC had almost no lag time (0.04 h) whereas the fructans had lags of 1.4 h. The maximum microbial N accumulation (a proxy for cell growth), tended to be 20% greater with PHL and INU than with GLC. The N accumulation rate with GLC (1.31 h<sup>-1</sup>) was greater than with PHL (0.75 h<sup>-1</sup>) and INU (0.26 h<sup>-1</sup>) which also differed. More microbial glycogen (+57%) was accumulated with GLC than with PHL, though accumulation rates did not differ (1.95 and 1.44 h<sup>-1</sup>, respectively); little glycogen accumulated with INU. Rates of organic acid formation were 0.80, 0.28, and 0.80 h<sup>-1</sup> for GLC, INU, and PHL, respectively, with PHL tending to be greater than INU. Lactic acid production was more than 7-fold greater for GLC than for the fructans. The ratio of microbial cell carbon to organic acid carbon tended to be greater with PHL (0.90) and INU (0.86) than with GLC (0.69) indicating a greater yield of cell per amount of substrate fermented with fructans. Reduced microbial yield with GLC may relate to the greater glycogen production which requires ATP, and lactate production which yields less ATP; together, these processes could have reduced ATP available for cell growth. Acetate molar proportion was less with GLC than with fructans, and less for PHL than for INU. Rumen microbes ferment PHL differently than other plant sugars or fructans.

**Key Words:** rumen fermentation, fructans, glycogen

**169 The effect of lactic acid bacteria as probiotics or silage inoculants on in vitro rumen digestibility, total gas and methane production.** Jennifer L. Ellis\*<sup>1,2</sup>, Andre Bannink<sup>3</sup>, Ida K. Hindrichsen<sup>4</sup>, Robert D. Kinley<sup>1</sup>, Wilbert F. Pellikaan<sup>1</sup>, Nina-Lotte Milora<sup>4</sup>, and Jan Dijkstra<sup>1</sup>, <sup>1</sup>Animal Nutrition Group, Wageningen University, Wageningen, the Netherlands, <sup>2</sup>Centre for Nutrition Modelling, Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada, <sup>3</sup>Animal Nutrition, Wageningen UR Livestock Research, Wageningen, the Netherlands, <sup>4</sup>Chr. Hansen A/S, Hørsholm, Denmark.

Lactic acid bacteria (LAB) included as a probiotic or silage inoculant may affect rumen fermentation, OM digestibility and methane (CH<sub>4</sub>) emissions in cattle. Therefore, 2 in vitro gas production trials were conducted to pre-screen several potential LAB inoculants at several inoculation levels, using different LAB mixtures, and on different silage substrates. In Experiment 1 the dose-response effects of 3 LAB inoculants added as probiotics (0.0,  $5 \times 10^5$ ,  $1 \times 10^6$  and  $5 \times 10^6$  cfu/mL) on in vitro total gas and CH<sub>4</sub> production were examined using grass silage as the substrate. In Experiment 2, 3 LAB inoculant mixtures were examined while varying the substrate. Substrates were inoculated with LAB before ensiling, and were ryegrass/clover (RCS), corn (CS) and ryegrass (RS) silage. Data were analyzed with proc MIXED of SAS with LAB inoculant  $\times$  dose as a fixed effect, and dose was analyzed via orthogonal polynomial contrasts (Experiment 1), and using substrate, inoculation and substrate  $\times$  inoculation as fixed effects (Experiment 2). Results showed that not all LAB affected in vitro fermentation. In Experiment 1, *L. plantarum* (LP) but not *L. lactis* (LL) or a 1:1 mixture of LL and LP, resulted in significant increases in OM digestibility ( $P = 0.023$ ), and there was a trend for several dose related responses. In Experiment 2, LAB showed both strain and substrate-specific responses. In RS, an inoculation of a mixture of *L. plantarum*, *L. buchneri* and *L. lactis* (LM1) increased OM digestibility, while inoculations of *L. buchneri* and *L. lactis* (LM2) and *L. plantarum*, *L. lactis* and *E. faecium* (LM3) decreased OM digestibility in RCS (inoculation  $P < 0.001$ ). These effects were generally mirrored by changes in gas and CH<sub>4</sub> production. In CS, no effects were observed on OM digestibility, total gas or CH<sub>4</sub> production. From these results we conclude that LAB



may be most effective in grass based silages (compared with corn) for altering OM digestibility, and that the LP treatment from Experiment 1, or the LM1 treatment from Experiment 2, may be most promising for evaluation in vivo.

**Key Words:** in vitro, lactic acid bacteria, cattle

**170 Influence of diet change frequency on growth, rumen fermentation, and behavior of prepubertal dairy heifers.** Tana S. Dennis<sup>\*1</sup>, Jason E. Tower<sup>1</sup>, Hans F. P. Schmitz<sup>1</sup>, Amanda M. Mosiman<sup>1</sup>, and Tamilee D. Nennich<sup>1,2</sup>, <sup>1</sup>Purdue University, West Lafayette, IN, <sup>2</sup>Famo Feeds, Freeport, MN.

The objective of this study was to evaluate the effects of diet change frequency on dairy heifer performance, rumen fermentation, and feeding behavior. Ninety Holstein heifers (141.8 ± 11.7 kg of BW) in 15 pens were randomly assigned to treatments designed to change diets using rapid (30 to 60% hay; 1STEP), moderate (30 to 45 to 60% hay; 2STEP), or gradual steps (30 to 40 to 50 to 60% hay; 3STEP). Heifers were fed for 84 d, and diets were changed every 2 wk starting on d 28. Weights were taken every 2 wk, and skeletal measurements and blood samples were collected monthly. Rumen fluid was collected esophageally 6 h after feeding (2 heifers/pen) to determine pH, NH<sub>3</sub>, and volatile fatty acids (VFA) 2 wk after each diet change. Behavior was evaluated using scan-sampling before and after diet changes for 30 min before and 120 min following feed delivery. Data were analyzed with pen as the experimental unit. On d 84, 3STEP heifers were heavier than 1STEP ( $P = 0.05$ ) and tended to be heavier than 2STEP ( $P = 0.06$ ). Daily gain was significantly improved for 3STEP after the second diet change ( $P < 0.01$ ), but lower after the last diet change compared with 1STEP and 2STEP ( $P = 0.05$ ). Feed intake was greater ( $P = 0.02$ ) for 3STEP from d 35 to 63, yet similar among treatments from d 63 to 84. Skeletal growth was similar with the exception of heart girth, which was greatest for 3STEP on d 84 ( $P = 0.01$ ). Blood glucose was greatest for 3STEP after the first diet change ( $P = 0.02$ ) and greatest for 2STEP after the last diet change ( $P = 0.04$ ). Rumen fermentation profiles were altered following the first diet change, as proportions of acetate ( $P < 0.01$ ) and isoacids ( $P = 0.10$ ) were greatest and total VFA, propionate ( $P < 0.01$ ), and valerate ( $P < 0.01$ ) were lowest for 1STEP. Heifers fed using 1STEP spent 51% more time feeding than 3STEP heifers 3 d after the first diet change ( $P < 0.01$ ). Time spent feeding was 18% ( $P = 0.01$ ) and 14% ( $P = 0.01$ ) greater 6 d before the second diet change for 1STEP and 2STEP, respectively, compared with 3STEP. Rapid diet changes appear to alter rumen fermentation and feeding behavior, which potentially reduces performance of growing dairy heifers.

**Key Words:** heifer, feeding, growth

**171 Effects of rumen inoculum adapted and unadapted to *Saccharomyces cerevisiae* fermentation product, culture pH, and starch fermentability on the biohydrogenation of unsaturated fatty acids in batch culture.** Yan Sun<sup>\*</sup>, Michael S. Allen, and Adam L. Lock, Michigan State University, East Lansing, MI.

The effect of rumen fluid (RF) inoculum either unadapted (U-RF) or adapted (A-RF) to *Saccharomyces cerevisiae* fermentation product (SCFP; Diamond V Original XPC) on the biohydrogenation (BH) of unsaturated fatty acids (FA) at 2 pH levels and starch sources with different fermentabilities (SF) were evaluated. Rumen inocula for batch cultures were collected at the end of each 4-wk feeding period in a crossover design ( $n = 6$ ). U-RF or A-RF cultures (4 replicates/treatment) were incubated for 24 h at pH 5.8 or 6.2 and included alfalfa hay (55% of DM), and either dry ground corn (DC) or high moisture corn (HMC) as starch sources (45% of DM). The alfalfa hay was treated with corn

oil (2% of DM) to increase the total unsaturated FA content of cultures. Effects of RF, culture pH, SF, and their interactions were determined. For main effects of treatments, A-RF compared with U-RF and pH 6.2 compared with pH 5.8 increased extent of BH for *cis*-9,*cis*-12 18:2 (41 vs. 38% and 47 vs. 32%, respectively; both  $P < 0.001$ ) and NDFD (14 vs. 12% and 16 vs. 10%, respectively; both  $P < 0.001$ ). Compared with DC, HMC increased BH extent for *cis*-9,*cis*-12 18:2 (41 vs. 39%,  $P = 0.06$ ), and decreased NDFD (12 vs. 14%,  $P < 0.001$ ). Overall, compared with U-RF, A-RF decreased *trans*-10,*cis*-12 18:2 (CLA) by 17% ( $P < 0.001$ ), and increased *trans*-10 18:1 by 10% ( $P < 0.001$ ). HMC compared with DC, and pH 5.8 compared with pH 6.2, increased content of CLA by 15 and 56%, respectively (both  $P < 0.01$ ), and increased *trans*-10 18:1 by 23 and 26%, respectively (both  $P < 0.001$ ). RF interacted with SF for CLA and *trans*-10 18:1 (both interactions  $P < 0.01$ ); in particular, for cultures containing HMC, A-RF decreased the content of CLA and increased the content of *trans*-10 18:1 compared with U-RF. Results demonstrate that under the conditions tested, rumen fluid from cows adapted to SCFP (A-RF) had positive effects on the extent of BH of unsaturated FA and NDFD. A-RF decreased CLA across culture pH, especially when HMC was the starch source.

**Key Words:** culture pH, dry ground corn, high moisture corn

**172 Effects of *Saccharomyces cerevisiae* fermentation products on performance of mid-lactation dairy cows.** Subash Acharya<sup>\*1</sup>, Jon P. Pretz<sup>1</sup>, Ilkyu Yoon<sup>2</sup>, Mark F. Scott<sup>2</sup>, and David P. Casper<sup>1</sup>, <sup>1</sup>South Dakota State University, Brookings, SD, <sup>2</sup>Diamond V, Cedar Rapids, IA.

This study was conducted to evaluate *Saccharomyces cerevisiae* fermentation products (Diamond V Original XPC and 2 prototypes) on production efficiency of mid-lactation dairy cows. Eighty mid-lactation (164.5 DIM ± 67.5) Holstein cows (56 multiparous and 24 primiparous) were blocked by parity, days in milk, and milk production, and randomly assigned to one of 4 treatments. Treatments consisted of (1) Control (C): corn silage and haylage based ration; (2) XPC: C ration with Original XPC added at 14 g/hd per day; (3) Prototype 1 (P1): C ration with Prototype 1 added at 5 g/hd per day; and (4) Prototype 2 (P2): C ration with Prototype 2 added at 19 g/hd per day. Treatments were mixed with dried distillers grains and then mixed into a TMR at 454 g/hd/d. The study lasted for 70 d. The first 14 d of the study (d -14 to 0) was used to train cows to use the Calan gate feeding system and cows were fed the C ration during this period. Treatment effects were continuously monitored for the 8 wk experimental period. Dry matter intakes (DMI) were similar ( $P \geq 0.10$ ) when cows were fed all treatments (25.7, 26.1, 25.1, and 26.2 kg/d for C, XPC, P1, and P2, respectively). Milk production (33.3, 34.4, 35.5, and 36.8 kg/d) was improved ( $P < 0.05$ ) for cows fed P2 compared with cows fed C, with cows fed other supplements being intermediate and similar. Feed efficiency (Milk/DMI) was improved ( $P < 0.08$ ) for cows fed P1 and P2, compared with cows fed C and XPC (1.30, 1.34, 1.49 and 1.41 kg/kg). Milk fat content was reduced ( $P < 0.05$ ) for cows fed P2 (4.17, 3.93, 4.08, and 3.85%) compared with cows fed C, with cows fed other treatments being intermediate. Milk protein and lactose percentages were similar ( $P \geq 0.10$ ) among treatments. Cows fed P2 had lower ( $P < 0.05$ ) molar percentages of ruminal acetate (63.8, 64, 63.1, and 62.3%) and greater ( $P < 0.05$ ) propionate (18.9, 19.3, 19.7, and 20.6%) than cows fed other treatments. Supplementing a dairy ration with *Saccharomyces cerevisiae* fermentation products can improve milk production and feed efficiency of mid-lactation cows.

**Key Words:** dairy cow, *Saccharomyces cerevisiae* fermentation product, milk production

## Small Ruminant: Nutrition

**173 Blood chemistry, milk yield and composition, and milk fatty acids profile of lactating Anglo-Nubian goats fed *Moringa oleifera* leaf as a protein source.** T. A. Morsy<sup>\*1</sup>, A. E. Kholif<sup>1</sup>, G. A. Gouda<sup>1</sup>, A. Z. M. Salem<sup>2</sup>, S. M. Kholif<sup>1</sup>, and A. M. Kholif<sup>1</sup>, <sup>1</sup>Dairy Science Department, National Research Centre, Giza, Egypt, <sup>2</sup>Facultad de Medicina Veterinaria y Zootecnia, Universidad Autónoma del Estado de México, Toluca, Estado De México, Mexico.

Low availability and increased price of protein sources are a serious problem for animal producers. Therefore there is a need to find alternative ingredients for protein sources. *Moringa oleifera* leaf contains about 25% crude protein (CP) with low degradability, which make it a good source of protein in ruminant diets. In a 4 × 4 Latin square design, 16 lactating Anglo-Nubian goats (36.2 ± 0.7 kg BW) were used to evaluate the effect of replacing sesame meal (259.0 g CP/kg DM; 279.1 g neutral detergent fiber (NDF)/kg DM) with *M. oleifera* (241.2 g CP/kg DM; 336.6 g NDF/kg DM) at 0% (control-M0), 50% (M50), 75% (M75) and 100% (M100). The experimental period consisted of 17 d for adaptation + 5 d for milk yield and sampling. All goats were sampled for blood at the last day of the experimental period 4 h after morning feeding. Goats were hand milked daily at 0900 and 2100 h for determination of milk yield. There was no effect of treatment on serum total proteins, albumin, globulin, glucose, creatinine, and triglycerides concentrations, with quadratically increasing GOT and GPT concentrations ( $P < 0.05$ ) and decreasing urea-N (linear,  $P = 0.0339$ ; quadratic,  $P < 0.0001$ ) and cholesterol (linear,  $P = 0.0009$ ; quadratic,  $P < 0.0001$ ) concentrations. Milk yield and energy corrected milk were linearly increased ( $P < 0.001$ ) when goats fed on Moringa vs. control. Milk yield was greatest for M75 which had also higher total solids, solids not fat, and lactose versus other treatments. Milk components yield were increased with goats fed Moringa compared with control ( $P < 0.01$ ). Moringa treatments increased ( $P < 0.01$ ) milk fat content from C14:1, C18:1n-9 *trans*, C18:1n-9 *cis*, C18:2 *trans*-10,*cis*-12, and C18:2 *cis*-9,*trans*-11 with decreased C16:0 concentrations. Moreover, decreased ( $P < 0.05$ ) saturated with increased unsaturated fatty acids and conjugated linoleic acids (CLA) were obtained when goats fed Moringa diets vs. control. It could be concluded that replacing 75% of sesame meal with *M. oleifera* as a protein source increased milk yield with enhancing milk content of healthy fatty acids (CLA and n-3), without detrimental effects on animal performance.

**Key Words:** milk fatty acid, milk composition, *Moringa oleifera*

**174 *Moringa oleifera* leaf as a protein source in the diet of Anglo-Nubian goats affects feed intake, digestibility, and ruminal fermentation.** A. E. Kholif<sup>\*1</sup>, G. A. Gouda<sup>1</sup>, A. Z. M. Salem<sup>2</sup>, T. A. Morsy<sup>1</sup>, and S. M. Kholif<sup>1</sup>, <sup>1</sup>Dairy Science Department, National Research Centre, Giza, Egypt, <sup>2</sup>Facultad de Medicina Veterinaria y Zootecnia, Universidad Autónoma del Estado de México, Toluca, Estado De México, Mexico.

Recently, the availability and price of concentrates especially of protein sources are a serious problem for animal producers especially small farms. Therefore there is a need to find alternative ingredients for protein sources. *Moringa oleifera* leaf contains about 25% crude protein (CP) with low degradability, which make it a good source of protein in ruminant diets. Therefore, in a 4 × 4 Latin square design, the effect of including *M. oleifera* leaf as a protein source in diets of Anglo-Nubian goats on intake, digestibility and ruminal fermentation given was evalu-

ated. Sixteen goats (36.2 ± 0.7 kg BW) were fed 4 diets (n = 4 goats) where sesame meal (259.0 g CP/kg DM; 279.1 g neutral detergent fiber (NDF)/kg DM) was replaced with *M. oleifera* (241.2 g CP/kg DM; 336.6 g NDF/kg DM) at 0% (control-M0), 50% (M50), 75% (M75) and 100% (M100). The experimental period consisted of 17 d adaptation + 5 d for feed intake measurements and feces collection. Water was always available to goats. All goats were sampled for rumen liquor at the last day of each experimental period after 0, 3 and 6h of morning feeding. Goats fed on M75 and M100 diets increased feed intake of all nutrients (linear,  $P < 0.001$ ; quadratic,  $P < 0.05$ ). Moreover, increased dry matter (quadratic,  $P = 0.0014$ ), organic matter (linear,  $P = 0.0117$ ), and crude fibers (linear,  $P = 0.0119$ ; quadratic,  $P = 0.0018$ ) digestibility were obtained with goats fed on M75. However, goats fed on Moringa (M50, M75, M100) had a linearly decreased CP digestibility ( $P < 0.0001$ ). Diets nutritive values as total digestible nutrients ( $P = 0.0049$ ), digestible CP ( $P = < 0.0001$ ), metabolizable energy ( $P = 0.0054$ ) and net energy of lactation ( $P = 0.0049$ ) were quadratically decreased with M50, M75 and M100 vs. M0. Goats fed on Moringa diets had increased ( $P < 0.05$ ) ruminal pH, volatile fatty acids, propionate and methane production vs. control. It could be concluded that inclusion of *M. oleifera* leaf as a protein source at 75% of sesame meal increased feed intake, nutrients digestibility and ruminal fermentation of Anglo-Nubian goats.

**Key Words:** digestibility, *Moringa oleifera*, ruminal fermentation

**175 Effect of dietary forage-to-concentrate ratios on urinary excretion of purine derivatives and microbial nitrogen yields in the rumen of Dorper crossbred sheep.** Tao Ma<sup>\*1</sup>, Kai-dong Deng<sup>2</sup>, Yan Tu<sup>1</sup>, Nai-feng Zhang<sup>1</sup>, Cheng-gang Jiang<sup>1</sup>, and Qi-yu Diao<sup>1</sup>, <sup>1</sup>Feed Research Institute, Chinese Academy of Agricultural Sciences/Key Laboratory of Feed Biotechnology of the Ministry of Agriculture, Beijing, China, <sup>2</sup>College of Animal Science, Jinling Institute of Technology, Nanjing, Jiangsu, China.

This study investigated the effects of various dietary forage-to-concentrate ratios (F: C) on urinary excretion of purine derivatives (PD) and microbial nitrogen (MN) yields from the rumen of Dorper 'thin-tailed Han crossbred sheep. Twelve Dorper × thin-tailed Han crossbred ram lambs (47.2 ± 1.0 kg) fitted with ruminal and duodenal cannulas were randomly assigned to 12 levels of dietary F:C in a balanced incomplete block experimental design (12 lambs × 4 periods). Digestibility trials were conducted and MN was estimated using either <sup>15</sup>N or PD as markers. Urinary excretion of allantoin and total PD increased ( $P < 0.05$ ) with decreasing F:C. Urinary excretion of uric acid or xanthine plus hypoxanthine was unaffected by F:C ( $P > 0.05$ ). MN yields estimated using <sup>15</sup>N as a marker were greater than those predicted from urinary PD (12.5 vs. 11.5 g/d,  $P < 0.05$ ), but the former was more variable than the latter (SE = 0.66 vs 0.45, respectively). A linear correlation existed between MN estimated by <sup>15</sup>N and urinary excretion of PD: MN (g/d) = -0.521 + 1.493 × PD (mmol/d) ( $r^2 = 0.86$ ). The purine nitrogen index (PNI: PD nitrogen/urinary N) was linearly correlated with nitrogen capture efficiency calculated from either <sup>15</sup>N or PD ( $r^2 = 0.60$  and 0.77, respectively). Results suggest that urinary PD is an accurate indicator of MN from the rumen of Dorper crossbred sheep, and PNI reflected the conversion of nitrogen degradation to MN in the rumen.

**Key Words:** forage-to-concentrate ratio, microbial N yield, purine derivative

**176 Effects of reducing dietary cation-anion difference plus vitamin D on plasma Ca and its modulators and growth performance of goats.** Wen-xuan Wu\*, Lun-qin Zhu, Xing-zhou Tian, and Ruo-yu Liu, *College of Animal Science, Guizhou University, Guiyang, Guizhou Province, China.*

This study was conducted to evaluate the effects of reducing dietary cation-anion difference (DCAD;  $\text{Na}^+\text{K}^-\text{Cl}^-\text{S}$ , mmol/kg DM) with vitamin D (VD) supplementation on plasma levels of Ca and its modulators, and ration palatability for goats. Eighteen healthy female goats were randomly allocated to 3 treatments with 6 goats per treatment and were fed 3 DCAD levels at 150, -150, -150 for control, treatment 1, and treatment 2, respectively. Anionic salts were included to reduce DCAD level for treatment 1, and VD was added in treatment 1 for treatment 2. Urine pH; plasma Ca, P, PTH, CT,  $1,25\text{-(OH)}_2\text{D}_3$ , vitamin D receptor (VDR), Ca-binding protein (CaBP-D9k) concentrations; and dry matter intake (DMI) were measured in the trial. Urine pH was reduced ( $P < 0.05$ ) as DCAD decreased for treatment 1 (6.71) and treatment 2 (6.67) over control (7.63). Compared with control, plasma Ca was increased for treatment 1 ( $P > 0.05$ ) and treatment 2 ( $P < 0.05$ ). There was no difference in plasma P and PTH for control, treatment 1, and treatment 2 ( $P > 0.05$ ). Treatment 2 had increased plasma  $1,25\text{-(OH)}_2\text{D}_3$  relative to control and treatment 1 ( $P < 0.05$ ) and tended to reduce plasma CT over treatment 1 ( $P < 0.1$ ). Moreover, reducing DCAD plus VD for treatment 2 resulted in higher plasma VDR and CaBP-D9k than that of control ( $P < 0.05$ ). Levels of DMI were unaffected among 3 treatments ( $P > 0.05$ ). These results suggested that plasma Ca homeostasis could be more available to maintain by reducing DCAD plus VD in the present study.

**Key Words:** dietary cation-anion difference, plasma Ca, goat

**177 Iodine supplementation of the pregnant ewe alters serum IgG concentration and expression of genes associated with antibody transfer in the ileum of the newborn lamb.** Fiona M. McGovern\*<sup>1</sup>, Torres Sweeney<sup>2</sup>, Francis P. Campion<sup>1</sup>, Marion T. Ryan<sup>2</sup>, Stephen Lott<sup>1</sup>, and Tommy M. Boland<sup>1</sup>, <sup>1</sup>*School of Agriculture and Food Science, University College Dublin, Dublin, Ireland,* <sup>2</sup>*School of Veterinary Medicine, University College Dublin, Dublin, Ireland.*

Ruminant neonates require the passive acquisition of maternal immunoglobulins (Ig) derived from colostrum, to confer immunity against infectious disease in early life. Excess iodine in the diet of the pregnant dam is known to negatively affect neonatal serum IgG concentrations. This experiment examined the mechanisms underpinning the reduced IgG in the progeny of iodine supplemented dams at 24h postpartum. In a randomized complete block design, twin bearing ewes were split into 2 dietary groupings ( $n = 32$  per group). The first group had no iodine supplement and the second was supplemented with 26.6mg per ewe per day of supplementary iodine in the form of calcium iodate for the final 4 weeks of gestation. Ewes were hand milked at 1, 10, and 18 h postpartum and colostrum intake of the lambs was recorded. At 24h postpartum a subset of lambs ( $n = 12$  per group) were euthanized. Duodenal and ileal sections were collected to determine morphology and the expression of a panel of genes associated with antibody transfer. At 24h postpartum and fortnightly until 28 d postpartum IgG concentration was quantified from the serum of the remaining progeny. Iodine supplementation had no effect on colostrum yield, composition or lamb colostrum intake within the first 18h postpartum ( $P > 0.05$ ). Despite this, the progeny from the iodine-supplemented ewes had lower serum IgG concentrations up until d 28 postpartum ( $P > 0.001$ ) when compared with the progeny of the non-supplemented dams. Progeny of supplemented dams had increased expression of *PIGR* and *FCAMR* while the expression of *B2M*, *THRB*

and *ALB* declined in the ileum when compared with non-supplemented progeny ( $P < 0.05$ ). A reduction in villus height and villus height: crypt depth ratio was observed in the ileum of the supplemented progeny ( $P < 0.01$ ). In conclusion, the results of this study presents novel data describing the relationship between maternal iodine nutrition and the subsequent failure of passive transfer leading to reduced serum IgG concentrations

**Key Words:** lamb, immunoglobulin G, ileal gene expression

**178 Metabolizable energy requirements of maintenance, energy efficiency of weight gain and fat deposition in Pelibuey and Katahdin ewes in tropical Mexico.** Jose Valentin Cardenas Medina\*, Pablo Ivan Duarte Arzapalo, Dahiavis Mena Arceo, and Olivier Santiago Ramos Trejo, *Instituto Tecnológico de Tizimin, Tizimin, Yucatan, Mexico.*

The objective was to estimate metabolizable energy requirements for maintenance ( $\text{ME}_m$ ), energy efficiency of weight gain (EEWG) and fat deposition, in Pelibuey and Katahdin ewes in Yucatan, Mexico. Eight non pregnant, non lactating, multiparous ewes, with average live weight and standard deviation of  $35 \pm 2$  kg, were distributed in a complete randomized design with a  $2 \times 3$  factorial arrangement, 2 breeds and 3 feed levels (100, 120 kcal of ME  $\text{kg}^{0.75}$  and ad libitum), during 3 periods of 28 d each; the diet were composed of 50% ground *Panicum maximum* hay and 50% of a concentrate, based on corn, wheat and soybean meal, containing 2.0 Mcal/ $\text{Kg}^{-1}$  of ME and 10% of CP. Feed intake was measured by offered-rejected daily food weighing, to asses ME intake (MEI), ewes were weighed every 14 d, to asses changes in live weight (CLW) and thickness of subcutaneous fat (SF) was measured between 12th and 13th dorsal vertebra. The  $\text{EM}_m$  was estimated by regressing values of CLW against MEI ( $Y = B_0 + B_1 \times X$ ), EEWG was estimate as gram of weight gain (WG) per Mcal of MEI. There were no differences ( $P > 0.05$ ) between breeds in  $\text{EM}_m$  ( $110 \pm 4$  and  $122 \pm 4$  kcal/ $\text{kg}^{0.75}$ , for Pelibuey and Katahdin; MEI [kcal/ $\text{kg}^{0.75}$ ] =  $119.748 + 0.264107 \times \text{WG}$ ,  $r = 0.81$ ) and EEWG ( $33 \pm 8$  and  $17 \pm 8$  g/Mcal of MEI, for Pelibuey and Katahdin); differences were found ( $P < 0.01$ ) in average SF despite ewe breed ( $5.6 \pm 0.2$  and  $4.7 \pm 0.2$  mm, for Pelibuey and Katahdin). There are not difference between breeds in energy requirements for maintenance and weight gain, but difference in body composition could be advantageous for Pelibuey ewes, fat reserves could be used with high efficiency for maintenance and lactation during dry season, in sheep systems in tropical Mexico.

**Key Words:** sheep, energy, requirement

**179 Effect of dry matter content and feeding level on dry matter digestibility and intake of perennial ryegrass fed to sheep.** Brian Garry\*<sup>1,2</sup>, René Baumont<sup>3</sup>, Tommy Boland<sup>2</sup>, Michael O'Donovan<sup>1</sup>, and Eva Lewis<sup>1</sup>, <sup>1</sup>*Teagasc, AGRIC Moorepark, Fermoy, Co. Cork, Ireland,* <sup>2</sup>*School of Agriculture and Food Science, University College Dublin, Dublin, Ireland,* <sup>3</sup>*INRA, UMR1213 Herbivores, Saint Genès Champanelle, France.*

Optimising animal performance from pasture requires a high intake of highly digestible grass. In vivo dry matter digestibility (IVDMD) of grass is affected by the level of dry matter intake (DMI). The objective of this experiment was to analyze the effect of dry matter (DM) content of grass on IVDMD at 2 feeding levels in wether sheep. A Latin square design, with 4 periods and 4 treatments, was used. There were 4 sheep per treatment per period ( $n = 16$ ) with 4 treatments in a  $2 \times 2$  factorial grass type; wilted or fresh grass offered at one of 2 feeding levels; ad



libitum or maintenance. Each 12-d period consisted of a 6 d adaptation phase and a 6 d measurement phase (MP). Grass was cut daily each morning. Approximately half the grass was artificially wilted for 5 h using a centrifugal fan at 18°C (wilted). The other half of the grass was stored at 4°C (fresh). The fresh and wilted grass was fed to the sheep daily at 4 p.m. The DM content of the wilted and fresh grass was 27.8% and 18.6%, respectively. Ad libitum sheep were offered 110% of the previous day's DMI. Maintenance fed sheep were fed 40 g grass DM/kg metabolic BW. Sheep were housed in stalls to allow for individual feeding and total fecal collection. During the MP in each period, a sample of the grass fed to and feces voided by each sheep was collected daily. The daily grass and feces samples were dried and bulked to give one sample per treatment and per sheep per period. Data were analyzed using a mixed model procedure in SAS. Period, feeding level, grass type and their interactions were fixed effects and sheep was a random effect. Wilted grass (757 g/kg) had greater IVDMD than fresh grass (746 g/kg;  $P < 0.01$ ) and maintenance level feeding (759 g/kg) resulted in greater IVDMD than ad libitum (744 g/kg;  $P < 0.001$ ). When fed ad libitum, wilted grass (1.50 kg DM/day) resulted in greater DMI than fresh grass (1.40 kg DM/day;  $P < 0.05$ ) but fresh grass had greater fresh weight intake than wilted grass ( $P < 0.001$ ). This indicates that reducing perennial ryegrass moisture content results in increased IVDMD and increased DMI in sheep fed indoors.

**Key Words:** digestibility, intake

**180 Effects of feeding varying levels of deoiled distillers dried grains with solubles on dry matter intake, rumen fermentation, blood chemistry profile, growth, feed efficiency and carcass quality of meat goats.** Jarvis G. Scott\*, Nar K. Gurung, Byeng R. Min, Errol G. Rhoden, and Wendell H. McElhenney, *Tuskegee University, Tuskegee, AL.*

The feeding value of deoiled distillers dried grains with solubles (D-DDGS) has been investigated for dairy, beef cattle, pigs and poultry, but no study has been conducted with meat goats. Objectives were to determine the effects of feeding varying levels of D-DDGS on dry matter intake, rumen fermentation, select blood metabolites, growth, feed efficiency and carcass quality. Twenty-four (24) Kiko wethers (26.67 ± 1.21 kg with initial BW and 5 to 6 mo of age) were randomly assigned to one of the 4 experimental diets (n = 6) containing 50% bermudagrass hay plus 50% concentrate mix with 0, 10, 20 or 30% D-DDGS in the diet on an as-is basis. The body weights were recorded every 4 weeks. The rumen volatile fatty acid were determined at the beginning, mid-point and at end of the trial while blood chemistry profiles were determined at the beginning and end of the trial. After 84 d, goats were slaughtered for carcass traits evaluation. Data were analyzed using the MIXED procedure of SAS. There was quadratic response in dry matter intake ( $P < 0.05$ ) with increasing level of D-DDGS but no differences ( $P = 0.49$ ) were detected in ADG (111.8, 128.4, 140.3 and 98.9 g/day for control, 10, 20 and 30% D-DDGS, respectively). Similarly, no significant differences ( $P > 0.05$ ) were observed for gain to feed ratios among treatments. Molar proportion of acetate (A), propionate (P), and A: P increased linearly ( $P < 0.05$ ) while the overall rumen pH response was quadratic ( $P < 0.05$ ). The blood chemistry profile showed that gamma-glutamyl transferase and glucose decreased linearly ( $P < 0.05$ ) with D-DDGS while the trend was reversed ( $P > 0.05$ ) with aspartate aminotransferase, creatine kinase and cholesterol. However, both rumen volatile fatty acid and blood chemistry profile were within the normal range. There were no significant differences ( $P > 0.05$ ) between treatments for percent transport loss, percent carcass shrink, 12th rib fat thickness, LMA and dressing percent. The findings suggest that up to 30% of D-DDGS can

be included in the diet for meat goats without any compromise in rumen fermentation, performance and carcass quality.

**Key Words:** goat, D-DDGS, performance

**181 Economic analysis of feedlot finished lambs receiving diets based on different *Cynodon* hay grasses.** Euclides Reuter Oliveira<sup>1</sup>, Flávio Monção<sup>2</sup>, Andreia Gabriel<sup>1</sup>, Jefferson Gandra\*<sup>1</sup>, Rayanne Souza<sup>1</sup>, Lais Moura<sup>1</sup>, Loan Silva<sup>1</sup>, Leandro Silva<sup>1</sup>, Vadim Carbonari<sup>1</sup>, and Thais Lemos<sup>1</sup>, <sup>1</sup>Universidade Federal da Grande Dourados, MS, Brazil, Dourados, MS, Brazil, <sup>2</sup>Universidade Estadual Julio de Mesquita, Jaboticabal, SP, Brazil.

This study aimed to evaluate the economic viability of the performance of crossbred lambs (1/2 Santa Inês+ 1/2 Suffolk), feedlot, fed different diets of hay *Cynodon dactylon*. The experiment was conducted at the agricultural sciences college, Federal University of Grande Dourados. Thirty non-castrated, crossbred Suffolk lambs were used, identified with ear tags, with an average age of 90 d, with average body weight (21.5 kg ± 1.61). The animals were blocked by weight and randomly assigned to one of the 6 hays. Hays of each cultivar (Jiggs, Vaquero, Tifton 68 Coast-cross, Tifton 85, and Russell) were harvested at 42 d of age or stage of maturity were fed as 60% of the diet with a standard concentrate making up 40% of the diet. The treatment diets were distributed in a fully randomized block with 5 replications. Means were analyzed as mixed models using the MIXED procedure of SAS software, 9.2 version, and the average compared by Scott Knot test at 5% probability. The intake of dry matter varied from 0.858 kg for Tifton 68 to 1.06 kg for Jiggs. There were significant differences between diets for total cost on feeding, and the Jiggs presented the highest values (\$17.11/animal). The animals fed Tifton 68 hay showed higher net income (\$41.84/ animal and \$44.70), when the animal was sold for slaughter in the refrigerator or alive for rearing in contrast to the other treatments. The animals fed diets with hay Tifton 68 had a higher rate of return (2.16%) and profitability (34.63%) compared with the other diets. In conclusion, the use of Tifton 68 hay in diets of lambs in finishing stage, feedlot, brings higher economic returns compared with other diets containing *Cynodon* hay grasses.

**Key Words:** economic, hay, lamb

**182 Effect of concentrate supplementation during early lactation on intake, rumen function and milk production of twin suckling ewes.** Francis P. Campion\*<sup>1</sup>, Fiona M. McGovern<sup>1</sup>, Eva Levicnik<sup>3</sup>, Aileen McCarron<sup>1</sup>, Philip Creighton<sup>2</sup>, and Tommy M. Boland<sup>1</sup>, <sup>1</sup>University College Dublin, Dublin, Ireland, <sup>2</sup>Teagasc Athenry, Co. Galway, Ireland, <sup>3</sup>University of Ljubljana, Ljubljana, Lithuania.

Ewe milk production peaks approximately 3 weeks postpartum, with intake potential peaking approximately 6 weeks postpartum. The aim of this study was to investigate the effect of concentrate supplementation on ewe DMI during early lactation. Fifty-four twin suckling ewes were allocated to one of 3 dietary treatments (n = 18) in a randomized block design on d 7 (+/- 2 d) of lactation as follows: GO: Ad-lib zero grazed grass, GC: Ad-lib zero grazed grass and 500g fresh weight (FW) of concentrate feed for 49 d postpartum and GC21: Ad-lib zero grazed grass and 500g FW of concentrate feed for 21 d postpartum. Ewes were penned with their progeny from 72 h postpartum until d 49 lactation. Lambs were offered free access to a creep area bedded with straw and were offered fresh water and zero grazed grass. Grass was harvested each morning using a single chop zero grazer from perennial ryegrass based swards with an average pre-grazing herbage mass of 1100kg DM/

ha with an average DM, protein and neutral detergent fiber content of 18.0, 18.4 and 45.0% respectively. Individual ewe intakes were recorded daily from d 7 to 49 of lactation. Rumen fluid samples were taken before and 7 d post-dietary introduction to assess rumen fermentation. Milk yield and composition was measured at 7 d intervals starting on d 14 ( $\pm$  2 d) of lactation. Statistical analysis was carried out using generalized least squares means in SAS v9.4. Ewes from GO treatment had higher grass DMI (1.96kg) compared with GC (1.65kg) ewes ( $P < 0.05$ ) and tended to have higher grass DMI than GC21 (1.84kg) ewes ( $P < 0.10$ ). However, total DMI did not differ between treatments ( $P > 0.05$ ). Rumen pH for GC and GC21 ewes decreased between d 0, 6.65, and d 7, 6.09 ( $P < 0.01$ ). Milk production tended to be lower during early lactation for GC21 (1.85kg) ewes compared with GC (2.11kg) ewes ( $P < 0.10$ ). No difference was observed between treatments in milk fat, protein or net energy ( $P > 0.05$ ). Concentrate supplementation did not lead to increased DMI but rather acted as a substitute for grazed grass.

**Key Words:** DMI, milk yield, rumen function

### 183 An investigation into the effect of crude protein intake, dry matter intake and body reserve mobilization on the ewe colostrum and IgG production in the first 18 hours postpartum.

Francis P. Campion<sup>\*1</sup>, Fiona M. McGovern<sup>1</sup>, Philip Creighton<sup>2</sup>, Alan G. Fahey<sup>1</sup>, and Tommy M. Boland<sup>1</sup>, <sup>1</sup>University College Dublin, Dublin, Ireland, <sup>2</sup>Teagasc Athenry, Co. Galway, Ireland.

Colostrum is essential to neonatal survival and future performance, providing energy and immunity in early life. Late gestation nutrition has been shown to influence colostrum production and immunoglobulin G yield (IgGY). This study aims to determine the key drivers of colostrum production, specifically the relationship with dry matter intake (DMI), crude protein intake (CPI), and body reserve mobilization during the final 6 weeks of gestation. Data from 7 late pregnancy nutrition studies carried out over 12 years using twin-bearing ewes were merged into a single data set. All studies recorded ewe colostrum production and IgGY at one, 10 and 18 h postpartum. Regression coefficients were estimated using PROC MIXED (SAS v9.4). The model included the fixed effects of ewe, year, ewe breed (Belclare, Leicester and Suffolk crosses) and time point as well as the continuous variables of gestation length, maternal age, average lamb birth weight (BW), live weight change (LWC) and body condition score change (BCSC) during late gestation. Colostrum volume changed by 0.02 l for every 10 g change in CPI ( $P < 0.01$ ), and 0.02 l for every 100 g change in DMI ( $P < 0.01$ ). Body reserve mobilization (LWC and BCSC) did not influence colostrum volume ( $P > 0.10$ ). Breed ( $P < 0.01$ ), time point ( $P < 0.01$ ), breed by time point interaction ( $P < 0.01$ ) and maternal age ( $P < 0.05$ ) had significant effects on the effect of CPI and DMI on the volume of colostrum produced by the ewe. The IgGY was also driven by CPI and DMI ( $P < 0.05$ ). Each 10g change in CPI gave a 0.50 g change in IgGY ( $P < 0.05$ ) and there was 0.80 g

change in IgGY for each 100 g change in DMI ( $P < 0.01$ ). Breed did not influence IgGY ( $P > 0.10$ ) despite a breed by time point interaction ( $P < 0.01$ ). Gestation length did not affect colostrum volume or IgGY ( $P > 0.10$ ). These results indicate that intake as opposed to body reserve mobilization is the major driver of colostrum production by the ewe.

**Key Words:** colostrum, immunoglobulin, gestation

### 184 Biochemical parameters of newborn goats raised from Saanen does supplemented with selenium and vitamin E during the transition period.

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The objective of this study was to evaluate biochemical parameters of newborn goats from does that were supplemented with Selenium and Vitamin E. Twenty 4 kids from Saanen does that were supplemented starting on the fourth month of pregnancy until birth. The animals received the treatments based on the dams treatments and were divided into 3 groups: Control milk (base diet with 50% forage and 50% concentrate); Se milk (2.5 mg of Se/kg Dm added to the diet) and Sev milk (2.5 mg of Se/kg DM and 1000 IU/day of vitamin E/kg DM). The kids started receiving ad libitum access to feed from 7 d postpartum. The experiment took 28 d. After birth blood samples were collected before ingesting of colostrum and 48 h, 7, 14, 21 and 28 d postpartum. Blood samples were analyzed for serum protein analysis, albumin, gamma-glutamyl transferase (GGT), total cholesterol, high density lipoproteins (HDL), low density lipoproteins (LDL), triglycerides, aspartate aminotransferase (AST), creatine phosphokinase (CK), glucose, lactate, urea, creatinine,  $\beta$ -hydroxybutyrate (BHB), nonesterified fatty acids (NEFA) and total antioxidant status (TAS). The experimental design was a complete randomized design with repeated measures, with 8 replications. Data were analyzed by ANOVA and the means were compared by Tukey test ( $P \leq 0.05$ ). There was no significant effect of treatment ( $P > 0.05$ ) for the parameters of GGT, LDL, triglycerides, CK, glucose, urea, creatinine, BHB, NEFA and TAS. For serum protein and albumin parameters the Sev treatment provided significant increase ( $P < 0.05$ ) when compared with the other treatments. There was a significant effects for Sev treatment ( $P < 0.05$ ) for lactate, total cholesterol, HDL and AST, showing, therefore, a better oxidant activity in the group treated with selenium and vitamin E.

**Key Words:** antioxidant, birth, nutrition

## Swine Species

**185 The effects of feeding frequency on pig performance, behavior, and tissue accretion rates.** Jessica D. Colpoys\*, Anna K. Johnson, and Nicholas K. Gabler, *Iowa State University, Ames, IA.*

Recent research suggests that feeding frequency may affect feed efficiency in pigs. Therefore, our objective was to compare 2 divergent feeding patterns and their effects on pig performance, tissue accretion, body composition and behavior. Following a 3 d acclimation to individual pens, 48 gilts were randomly blocked by body weight ( $55.9 \pm 5.2$  kg on test BW) into 2 feeding treatments; 1) ad libitum access or 2) twice daily gilts that were allowed to eat ad libitum between 0800 and 0900 h and again from 1700 to 1800 h. Performance was recorded weekly for 55 d and ADG, ADFI, and G:F was recorded and calculated. Body composition was assessed in 12 gilts per treatment using dual x-ray absorptiometry (DXA) at day -3 and 55, and tissue accretion rates were calculated. Video was recorded on d 51 and continuously watched from 7:00–19:00 h to assess the time pigs spent standing, sitting, lying and eating. Performance and DXA data were analyzed using the Mixed procedure and behavior data were analyzed using the Glimmix procedure of SAS. The model included the fixed effect of treatment, covariate of on test BW, and pig as the experimental unit. Gilts fed twice daily had lower ADG and ADFI compared with ad libitum gilts ( $P \leq 0.01$ ); however, no treatment difference in G:F was observed ( $P = 0.41$ ). No treatment differences were observed in body composition at d 55. However, gilts fed twice daily had lower fat (twice = 297 vs. ad libitum = 339 g/d), lean (twice = 673 vs. ad libitum = 737 g/day), and protein (twice = 136 vs. ad libitum = 149 g/d) daily accretion rates compared with ad libitum gilts ( $P = 0.01$ ). No difference in bone accretion was observed. Gilts fed twice daily spent less time lying and eating compared with ad libitum gilts ( $P \leq 0.05$ ). However, treatments did not differ in duration of time spent sitting or standing ( $P \geq 0.19$ ). In conclusion, twice daily feeding resulted in reduced growth, tissue accretion, and lying behavior compared with ad libitum feeding. However, feeding frequency did not alter feed efficiency. This pig performance can be partly explained by the differences in caloric and nutrient intake.

**Key Words:** feed efficiency, feeding frequency, pig

**186 Relationships between nursery stocking density and feeder space allowance with subsequent gilt estrous characteristics.** E. B. Cook\*, W. L. Flowers, and M. T. Knauer, *North Carolina State University, Raleigh, NC.*

The objective of the study was to determine the effect of nursery stocking density (STOCK) and feeder space (FS) on gilt estrous traits. Composite Landrace  $\times$  Large White PIC gilts ( $n = 230$ ) were weaned at 37 d of age and 9.6 kg. Gilts were randomly assigned to a nursery STOCK of 0.42 or 0.21 m<sup>2</sup> and a FS of 14.35 or 7.175 cm per pig in a 2  $\times$  2 factorial arrangement. After 33 d in the nursery, gilts were moved to the finisher weighing 23.7 kg. In the finisher, gilts were remixed and housed 15 per pen (0.84 m<sup>2</sup> per pig). Starting at 181 d of age, gilts were penned daily with a mature boar for 7 min and estrous symptoms recorded. At 188 d of age, gilts were allotted 7 min of fence-line boar contact daily until breeding. Measurements of gilt reproductive performance included: whether or not a gilt reached puberty (Pubertal), age at puberty, puberty weight, vulva width at puberty (Vulva\_Width1), length of estrus at puberty (LOE1), vulva width at second estrous (Vulva\_Width2), and length of estrus at second estrous (LOE2). Data were analyzed in SAS using PROC MIXED for continuous traits and PROC GLIMMIX for

the categorical trait Pubertal. Fixed effects included STOCK, FS, the interaction between STOCK and FS, nursery room and age at first boar exposure. Stocking density and FS LSMEAN estimates for gilt estrous traits are shown in Table 1. Length of estrus at puberty tended ( $P = 0.08$ ) to be greater for pigs stocked at 0.21 m<sup>2</sup> when compared with 0.42 m<sup>2</sup> (1.74 vs. 1.57 d). Yet no other estrous characteristics were affected by STOCK or FS ( $P \leq 0.22$ ). Results suggest nursery STOCK and FS have little effect on subsequent gilt estrous characteristics.

**Table 1 (Abstr. 186).** Effect of nursery stocking density and feeder space on subsequent gilt estrous characteristics

Trait	Stocking density, m <sup>2</sup>			Feeder space, cm/pig		
	0.42	0.21	SE	14.35	7.175	SE
Pubertal, %	79.4	74.5	4.0	78.7	75.3	4.0
Age at puberty	203.5	202.5	2.7	203.9	202.1	2.7
Puberty weight, kg	138.1	135.7	5.4	136.7	137.1	5.4
Vulva_Width1, cm	38.6	37.7	0.72	38.3	38.1	0.72
LOE1, d	1.57	1.74	0.10	1.65	1.65	0.10
Vulva_Width2, cm	38.2	38.4	1.11	37.9	38.6	1.10
LOE2, d	1.89	1.90	0.15	1.94	1.85	0.15

**Key Words:** estrus, gilt, nursery

**187 Relationships between nursery stocking density and feeder space allowance with gilt growth performance.** E. B. Cook\*, W. L. Flowers, and M. T. Knauer, *North Carolina State University, Raleigh, NC.*

The objective of the study was to determine the effect of nursery stocking density (STOCK) and feeder space (FS) on gilt growth performance. Composite Landrace  $\times$  Large White PIC gilts ( $n = 500$ ) were weaned at an average age and weight of 39.3 d and 9.6 kg, respectively. Gilts were randomly assigned to either 5 or 10 pigs per pen to achieve a STOCK of 0.42 or 0.21 m<sup>2</sup> and FS of 14.35 or 7.175 cm per pig in a 2  $\times$  2 factorial arrangement. In 2013 and 2014 pigs remained in the nursery for 33 and 45 d, respectively, and left at an average weight of 23.7 and 39.0 kg, respectively. In the finisher, pigs were randomly placed into groups of 15 gilts per pen (0.84 m<sup>2</sup>). Data were analyzed in SAS using PROC GLM. Fixed effects included STOCK, FS, year, nursery room and the interaction between year and room. All interactions between STOCK and FS were  $P \geq 0.24$ . In 2013 nursery ADG did not differ ( $P = 0.27$ ) between STOCK. However, in 2014 nursery ADG was greater ( $P < 0.01$ ) for pigs stocked at 0.42 m<sup>2</sup> when compared with 0.21 m<sup>2</sup>. Overall nursery ADG was greater ( $P < 0.01$ ) for pigs stocked at 0.42 m<sup>2</sup> when compared with 0.21 m<sup>2</sup> and numerically greater ( $P = 0.21$ ) for pigs housed with 14.35 cm of FS when compared with 7.175 cm. Overall finishing ADG tended ( $P = 0.08$ ) to be greater for pigs previously housed with 7.175 cm of FS when compared with 14.35 cm. In 2013 wean-to-finish ADG did not differ ( $P = 0.84$ ) between STOCK. However, in 2014 wean-to-finish ADG tended ( $P < 0.08$ ) to be greater for pigs stocked at 0.42 m<sup>2</sup> when compared with 0.21 m<sup>2</sup>. Overall wean-to-finish ADG was greater ( $P = 0.03$ ) for pigs stocked at 0.42 m<sup>2</sup> when compared with 0.21 m<sup>2</sup>. Results suggest nursery FS, but not STOCK, affected subsequent finishing growth rate.

*Contd.*



**Table 1 (Abstr. 187).** Effect of nursery stocking density and feeder space on gilt growth performance

ADG	Stocking density, m <sup>2</sup>			Feeder space, cm/pig		
	0.42	0.21	SE	14.35	7.175	SE
2013 nursery	0.431	0.409	0.019	0.428	0.411	0.020
2014 nursery	0.693	0.619	0.016	0.665	0.647	0.017
Overall nursery	0.563	0.513	0.013	0.547	0.530	0.013
2013 finisher	0.906	0.893	0.015	0.891	0.908	0.015
2014 finisher	0.869	0.877	0.012	0.865	0.881	0.013
Overall finisher	0.887	0.885	0.01	0.878	0.894	0.01
2013 wean-to-finish	0.804	0.787	0.013	0.791	0.799	0.013
2014 wean-to-finish	0.816	0.797	0.011	0.803	0.810	0.011
Overall wean-to-finish	0.810	0.792	0.008	0.798	0.805	0.008

**Key Words:** gilt, growth, nursery

**188 Apparent total-tract digestibility (ATTD) of total dietary fiber (TDF) between and within high fiber feed ingredients in finishing pigs.** Zhimin Huang\*, Pedro Urriola, and Gerald Shurson, *University of Minnesota, St. Paul, MN.*

Fibrous ingredients, such as distillers dried grains with solubles (DDGS), are a price competitive source of energy in swine diets, but TDF may decrease nutrient digestion and absorption as well as energy utilization. However, we know little about the effects of ATTD of TDF on nutrient digestibility and energy utilization. Therefore, the objective of this study was to evaluate the effect of different fiber sources on ATTD of TDF and GE of finishing pigs. A total of 34 barrows and 2 gilts (Large White × Danish Landrace) with an average BW of 84.0 ± 7.2 kg were used in a changeover design with 2 consecutive 13-d periods, and involving 10-d of adaptation and 3-d of fecal and urine collection. During each period, the 36 pigs were allotted to 4 blocks by BW, 9 pigs in each block were fed 9 different experimental diets. Diets were formulated to contain similar concentration of TDF, from 3 sources of 3 ingredients (wheat straw, WS; soybean hulls, SBH, and DDGS). TiO<sub>2</sub> (0.5%) was used in diets as an indigestible marker. The direct procedure was used to measure ATTD of DM, GE, CP, acid ether extract (AEE), and TDF in the diets. Results showed that ATTD of DM, GE, and TDF of pigs fed the diet with SBH (87.4, 86.8%, and 78.9%) were greater ( $P < 0.01$ ) than those fed DDGS (82.6, 80.1%, and 43.0%), which were greater ( $P < 0.01$ ) than those fed WS (75.1, 75.8%, and 26.7%). The ATTD of DM and GE were ( $R^2 = 0.84$  and  $R^2 = 0.85$ ) correlated with ATTD of TDF among diets. Between sources of the same ingredient, we observed differences ( $P < 0.01$ ) in ATTD of TDF among 3 DDGS sources (36.0, 43.1, and 49.8%), but not for WS, or SBH. Among sources of DDGS, the correlation between ATTD of DM and GE to ATTD of TDF was poor ( $R^2 = 0.62$  and  $R^2 = 0.51$ ). In conclusion, ATTD of TDF is a good predictor of ATTD of DM and GE among fibrous ingredients, but it is less predictive among sources of the same ingredient because ATTD of TDF does not affect ATTD of GE, CP, or AEE.

**Key Words:** apparent total-tract digestibility, pig, total dietary fiber

**189 Net portal-drained viscera flux of volatile fatty acids in Iberian and Landrace pigs.** Gonzalez-Valero Lucrecia<sup>1</sup>, Rojas-Cano Maria Luz<sup>1</sup>, Lachica Manuel<sup>1</sup>, Ranilla Maria Jose<sup>1</sup>, and Fernandez-Figares Ignacio\*<sup>2</sup>, <sup>1</sup>Estacion Experimental del Zaidin-CSJC, Granada, Spain, <sup>2</sup>Universidad de Leon. Dept. Produccion Animal, Leon, Spain.

Iberian pigs are traditionally reared outdoors where they feed on acorns and herbage. The capacity of volatile fatty acids (VFA) absorption could be of importance for these animals although it has never been determined. The aim of the present work was to evaluate the capacity of VFA absorption in Iberian compared with Landrace gilts fed a standard corn-soybean meal diet (160g/kg; 14.25 MJ ME/kg DM), using arterio-venous preparations. Iberian and Landrace gilts (30 kg BW; n = 4 Landrace and 6 Iberian) were used. Diets were formulated with protein of high biological value and offered at 85% ad libitum. Gilts were fasted for 24 h prior surgery. Catheters were implanted in the portal and mesenteric veins and carotid artery. Then, pigs were placed in metabolism cages and fed the diet for 6 d. On d 7, *p*-aminohippuric acid (PAH) was continuously infused through the mesenteric vein and pigs were offered 25% of their daily intake. Blood was withdrawn simultaneously from portal vein and carotid artery at 0, 0.5, 1, 1.5, 2, 2.5, 3, 3.5, 4, 5 and 6 h after feeding, centrifuged for plasma harvest and stored at -20°C until analysis for PAH and VFA. Compared with Landrace, Iberian gilts had greater net portal absorption (μmol/min) of acetic acid (252 vs. 178;  $P = 0.000$ ), butyric acid (17.0 vs. 12.7;  $P = 0.007$ ) and isovaleric acid (10 vs. 4.7;  $P = 0.010$ ), and lower of valeric acid (-8.5 vs. 12.9;  $P = 0.007$ ), than Landrace gilts. No differences ( $P > 0.10$ ) in net portal absorption of propionic (146 vs. 153), isobutyric (13 vs. 9) and caproic (4.5 vs. 5.5) acids were found. The greater capacity of net portal absorption of VFA in Iberian pigs could constitute a meaningful contribution to the energy requirement for maintenance and constitute an advantage when fed outdoors fibrous rich feeds.

**Key Words:** net portal absorption, volatile fatty acids, Iberian pig

**190 Effects of a phytogetic feed additive in a protein-reduced diet in growing/finishing pigs.** Carina Schieder\*<sup>1</sup>, Christiane Schwarz<sup>2</sup>, Christine Hunger<sup>1</sup>, Barbara Rueel<sup>1</sup>, Jose Soto<sup>3</sup>, and Karl Schedle<sup>2</sup>, <sup>1</sup>Biomim Holding GmbH, Herzogenburg, Austria, <sup>2</sup>University of Natural Resources and Life Sciences, Vienna, Institute of Animal Nutrition, Products, and Nutritional Physiology, Vienna, Austria, <sup>3</sup>Biomim USA Inc., San Antonio, TX.

The experiment was conducted to evaluate the efficacy of a phytogetic feed additive (PFA) on body weight gain, carcass composition and quality of 72 fattening pigs [(Large White × Landrace) × Piétrain]. Animals were distributed into 3 treatment groups with 3 replicates of 8 pigs per replicate, with an initial weight of 32.2 ± 0.4 kg. Control group (CON) and one phytogetic group (PFA+normal; Digestarom® Finish 150 ppm, BIOMIN PhytoGenics GmbH, Germany) received a diet containing 17% and 15% CP in the growing and finishing period, respectively. Another phytogetic group (PFA+low; Digestarom Finish 150 ppm) received a ration with a reduced protein content (-0.4% CP) throughout the whole growing and fattening period. Feed and water was provided ad libitum. At 73.2 ± 0.6 kg of BW, feed changed from growing to finishing diet. Body weight was measured weekly and individual feed intake was recorded daily. Pigs were slaughtered at 117.5 ± 0.2 kg and carcass composition and quality were evaluated. Data were analyzed with procedure GLM (SAS 9.4) and a multiple comparison was conducted with Tukey's range test. Results for the whole fattening period show significantly improved gain:feed ratio ( $P < 0.05$ ; 0.389 and 0.388 in PFA+normal and PFA+low, respectively, compared with 0.368 in CON). Average daily feed intake was 2298 g/d in the CON compared with 2288 g/d in the PFA+normal and 2275 g/d in the PFA+low group. Daily weight gain tended to be higher ( $0.05 < P < 0.10$ ) in the PFA+normal and PFA+low group (889.0 and 881.7 g/d) compared with CON (844.5 g/d). Carcass composition and quality parameters did not differ significantly ( $P > 0.05$ ) between treatments besides conductivity of meat which has been

improved for PFA+low group compared with CON 1 h post mortem (4.35 vs. 5.02 mS/cm, respectively). Overall, the supplementation of a PFA improved the performance of pigs and a protein reduction was efficiently compensated through the supplementation of PFA.

**Key Words:** fattening pig, nutrient-sparing, phytogetic feed additive

**191 Effect of milky derived flavor inclusion in creep-feed diets on suckling piglet performance and litter homogeneity.** Laia Blavi\*<sup>1</sup>, David Solà-Oriol<sup>1</sup>, Francisco Javier Crespo<sup>2</sup>, María del Mar Serra<sup>2</sup>, and José Francisco Pérez<sup>1</sup>, <sup>1</sup>*Animal Nutrition and Welfare Service, Department of Animal and Food Sciences, Universitat Autònoma de Barcelona, Bellaterra, Spain*, <sup>2</sup>*Interquim S.A (Ferrer Health Tech), Barcelona, Spain*.

Hyper-prolific sows provide a higher number of piglets born per litter and year but, this promotes greater within-litter birth weight variation with high percentage of small animals. It was hypothesized that the inclusion of a milky derived flavor into the creep-feed (CF) would improve the number of eaters, creep-feed intake, piglet performance and litter homogeneity. A total of 35 litters (418 piglets) of Landrace × Large White sows were used. At cross-fostering (max 48 h after birth) all litters were standardized and were distributed into 3 experimental treatments; a basal CF (2480 kcal/kg, 1.53% digestible Lys) diet without flavor (T1), and 2 different milky derived flavors (T2 and T3) according to number of piglets, litter weight and coefficient of variation (CV). Creep-feed was offered from d 2 to d 28 and indigo carmine (5g/kg) was used as indigestible marker to register the number of creep-feed eaters on d 7, 14, 21 and 28. Creep-feed disappearance and individual BW were registered on d 2, 7, 14, 21 and 28 to calculate the within litter CV and the average daily weight gain (ADG) and mortality rate. No differences ( $P > 0.10$ ) in BW (6.9 kg), CF disappearance (1571 g) and the number of eaters (4.1 piglets per litter) were observed among treatments. However, lower ( $P < 0.05$ ) CV was observed for piglets fed the T2 (0.16 and 0.17%) than those fed the T3 (0.21 and 0.20%) and T1 (0.25 and 0.26%) diets on d 21 and 28, respectively. In addition, animals categorized as small (<180 g/d at 28 d) in T2 presented higher ADG ( $P < 0.001$ ) than T1 (153.5 g/d vs 119.1 g/d, respectively from d 0 to 28). Higher ( $P < 0.01$ ) piglet mortality was observed in litters fed T1 than T2 and T3 (6.9, 3.0 and 4.1%, respectively). These results suggest that the use of a milky derived flavor in the creep-feed improves growth in small piglets and may reduce the CV of the BW within the litters and piglet mortality obtaining higher homogeneity BW at weaning.

**Key Words:** flavor, creep-feed, piglet

**192 Effects of cathelicidin-BF on growth performance, intestinal morphology and inflammation in weanling piglets.** Hongbo Yi\*, Caihua Yu, Haiwen Zhang, Huahua Du, and Yizhen Wang, *Institute of Feed Science, College of Animal Sciences, Zhejiang University, Hangzhou, Zhejiang, China*.

The objective of this study was to investigate the effects of cathelicidin-BF (C-BF) derived from the Chinese endemic genera *Bungarus fasciata* on postweaning diarrhea, growth performance, intestinal morphology and inflammation in weanling piglets. Eighteen piglets (Duroc × Landrace × Yorkshire) were weaned at 21 d with an initial BW of  $6.63 \pm 0.13$  kg and randomly assigned to one of 2 treatments to give 3 pens per treatment and 3 piglets per pen. Piglets were intraperitoneally injected with normal saline (control group) or 0.55 mg/kg BW C-BF (C-BF group) once a day and offered diets and water on an ad libitum basis for 7 d. Diarrheal index was scored twice daily according a 4-point system (0,

normal; 1, soft feces; 2, mild diarrhea; and 3, severe diarrhea). The ADG, ADFI and G:F were calculated at the end of study. All piglets were euthanized to determine serum immunoglobulin, serum cytokines, intestinal morphology and intestinal inflammation. Our data showed that C-BF treatment increased ADFI (245.4 vs. 157.9 g/d,  $P < 0.05$ ) and ADG (181.9 vs. 112.9 g/d,  $P < 0.05$ ) compared with control group. Meanwhile, C-BF treatment reduced diarrheal index compared with control (0.47 vs. 1.67,  $P < 0.05$ ). Serum IgG, IL-6, IL-22 and TNF- $\alpha$  production were decreased ( $P < 0.05$ ) by C-BF treatment. C-BF increased ( $P < 0.05$ ) villus height of jejunum and reduced ( $P < 0.05$ ) crypt depth of ileum. We demonstrated that C-BF reduced ( $P < 0.05$ ) the expression of the inflammatory cytokines TNF- $\alpha$ , IL-6 and IL-8, but increased the expression of the anti-inflammatory cytokine IL-10 ( $P < 0.05$ ) in the jejunum. Furthermore, we demonstrated that C-BF suppressed inflammation by downregulating the nuclear factor- $\kappa$ B (NF- $\kappa$ B) signaling pathway in the jejunum. In summary, these data suggest that intraperitoneal injections of C-BF ameliorated postweaning diarrhea and improved growth performance along with suppressing intestinal inflammation by downregulating the NF- $\kappa$ B signaling pathway in weanling piglets.

**Key Words:** cathelicidin-BF, weanling piglet, intestinal inflammation

**193 Improving growth performance of weaning pigs with different dosages of combined natural growth promoter products.** Jose A Soto\*<sup>1</sup> and Attila Kovacs<sup>2</sup>, <sup>1</sup>*Biomin America Inc., San Antonio, TX*, <sup>2</sup>*Biomin Holding GmbH, Herzogenburg, Austria*.

A combination of organic acids OA (formic, propionic and acetic), phytochemical (PC) and permeabilizing substance (PS) which alters the permeability of the outer membrane in gram-negative bacteria, has shown synergistic effect on the inhibition of bacterial growth in vitro as well as improved growth performance in weaning pigs in vivo. The aim of the trial was to determine the efficacy of the combination of OA, PC and PS in the form of combined natural growth promoter (NGP) Biotronic Top3, at 3 different levels (0.1, 0.15, and 0.2%) on the growth performance of wean pigs. 600 pigs weaned at 28 d were assigned to 4 treatments: (1) basal diet, (2) basal diet combined with 0.1 NGP, (3) basal diet combined with 0.15 NGP and (4) basal diet combined with 0.2% NGP with 6 replicates by treatment. Starter diet from d 28 to d 56 (ME 14.00 MJ/kg, CP 17%, Met 0.45% and Lys 1.40%) and grower diet from d 56 to d 71 (ME 13.4 MJ/kg, CP 17.50%, Met 0.36% and Lys 1.20%) were fed. BW of the animals was recorded at d 28, 56 and 71. ADG and FCR were calculated. Dietary supplementation of NGP at inclusion levels of 0.1, 0.15 and 0.2% enhanced growth performance. The BW (kg) was improved on d 56 (quadratic  $P < 0.01$ ; 17.2<sup>a</sup>, 18.4<sup>b</sup>, 18.7<sup>b</sup> and 18.9<sup>b</sup> for 0, 0.1, 0.15 and 0.2% of NPG, respectively) and d 71 (quadratic  $P < 0.01$ ; 26.9<sup>a</sup>, 28.1<sup>b</sup>, 28.3<sup>b</sup>, and 28.3<sup>b</sup> for 0, 0.1, 0.15 and 0.2% of NPG, respectively) as well as feed intake (g/d) (quadratic  $P < 0.01$ ; 762<sup>a</sup>, 804<sup>b</sup>, 816<sup>c</sup>, and 818<sup>c</sup> for 0, 0.1, 0.15 and 0.2% of NPG, respectively). ADG (g) ( $P < 0.1$ ) was only numerically different among treatments (439, 458, 465, and 465 for 0, 0.1, 0.15, and 0.2% of NPG, respectively). There were no differences in FCR ( $P > 0.1$ ). Diet supplemented 0.15% combined NGP had the highest final BW and ADG tended to be greater compared with other groups. The supplementation with the combined NGP at a level of 0.15% was most beneficial.

**Key Words:** natural growth promoter, organic acids, phytochemical

**194 The effects of exogenous protease enzyme on growth performance, nutrient digestibility, blood profiles, fecal microflora, fecal gas emission, and fecal score in weanling pigs.** Glenmer B. Tactacan\*<sup>1</sup>, Se-young Oh<sup>1</sup>, Jin H. Cho<sup>2</sup>, and In H. Kim<sup>3</sup>, <sup>1</sup>*Innovation and Development Department, Jefe Nutrition, Saint-Hyacinthe, Quebec, Canada,* <sup>2</sup>*Department of Animal Science, Chungbuk National University, Cheongju, Chungbuk, Korea,* <sup>3</sup>*Department of Animal Resources and Science, Dankook University, Cheonan, Korea.*

Although exogenous protease enzymes have been used in poultry quite extensively, this has not been the case in pigs. In general, due to their better fermentative capacity and longer digesta transit time in the gut, pigs have greater capacity to digest nutrients than poultry. In young pigs however, the digestion of nutrients, particularly of dietary proteins is marginally incomplete. Therefore, a study was conducted to elucidate the effects of a commercial protease enzyme supplemented in weanling pig diets. The indices of growth, nutrient digestibility, blood profiles, fecal microflora, fecal gas emission and fecal scores were measured during the study. A total of 50 weanling pigs ( $6.42 \pm 0.60$  kg) at 28 d of age were randomly assigned to receive 1 of 2 dietary treatments: (1) basal diet (corn-soy based) with no supplemental protease, and (2) basal diet + 200 g/ton protease for 42 d. A completely randomized block design consisting of 5 replicate pens per treatment with 5 pigs per pen was used. Growth performance in terms of bodyweight and average daily gain in pigs fed with protease enzyme ( $27.04$  vs.  $25.75 \pm 0.25$  kg and  $491$  vs.  $460 \pm 6$  g;  $P < 0.05$ ) was increased significantly, but gain per feed was similar between treatments. Compared with the control, protease supplementation increased ( $P < 0.05$ ) the apparent total-tract nutrient digestibility ( $84.66$  vs.  $81.21 \pm 0.91$  DM and  $84.02$  vs.  $80.47 \pm 0.95$  N) and decreased ( $P < 0.05$ ) the  $\text{NH}_3$  emission ( $2.0$  vs.  $1.2 \pm 0.14$  ppm) in the feces. Except for a decreased ( $P < 0.05$ ) in blood creatinine level, no differences were observed in RBC, WBC, lymphocytes, urea nitrogen, and IgG concentrations between treatments. Fecal score and fecal microflora (*Lactobacillus* and *E. coli*) were also similar between the control and the protease-supplemented diets. Overall, the supplementation of protease enzyme in weanling pigs resulted to improved growth rate and nutrient digestibility. Exogenous protease enzyme reduced fecal  $\text{NH}_3$  emission, thus, potentially serving as a tool in lowering noxious gas contribution of livestock production in the environment.

**Key Words:** protease, nutrient digestibility, weanling pig

**195 Maternal betaine supplementation during gestation attenuates hepatic cell cycle and proliferation through epigenetic regulation of the STAT3-dependent pathway in newborn piglets.** Demin Cai\*, Mengjie Yuan, Yimin Jia, Yun Hu, and Ruqian Zhao, *Nanjing Agricultural University, Nanjing City, China.*

Betaine, which donates methyl groups through methionine metabolism for DNA and protein methylation, is critical for epigenetic gene regulation especially during fetal development. Here we fed gestational sows (8 per group) with control or betaine supplemented diets (3 g/kg, from the first day of gestation to parturition) to explore the effects of maternal betaine on hepatic cell proliferation in neonatal piglets. Newborn piglets were individually weighed immediately after birth and the piglets from the same litter were kept together in the warm creep area. One male piglets of the mean body weight were selected per litter and killed for tissue sampling before suckling. Neonatal piglets born to betaine-supplemented sows demonstrated a reduction of cell number and DNA content ( $P < 0.05$ ) in the liver, which was associated with significantly downregulated hepatic expression of cell cycle regulatory genes ( $P < 0.05$ ), cyclin 2 (CCND2) and presenilin1 (PSEN1). Moreover, STAT3 binding to the promoter of *CCDN2* and *PSEN1* genes was lower ( $P < 0.05$ ) in betaine-exposed piglets, which was accompanied by strong reduction ( $P < 0.05$ ) of STAT3 mRNA and protein expression along with its phosphorylation at Tyr<sup>705</sup> and Ser<sup>727</sup> residues ( $P < 0.05$ ). Also, prenatal betaine exposure significantly attenuated upstream kinases ( $P < 0.05$ ) of STAT3 signaling pathway (phospho-ERK1/2, phospho-SRC and phospho-JAK2) in the liver of neonates. Furthermore, the repressed *STAT3* expression in the liver of betaine-exposed piglets was associated with DNA hypermethylation ( $P < 0.05$ ) and more enriched repression histone mark H3K27me3 ( $P < 0.05$ ) on its promoter, together with significantly upregulated expression ( $P < 0.05$ ) of H3K27me3 and enhancer of zeste homolog 2 (EZH2) protein, as well as miR-124a ( $P < 0.05$ ) which targets STAT3. Taken together, our results suggest that maternal dietary betaine supplementation during gestation inhibits cell proliferation in the liver of neonatal piglets through epigenetic regulation of hepatic *CCDN2* and *PSEN1* genes via a STAT3-dependent pathway.

**Key Words:** cell cycle and proliferation, STAT3, epigenetic regulation



## ADSA-SAD (Student Affiliate Division) Undergraduate Competition: Dairy Foods

### 196 Organic versus conventional milk production systems.

Savannah B. Meade\* and Jeffrey M. Bewley, *University of Kentucky, Lexington, KY.*

In the past decade, organic farming has increased in popularity. Many consumers who support organic farming believe that organic products differ in composition from conventionally produced products. Additionally, organic milk supporters often believe that organic dairy farming is more beneficial to cows and the environment. The milk is produced without antibiotics, added hormones, synthetic chemical treatment or genetic modification. This leads many consumers to believe that organic milk has potential human health benefits. Milk composition is affected by many factors. Studies comparing organic vs. conventional milk are sparse. A recent *Journal of Dairy Science* review covers the individual factors that affect milk composition and provides an overview of a few studies that compare organic and conventional milk. Factors influencing milk composition, such as nutrition, breed, and stage of lactation, have been studied individually, but are difficult to compare simultaneously. Some studies have shown that organic milk has higher levels of omega-3 fatty acids and conjugated linoleic acid, but it is unclear as to whether these differences are truly beneficial. Milk fatty acids are often researched when considering organic vs. conventional milking systems because fatty acids respond rapidly and are very sensitive to changes in the diet of cattle. Studies also discuss the use of antibiotics, the presence of hormones, and genetic modification factors that many organic consumers base their purchases on. More research needs to be conducted to determine whether one production system is more beneficial to human health than the other. Organic consumers pay a higher market price for their products, and as of now, no proven benefits for purchasing organic milk have been demonstrated. No major health differences between organic and conventional dairy products have been proven.

**Key Words:** organic milk production, conventional milk production, milk composition

### 197 Postmortem factors relevant to veal quality. Sloane Garcia\* and Chad Carr, *University of Florida, Gainesville, FL.*

Veal represents a relatively minor percentage of the meat industry as a whole. During the past half century the veal industry has undergone considerable changes in production practices. Currently, grain-fed and heavier calves are the norm and many farms are converting to individual pens as opposed to group housing which, in addition to improving the conditions for the animals, has also improved the public's perception of the industry. (1) It is generally accepted that meat quality is the most important indicator of financial returns on the product. In today's world, the industry places strong emphasis on lean color in spite of the absence of scientific evidence validating a direct effect on quality. (2) This presentation will discuss the effects of post-mortem factors on veal quality. Factors associated with meat quality include carcass grade, aging, and packaging. According to the USDA, milk fed calves produced carcasses that were lighter in lean color and fatter, however these graded scores do not directly correlate with palatability. (1) Color determination plays a large role in quality determination, but data continues to show a poor relationship between color and taste. Attempts at quality improvements should evaluate factors such as aging and packaging for determining veal quality. Aging veal results in increases in tenderness, decreases in shear force, and increases in lightness and redness. Type of

packaging and the time interval between slaughter and packaging has a profound effect on meat quality and shelf life. For example, packaging veal within 24 hours post-mortem yielded a product that was lighter and had a longer shelf life than similar meat that was packaged later. (3) In conclusion, the veal industry should be evaluating factors that directly affect quality of the product. Less emphasis should be placed on color when, ultimately, taste and tenderness of the meat should be the determining factors in assessing the value of the product. Public education may be in order because often the consumer's perception of veal quality is based solely on the color of the meat in spite the lack of scientific evidence correlating veal color with meat quality.

**Key Words:** dairy, food, veal

### 198 Probiotics in yogurt and human health. Erin M. Sole\* and Gustavo J. Lascano, *Clemson University, Clemson, SC.*

Gastrointestinal diseases are becoming increasingly prevalent in the medical society today with approximately 60 to 70 million Americans affected by gastrointestinal diseases every year. One method of combating these diseases is through the use of probiotics. Therefore, the aim of this presentation is to provide a review of the literature and a summary of what probiotics and yogurt are, how probiotics are selected, factors that influence the effectiveness of probiotics in yogurt, and how probiotics can benefit human health. A systematic literature research was conducted to further investigate this topic. Databases used included Google Scholar and the *Journal of Dairy Science*. One of the primary vectors for human consumption of probiotic is through fermented dairy products, especially yogurt. The bacteria of the genera *Lactobacillus* and *Bifidobacterium* are added to the yogurt either before or following fermentation and have been proven to positively affect gut health by improving the balance between beneficial and harmful bacteria. Although probiotics are not essential for survival, probiotics are highly recommended as they improve gut health, especially under conditions that decrease the normal microflora of the intestines, such as chronic and acute diseases, antibiotics, and age.

**Key Words:** probiotic, yogurt, microbiology

### 199 Protein pricing and promoting: A renewed outlook on milk. Jessica M. Sentelle\*, David R. Winston, and Benjamin A. Corl, *Virginia Tech, Blacksburg, VA.*

Fluid milk consumption in the United States has steadily declined over the past several years. Whole milk consumption has declined the most by product as seen by data from USDA ERS. As consumers' tastes and preferences change, innovation in the dairy food industry must occur to sustain demand for dairy products. Recent promotions, such as "milk life" promote the nutritional aspects of milk. Fairlife ultra-filtered milk was recently introduced to capitalize on a growing demand for high protein drinks for the physically active consumer. After developing a patented cold-filtration process for milk, Select Milk Producers started a partnership with Coca-Cola to develop and distribute their dairy based protein drinks. Their products are offered in 4 varieties (whole, reduced fat, fat free, and chocolate); they are priced higher than conventional fluid milk. To obtain an ultra-filtered milk product, the founders of Fairlife use a patented cold filtration process that first concentrates the milk,

and then sterilizes the concentrate. The process differs from ultra-high temperature (UHT) processing and manages to avoid the characteristic burnt flavor of UHT milk (Maron and Corby, 2012). Milk components are separated to obtain a final product with twice as much protein, more calcium, less sugar, and no lactose. In a systematic review, Pasiakos et al. suggests that protein supplementation can enhance muscle mass and performance with adequate exercise procedures. This shows that high protein dairy drinks will be beneficial for active consumers. Ultrafiltration allows the separation of lactose to pick up a new class of lactose-intolerant consumers as well. In conclusion, consumption of fluid dairy products is steadily declining. As consumers' tastes and preferences change, innovation in the dairy food industry must occur to sustain demand for dairy products. Ultrafiltration and the new cold filtration process allow the separation and concentration of milk components to create a specialized product for different consumers. Pricing and promotion of new dairy products is very important to convey the proper message and increase demand.

**Key Words:** milk protein, lactose, ultrafiltration

**200 Reinventing sweetness in dairy products.** Halee L. Wasson\* and Dale R. Olver, *Pennsylvania State University, University Park, PA.*

In recent years, high levels of sugars in dairy products have created health concerns among consumers. According to the Mid-Atlantic Dairy Association, the average American consumes less than 2 servings of dairy each day. This indicates that many school children and others are missing out on the vital nutrient package provided by dairy products. In fact, chocolate milk has been removed from some school lunch programs due to its high sugar content. As a result, dairy researchers and manufacturers are challenged to identify acceptable alternatives for sweetening. Recent studies have focused on chocolate milk and ice cream to identify methods that reduce sucrose content while maintaining flavor. Research at North Carolina State University demonstrated that reductions up to 30% of sugar content in chocolate milk did not influence acceptability among young consumers. These researchers also investigated ways to use lactose hydrolysis to reduce sucrose content in chocolate milk. However, they found that the additional lactose needed to achieve the sweetness of sucrose did not result in a reduction of calories. Reducing the sugar content in ice cream can be even more challenging. This is because along with being a sweetener, sucrose plays a vital role in Freezing Point Depression Factor (FPDF). The FPDF

is important to ensure the desired hardness is reached to maintain ice cream's scoopable properties. Researchers have studied the effects of using erythritol and maltodextrin as sucrose substitutes in ice cream. Erythritol is a sugar alcohol known for its zero calorie status, but it has a high FPDF. Maltodextrin has a similar molecular weight, solubility, and FPDF as sucrose. With these alternative sweeteners and others, it is still possible for ice cream to maintain key attributes such as being scoopable or holding its shape on a stick. The findings of these studies present potential solutions to alter the sugar content in dairy products while maintaining functionality and flavor.

**Key Words:** chocolate milk, ice cream, lactose hydrolysis

**201 How fair is Fairlife?** Sarah Genest\* and Cathleen C. Williams, *Louisiana State University, Baton Rouge, LA.*

Coca-Cola is now distributing a new dairy product called Fairlife. It is milk that has 50% more protein, 30% more calcium, and half the sugar of regular milk. It is also lactose free. Recently, the consumption of soft drinks as well as milk has been declining. In the past 10 years, consumption of soft drinks has decreased 20%. Since 1970, consumption of milk has decreased by 37%. Coca-Cola believes that the recent interest in healthier food and drinks will drive consumers to buy Fairlife milk. A Coca-Cola executive even stated that she believed that Fairlife milk would make it "rain money." Milk is composed of water, fat, lactose, protein, and vitamins and minerals. Fairlife makes milk with more protein and calcium and less sugar by using cold filtration. The milk is pasteurized at a higher temperature than regular milk, but for a shorter time period. Then the cold filtration is used to separate the components of milk based on their molecular size. After this is completed, a larger ratio of protein and calcium are added into the milk. Lactase, an enzyme that breaks down lactose is also added. This ensures that Fairlife milk is lactose free. Fairlife was created by Sue and Mike McCloskey. They founded Select Milk Producers which is made up of family-owned farms. Their motto is "from grass to glass." They grow crops and formulate their own cow feed. The cows used to produce milk have freestanding stalls and are protected from harsh weather conditions. After the milk is collected, it is then loaded on Fairlife trucks and it brought to the plant in Michigan with only milk from that one dairy. It is processed and bottled in the same plant and then distributed to stores nationwide.

**Key Words:** cold filtration, milk, Fairlife

## ADSA Foundation PhD Symposium: Meeting the present and future demand for employees with a PhD

**202 Current and anticipated supply of people with PhDs.** J. R. Knapp\*, *Fox Hollow Consulting LLC, Columbus, OH.*

There is a growing concern among employers in academia and industry regarding the adequacy of PhD training programs for animal scientists in the US in terms of the number of graduates as well as their preparedness. Data from the National Center for Education Statistics and USDA Food and Agricultural Education Information System show that the number of PhDs awarded from animal, dairy, and poultry science departments has declined by nearly 40% in the past 2 decades. Currently, ~150 PhDs are awarded per year in animal science, 80 to 90 of whom are US citizens. While some non-citizens may seek employment and stay in the US, the majority return home. The decline in graduate students is associated with decreased funding for animal agricultural research, the declining numbers of animal science faculty at land grant universities, increased undergraduate enrollment, and the expansion of departmental programming to meet changing societal demands. Although a recent National Research Council publication noted that agricultural research funding to the land-grant universities has been flat in real dollars, the reality is that less funding is available to principal investigators (PIs) to support their research programs and graduate training now than in the past due to increased indirect cost rates charged by universities as well as expansion in administrations. Also, whereas graduate assistantships from formula funds and tuition waivers were the normal way of funding graduate training in the 1980s, today PIs often must cover tuition, stipend, and benefits from their research funds at costs of \$50,000 to \$70,000 per graduate student per year. The likelihood of increased federal funding for agricultural research and graduate training in the future is low, and new approaches to funding and operating graduate training are needed. Industry will need to take a larger role in these approaches. Also, significant redundancy exists among the land-grant agricultural experiment stations; regionalization would lead to more efficient use of limited research funding. Consistent availability and effective use of funding for graduate training will be needed to increase the number of animal science PhDs to a sustainable level for our field.

**Key Words:** graduate training, research funding, future of agriculture

**203 What can industry and academia do to help maintain viability of dairy/animal science departments at land-grant institutions?** Kenneth G. Odde\*, *Kansas State University, Manhattan, KS.*

Animal and dairy science departments at land-grant universities have long served their livestock industries through teaching, research and extension. The number of livestock producers has declined dramatically, and the number of students with farm and ranch backgrounds majoring in animal or dairy science has declined simultaneously. Additionally, Colleges of Agriculture and Departments of Animal Science and Dairy Science have become smaller and less influential components of land-grant universities. As the livestock producer constituency has declined in size, the power of livestock organizations to influence state funding levels and priorities for that state funding has also diminished. Other forces affecting departments include increased emphasis on federally funded research, rising standards for scholarship productivity for faculty members to achieve tenure and promotion, loss of livestock units in proximity to campus, loss of faculty positions, particularly tenure-track positions, greater investment of scarce resources in compliance, and greater emphasis on universities protecting intellectual property and capturing full research costs. The potential exists for stronger relationships between industry and university departments that will ultimately strengthen departments. The strong relationship requires that departmental leadership fully understands the needs and constraints of industry. Likewise, industrial organizations must also understand the nature of universities, and the variables that affect departmental and faculty behavior.

**Key Words:** animal science, dairy science, animal industry

**204 Graduate student perspective: Concerns with pursuing a PhD.** Curtis W. Park\*, *North Carolina State University, Raleigh, NC.*

College students are in a time of life that requires a lot of decisions that will affect their future careers. Attending graduate school and whether to pursue a PhD is a critical decision students have to make. In order for more students to pursue a PhD in a dairy science related field, more



must be done to inform them of the job opportunities available. Some students are intimidated by getting a PhD simply because they do not know what is required for the degree. A survey was conducted with both the ADSA Student Affiliate Division (undergraduates) and the ADSA Graduate Student Division to assess what students consider when making this important decision. The results from this survey will be discussed, as well as insights on how to better engage students early in their educational career, to help those who are thinking of pursuing a PhD in a dairy science-related field.

**Key Words:** PhD, graduate student

**205 Industry–university partnerships in research and graduate student training.** Michael L. Day\*, *Department of Animal Sciences, The Ohio State University, Columbus, OH.*

Industry partnerships for graduate education provide opportunities that benefit students' future careers, a means to meet industry needs for employees with advanced research and technical expertise, and a method to train adequate numbers of scientists for academia and industry, especially given escalating costs of graduate education. The OSU Office of Sponsored Programs (OSP; with M. L. Day as PI) and Select Sires Inc. (SSI) have had a cooperative training partnership program to train graduate students in bovine reproduction, with an emphasis in male reproduction, since 2010. The partnership was the result of recognition by the PI and SSI that the number of students trained in male reproduction was limiting and that needs for scientists with this expertise in industry and academia were unlikely to be fulfilled. This partnership provides a platform to leverage resources of OSU and SSI to study male reproduction. Inherent to all partnerships, especially given that the training affects the student's career, having a thoughtful agreement is crucial to protect interests of all parties. Hence, the resources of OSP were instrumental to develop this program. Key aspects of the agreement give guidance as to intellectual property, inventions, patents, publication of results, and so on. Costs of the graduate stipend and tuition are equally shared by SSI and the Animal Sciences department. Consensus between the PI and SSI scientists is necessary for admission into the program. One member of the student's advisory committee must be from SSI and the specific research hypotheses are determined by consensus to ensure needs of the student, PI, and SSI are met, and to affirm the research is comprehensive and high quality. Supply and travel costs for research are paid by SSI and matched "in-kind" by OSU. An important aspect of the agreement, for the student's protection, is that they are not obligated to be employed by SSI upon graduation. Two graduate degrees, with a third in progress, have resulted from this program. This model of university–industry collaboration has been very effective in producing young scientists to meet emerging needs in the area of male reproduction.

**Key Words:** graduate education, industry, partners

**206 Opportunities for PhD student training support at the National Science Foundation.** Steven Ellis\*, *National Science Foundation, Arlington, VA.*

In 1950, the National Science Foundation (NSF) was created to "... promote the progress of science; to advance the national health, prosperity, and welfare; and to secure the national defense." The NSF has been a strong supporter of graduate education throughout its history. For example, some of the NSF's first awards were to support graduate training, and extensive support for PhD training is still offered through the NSF Research Traineeship program (NRT) and the Graduate Research

Fellowship Program (GRFP). In 2015, the NRT expects to award almost \$37.9M and the GRFP is expected to award roughly \$333M in new and continuing awards (pending availability of funds). Additional support for the costs associated with graduate and postdoctoral training is available through fellowships and awards funded by core programs, special solicitations, and related activities in the research directorates. Understanding the NSF structure, priorities and merit review process are critical to securing support through these programs because each funding agency has a distinct set of goals and objectives. Researchers associated with the ADSA (or other member societies associated through FASS) can find very relevant funding opportunities in all of the NSF research directorates. However, those submitting proposals should recognize and emphasize the basic, fundamental aspects of their research proposals and activities. Merit review at the NSF centers around the intellectual merits of a proposal, and the broader impacts of the proposed work. So, although NSF does not have an explicit emphasis on food production or agricultural efficiency, animal production systems are potentially excellent experimental systems to reveal basic biologic processes, advance complex technical or engineering principles, and even elucidate economic factors. Putting any proposed effort in context with an over-arching and broadly relevant scientific scope can result in a compelling and fully competitive research proposal that includes support for student training. Such activities also represent excellent student training opportunities that will supply the future demand for PhDs with interdisciplinary skill sets.

**Key Words:** research funding, National Science Foundation (NSF)

**207 Research funding opportunities available through Foundation For Food and Agricultural Research.** Christopher Mallett\*<sup>1,2</sup>, <sup>1</sup>*Cargill, Wayzata, MN,* <sup>2</sup>*Foundation for Food and Agricultural Research, Washington, DC.*

The objective of this presentation will be to give an overview of the Foundation for Food and Agricultural Research (FFAR) and research funding opportunities available through the foundation. Authorized by Congress as part of the 2014 Farm Bill, the foundation operates as a non-profit corporation seeking and accepting private donations to fund research activities that focus on problems of national and international significance. The US Congress provided \$200 million for the Foundation, which must be matched by non-federal funds as the Foundation identifies and approves projects. The foundation will leverage public and private resources to increase scientific and technological research. Research funded by the FFAR is addressing issues including plant and animal health; food safety, nutrition and health; renewable energy, natural resources, and environment; agricultural and food security; and agriculture systems and technology.

**Key Words:** USDA, Foundation for Food and Agricultural Research (FFAR), research funding

**208 Dairy Management Inc.'s role in funding PhD research.** David R. McCoy\*, *Dairy Management Inc., Rosemont, IL.*

Dairy Management Inc. (DMI) is funded by America's nearly 49,000 dairy farmers, as well as dairy importers. DMI and its related organizations work to increase demand for dairy through research, education and innovation. US dairy farmers provide 15 cents per hundred pounds of milk, and importers provide 7.5 cents per hundred weight equivalent to the National Dairy Promotion and Research Program. Part of those funds go to DMI for use in funding dairy promotions as well as research. The research funds are used to provide for graduate student salaries

and supplies at several universities. The legislation that established DMI limits us to studies on the utilization of milk and dairy products and the creation of new products, so that the utilization of milk and dairy products may be encouraged, expanded, improved or made more acceptable. DMI also has developed consortia of industry, academia, and government agencies to leverage more funding into dairy research. Through the combination of dairy farmer/ importer contributed funds and consortia project funding, DMI and the associated state and regional check-off programs contract for a significant portion of the dairy foods research in the US

**Key Words:** funding, Dairy Management Inc.

**209 Funding opportunities for PhD programs in animal, dairy and poultry science at the USDA National Institute of Food and Agriculture.** Mark A. Miranda\*, Adele M. Turzillo, and Ray Ali, *USDA National Institute of Food and Agriculture, Washington, DC.*

A variety of opportunities exists to support PhD programs through funding provided by the USDA National Institute of Food and Agriculture (NIFA). NIFA competitive grant programs generally have broad eligibility and, thus, are readily available to support PhD programs across a wide range of institutions and programs, although eligibility is restricted to entities within the US. A major funding opportunity for PhD students is the Predoctoral Fellowship Program of the Agriculture and Food Research Initiative (AFRI) Food, Agriculture, Natural Resources and Human Sciences Education and Literacy Initiative. This

program provides support for up to 2 yr and \$79,000 in total costs/ grant for stipends, tuition, fees, fringe benefits, supplies, and travel. Individual students apply for the fellowships, thereby gaining valuable experience in preparing and submitting a grant proposal. Funding for PhD programs, including allowances for international training, is also available through NIFA's Food and Agricultural Sciences National Needs Graduate and Postgraduate Fellowship Grants Program. In that program, mentors apply for funding to support one or more fellowships totaling \$79,500/fellow over 3 yr, with grant awards limited to 3 yr and \$262,500 in total costs. Support for PhD students can also be included in applications to most other NIFA competitive grant programs. Those with greatest relevance to animal agriculture include, but are not limited to, the AFRI Foundational Program, AFRI Food Security Challenge Area, Organic Research and Extension Initiative, Organic Transitions Program, Biotechnology Risk Assessment Grants Program, and Beginning Farmers and Ranchers Development Program. Information about these and other NIFA funding opportunities is available at <http://www.nifa.usda.gov/fo/funding.cfm>. Funding support for PhD students may also be available through capacity-funded research and extension programs provided by NIFA to institutions (e.g., Hatch, Evans-Allen, Smith-Lever); however, these funds are subject to legislative and institutional restrictions, and thus, may not be readily available to support PhD programs at all institutions.

**Key Words:** federal funding, competitive grants, PhD program

# ADSA Multidisciplinary and International Leadership Keynote (MILK) Symposium: Global dairy perspective—Production, processing, people, politics, and priorities

**210 Role of milk and milk production in reducing poverty and malnutrition in emerging market countries.** Jim Yazman\*, *US Agency for International Agriculture, Washington, DC.*

Milk is the food that unites humans with the more than 5,000 other members of the Mammalia class. Milk supplies critical nutrients for newborns and is valued as a dietary component for all age groups. Expanding global trade in milk and milk components reflects an important role as a traditional foodstuff and in food product innovation. The rapidly expanding and urbanized middle class in emerging market countries is driving increased demand for milk and dairy products. The international development community and government partners are working to improve food security for vulnerable populations, including reducing high rates of malnutrition in adolescent, pregnant and nursing women and infants through diversification of diets. Milk and milk products are recognized by nutritionists as key components of diversified diets. Families grazing livestock across semi-arid lands often manage milking animals to supply milk for women and young children. In higher-potential areas, conversion of forage and crop by-products to milk and dairy products is a key income generation strategy for the poor, with women often controlling milk income. The US Agency for International Development (USAID) and other development partners recognize the potential of milk production and marketing to transform the lives of the rural poor. The Feed the Future initiative ([www.feedthefuture.gov](http://www.feedthefuture.gov)) is the US Government's contribution to the global effort to improve income, food security and nutrition in poor households. Implemented in Africa, Asia and Latin America and the Caribbean, Feed the Future strategies enhance household income by linking families to dairy value chains while increasing the availability, access and utilization of milk as a nutrient-rich animal-source food. Pastoralists as well as sedentary smallholder producers are assisted to increase milk-derived income as well as produce milk for home consumption. Dairy producers in emerging market countries face several challenges in an increasingly globalized dairy market: accessing technology and services; meeting raw milk quality standards; and control of production diseases, especially mastitis.

**Key Words:** food security, malnutrition

**211 Meeting 2050 global milk demand while freezing the environmental footprint of dairy production.** Roger A. Cady and Howard B. Green\*, *Elanco, Greenfield, IN.*

The Food and Agriculture Organization (FAO) of the United Nations projects that the global milk demand in 2050 will exceed 1 billion metric tonnes (MT), an increase of nearly 60% compared with 2010 production levels. The increase in milk solids demand will be 50 million MT, more than any other animal-sourced food except poultry products. The primary sources of human consumed dairy products are cattle and water buffalo (96%). Combined global milk production from these 2 species more than doubled from 1961 to 2010 (331.5 million MT to 692.0). However, milk production has not kept pace with human population growth, resulting in 6% less bovine milk produced per capita than in 1961. Milk production must grow more in the next 35 years than it has in the past 50. Furthermore, growth must occur without increasing the environmental footprint of milk production. Between 1961 and 2010, milk supply from dairy cattle increased primarily due to increased cows (54%). Since 2007, 82% of supply increase is due to a larger cattle

population. That contrasts to an average annual yield increase of only 9.7 kg/cow/yr of cattle milk since 1961. Every additional cow to the population adds between 9 and 13 MT of CO<sub>2</sub>e (accounts for additional replacement heifers and bulls) to the atmosphere annually. To meet 2050 demand and freeze the environmental impact of milk production requires increasing yield by at least 45 kg/cow/yr annually between now and 2050. This compares to sustained annual increases in developed countries of 130 kg/cow/yr and over 200 kg/cow/yr in some countries with developing dairy industries. In regions where milk production increases are limited, increased trade will be required to source milk from more resilient production regions.

**Key Words:** dairy, environment, sustainability

**212 Global dairy: African perspective.** Pieter H. Henning\*<sup>1</sup> and Lourens J. Erasmus<sup>2</sup>, <sup>1</sup>*Meadow Feeds, Johannesburg, South Africa*, <sup>2</sup>*University of Pretoria, Pretoria, South Africa.*

Africa is often seen as a “dark continent” yet it has the potential to be a continent of “milk and honey.” The objectives of this presentation are to describe the dairy industry in Africa, highlight the challenges faced, and propose how they may be met. The human population of Africa is just over 1 billion. The 8 leading “dairy” countries in Africa together have approximately 37 million dairy cows. Average milk production varies from 0.12 to 6.45 tons/cow/year, and annual per capita consumption ranges from 8 to 241 kg milk equivalent (ME). Comparative figures for the United States are 9.2 million dairy cows, 9.44 tons/cow/year and 259 kg ME/capita. Growing urbanization and westernization are increasing the demand for milk and dairy products in Africa. This is further supported by a rapidly expanding supermarket industry. There is good potential for even more growth in demand. Africa is currently a net-importer (ca 6 million tonnes/year) of dairy products. Cows, though, are abundant in Africa and there is great scope to increase milk production from the continent's cow population. Growth in dairy farming will also contribute to improved livelihood for many African people. However, serious challenges in respect of infrastructure, animal husbandry practices, genetic material, animal nutrition and cow health have to be met if Africa's dairy potential is to be realized. Africa has the intellectual potential but needs to learn and apply suitable technical knowledge and skills. The internet, cell phone, and tablet have revolutionized communication and can play a significant role in the transfer of this knowledge and skills to the emerging African dairy farmer. Politics still remain a stumbling block in some African countries. Two priorities for the dairy industry in Africa are infrastructure creation (from milk collection to final products) and the transfer of appropriate technical knowledge and skills to create successful dairy farmers, whether small or big. The latter priority, especially, poses a challenging opportunity for first-world dairy organizations, such as ADSA, and its members.

**Key Words:** Africa, dairy industry, technical challenge

**213 Dairy in China: Present status and future prospects.** Jiaqu Wang\*, *Chinese Academy of Agricultural Sciences, Beijing, China.*

China's dairy industry has made tremendous progress in the past 20 years, with cows and milk production growing >10% /yr. In 2000,



total number of dairy cattle was <5 million producing 8.27 million tons of milk. In 2014 there were 14.98 million producing 37.25 million tons of milk. Dairy was the most rapid growing section among all livestock production. Herd size shifted from 5 cows in 1995 to a larger present size, with 40% of dairy cows in herds >50 cows. Over 70% of milk is produced and processed in northern China. Dairy industry international trade has shown strong activities since China became a WTO member. Animal importation increased from 37453 cows in 2009 to 99348 cows (2011) and 195000 cows (2014). Importation of dairy products grew more significantly. In 2009, China imported 596999 tons of dairy products, but increased to 1,880,000 tons in 2014. 88% was dry products, mainly milk powder and whey. Seventy percent of the imported milk powder was from New Zealand with Australia, EU and USA also important dairy trade sectors. China exported 20000 to 25000 tons of dairy products annually in the past 5 years. Annual urban dairy product consumption increased from 15 kg/ person (2000) to 23 kg/person (2012), while consumption in rural areas remains relatively low and stagnant (<10 kg/person). Dairy processing plants are becoming larger with Yili and Mengniu processing 10,000 tons of raw milk daily. Currently, raw milk production is from 3 models: Cropping area model (70% of milk; small family farms, local feeds, local raw milk stations), suburban area model (20%; near stable consumer markets) and pasture model (10%). Average annual productivity is 5500 kg/cow. Low productivity means low efficiency and high cost. It is reflected in high raw milk prices (\$0.8/L in 2013). China will be the world market center of dairy products in the future due to the rapid growing demand of the huge population. More international trade of dairy products will encourage information and technology exchanges, which will help improve quality and efficiency of domestic production. China's dairy industry must be ready for international competition and cooperation. Standards, policies and legislations will be critical for such prospects.

**Key Words:** China, dairy production, dairy product

**214 Brazil: Recent growth, importance and future of dairy markets.** Marcelo Pereira de Carvalho\*, *AgriPoint Consultoria, Piracicaba, SP, Brazil.*

This presentation will offer an overview of recent and expected future developments of milk and dairy markets in Brazil, which have expanded

significantly in the last 15 years. Since 2000, per capita consumption moved from 122 to 176 kg/year in 2014 (in milk-equivalent terms). As population grew from 173 million to 203 million in the same period, total market increase almost 70% or 14,6 billion kg in volume. This growth was accomplished mainly by domestic production, which expanded from 19,7 billion kg (2000) to 34,3 billion (2013). From 2001 to 2013, the amount of milk delivered to dairy companies for processing increase 6,5x more than the amount unprocessed, a rate of growth of 5,4% per year. Cheese production increased from 440.000 metric tons in 2000 to nearly 1 million tons in 2013, a growth of 6,6% per year, reaching 5 kg/person/year. In the last 10 years, fresh dairy products doubled its market size, from 780.000 tons in 2003 to 1.450.000 tons in 2013, reaching 7,5 kg/person/year. Fluid milk market grew 2,3% per year, with a gradual replacement of raw and pasteurized milk by UHT milk. Income growth and better income distribution were the most important drivers for this growth. Since 1994, minimum wage grew 2x more than milk prices. From 2004 to 2013, salaries grew 72% over inflation, with 17 million new jobs created. Middle class expanded from 33% to 42% of total population. What are the challenges? Economic performance is lagging behind what Brazil experienced during the last decade and past income gains are being challenged. Also, more strict regulation regarding labeling and claims may negatively affect innovation. There have been milk quality problems, suggesting weak supply chain coordination. Productivity, production per farm, cost of production and overall farm efficiency also factors that need to be greatly improved. Anti-dairy groups are not an important factor yet, but there is a growing number of health professionals not recommending dairy products. Brazil is a large and still growing dairy market, but a more challenging environment demands a more active approach to ensure market expansion.

**Key Words:** consumption, markets, trend

# ADSA Southern Section Symposium: Maximizing forage quality in the Southeast

**215 Improving corn silage quality in the Southeast and throughout the United States.** Donna M. Amaral-Phillips\*, *University of Kentucky, Lexington, KY.*

Corn silage has been called the king of stored forages for good reason. Dry matter yields and milk production potential per acre consistently surpass other forage crops while allowing farmers to harvest large amounts of quality feed over a relatively short harvest period. Corn raised for silage can be used in combination with other forage crops to allow for the implementation of double or even triple cropping systems. Thus, increasing the number of dairy cattle supported by each acre of farmland and increasing utilization of nutrients applied to soils through manure applications (i.e., phosphorus). Seed companies have improved the genetics of corn hybrids used for silage, especially as they relate to NDF digestibility. These genetic improvements have increased the nutritional value of these crops, which is even more important under the extended heat stress conditions and resulting decreased dry matter intakes of the Southeast. Environmental temperatures in excess of 32°C and limited rainfall during tasselling and pollination decrease grain development in the corn plant, resulting in a lower energy crop and presenting challenges for meeting the energy needs of high producing dairy cows. Harvesting corn for silage at the proper dry matter content and particle size can affect packing density and quality of silage available at feedout. Starch digestibility can be improved with the proper use of kernel processors on silage choppers. Silage management at harvest and feedout also affects feed quality and dry matter losses.

**Key Words:** corn silage, forage, corn

**216 The effect of forage quality on health and performance of dairy cattle.** Adegbola T. Adesogan\*, *University of Florida, Gainesville, FL.*

Forages constitute a major portion of the diet of dairy cattle; therefore, factors that affect forage quality can determine the performance and health of the cows. The quality of southeastern forages is relatively low because the prevailing high temperatures favor fiber deposition. This factor coupled with heat stress reduces the productivity of dairy cows in the southeast. Consequently, strategies that increase the quality and utilization of southeastern forages are needed critically. Though effective at increasing forage utilization, strategies like kernel processing and chemical treatment have not been widely adopted. Other approaches such as application of fibrolytic enzymes have given variable results but strategic supplementation with yeast or yeast culture is often effective. Recent studies have shown that milk production was increased by replacing conventional corn silage with shredlage, BMR hybrids or mutant hybrids with lower ferulic acid concentrations. Similar research is needed for other high yielding warm-season grasses adapted to the southeast. Forage-related factors that can reduce the performance and health of dairy cows can be classified as those occurring during the growth or storage of the forage. Compounds such as nitrates, glycosides and alkaloids can accumulate in growing forages during inclement weather or under inadequate management. In addition, forage stressors such as disease, hail, pests and lodging can result in mycotoxin contamination by invasive fungi, which can reduce milk production and safety. In addition, poorly managed silage can reduce milk safety and predispose cows to low milk production and diseases due to accumulation of harmful compounds like mycotoxins, nitrates and biogenic amines or pathogens

like *Listeria*, *Bacillus*, *Clostridia* and molds. Microbial inoculants and chemical additives can increase forage preservation, prevent accumulation of harmful compounds and inhibit growth of pathogens but their effects on milk production by dairy cows have been variable.

**Key Words:** forage quality, milk, health

**217 Warm season annual grasses for dairy cattle.** J. K. Bernard\*, *University of Georgia, Department of Animal and Dairy Science, Tifton, GA.*

Warm season annual grasses are commonly used by dairy producers in their forage programs for grazing, green chop, hay, or silage production. Most warm season annual grasses require less water to grow and are more drought and heat tolerant than corn. These characteristics along with improved varieties that have improved DM yield and higher fiber digestibility have increased the interest in their use for dairy cattle. The incorporation of the brown-midrib 6 gene (*bmr*) in varieties increases in fiber digestibility and energy to support production compared with non-*bmr* varieties. Warm season annual grasses are susceptible to nitrate toxicity during drought conditions. Prussic acid toxicity is an issue for members of the sorghum family during early growth and after stress from frost. Of the warm season annuals, forage sorghum is used more frequently for silage production. It is susceptible to lodging, but new brachytic dwarf varieties have shorter internodes, reducing lodging potential while providing forage with greater leaf to stem ratio. Diets based on *bmr*-6 forage sorghum silage have supported milk yield comparable to that of corn silage based rations when balanced for fiber content. Pearl millet silage support similar DMI and milk yield, and higher milk fat percent compared with corn silage; however, other trials reported reduced ADG or milk yield when fed to growing heifers or lactating cows. Differing responses may be partially attributed to differences in fermentation of the silages produced from pearl millet, which is more challenging to ensile if water-soluble carbohydrate concentrations are low. Data on *bmr* sorghum-sudangrass are limited and suggest that these forages can support solids-corrected milk yield similar to that of corn silage-based diets. Warm season annual grasses can be effectively incorporated into forage systems for dairy cattle. Improved varieties, especially *bmr* varieties, can support acceptable milk yield.

**Key Words:** dairy cattle, warm season annual, milk yield

**218 Forage systems for Southern dairy production.** John G. Andrae\*, *Clemson University, Clemson, SC.*

Forages are the major component of lactating dairy rations and make up an even greater proportion of replacement heifer and dry cow diets. Due to the wide range in climate, soils, irrigation availability, and cow management (i.e., confinement vs. grazing vs. hybrid systems), a "cookie-cutter" approach to designing forage systems for southern US dairy production is impossible. Consequently, forage systems are driven by local conditions and management preferences and often vary markedly even within a community. Traditional forage systems comprised of corn, alfalfa and/or small grains silage are common regionwide; however, rising fertilizer input costs and risks associated with drought and irrigation water have increased plantings of forage sorghum and pearl millet. Tropical corn hybrids, Tifton 85 bermudagrass, and adapted

non-dormant alfalfa varieties have also improved silage and green chop options for confinement forage systems. Baleage as a production practice has increased the feasibility of harvesting high quality forage when traditional silage equipment is prohibitively expensive or where custom harvesters are unavailable. Grazing dairies have also recently increased some areas. Forage systems for these dairies vary depending on region and range from non-toxic tall fescue, alfalfa, bermudagrass and orchardgrass based perennial systems to annual systems including ryegrass, crabgrass, pearl millet and sorghum × sudangrass. Systems regionwide will be reviewed to provide an overview of their diversity and uses.

**Key Words:** forage, silage, grazing

**219 Environment and crop management as determinants of forage yield and quality in the Southeast.** Gonzalo Ferreira\*, *Virginia Polytechnic Institute and State University, Blacksburg, VA.*

After the summer and spring drought of 2012, increasing interest has emerged to use alternative species to maize (*Zea mays*) to produce forages in dairy production systems. Sorghum species (*Sorghum* sp.) are characterized for having greater water use efficiency, water potential, and transpiration and photosynthetic rates under drought conditions. In addition to this, sorghum shows higher turgor potential than corn at the same water status. Despite all these attributes, the land surface planted with corn for silage is substantially greater than that of sorghum for

silage in the Southern region. Potential evapotranspiration is substantially greater in the Southern than in the Midwest region. Therefore, to minimize the adverse effects of drought and heat stress, in the Southern region adequate crop management should be prioritized. Drought stress around silking diminishes kernel development by negatively affecting pollination, mainly through an increased anthesis-to-silking interval. Kernel development is also negatively affected when high temperatures ( $\geq 35^{\circ}\text{C}$ ) occur during the lag phase of kernel development. High temperatures immediately after silking limit starch accumulation within the kernels, and increases the rate of kernel abortion as well. Therefore, in regions with extended periods of temperatures greater than  $35^{\circ}\text{C}$ , choosing early maturity corn hybrids, or delaying planting date, should be considered to avoid drought and heat stress during silking and kernel development. Planting density can also affect yield of corn for silage, although this effect depends on the crop rotation system and time of harvesting. Corn silage yield increased at high corn planting densities when an extended fallow and abundant precipitations occurred during the crop cycle. However, no differences in corn silage yields were observed when different corn planting densities were used in more intensive (i.e., double-crop) rotation systems. The effects of corn planting density on nutritional quality are also dependent on environment, although these effects are likely dependent on harvesting time. In summary, adequate crop management strategies should be carefully considered to ensure and maximize yield and quality of corn silage in the Southern region.

**Key Words:** corn silage, drought stress, heat stress



# ADSA-SAD (Student Affiliate Division) Undergraduate Competition: Dairy Production

**220 The effects of heat stress on reproductive fertility: An effective solution.** Alexandra T. Lemus\* and Peter J. Hansen, *University of Florida, Gainesville, FL.*

By virtue of its high metabolic heat production (4–5 times maintenance), the lactating dairy cow is very sensitive to heat stress. A rise in body temperature of 1–2°C can have negative effects on various aspects of reproductive function. Heat stress can reduce luteinizing hormone and steroid hormone secretion, alter folliculogenesis, disrupt oocyte quality and inhibit embryonic development. The net result is reduced expression of estrus and low fertility. Effects on oocyte quality take place over a broad time frame. The ovarian follicle requires about 4 mo to develop from the primary stage to ovulation. The effects of heat stress on the oocyte within the follicle can persist for as long as 103 d. This stress in turn alters the integrity of the follicle and consequently makes it more difficult for the fertilized oocyte to develop into a blastocyst. The newly formed embryo is also susceptible to maternal heat stress. After 2–3 d, however, the embryo becomes increasingly resistant to heat stress and by d 7, when embryo transfer is ordinarily performed, elevated temperature has little effect on the embryo. Still another effect of heat stress is the depression of estrus behavior where both the duration and the intensity of estrus decrease resulting in an increased percentage of missed estruses. The combined effects of reduced detection of estrus and reduced fertility after insemination result in lower conception rates during warm months. These negative effects of heat stress can be bypassed by incorporation of 2 reproductive technologies. Ovulation synchronization protocols such as Ovsynch allow farmers to predict the time of ovulation without the need for estrus detection. Fertility is improved when synchronization is performed in conjunction with embryo transfer. Embryo transfer bypasses causes of infertility due to loss of oocyte competence and damage of the early embryo due to elevated maternal body temperature. The d 7 embryo used for transfer has acquired resistance to heat stress; therefore, maternal heat stress no longer affects the embryo.

**Key Words:** fertility, embryo transfer, synchronization

**221 Effects of colostrum and milk intake on future performance in dairy calves.** Katherine M. Kelly\*, Donna M. Amaral-Phillips, and Jeffery M. Bewley, *University of Kentucky, Lexington, KY.*

The first 56 d of a calf's life are critical for improving future milk production. Doubling birth weight within the first 56 d increases future milk production. Colostrum harvested from the mother within 10 h after calving loses 27% of IgG (Jaster, 2004). Consuming 4 L of colostrum compared with 2 L improves average daily gain (ADG), which has a direct effect on milk production (Soberan et al., 2012). Insulin found in colostrum affects gut absorption and growth (Van Amburgh et al., 2011). Without immunity from colostrum, calves that are given antibiotics, due to illness, are 2.5 times more likely to die within the first 2 years of life (Soberan et al., 2012). Milk intake also affects ADG. Calf metabolizable energy (ME) requirement is about 2.34 Mcal/d for a 50 kg calf, to support maintenance. Conventional feeding systems provide the calf with reduced amounts of milk so the calf will begin eating calf starter sooner. Conventional feeding systems provide the calf with enough ME for maintenance (Drackley, 2011). Higher intakes of whole milk provide the calf with enough dietary fat to support maintenance and growth

(Van Amburgh, 2011). Increased growth increases milk production in Holstein calves during their first lactation and decreases age at first breeding (Khan et al., 2010). Through increased colostrum and milk intakes, milk yields and production life can be increased.

**Key Words:** calf performance, colostrum, milk intake

**222 The effects of feeding heat-treated colostrum to dairy calves.** Rebecca N. Klopp\* and Dale R. Olver, *Pennsylvania State University, University Park, PA.*

Heat-treated (HT) colostrum offers many benefits when fed to dairy calves. Calves are born agammaglobulinemic because the placenta does not allow antibodies to pass from the dam to the fetus. As a result, neonatal calves rely on passive absorption of antibodies from colostrum in the first few hours after birth to protect them from infectious diseases. The most prevalent of the colostrum antibodies is IgG. This antibody is commonly used as an indicator to show that immunity through passive absorption was achieved. In the past, commercial pasteurizers were used to process both waste milk and colostrum, and they achieved temperatures of at least 63°C to reduce risks of microbial contamination from collection and storage. However, heating colostrum to this temperature reduced IgG concentrations and increased viscosity. Recent studies have demonstrated that heating colostrum to a lower temperature (60°C) for up to 120 min drastically decreased bacterial populations while still maintaining IgG levels and viscosity. Research also shows that calves have a reduced risk for illness when fed HT colostrum compared with fresh colostrum because the heat treatment process significantly reduces total coliform counts. Calves that are fed HT colostrum will more efficiently absorb immunoglobulins compared with calves fed unheated colostrum. Plasma samples taken from calves showed that IgG concentrations in calves that were fed HT colostrum increased by 18.4% with a 21% increase in apparent efficiency of absorption compared with calves fed fresh colostrum. Heat treating colostrum is a continually improving technique resulting in an increased rate of IgG absorption in dairy calves.

**Key Words:** heat-treated colostrum, calves

**223 Rumen development in dairy calves.** Morgan Richard\* and Cathleen C. Williams, *Louisiana State University, Baton Rouge, LA.*

At birth, a dairy calf's rumen is not yet developed physically or metabolically. Thus, the calf functions as a monogastric animal until 4 to 8 weeks of life. In order for a calf to transition to a ruminant, the rumen must be able to support fermentation. This development of the rumen is necessary for successful weaning and is primarily affected by dietary change. The rumen changes physically by increasing mass and growth of papillae. At birth, a calf's rumen is only 25% of the stomach capacity, but a mature cow's rumen can compose up to 80% of its stomach. The physical stimulation of feed in the rumen can cause increases in rumen weight and muscular development. However, physical bulk alone has not been proven to promote papillary development. When calves drink milk they stimulate the opening of the esophageal groove. This closure keeps milk from entering the reticulo-rumen and prevents fermentation in the rumen. Calves should be given free choice water to produce the liquid environment needed for microbial growth. Besides liquid and muscular

action, the ingredients needed in rumen development consist of bacteria, absorptive ability of the tissue and substrate. A newborn calf has a sterile rumen, but by 2 d after birth microbes are colonized. These microbes influence growth and development. The earlier dry feed and forage is given to calves, the earlier microbial development occurs. Higher rumen metabolic activity and increased Volatile Fatty Acid concentrations are a direct reflection of the nutrient substrate required for rumen microbes. The fermentation end products of butyrate and propionate are responsible for the growth of ruminal papillae. In conclusion, early consumption of dry feed develops the calf rumen physically and metabolically by promoting growth of rumen epithelium and mobility.

**Key Words:** dairy calves, rumen development

**224 The effect of automated calf feeders on calf welfare.** Kelly H. Leatherman\*, David R. Winston, and Robert E. James, *Virginia Tech, Blacksburg, VA.*

The dairy calf usually spends its first 6 to 8 weeks in individual pens. The reasons for this practice are to provide individual calf care and to prevent disease. However, there is a push from consumers to consider the welfare of dairy calves by moving away from individually housed, limit fed calves to a system of group housed calves fed ad libitum. The inclusion of an automated calf feeder into a farm's calf rearing program would, if managed correctly, provide the opportunity to increase the overall welfare of dairy calves. Group housed calves tend to be more relaxed when introduced to new situations and are quieter during the weaning process as opposed to their individually housed counterparts (Vieira et al., 2010). By using automated calf feeding systems, calf growers can offer more milk, more often to calves. Calves on automated calf feeders with an ad libitum milk feeding program consume up to 10 kg of milk per day (Jensen and Weary, 2002) which is similar to how a calf would nurse if left with the cow. Calf growing programs that incorporate an automated calf feeder can greatly increase the welfare of pre-weaned calves though increased nutrition and reduced stress.

**Key Words:** automatic calf feeder, calf management

**225 Effectiveness of DHIA herd testing frequency on management decisions and dairy herd performance.** Lauren E. G. Clemency\*, Kasimu Ingawa, Steven Washburn, John Clay, and Shannon Davidson, *North Carolina State University, Raleigh, NC.*

Statistical analyses indicate that more data points usually improve the reliability of results; hence the objective of this study is to test the hypothesis that higher testing frequency of a dairy herd by Dairy Herd Improvement Associations (DHIA) affects management decisions and herd performance. Higher frequency of DHIA testing provides more information that can be used to make management decisions, thereby enhancing the decision making process; consequently, resulting in improved dairy herd performance. DHIA's are a farmer-owned organizations originating in 1905 as Newaygo Dairy Testing Association, the first Cow Testing Association (CTA) in the US; it assumed its current name, Dairy Herd Improvement Association in 1927. Throughout its more than 100-year history, the core focus of DHIA has not changed from being

the means for dairy cattle genetics improvement in the United States. Basically, dairy farm data are periodically collected on each cow and processed into reports which dairy farmers use to manage their herds. Such records include milk and fat weights, percent butterfat, milk fat price, reproduction parameters, weights of different roughages and grains fed, etc. Values of performance variables are computed to determine periodic profits or losses for each cow in the herd. The National DHIA reports a declining trend in DHIA dairy herd testing participation in the last 11 years; in 2004, about 25,077 dairy herds were on test compared with 17,875 as of 1/1/2015. However, number of dairy cows on DHIA test increased during the same period which indicates shrinking number of dairy farms but an increase in dairy farm size. The report also indicates the Holstein breed to be the predominant breed of DHIA testing. A preliminary study by Minnesota DHIA indicated a production increase resulted from a higher frequency of DHIA dairy herd testing, however, variation due to management approach was not considered. In this study, higher frequency of dairy herd DHIA testing resulted in more valuable herd management information and that management approach should be considered when analyzing how the information should be used to manage dairy herds.

**Key Words:** DHIA, testing, reports

**226 Hyperkeratosis: A costly consequence of milking equipment.** Kayla J. Alward\* and Jillian F. Bohlen, *University of Georgia, Athens, GA.*

Milking a cow causes stress to the teats and damage that is normal, but that can become severe due to variations in cow genetics, management, and equipment. Problems with the teats can be assessed through teat conditioning and may be the result of regular milking or faulty equipment. One alteration of the teat due to milking is hyperkeratosis, which is a thickening of the skin that lines the teat canal, characterized by excessive keratin growth. This excessive growth leaves an area for bacteria to hide and breed, which cannot be sanitized, and increases the risk of mastitis or intramammary infections (IMIs). The severity of the hyperkeratosis is drastically increased due to milking equipment factors. Duration of milking or over-milking, milk flow rate, liner closed phase, vacuum level and the amount of time the cups are on the teats all contribute to hyperkeratosis. The total time that the milk flow rate is less than 1.0 kg/min per day is the main contributing factor to hyperkeratosis severity. Faulty milking equipment has the potential to further exacerbate issues with hyperkeratosis by exposing the teat end to a higher number of bacteria than normal. Once present, these IMIs decrease producer profits by reducing total milk production, increasing somatic cell counts, treatments, cull rates, and milk lost due to treated cows. Although average somatic cell count (SCC) has fallen in herds over the past 20 year, national estimates still cite a to \$300 per cow per year loss to elevated SCC. Comprehensive teat assessment and milking equipment checks are an effective way to help minimize incidences of hyperkeratosis. Understanding how and when to do both may help producers to reduce profit loss to IMIs as a result of hyperkeratosis due to milking equipment.

**Key Words:** hyperkeratosis, milking equipment, mastitis

## ADSA-SAD (Student Affiliate Division) Undergraduate Competition: Original Research

**227 Use of green vegetative index maps to predict nutritional quality variation of corn silage.** Eleonor L. Cayford\*<sup>1</sup>, Leyang Feng<sup>2</sup>, Shao Yang<sup>2</sup>, and Gonzalo Ferreira<sup>1</sup>, <sup>1</sup>*Department of Dairy Science, Virginia Polytechnic Institute and State University, Blacksburg, VA*, <sup>2</sup>*Geography Department, Virginia Polytechnic Institute and State University, Blacksburg, VA*.

The objective of this study was to evaluate the use of remote sensing techniques to anticipate the nutritional variation of corn silage. Cornfields (i.e., fields) from 3 commercial dairy farms located in Pittsylvania, Montgomery, and Washington counties in Virginia were used. Fields had an approximate surface of 25, 10, and 54 ha, respectively. Landsat images were obtained from the US Geological Survey online EarthExplorer system for the months Apr to Sep from 2000 to 2014. The spatial resolution of the images is 30 m. Visual assessment of cloud contamination in and around the 3 fields was made using red band ( $R_{red}$ ) and near infrared band ( $R_{nir}$ ) imagery. Images were then made into subsets using the field boundaries to calculate the normalized difference vegetation index (NDVI). Normalized difference vegetation index maps were derived for all images as follows:  $NDVI = (R_{nir} - R_{red}) / (R_{nir} + R_{red})$ . This NDVI equation produces values in the range from -1 to 1, where positive values indicate vegetated areas and negative values represent non-vegetated areas, such as water, clouds, or snow. The NDVI values for each pixel of all images were calculated under ArcGIS/Arcpy environment. Low, mid, and high NDVI values within a field were represented by red, yellow, and green pixels, respectively. Coordinates for a single red, yellow, and green area within each field were obtained. At harvesting time, each of the 3 selected areas within each field was reached using a GPS, and 3 samples composed of 8 plants were cut 15 cm above ground (3 areas  $\times$  3 samples = 9 samples per field). Whole-plants were weighed, chopped, mixed, and ensiled in bags for 60 d. Nutritional composition of corn silage was performed by wet chemistry. Data was analyzed as a randomized complete block design, where field and NDVI were blocks and treatments, respectively. Dry plant biomass was similar among NDVI areas (270 g/plant;  $P > 0.39$ ). Concentrations of DM (28.3%;  $P > 0.25$ ), ash (4.55;  $P > 0.38$ ), CP (10.5%;  $P > 0.29$ ), NDF (41.3%;  $P > 0.49$ ), and ADF (25.3%;  $P > 0.89$ ) did not differ among NDVI areas. In conclusion, differences in NDVI in cornfields did not correspond with differences in nutritional composition of corn silage.

**Key Words:** corn silage, nutritional variation, normalized difference vegetation index

**228 Processed water and its effect on daily intake and growth in dairy calves.** Patrick J. Neff\*<sup>1</sup>, Matt C. Claeys<sup>1</sup>, and Tamilee D. Nennich<sup>1,2</sup>, <sup>1</sup>*Purdue University, West Lafayette, IN*, <sup>2</sup>*Famo Feeds, Freeport, MN*.

There are potentially bacteria and minerals in well water that could reduce the growth and health of preweaned calves. The objective of this study was to determine the effects of providing dairy calves processed drinking water on calf growth, starter intake, and water intake, as well as determine the efficacy of a novel water processing system on water mineral levels. Sixteen Holstein heifers ( $49.9 \pm 7.7$  kg of BW) were assigned to 1 of 2 drinking water treatments in a randomized complete block design and blocked by birth date. Treatments were either conventional well water (CONV) or processed water using a novel processing

system (PRO). Water was sampled 1 $\times$ /mo for analysis of minerals. Calves were fed 1.9 L of a 22:20 milk replacer with 0.70 kg/d of powder being fed 2 $\times$ /d and a 20% CP texturized calf starter. Body weights, hip width (HW), hip height (HH), and heart girth (HG) were measured every 2 wk. Calves began treatments at 2 wk, were weaned at 8 wk, and ended the study at 10 wk of age. Starter and water intake were determined daily. Data were analyzed as repeated measures using PROC MIXED in SAS with calf as the experimental unit. Water mineral content was similar between treatments with sulfates averaging 45.3 ppm in CONV and 35.0 ppm in PRO, chlorides averaging 14.0 and 11.7 ppm in CONV and PRO, respectively, total dissolved solids of 330.3 ppm in CONV and 259.3 ppm in PRO, and sodium levels of 6.3 for CONV and 5.0 ppm for PRO, respectively. In this study, growth results were similar for calves regardless of treatment. The average BW for calves over the study was 155.2 kg for CONV and 156.8 kg for PRO ( $P = 0.63$ ) and ADG was 0.79 and 0.83 kg/d for CONV and PRO ( $P = 0.43$ ), respectively. Starter intake for CONV averaged 1.2 kg/d and 1.3 kg/d for PRO ( $P = 0.74$ ) with feed efficiencies averaging 1.01 kg starter intake/kg gain overall ( $P = 0.86$ ). Average water intakes were 2.0 and 2.2 kg/d for CONV and PRO ( $P = 0.50$ ), respectively. The HH averaged 89.9 cm ( $P = 0.80$ ), HW averaged 20.9 cm ( $P = 0.43$ ), and HGC averaged 96.5 cm ( $P = 0.67$ ). In this study, both treatments showed similar results for water mineral levels, water intake, starter intake, and growth.

**Key Words:** dairy calves, water, growth

**229 Calving detection in dairy cattle using a novel vaginal temperature device.** Megan C. Hardy\*, Denise L. Ray, Joey D. Clark, and Jeffrey M. Bewley, *University of Kentucky, Lexington, KY*.

Parturition detection and dystocia prevention represent major challenges to dairy farmers. A potential solution is a vaginal thermometer used to detect calving. The objective of this study was to assess the Vel'Phone vaginal thermometer (Vel'Phone, Medria, Chateaubourg, France) for calving prediction accuracy and to characterize temperature and behavior before calving time. Primiparous ( $n = 40$ ) and multiparous ( $n = 55$ ) Holstein cows were enrolled in the study, conducted at the University of Kentucky Coldstream Dairy from May 2013 to August 2014. The Vel'Phone thermometer was vaginally inserted into each animal before the expected calving date. Vel'Phone service provided text messages displaying daily temperature reports, as well as impending calving and thermometer expulsion alerts. The Vel'Phone collected temperature data to investigate precalving temperature decreases. Alert timing data were collected to assess the Vel'Phone's calving prediction ability. Video monitoring recorded calving time and behavior (Barn Cams, Oconomowoc, WI). The MEANS procedure of SAS 9.3 (SAS Institute, Inc., Cary, NC) was used to calculate the average time between impending calving alerts, expulsion alerts, and calving time. The FREQ procedure of SAS created alert-frequency distributions. The GLM procedure of SAS was used to assess the effects of calf weight, calf sex, parity, calving ease, and season on precalving temperature decreases. A gradual temperature decrease of 0.6°C was seen at 48 h before parturition. Calf weight, calf sex, parity, calving ease, and season did not have a significant ( $P < 0.05$ ) effect on precalving temperature decreases. Results indicate temperature



decreases 48 h before parturition and thermometer expulsion alerts could be useful in calving detection technology.

**Key Words:** vaginal temperature, calving detection, precision dairy technology

**230 Consideration of *DGATI* interactions with DNA markers improved genetic predictions.** Amber N. Gabel\* and Chad D. Dechow, *The Pennsylvania State University, University Park, PA.*

*DGATI* is a major gene influencing yield in cattle with effect magnitudes that vary by breed, implying that *DGATI* interacts with other parts of the genome. This study aims to determine whether consideration of interactions between *DGATI* and single nucleotide polymorphisms (SNP) could improve the accuracy of genomic predictions for fat yield in Holsteins. The initial data set included 1,143 305-d fat yield records from 358 Holstein cows in 11 herds that were genotyped for *DGATI* and 45,187 SNPs. A series of analyses included a random animal effect, a fixed *DGATI* effect, a single fixed SNP effect, and a fixed interaction of *DGATI* with the same SNP. This analysis was repeated for each SNP, 191 of which significantly interacted with *DGATI* ( $P < 0.05$ ). The significant SNP were then included in a single analysis and backward eliminated until a group of 41 significant *DGATI* × SNP interactions remained. To evaluate whether consideration of the interactions improved genetic prediction, 5 data sets were created where all data from 2 to 3 herds was eliminated. Breeding values were subsequently estimated for those cows whose data were excluded. A sixth validation data set was created where data from the youngest cows was excluded. Four breeding values were estimated for each validation data set: EBVBASE (random animal effect only), EBVDGAT1 (EBVBASE plus *DGATI* effects), EBVSNP (EBVDGAT1 plus SNP effects), and EBVI (EBVSNP plus *DGATI* × SNP effects). Validation models were more significant when 305-d yield was regressed on EBVI ( $F = 37.41$ ) than on EBVSNP ( $F = 14.03$ ), EBVDGAT1 ( $F = 14.51$ ), or EBVBASE ( $F = 0.84$ ) for the young cow validation data. Validation models were also most significant when 305-d fat yield was regressed on EBVI in 4 of the 5 validation groups where data were excluded from specific herds. Similarly, the correlation between EBVI and EBV from a full model with all data included was highest in 4 of the 5 herd validation analyses (mean = 0.48, range = 0.29 to 0.64) and for the young cow evaluation (0.54), whereas the lowest was always EBVBASE (range = -0.10 to 0.27). Consideration of SNP interactions with *DGATI* may yield more accurate genomic predictions for fat yield.

**Key Words:** *DGATI*, fat

**231 Effectiveness of treating subclinical ketosis in dairy cows.** Albert J. Brown\*<sup>1</sup>, Maurice L. Eastridge<sup>1</sup>, Leon D. Weaver<sup>2</sup>, and K. J. Chapman<sup>2</sup>, <sup>1</sup>*The Ohio State University, Columbus, OH*, <sup>2</sup>*Bridge-water Dairy, Montpelier, OH.*

Ketosis is a major metabolic disorder of dairy cattle in the United States. There are various viewpoints on whether it is cost effective to treat subclinical cases of ketosis (SCK) with propylene glycol (PPG) and dextrose in comparison to treating animals that become clinically ketotic. For this trial, there was a control group and 2 treatment groups. Control cows had <1.2 mM/L β-hydroxybutyrate (BHBA) and did not receive treatment. Treatment 1 cows were deemed SCK, defined by a blood BHBA of 1.2 to 2.9 mM/L and received 250 mL 50% dextrose solution intravenously and 300 mL PPG orally for 3 d. Treatment 2 cows also were SCK (same criteria as Trt 1) but did not receive the PPG and dextrose. Cows with > 2.9 mM/L BHBA were not enrolled in the trial.

To determine treatment, blood was drawn from the tail vein/artery at 4 d in milk (DIM) and tested for BHBA using a Precision Xtra Meter (Abbott Laboratories, Abbott Park, IL). NEFA and BCS (1 = thin, 5 = fat) were recorded -14 to -3 d prepartum. Data were analyzed using the PROC GLM procedure with repeated measures of SAS (2012), with significance at  $P < 0.05$  and trends at  $P < 0.10$ . Prepartum NEFA were similar between Trt 1 and 2 but lower for control (316, 300, and 240 μEq/mL, respectively). BHBA at 4 DIM was similar for Trt 1 and 2 but lower for control (1.66, 1.69, and 0.70 mM/L, respectively), with a similar pattern at 11 DIM (1.34, 1.46, and 0.69 mM/L, respectively). BCS at 11 DIM was higher for Trt 1 cows (3.45) than control (3.33) and Trt 2 cows (3.31). Milk yield during the first 90 DIM was similar for control and Trt 1 cows (42.7 vs. 42.1 kg/d, respectively), but milk yield was lower for Trt 2 cows (41.2 kg/d) compared with control cows and tended ( $P = 0.06$ ) to be lower than for Trt 1 cows. Cows with SCK postpartum were showing signs of altered energy metabolism prepartum. Treatment for SCK had minimal effect on BHBA, appeared to increase BCS, and tended to increase milk yield. Additional data on health events and cost of treatments will provide further evidence to the effectiveness of treatment of SCK.

**Key Words:** ketosis, subclinical ketosis, propylene glycol

**232 Assessing the impact of bovine fecal contamination in water on health and management practices.** Sarah J. Thomsen\*<sup>1</sup>, Jillian F. Bohlen<sup>1</sup>, and J. Brooks Crozier<sup>2</sup>, <sup>1</sup>*University of Georgia, Athens, GA*, <sup>2</sup>*Roanoke College, Salem, VA.*

The objective of the experiment was to determine the survival and then relationship between *E. coli* O157:H7 and a bovine fecal marker in river water in a set of sequential experiments. Microcosms of river water and sediment were created in 500-mL sampling bottles. In the first module, *E. coli* O157:H7 was inoculated into water samples (125 mL sediment, 400 mL water); in another experimental setup, 1 g of dairy cattle manure was inoculated into the microcosms (150 mL sediment, 300 mL water). Both setups had control groups. Throughout a period of one week, samples were collected; on d 7, an initial sample was collected and all were shaken to represent a disturbance in the water and sampled again. All samples underwent DNA extraction and polymerase chain reaction, utilizing the specific markers *eae* gene in *E. coli* and bovine *Bacteroides* in the manure to indicate presence or absence. In both experiments, data supported presence of the desired markers after inoculation 24 h, but were absent by the end of the week. However, both experimental setups were also positive for the markers 2 out of 3 times in the samples when shaken after one week. This data supports the theory that markers remain present in the sediment even after one week. These data were then used to consider different management practices to better human and herd health, in addition to farm efficiency, with the hope to reduce fecal contamination into water sources. The goal of the experiment was to examine the survival of a harmful strain of *E. coli* and a bovine fecal marker. The bovine fecal marker was used as pathogenic fecal coliforms like *E. coli* can be transferred through bovine feces. This makes bovine feces not only a home but a mobile mechanism by which harmful and zoonotic pathogens may contaminate water supplies. These contaminations may be direct or by indirect sources such as runoff. These realities generate health concerns for both humans and animals with a need to minimize fecal deposition and access to water sources through best dairy management practices.

**Key Words:** *E. coli*, water, fecal contamination

# Animal Behavior and Well-Being Symposium: Novel and multidisciplinary approaches to animal welfare

**233 Opportunities and challenges of interdisciplinary approaches to quantifying welfare.** Peter D. Krawczel\*, *The University of Tennessee, Department of Animal Science, Knoxville, TN.*

There is a growing interest within the United States in ensuring management systems and strategies for animal agriculture are not only highly productive and cost efficient, but also provide for the welfare of animals raised within them. The benefit of this situation is the chance for an empirical approach to evaluating welfare across animal agriculture to make a valuable contribution to the public discourse. The overall objective of this presentation will be to address the challenges and opportunities to using interdisciplinary approaches to quantifying welfare. To narrow the scope of this paper, and provide a more cohesive narrative, examples from dairy production will be used to demonstrate the main concepts. The discussion of opportunities will focus on the interaction between traditional approaches to measuring welfare, such as behavior and productivity, and novel aspects of sleep, immune function, reproduction, or sociology. Sleep research on dairy cows provides a means to demonstrate how biologists, ethologists, and engineers can collaborate to redefine how the lying behavior of a dairy cow is assessed. Moving beyond ideas of immunosuppression and into the approaches to evaluate dysfunction of immune function may provide a more accurate assessment of the effect of a management strategy on a dairy cow or calf. End points commonly used to evaluate reproductive parameters, such as return to cyclicity in early lactation, could provide a means to evaluate the welfare of postpartum dairy cows. Finally, incorporation of sociologists provides a means to understand producers' attitudes toward disease, pain, housing, and other factors that can alter dairy cow welfare. The discussion of the challenges of engaging in a multidisciplinary approach will focus on pain mitigation and immune function. These areas will be used as examples of the difficulties that might be encountered when the collection of one response variable affects another.

**Key Words:** welfare assessment, multidisciplinary, dairy cow

**234 Animal welfare as a source of confounding and variation in science.** Amy L. Stanton\*, *University of Wisconsin-Madison, Madison, WI.*

It has been well established that chronic stress has a negative impact on the welfare of animals under our care. From the stereotypic behaviors observed in high numbers in zoo and laboratory animals to the decreased milk let-down observed in roughly handled dairy cows the effect of housing and handling practices have been demonstrated clearly by the animals in our care. Although the impact of rough handling and barren environments have been studied many of the more subtle effects that are unique to animal science, especially non-traditional laboratory species, have not been examined as closely. The objective for this paper is to review the effect of common data collection systems and environments on management, behavior, and welfare of agriculture research animals and through this introduce a source of variation. The 3 R's of ethical animal science—Replace, Reduce, and Refine—are used to minimize the impact of research on animals and to reduce the number of animals used in research. Animal behavior and the associated welfare consequences may be sources of variation in many research studies. As an example, to collect physiological measures it is often necessary for animals to be housed in systems that vary greatly from non-research animals. These different housing and handling methods are not studied as extensively

in animal agriculture and may be having more of a negative effect on welfare than expected. For this reason, it is important that we understand the effect that different research practices have on animals and accompanying research results. Many of our agriculture species are social animals that have evolved as a prey species. However, many studies require isolation and extensive human observation. Both of which alter animal behavior and welfare. Alternatively, measurement devices, such as feeding gates, may affect normal behavior. Differences in social dominance, prior experiences, and temperament can alter the ability of animals to learn to interact with these systems. For these reasons, an understanding of animal behavior is essential to minimize welfare effects and the accompanying variation in study results.

**Key Words:** welfare, behavior, research

**235 Interaction between coping style/personality, social stress, and disease risk.** J.M. Koolhaas\*, *University of Groningen, Groningen, the Netherlands.*

Ecological studies in feral populations of mice, fish and birds start to recognize the functional significance of phenotypes that differ individually in their behavioral and neuroendocrine response to environmental challenge. Within a species, the capacity to cope with environmental challenges largely determines the individual survival in the natural habitat. Recent studies indicate that the individual variation within a species may buffer the species for strong fluctuations in the natural habitat. A conceptual framework will be presented that is based on the view that the individual variation in aggressive behavior can be considered more generally as a variation in actively coping with environmental challenges. Highly aggressive individuals adopt a proactive coping style, whereas low levels of aggression indicate a passive or reactive style of coping. Similar coping styles have now been identified in a range of species including cattle, pigs and horses. They can be considered as trait characteristics that are stable over time and across situations. Evidence will be presented that a proactive coping style is best in a stable environment; these animals heavily rely on predictions. Reactive coping is more suited to variable environmental conditions because it is characterized by a continuous use of environmental input. Because the 2 extreme coping styles are adapted to different environmental conditions, there is differential stress vulnerability. Serious health problems may develop when coping fails. Social stress studies show that proactive individuals are resilient under stable environmental conditions but vulnerable when outcome expectancies are violated. Reactive individuals are in fact rather flexible and seem to adapt more easily to a changing environment. The health consequences of this interaction between individual coping style and social environment will be illustrated with examples from the cardiovascular system and the immune system. It will be argued that understanding animal welfare and the individual vulnerability to stress related disease requires a fundamental understanding of the functional individual variation as it occurs in nature and the underlying neurobiology and neuroendocrinology.

**Key Words:** coping style, welfare, individual differentiation

**236 Of nature and nurture: The role of genetics and environment in behavioral development.** T. Bas Rodenburg\*, *Behavioural Ecology Group, Wageningen University, Wageningen, the Netherlands.*

The behavioral characteristics of an individual are determined by its genes and by its physical and social environment. Not only the individual's early life and current environment is of importance, but also the environment of previous generations. Through epigenetic processes, stress in parents and even grandparents can translate in changes in behavioral and physical characteristics of the offspring. This can also result in an increase in damaging behavior, such as feather pecking in laying hens. We have recently shown that stressed flocks of laying hen parent stock lay smaller eggs and that the offspring that hatched from these eggs were more fearful and showed more severe feather pecking already at one week of age. This effect depended on the genetic background of the hens: it was much more pronounced in the white laying hens than in the brown birds. Apart from epigenetics, also maternal hormones excreted before egg-laying or during pregnancy have effects on behavioral development of the offspring. Recent studies even indicate that epigenetic and hormonal effects may go hand in hand, where parental

stress leads to changes in gene expression in genes involved in hormonal responses. The environment in which an individual is born and in which it spends its first weeks of life also has considerable impact on behavioral development. Absence or presence of maternal care has been shown to have strong effects in laying hens: we found that maternal care resulted in birds that were less fearful and developed less damaging behavior when they were adult. In these experiments, we also studied effects of genetic selection on low mortality in group housing. We found that effects of selection on low mortality and of maternal care were additive in most cases, with birds selected for low mortality and reared with a foster mother having the best performance. This illustrates that in our approaches to breed and rear animals for good performance in group housing systems, an approach where genetic selection is combined with improvement of rearing and housing conditions of both parent stock and offspring has the largest chance of success.

**Key Words:** behavioral development, genetic selection, early-life environment



## Animal Health: Swine health and transition cows

**237 Protected sodium butyrate may reduce *Salmonella* spp. excretion in contaminated fattening pig farms.** M. Puyalto\*<sup>1</sup>, R.C. Mainar-Jaime<sup>2</sup>, S. Andres-Barranco<sup>3</sup>, E. CREUS<sup>4</sup>, and J. J. Mallo<sup>1</sup>, <sup>1</sup>Norel S.A., Spain, <sup>2</sup>Department of Animal Pathology, University of Zaragoza, Zaragoza, Spain, <sup>3</sup>AgriFood Research and Technology Centre of Aragon, Spain, <sup>4</sup>Agrofestiic S.L., Spain.

The objective of this study was to assess the level of *Salmonella* shedding and exposure in a fattening unit when protected sodium butyrate was added to the diet of the pigs. The study was carried out in a commercial *Salmonella*-infected fattening unit (8 pens, 110 pigs). Feed with 70% sodium butyrate protected with vegetable fat (3 kg/t) was administered to animals from 4 randomly selected pens throughout the whole fattening period (4 mo) (TG). Pigs from the remaining 4 pens were fed with regular diet without additive (CG). Individual serum and fecal samples were collected at 30, 60, and 90 d of fattening period and at slaughter. Bacteriology on fecal samples was performed following the ISO 6579:2002 protocol. Serum samples were analyzed by means of an indirect ELISA and 3 cutoff values were used (OD%  $\geq 10$ ,  $\geq 20$  and  $\geq 40$ ). Chi-squared analyses were performed to compare microbiological and serological results between groups at different time periods. A difference was considered significant when the one-tail *P*-value was  $\leq 0.05$ . In addition, a repeated measures analysis was used to estimate differences in mean OD%, after taking into account sampling times and the interaction treatment  $\times$  time. The levels of shedding were significantly lower for TG when compared with CG for all samplings but the second one (60 d). No significant differences between groups were observed when cut-off values of OD%  $\geq 10$  or %OD  $\geq 20$  were used. However, when OD%  $\geq 40$  was considered, significant differences in seroprevalence were observed for the sampling just before slaughter (CG 89.6% vs. TG 48%). Overall, a lower mean OD% value was observed for samplings at 60 and 90 d, and at slaughter in the TG (71, 66, and 46%, respectively) compared with the CG (88, 87, and 83%, respectively). The results indicate that the use of protected sodium butyrate at 3 kg/t may reduce the shedding of *Salmonella* spp. under this farm conditions, and therefore the risk of contact of the animals with this pathogen, as suggested by serological results.

**Table 1 (Abstr. 237).** Number of positive *Salmonella* animals/no. of total animals (% positive *Salmonella*)

Treatment	30 d	60 d	90 d	Slaughter
CG	19/29 (65.5%)	5/21 (23.8%)	7/21 (33.3%)	36/48 (75%)
TG	12/28 (42.8%)	2/21 (9.5%)	1/22 (4.5%)	29/50 (58%)
<i>P</i> (one-tail)	0.04	0.2	0.01	0.05

**Key Words:** *Salmonella* spp., sodium butyrate protected

**238 Rapid cooling after acute hyperthermia alters intestinal morphology and may negatively affect pig health.** J. S. Johnson\*, A Sapkota, and D.C. Lay Jr., *USDA-ARS Livestock Behavior Research Unit, West Lafayette, IN.*

Heat stress (HS) reduces livestock welfare and productivity and can negatively affect pig health. The study objective was to determine the effects of 2 HS recovery methods (rapid vs. gradual cooling) on pig welfare after acute hyperthermia. In 4 repetitions, 36 barrows (88.7  $\pm$  1.6 kg BW) were exposed to thermoneutral conditions (TN; n = 3/rep;

19.5  $\pm$  0.1°C) for 6h, or HS (36.4  $\pm$  0.1°C) for 3h, followed by a 3h recovery period of rapid cooling (HSRC; n = 3/rep; immediate TN exposure and water dousing) or gradual cooling (HSGC; n = 3/rep; gradual decrease in HS room temperature to TN). To evaluate heat dissipation, a thermal circulation index was calculated using gastrointestinal tract ( $T_{GI}$ ), ambient ( $T_A$ ), and skin ( $T_{SKIN}$ ) temperatures collected every 15 min [TCI;  $(T_{SKIN} - T_A)/(T_{GI} - T_{SKIN})$ ]. In repetitions 1 and 2, blood was collected at 60, 180, 210 and 240 min for lipopolysaccharide (LPS) analysis, and then pigs were euthanized at 360 min and duodenum, ileum and colon tissue was collected to determine intestinal morphology. HS treatment reduced ( $P < 0.02$ ) TCI in HSRC (-0.69) and HSGC pigs (-0.64) compared with TN controls (1.31); however, during recovery HSRC reduced TCI ( $P < 0.01$ ; 53.5%) compared with HSGC and TN treatments. In the duodenum and ileum, HSRC and HSGC treatment reduced villus height ( $P < 0.01$ ; 29.4 and 21.1%, respectively) and increased crypt depth ( $P < 0.01$ ; 56.9 and 32.2%, respectively) and villus width ( $P < 0.01$ ; 30.5% and 32.5%, respectively) compared with TN controls. In addition, HSRC reduced duodenal and ileal villus height ( $P < 0.01$ ; 27.6 and 38.2%, respectively) and increased crypt depth ( $P < 0.01$ ; 43.8 and 35.2%, respectively) compared with HSGC pigs. While no colon crypt depth differences were observed for HSRC and HSGC pigs (156.8  $\mu$ m), crypt depth was reduced ( $P < 0.01$ ; 37.1%) in HSRC and HSGC pigs compared with TN controls. During HS, circulating LPS was similar for all treatments ( $P < 0.57$ ; 56.2 EU/mL); however, during recovery HSRC pigs had increased circulating LPS ( $P < 0.05$ ; 68.5 and 52.4%, respectively) compared with TN and HSGC pigs. In summary, rapid cooling after acute hyperthermia reduces heat dissipation capacity, damages intestinal tissue, and increases circulating LPS compared with gradual cooling.

**Key Words:** heat stress, pig, recovery

**239 Mycotoxin-contaminated diets affect immunity parameters of piglets.** Simone Schaumberger\*, Sabine Masching, and Ursula Hofstetter, *BiomIn Holding GmbH, Herzogenburg, Austria.*

Important aspects of more than one multi-mycotoxin in feed, are the synergistic and additive effects which may result in negative effects on immunity and liver health of animals. The aim of the feeding trial was to investigate the effects of a multi-mycotoxin contaminated diet on immune parameters and liver health in weanling piglets. Twenty-four female weanling piglets were randomly assigned to 2 groups with 12 piglets each. Piglets were assigned to a vaccination program including hog cholera, pseudorabies and foot and mouth disease. Naturally mycotoxin contaminated corn was used to prepare the feed. Groups were as following: negative control group (A) and a mycotoxin contaminated (ZEN 1183 ppb, DON 1740 ppb, FUM 988 ppb) feed group (B). Parameters evaluated included antibody titers for pseudorabies referred to as sample to negatives ratio (S/N 0.6 was defined as positive), plasma CD4+/CD8+, IL-2, TNF- $\alpha$ , IgA, IgG, IgM, total plasma protein (TP) and liver enzymes (ALT, AST). The ratio of S/N in group B was significantly higher compared with group A at d 42 ( $P < 0.05$ ). There were no significant differences of plasma CD4+/CD8+ among the 2 groups. The plasma IL-2 levels of group B were significantly lower than group A at d 14, 28 and 42 ( $P < 0.05$ ). The plasma TNF- $\alpha$  level of group B was significantly lower than group A at d 28 and 42 ( $P < 0.05$ ). The IgA, IgG levels of group B were significantly lower than group A at d 28 and 42 ( $P < 0.05$ ). There was no significant difference of IgM among the 2 groups. Plasma TP levels of group B were significantly

lower than group A at d 14, 28 and 42 ( $P < 0.05$ ). Liver enzymes AST and ALT of group B were significantly higher compared with group A at d 14, 28 and 42 ( $P < 0.05$ ). To conclude, the combination of mycotoxins can damage the liver and impair immune response in weanling piglets. The decreased antibody titer against pseudorabies and the increase of S/N ratio in the group fed with mycotoxin-contaminated diet, suggests animal's decreased protection against pseudorabies when mycotoxins are present. This points to the need for greater awareness of a multiple mycotoxin contamination in feed and the protection of animals against said toxins.

**Key Words:** mycotoxin, piglet, immunity

**240 Butyrate enhances disease resistance of piglets through up-regulated gene expression of endogenous host defense peptides.** Haitao Xiong\*, Bingxiu Guo, and Yizhen Wang, *College of Animal Sciences of Zhejiang University, Hangzhou, Zhejiang, China.*

Dietary substances can manipulate the expression of endogenous host defense peptides (HDPs), which may provide a promising strategy for disease control and prevention, especially for antibiotic-resistant infections. We hypothesized that butyrate can induce HDP expression, which likely contributes to the elimination of *Escherichia coli* O157:H7 in the intestine and reduces the severity of inflammation caused by *E. coli* O157:H7 challenge. Piglets treated with or without butyrate (2 g/kg of diet) 2 d before *E. coli* O157:H7 challenge was employed to investigate the relationship among porcine HDP expression, status of inflammation and *E. coli* O157:H7 load in feces. The effects of butyrate on HDP gene expression, antibacterial activity of 3D4/2 macrophages and IPEC-1 cells, and phagocytic capacity of 3D4/2 and polymorphonuclear (PMN) leukocytes in vitro were also examined. Data were represented as mean  $\pm$  SEM. One-way ANOVA or Student's *t*-test was used to compare differences between treatment groups,  $P < 0.05$  was considered statistically significant. Results showed that butyrate treatment (1) alleviated clinical symptoms of *E. coli* O157:H7-induced hemolytic uremic syndrome and severity of intestine inflammation, (2) significantly reduced *E. coli* O157:H7 load in feces 24 h after the last inoculation ( $P < 0.01$ ), (3) significantly upregulated multiple HDPs, such as pBD2, pBD3, PMAP23, PR-39, and PG1-5 ( $P < 0.05$ ), but not all HDPs, in vitro and in vivo, and (4) enhanced the antibacterial activity of macrophage 3D4/2 and IPEC-1 and the phagocytic capacity of 3D4/2 and PMN cells in vitro. Our findings indicate that butyrate enhances disease resistance, promotes clearance of *E. coli* O157:H7, and alleviates clinical symptoms of hemolytic uremic syndrome and inflammation partially by affecting endogenous HDP expression.

**Key Words:** butyrate, *E. coli* O157:H7, host defense

**241 The effect of antiseptic compounds on umbilical cord healing and infection rates in neonatal piglets from a commercial facility.** Amanda L. Robinson\*, Jessica D. Colpoys, Glenn D. Robinson, Elizabeth A. Hines, Leo L. Timms, Erika M. Edwards, Kenneth J. Stalder, Anna K. Johnson, and Howard D. Tyler, *Iowa State University, Ames, IA.*

Umbilical cord antiseptics are often not used in swine production systems. The objective of this study was to determine if treating the umbilical cord with antiseptics reduces infection and enhances healing within the first 48 h after birth in newborn piglets. A total of 421 mixed sex commercial piglets from a breed-to-wean sow farm were enrolled. Piglets were alternately assigned by birth order within a litter

to 4 treatment groups; (1) iodine (2%), (2) trisodium citrate (10%), (3) a dry dip created using an antibacterial peptide (nisin) mixed with talc, and (4) no treatment. All treatments were applied within 1 h of birth. At birth, stall conditions (wet/dry and clean/dirty) were evaluated on a 3-point scale (3 = most dirty or most wet and 1 = dry or clean). Prior to treatment, diameter of the umbilical cords (as an indicator of cord drying and healing) was determined using digital calipers. As a potential indicator of umbilical infections, surface temperature of the umbilical stump, along with a reference point at the midpoint of the sternum, was measured using a dual laser infrared thermometer. These measurements were repeated at  $24 \pm 1$  h of age and at 48 h of age. In addition, umbilical stump redness and swelling (indicators of infection) were evaluated visually at 24 and 48 h. All data were analyzed using mixed model methods. Models included the fixed effects of umbilical diameter at birth, sex (female or male), stall conditions and treatment. No treatment differences were noted between dips on change in diameter of the umbilical cord during the first 24 h ( $6.60 \pm 0.057$  mm at birth vs.  $3.25 \pm 0.072$  mm at 24 h). There was no difference in umbilical cord stump surface temperature, redness or swelling at 24 h or 48 h. Stall conditions at birth did not affect the change in umbilical diameter, surface temperature of the umbilical stump, or visual indications of infection. In conclusion, there was no benefit observed when applying an antiseptic treatment on piglet umbilical cords to improve healing or reduce the incidence of infections during the first 48 h of life.

**Key Words:** piglet, umbilical cord, antiseptic

**242 Porcine hepcidin protects piglet intestinal epithelial cells by aggregating *Escherichia coli* K88.** Huahua Du\*, Dan Liu, and Zhenshun Gan, *Zhejiang University, Hangzhou, China.*

Hepcidin is a liver-expressed iron-regulating hormone that also is an antimicrobial peptide. Though the iron regulatory function of porcine hepcidin (pHepc) has been extensively investigated, the studies on the relationship between pHepc and bacteria are limited. The aim of current study was to evaluate its antibacterial activity against *Escherichia coli* K88 (*E. coli* K88) and investigate the effect of pHepc on bacterial infection in vitro. The antibacterial activity against pathogen bacteria was evaluated via radial diffusion, colony forming count, transmission electron microscopy (TEM) and DNA binding assays. Invasion assay and immunofluorescence microscopy were employed to determine its effect on bacterial infection. The results showed that pHepc exerted an iron-independent bacteriostatic activity against *E. coli* K88 in a dose-dependent manner. pHepc-treated *E. coli* K88 exhibited longer cells and cytoplasm unevenly distributed. To determine whether pHepc inhibited bacterial infection, piglet intestinal epithelial cells were challenged with pHepc-treated or untreated *E. coli* K88. The ability of pHepc-treated bacteria to invade IPEC-1 epithelial cells was impaired. pHepc significantly reduced 33.5% amount of *E. coli* K88 which adhered or invaded to cells ( $P < 0.05$ ). Another human epithelial colorectal adenocarcinoma cell Caco-2 exhibited the same inhibition pattern by pHepc treatment ( $P < 0.05$ ). By scanning electron microscopy (SEM), pHepc-treated *E. coli* K88 was aggregated, and higher magnification revealed a net-like meshwork of fibrils emanating from the bacterial surface that entangled the bacteria. It suggested that pHepc could afford protection against infection of *E. coli* K88 by a novel aggregation strategy, which will contribute broadly to piglet innate immunity.

**Key Words:** porcine hepcidin, *E. coli* K88, aggregation

**243 Limited sun exposure increases 25(OH)D serum concentration and affects mRNA expression of the vitamin D enzymes in liver and kidney of growing pigs at high altitude.** Samanta R. Fensterseifer\*<sup>1</sup>, D. Enette Larson-Meyer<sup>2</sup>, Bennett C. Ingold<sup>2</sup>, Kathleen J. Austin<sup>1</sup>, Kacey C. Myers<sup>1</sup>, and Brenda M. Alexander<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Wyoming, Laramie, WY, <sup>2</sup>Department of Family and Consumer Sciences, University of Wyoming, Laramie, WY.

Traditional confinement production management limits exposure of pigs to sunlight. With vitamin D primarily obtained from the diet, producers risk suboptimal vitamin D status. Ten weaning pigs (67.6 ± 6.1 kg; 76d ± 11) were randomly divided into control (CON, n = 5, traditional swine confinement) or sun (SUN, n = 5, exposed to one hour of sunlight at solar noon for 2-weeks during the summer solstice and fall equinox) groups. Blood samples were collected via cranial cava venipuncture before and after the 2-week sunlight exposure to determine serum concentration of 25-hydroxyvitamin D [25(OH)D]. Following fall sun exposure, pigs were slaughtered and liver and kidney tissue collected and frozen. Expression (mRNA) of vitamin D binding protein (VDBP), vitamin D receptor (VDR) and the enzymes CYP2R1 and CYP27B1 were determined. Serum 25(OH)D concentration did not differ ( $P = 0.1$ ) before summer sunlight exposure (39.5 ± 2.6 ng/mL). Sunlight exposure increased ( $P = 0.001$ ) serum concentration of 25(OH)D by 54.5% in SUN pigs during summer exposure, with 25(OH)D 87.5% greater ( $P = 0.001$ ) compared with CON pigs (66.5 ± 4.8 vs 35.5 ± 2 ng/mL, respectively). Serum 25(OH)D concentration returned to CON values ( $P = 0.3$ ) by the fall exposure period (45.1 ± 2.1 ng/mL). Following fall sun exposure, serum 25(OH)D concentration was again increased ( $P < 0.001$ ) in SUN compared with CON pigs (80.8 ± 3.3 vs. 39.9 ± 3 ng/mL). CYP27B1 mRNA expression was decreased in kidney ( $P = 0.03$ ) but increased in the liver ( $P = 0.05$ ) of SUN pigs when compared with CON pigs. The relative mRNA expression of VDBP, VDR and CYP2R1 did not differ ( $P > 0.05$ ) among pigs. In conclusion, 2-week sunlight exposure at high altitude during the summer solstice and fall equinox increases serum concentration of 25(OH)D and influences expression of CYP27B1 in kidney and liver of sun exposed pigs. Limited sun exposure enhances vitamin D status in confinement-raised pigs and may improve health and production outcomes dependent on vitamin D.

**Key Words:** vitamin D, sun exposure, pig

**244 Relationship between left displacement of the abomasum and daily milk yield in high-producing Chilean dairy cows.** Pedro G. Melendez\*<sup>1</sup>, Catalina Romero<sup>2</sup>, Maria P. Marin<sup>2</sup>, Mario Duchens<sup>3</sup>, Patrick Pithua<sup>1</sup>, and Pablo J. Pinedo<sup>4,5</sup>, <sup>1</sup>University of Missouri-Columbia, Columbia, MO, <sup>2</sup>University Santo Tomas, Viña del Mar, Chile, <sup>3</sup>University of Chile, Santiago, Chile, <sup>4</sup>Texas A&M AgriLife Research, Amarillo, TX, <sup>5</sup>Texas A&M University System, College Station, TX.

Left displacement of abomasum (LDA) is a metabolic disease affecting dairy cows within the first 30 DIM. Dairy farms in central Chile, consisting of Holstein cows maintained under confinement, use advanced technology including computerized record keeping systems. These farms have well-established postpartum health monitoring programs and LDA is regularly diagnosed. However, due to a scarcity of immediate veterinary assistance, many cows with LDA are treated conservatively by administering oral fluids, anti-inflammatory drugs and/or antibiotics. The hypothesis was that cows with LDA treated conservatively would produce less milk than cows with LDA treated surgically. The objective was to compare daily milk production among cows with LDA that received a conservative treatment, had surgery (right omentopexy or

toggle suture) or were not affected (negative controls). Data between 2010 and 2012 from 4 high-producing dairies in Central Chile were analyzed. Cases (n = 146) were categorized as treated surgically (DAs, n = 72), treated with toggle suture (DA<sub>t</sub>, n = 16), or treated conservatively (DA<sub>c</sub>, n = 58). For each case, 2 healthy contemporary cows (controls) were selected at random and matched by farm, parity and DIM (±15 d; n = 289). A mixed model for repeated measures was developed for the analysis of daily milk yield up to 90 DIM with the corresponding covariance structure based upon the best goodness of fit criteria. The mean and median DIM for LDA diagnosis was 13 and 9 d, respectively. Average daily milk up to 90 DIM was 43.0 kg/d for controls, 30.4 kg/d for DAs, 30 kg/d for DA<sub>t</sub>, and 14.2 kg/d for DA<sub>c</sub>. Controls produced more milk than DAs, DA<sub>t</sub>, and DA<sub>c</sub> group ( $P \leq 0.01$ ). DAs and DA<sub>t</sub> produced a similar amount of milk ( $P > 0.05$ ), but more milk than the DA<sub>c</sub> group ( $P \leq 0.01$ ). It is concluded that cows with LDA receiving conservative treatment produced substantially less milk than control cows (30 kg/d) and cows with LDA treated with surgery or toggle suture (16 kg/d). Furthermore, control cows produced more milk than cows with LDA treated with surgery or toggle suture (13 kg/d). In light of these results, it is recommended that all cows developing LDA should be treated surgically or with toggle suture.

**Key Words:** displaced abomasum, treatment

**245 Comparison of hematological parameters in dairy cows during periparturient period.** Samir Kumar Dash\*<sup>1</sup>, Hemant Dasharath Kadam<sup>1</sup>, Asmita Anant Kulkarni<sup>1</sup>, Mohua Das Gupta<sup>1</sup>, Narayan Laxman Phadke<sup>1</sup>, and Arun P. Phatak<sup>2</sup>, <sup>1</sup>BAIF Development Research Foundation, Central Research Station, Uruli Kanchan, Pune, Maharashtra, India, <sup>2</sup>Waterford, CA.

The periparturient period ranging from 3 wk before calving to 3 wk after calving presents enormous physiological challenges for the dairy cows. It is a period that is affected by metabolic stressors, major changes in endocrine status and altered immune function which together results in an increased risk of diseases including hypocalcaemia, ketosis, hepatic lipidosis, laminitis, mastitis, retained placenta and metritis. The present investigation was undertaken in Central Cattle Breeding Farm, Central Research Station, BAIF, India to study the hematological changes in puerperal period in 30 cows of HF pure, JE pure and their crosses under tropical conditions to compare the Hgb, TLC and CBC/diff. count 10 d before and 10 d after parturition to assess the extent of metabolic changes, infection and stress. The results were analyzed using ANOVA. The study revealed a nonsignificant difference and decrease in the Hgb count from 11.41 ± 0.30 to 10.59 ± 0.30 which may be due to the increased blood supply to fetus and milk adipose tissues with hemorrhagic loss during calving. The TLC count displays a significant ( $P < 0.05$ ) increase from 8.57 ± 0.39 to 10.00 ± 0.58 in postpartum period confirming the reports of previous studies which states that there is a decrease in immunity level due to stress and rise in infection. Neutrophil count increased from 47.03 ± 1.77 to 51.27 ± 2.14 nonsignificantly as the infection in postpartum period may rise. Eosinophil count, Lymphocyte and monocyte level did not reveal any significant changes. It can be concluded that due to lower hemoglobin, hemorrhagic stress and lower immunity due to rise in infection, the animal's health and production are always under threat; hence careful attention is required during the periparturient period.

*Contd.*



**Table 1 (Abstr. 245).** Blood parameters in Holstein cows 3 wk pre- and post-calving

Parameter	Hgb (g %)	TLC ( $\times 10^3$ )	Neutrophil (%)	Eosinophil (%)	Lymphocyte (%)	Monocyte (%)
Pre	11.41 $\pm 0.30$	8.57 $\pm 0.39$	47.03 $\pm 1.77$	6.07 $\pm 0.43$	44.03 $\pm 1.86$	2.73 $\pm 0.17$
Post	10.59 $\pm 0.30$	10.00 $\pm 0.58$	51.27 $\pm 2.14$	5.57 $\pm 0.42$	40.70 $\pm 2.18$	2.53 $\pm 0.20$
“F”	0.0572	0.0454	0.1299	0.5808	0.2494	0.5318

**Key Words:** tropical, puerperal, immunity

**246 Characterizing the effect of feed restriction on biomarkers of leaky gut.** Sara K. Stoakes<sup>\*1</sup>, Erin A. Nolan<sup>1</sup>, David J. Valko<sup>1</sup>, Mohannad Abuajamieh<sup>1</sup>, Jake Seibert<sup>1</sup>, Maria V. Sanz Fernandez<sup>1</sup>, Patrick J. Gorden<sup>1</sup>, Howard B. Green<sup>2</sup>, Katie M. Schoenberg<sup>2</sup>, William E. Trout<sup>2</sup>, and Lance H. Baumgard<sup>1</sup>, <sup>1</sup>Iowa State University, Ames, IA, <sup>2</sup>Elanco Animal Health, Indianapolis, IN.

Study objectives were to determine the magnitude of feed restriction (FR), which causes “leaky gut” and characterize the temporal consequences on production and blood variables. Twenty-three lactating Holstein cows (157  $\pm$  46 DIM; 713  $\pm$  54 kg BW; parity 2–4) were enrolled in 2 experimental periods. Period 1 lasted 5 d (ad libitum feed intake) and served as baseline for period 2 (P2), which lasted 5 d during which cows received one of 5 dietary treatments: 100% of ad libitum feed intake (AL; n = 3), 80% of ad libitum feed intake (AL80; n = 5), 60% of ad libitum feed intake (AL60; n = 5), 40% of ad libitum feed intake (AL40; n = 5), or 20% of ad libitum feed intake (AL20; n = 5). As the magnitude of FR increased, milk yield, MUN, and milk lactose content decreased linearly ( $P < 0.01$ ) whereas milk fat content linearly increased ( $P = 0.02$ ). Heart rate declined linearly with increasing FR ( $P < 0.02$ ) while respiration rate declined similarly for all FR treatments ( $P = 0.02$ ). Body weight loss increased linearly with increased FR ( $P < 0.01$ ). Both plasma insulin and BUN decreased, whereas NEFA increased linearly with greater FR ( $P < 0.01$ ). Lipopolysaccharide binding protein, haptoglobin, and serum amyloid A tended to increase linearly with increasing FR ( $P = 0.09$ ,  $P = 0.07$ ,  $P = 0.10$ , respectively). Circulating lymphocytes increased with increasing FR ( $P = 0.04$ ). AL40 and AL-fed cows were killed to determine the effect of FR on intestinal histology. Jejunum villus width, jejunum goblet cell area, ileum height and ileum crypt depth were or tended to be reduced ( $P = 0.03$ , 36%;  $P = 0.02$ , 52%;  $P = 0.06$ , 22%;  $P = 0.03$ , 28%, respectively) in AL40 cows compared with AL controls. Liver weight tended to be decreased (15%;  $P = 0.07$ ) in AL40 compared with AL cows. Liver fat and moisture percentages did not differ between treatments. In summary, FR tended to increase circulating acute phase proteins, which we speculate is due to an increase in leaky gut as demonstrated by the deterioration in intestinal architecture. Consequently, FR appears to be a simple and viable model to study intestinal integrity and barrier function.

**Key Words:** lipopolysaccharide, intestinal integrity, feed restriction

**247 Neutrophil function is dysregulated over the transition period.** Mallory A. Crookenden<sup>\*1</sup>, Caroline G. Walker<sup>1</sup>, Axel Heiser<sup>6,5</sup>, Juan J. Looor<sup>2</sup>, Kasey M. Moyes<sup>3</sup>, Jane K. Kay<sup>1</sup>, Susanne Meier<sup>1</sup>, Alan Murray<sup>5</sup>, Venkata S. R. Dukkipati<sup>5</sup>, Murray Mitchell<sup>4</sup>, and John R. Roche<sup>1</sup>, <sup>1</sup>DairyNZ, Hamilton, New Zealand, <sup>2</sup>University of Illinois, Champaign-Urbana, IL, <sup>3</sup>University of Maryland, College Park, MD, <sup>4</sup>University of Queensland, Brisbane, Queensland, Australia, <sup>5</sup>Massey University, Palmerston North, New Zealand, <sup>6</sup>AgResearch, Palmerston North, New Zealand.

There is a high incidence of infectious and metabolic disease in the transition period. During this time, there is a period of observed immunosuppression that appears to be ‘natural’. The effect of parturition on neutrophil function is of particular interest; we hypothesized that neutrophil dysfunction occurs around the transition period, as evidenced by altered gene expression. To determine this, we extracted neutrophils from peripheral blood of 45 cows at 5 time points: 1 wk pre-calving (–1 wk), day of parturition (d 0), and post-calving at wk 1, 2, and 4. Key pathways of neutrophil function were investigated by qRT-PCR. The ‘bovine immune panel’ (BIP) included 96 targets for detection with Fluidigm 96.96 integrated fluidic circuits and the Biomark HD system. Data were analyzed using SAS 9.3; the effect of time was analyzed using a mixed model approach to repeated measures ANOVA and Tukey’s *t*-test for pairwise comparisons between weeks. Data were submitted for pathway enrichment analysis using the protein analysis through evolutionary relationships tool. The top 3 pathways enriched from gene targets on the BIP included inflammation signaling (14-fold enrichment,  $P < 0.001$ ), apoptosis signaling (19-fold enrichment,  $P < 0.001$ ), and interleukin/cytokine signaling (18-fold enrichment,  $P < 0.001$ ). Biological processes enriched at –1 wk, d 0, and 1 wk post-calving included the immune system process, response to stimulus, and the immune response. The immune response process was most highly enriched (8-fold enrichment,  $P < 0.05$ ) at 1 wk post-calving, which coincided with greatest NEFA concentration (1.08  $\pm$  0.52) and, therefore, greatest NEB. No biological processes were enriched at 2 wk or 4 wk post-calving. Results indicate that the gene expression profile of neutrophils is altered over the transition period. This provides insight into neutrophil activity around parturition and the increased susceptibility to disease during this time.

**Key Words:** neutrophil, transition period, dairy cow

**248 The effects of grain-induced subacute ruminal acidosis on blood plasma proteomic characterization in dairy cows.** S. C. Li<sup>\*1</sup>, A. M. Danscher<sup>2</sup>, P. Azevedo<sup>1</sup>, P. H. Andersen<sup>3</sup>, P. Ezzati<sup>4</sup>, J. A. Wilkins<sup>4</sup>, E. Khafipour<sup>1</sup>, and J. C. Plaizier<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Manitoba, Winnipeg, Manitoba, Canada, <sup>2</sup>Department of Large Animal Sciences, University of Copenhagen, Copenhagen, Denmark, <sup>3</sup>Department of Clinical Sciences, Swedish University of Agricultural Sciences, Uppsala, Sweden, <sup>4</sup>Manitoba Centre for Proteomics & Systems Biology, Winnipeg, Manitoba, Canada.

Subacute ruminal acidosis (SARA) resulting from excessive grain feeding to dairy cows is accompanied by an immune response and has been associated with impaired liver function and blood composition. We, therefore, investigated the proteome in blood plasma to discover potential biomarkers for SARA. Four lactating Danish Holstein cows were used in a study that included a 4-week control period followed by a one-week SARA challenge period. During the control period, all cows received a total mixed ration (174 g crude protein/kg DM; 192 g starch/kg DM; 6.28 MJ NE<sub>L</sub>/kg DM) ad libitum. During the SARA period, 40% of the ration was gradually substituted with grain pellets (50:50 wheat:barley) within 3 d to induce SARA. This SARA induction diet was then fed for another 4 d. Rumen pH was monitored with indwelling pH probes to verify that cows had rumen pH lower than 5.6 for 3 h daily during the SARA induction. Blood samples were obtained from the jugular vein at 7 h post-feeding on d 4 before SARA induction and on the second of 4 SARA-induction days. The blood plasma samples were then processed for the enrichment of low molecular weight serum proteins using acetonitrile precipitation before mass spectrometry-based proteomic analysis on a nano-RPLC-MS/MS system. The analysis

identified 14 high-confidence proteins with significant quantitative differences between control and SARA periods as defined by MS intensity, of which 9 were potential differentially expressed (PDE) proteins LASP1, TMSB4, and the precursors of KNG1, APOE, SERPINA3–8, CRP, SERPINF2, SERPIND1 and CATHL4 reportedly involved in acute phase response, cytoskeletal proteins, blood coagulation, cholesterol transport, liver disease, and innate immune defense molecules. Among these PDE proteins, LASP1 and the precursors of APOE, SERPINA3–8,

SERPINF2, SERPIND1 and CATHL4 were upregulated and others were downregulated ( $P < 0.05$ ). These results suggest that it may be possible to identify proteins that are differentially expressed during SARA. Studies are ongoing to validate the possible candidates and identify additional proteins for the understanding of ethological concepts of SARA.

**Key Words:** dairy cow, SARA, plasma proteome

# Animal Health Symposium: Understanding and reducing the impact of various stressors on immune responses and health of cattle

**249 Understanding what stresses a dairy cow and the effect on immunity.** Michael A. Ballou\*, *Texas Tech University, Department of Animal and Food Sciences, Lubbock, TX.*

Dairy cows experience stress at many times in their life and these stressful events increase the risk for negative health and productive outcomes. A better understanding of what causes stress and the impacts on the immune system will aid in developing management strategies that either reduce stress or attenuate the risk for disease. Cows are creatures of habit and this consistency gives her control. Take away this control and the more stressed she becomes. Stress generally suppresses many inflammatory leukocyte, neutrophil, and lymphocyte responses that create holes in the immune system, which microorganisms capitalize on to cause disease. An early lactating dairy cow is a great example of homeostasis, whereas she is able to adapt to a new physiological state and environmental conditions. However, when that change is abrupt and/or dramatic, such as parturition and initiation of lactation, the cow can become stressed because the adaptation lags. The metabolic demands during the initiation of lactation play a major role in the dysfunctional immune system, and the effects are likely both direct and indirect. Direct effects are considered if nutrient demands of leukocyte responses are not met / prioritized or an increase in a metabolite, such as NEFA or BHBA, inhibits leukocyte function. Another example of a direct effect would be the negative effect of hypocalcemia on neutrophil function. The indirect effects are considered the physiological adaptations that need to occur in the cow that causes stress because they are a significant deviation and happen abruptly. Since some of the stress during the transition period is unavoidable, the focus should be to limit microbial exposure and any additional stressors that may have additional negative effects on immunity. Some factors that can contribute to additional stress include: high stocking density, poor cow comfort, dirty environment, comingling cows and heifers, and poor feed and bunk management. It is called the transition period because many changes are occurring and this increases the risk for stress. The stressors are multifactorial; therefore, a systematic approach to improving transition cow health is required.

**Key Words:** dairy, health, stress

**250 Stress, immunity, and management of calves.** Lindsey E. Hulbert\* and Sonia J. Moisés, *Department of Animal Sciences and Industry, Kansas State University, Manhattan, KS.*

Despite many advances in management and housing of dairy calves, 1 in 10 US dairy heifers die before weaning. A better understanding of the internal and external stimuli that contribute to the calf's physiological and behavioral responses to stressors is needed to reduce the risk of morbidity and mortality. Feeding calves their first meal is crucial, as successful passive transfer reduces mortality by 60%. There appears to be sexually dimorphic immune and stress responses in young cattle, but more research is needed to determine if this is caused by human-bias for female calves. After that first feeding, 1 in 10 heifers and most bull-calves in the US are transported to specialized calf-raising facilities, yet there is a lack of information of the newborn calf stress response during transit and handling. Whether calves are raised off-site or at a calf-ranch, individual housing systems are commonly used in the US to reduce the risk of pathogen exposure and provide individual feeding and healthcare. However, there may be health, growth, and social benefits for calves in alternative systems that have increased space-allowance

or group housing. Disbudding and castration are typically performed at an early age for dairy calves, during the pre-wean stage. These stressors often take place when the calf has decreased passive-transfer Ig and immunity is developing. There is limited availability of pain-mitigation through anesthetics and analgesics, but there is evidence that analgesics attenuate suppressed leukocyte function during these procedures. Milk replacer (MR) and milk quality may alter immunity. Solid-feed intake is a primary measure for determining "weaning-readiness," but some MR formulas may influence the calf's oral behaviors before weaning, therefore alternate weaning-methods may need to coincide with alternate MR formulas. The calf's behavioral and stress response at weaning may influence its immunity during the transition from individual to group housing (comingling). Alternate comingling strategies and nutritional supplements may help with this transition, but more research is needed to explore feasible alternatives. Optimizing the calf's health and well-being at these early-stages may improve its long-term health and behavioral strategies.

**Key Words:** calves, stress, immunity

**251 Effects of late-gestation heat stress on immunity and performance of calves.** Geoffrey Dahl<sup>1</sup>\*, Ana Monteiro<sup>2</sup>, and Sha Tao<sup>2</sup>, <sup>1</sup>*University of Florida, Gainesville, FL,* <sup>2</sup>*University of Georgia, Tifton, GA.*

Heat stress effects during lactation are well characterized and include reductions in dry matter intake, milk yield and metabolic shifts that reduce the efficiency of milk production. Similarly, when dry cows are heat stressed they experience lower intake, reduced mammary growth and compromised immune function that ultimately results in a poorer transition into lactation and lower milk yield. Recently, we have focused on the effects of late gestation heat stress on calf survival and performance, with a series of studies examining preweaning growth and health, and later reproductive and productive responses, in an attempt to quantify acute and persistent effects of in utero stress. Calves born to dams heat stressed when dry have lower body weight at birth, are shorter at weaning, and do not achieve the same level of weight or height accumulation to 12 mo of age observed in calves from dams that are cooled when dry. Some of the reduced growth may result from the lower immune status observed in calves heat stressed in utero, which begins with poorer apparent efficiency of immunoglobulin absorption and extends to lower survival rates through puberty. However, heat stressed calves also have permanent shifts in metabolism that may lead to greater peripheral accumulation of energy and less lean growth relative to those from cooled dams. Comparing reproductive performance in calves heat stressed versus those cooled in utero, we observe that the cooled heifers require fewer services to attain pregnancy and become pregnant at an earlier age. Tracking the milk production in calves that were heat stressed in utero versus those cooled in late gestation revealed a significant reduction of yield in the first lactation, approximately 5 kg/d through 35 weeks of lactation, despite similar bodyweight and condition score at calving. These observations indicate that a relatively brief period of heat stress in late gestation dramatically alters the health, growth, and ultimate performance of dairy calves. Thus, it is critical to effectively manage heat stress of dry cows to avoid negative effects on the calf.

**Key Words:** in utero heat stress, growth, health



**252 Social stressors and their effects on immunity and health of periparturient dairy cows.** Ricardo C. Chebel\*<sup>1,2</sup>, Paula R. B. Silva<sup>2</sup>, Karen Luchterhand<sup>2</sup>, and Marcia Endres<sup>2</sup>, <sup>1</sup>*University of Florida, Gainesville, FL*, <sup>2</sup>*University of Minnesota, St. Paul, MN*.

Management practices during the periparturient period have been the focus of much research recently because during this period immune function, metabolism and health of cows are severely challenged. Responses to stress are often classified as behavioral, immunological, neuroendocrine, and autonomic. In production systems, understanding all facets of stress response is important to correctly predict the consequences of stressors to the health and performance of animals and to prevent costly managerial changes that have minimal impact on animal well-being and performance. Common stressors faced by periparturient animals are: regrouping, stocking density, and, for nulliparous animals, commingling with parous animals. In conventional dairies, feeding strategies during the periparturient period often require several group changes during the most challenging period of an animal's life. Traditional weekly regrouping of prepartum cows increases competitive behavior at the feed bunk but it does not affect innate and adaptive immunity, metabolic parameters, health and production, as long as stocking density is not overwhelming and nulliparous and parous animals are housed separately. Stocking density of prepartum animals is often overlooked because these are non-productive animals. Although severe overstocking (200% of feeding space) of commingled nulliparous and parous pregnant animals produces important neuroendocrine and metabolic changes, when prepartum nulliparous and parous animals are housed separately, stocking densities of up to 100% of feed space do not seem to affect innate and adaptive immunity, metabolic parameters, and performance. In recent experiments, when animals were ranked based on feed bunk displacement, submissive animals were more likely to be diagnosed with metritis than dominant animals despite not presenting significant differences in metabolic parameters. With the advent of new technologies that monitor rumination, activity, and lying behavior, it may be possible to more easily identify submissive animals and create strategies to prevent diseases.

**253 Metabolic and physiological stressors during the periparturient period and effects on immunity and health of dairy cows.** José E. P. Santos\*, Eduardo S. Ribeiro, and Natalia Martinez, *University of Florida, Gainesville, FL*.

During early postpartum, high-producing dairy cows undergo a period of extensive tissue catabolism because of negative nutrient balance.

Homeostatic controls assure that nutrients are partitioned to favor lactation at the same time that homeostasis secures survival. However, unrestrained metabolic disturbances often lead to diseases which, in turn, dramatically decrease both productive and reproductive performance. In early lactation, dairy cows are more susceptible to diseases, particularly those that affect the uterus, such as metritis, and the mammary gland, such as mastitis. It is thought that calving and the increase in nutrient demands with the onset of lactation affect the immune system causing a temporary dysregulation in immune function. Humoral and cellular immunity are usually depressed at the same time that inflammation is enhanced with parturition. Negative nutrient balance has been associated with compromised immune function in dairy cows, and low concentrations of glucose, insulin, and insulin-like growth factor (IGF)-1 associated with elevated concentrations of nonesterified fatty acids and ketone bodies can have disruptive and detrimental effects on immune cells. Reductions in blood concentrations of Ca and antioxidant vitamins that typically occur around parturition are also linked with impaired immune competence and increased risk of uterine diseases. Nevertheless, experimentally-induced negative nutrient balance alone have minor effects on the leukocyte function, does not seem to affect the clinical symptoms associated with an intramammary endotoxin infusion, and have minor effects on immunocompetence of cows challenged to develop mastitis. The disagreement between experimental models that use nutrient restriction to study negative nutrient balance and immunity and the observations of immunosuppression in periparturient dairy cows suggests that changes in immune function are complex; they likely involve nutritional imbalances associated with the physiological and endocrine state of the cow that leads to the dysregulation of the immune system and increased risk of diseases.

**Key Words:** dairy cow, health, immunity

# Bovine tuberculosis (TB) and paratuberculosis (Johne's disease) Symposium: What we know and what we need to know

**254 A three-year study of bovine tuberculosis in an enzootic area, the Nile Delta.** Adel M. Talaat\*<sup>1</sup>, Hazem A. Abdelaal<sup>1</sup>, and Essam A. Nasr<sup>2</sup>, <sup>1</sup>University of Wisconsin-Madison, Madison, WI, <sup>2</sup>Veterinary Serum and Vaccine Research Institute, Cairo, Egypt.

Bovine tuberculosis (BTB) is a chronic infectious disease characterized by the formation of granulomatous lesions in organs, mainly lungs and lymph nodes. BTB is caused by slowly growing bacilli, mainly *M. bovis*. In developed countries, BTB in animals is significantly in decline with rates that reached down to <0.001% in cattle herds in the United States. In developing countries, BTB represent a major problem with a prevalence that could reach up to 10 to 15% of cattle herds in some parts of Africa. In Egypt, BTB transmission among animals and from animals to humans represents a major problem because of the complexity of animal husbandry and animal trade. In this study, our teams in both the United States and Egypt used the single intradermal comparative skin test (SICST) to examine the herd level prevalence of BTB in 5 different regions within the Nile Delta of Egypt. During the years of 2012–2015, several farms were visited where SICST was conducted on all animals in the herd alongside collecting all of the historical background of disease prevalence in these farms. As expected, farms with a known history of risky management behavior (e.g., introduction of new animals without testing, or not testing at all for BTB) were associated with high level of BTB prevalence that reached up to 45% in some herds. Interestingly, clinical health condition of animals and milk yield were not associated with the BTB herd-level prevalence. However, where BTB was enzootic, the highest percentage of SICST-positive animals (average 14%) were among the 6–12 mo of age while newly testing farms identified the highest percentage of SICST positive (60%) in animals above 24 mo of age. This result highlights the importance of periodical testing and intervention to control BTB in dairy herds. Overall, the prevalence of BTB among total number of 2799 examined animals was 7.3% during the 3 years screening project. Recommendation to reduce the BTB in these herds included the frequent testing of animals using SICST, especially before restocking of a dairy herd.

**Key Words:** bovine tuberculosis, prevalence, enzootology

**255 Host genomics—What have we learned?** Holly L. Neibergs\*, Washington State University, Pullman, WA.

The study of the genomic variation in cattle has been undertaken to identify loci that allow the prediction of cattle susceptible to *Mycobacterium avium* ssp. *paratuberculosis* (MAP) infection. The application of this information is to assist in disease prevention through selective breeding of cattle that are less likely to become infected should MAP exposure occur. A genetic predisposition to MAP infection in cattle was established through differences in prevalence of MAP infection found across breeds and across sires. Heritability estimates for susceptibility to MAP infection have been computed from family and genomic studies and typically range from 0.01 to 0.18, although higher estimates have been reported. Candidate gene and genome wide association studies have both been used to identify the loci associated with MAP infection. Phenotypes of susceptible and non-susceptible cattle have been based on serum ELISA, milk ELISA, fecal culture, tissue culture, PCR of tissue or feces or a combination of these diagnostic methods. The disparity in the sensitivity of the diagnostic methods affects the reliability of the controls, and the stage of MAP infection at which the animal is diag-

nosed. These differences make comparisons across studies difficult, as genomic studies are sensitive to phenotypic differences. Consequently, few studies have identified the same susceptibility loci in independent cattle populations. This lack of agreement across studies is most likely due to small sample sizes (most studies have less than 500 cases and 500 controls), the lack of consistency in the definition of phenotypes, and the polygenic nature of the disease which could result in different genes having a different level of effect within and across breeds. Identifying loci associated with MAP infection offers a permanent preventative strategy to lower the prevalence of bovine paratuberculosis and to have a better understanding of the disease etiology. To obtain concordant results, larger studies with a consistent phenotype are needed and must be followed with independent validation and functional studies. This will allow genomic prediction to deliver its translational promise to assist in preventing MAP infection in cattle.

**Key Words:** host genomics, bovine paratuberculosis

**256 Johne's disease and bovine tuberculosis: Updates on control and prevention.** Scott J. Wells\*<sup>1</sup>, Kimberley L. VanderWaal<sup>1</sup>, Catalina Picasso<sup>1</sup>, Eva A. Enns<sup>1</sup>, Meggan Craft<sup>1</sup>, Federico Fernandez<sup>2</sup>, Andres Gil<sup>3</sup>, Julio Alvarez<sup>1</sup>, Lucas Dominguez<sup>1</sup>, and Andres Perez<sup>1</sup>, <sup>1</sup>University of Minnesota, St Paul, MN, <sup>2</sup>Ministerio de Ganadería, Agricultura y Pesca, Montevideo, Uruguay, <sup>3</sup>Facultad de Veterinaria, Universidad de la Republica, Montevideo, Uruguay.

While improved understanding of within-herd transmission of *Mycobacterium avium* ssp. *paratuberculosis* through conduct of longitudinal herd control studies has led to identification of effective long-term herd control measures, between-herd transmission continues unabated in many cattle herds through movements. Cattle movements serve as one important contributor to transmission of *Mycobacterium bovis* between herds, pointing to the need to better understand the role of cattle movements in disease transmission. Although complete cattle movement and traceability information is not available in the United States, the origin and destination farms for cattle movements in some countries, such as Uruguay and Spain, are captured in official cattle traceability systems. This information can be critical to understanding the risk of disease spread in the endemic cattle populations, which could be adapted to improve understanding of transmission in US cattle. In Uruguay, while the prevalence of *M. bovis* in cattle has been low since implementation of an active surveillance program, multiple outbreaks were reported during 2011–2013. In our study, Uruguayan cattle movements were evaluated using network analysis to understand cattle flows and identify high-risk farms for surveillance and control. Farms represented nodes in the network, which were linked based on the frequency and number of cattle moved. The distribution of in-movements and out-movements from farms was severely right-skewed, and the 10% most highly connected farms accounted for 72–83% of cattle moved each year. In comparison, during the time when bovine tuberculosis was identified in Minnesota (2008 to 2011), 20% of farms performed 90% of the movements. This heterogeneity in movement patterns indicates that some farms are likely critical for disease spread. In Uruguay, farms that experienced outbreaks of *M. bovis* during this period engaged in substantially more movements than the population average. Results from these studies highlight the importance of collecting livestock movement data, and of integrating

and coordinating actions for infectious diseases to understand the vulnerability of livestock trade networks to infectious diseases.

**Key Words:** bovine tuberculosis, cattle, movement

**257 MDA outreach: Communicating for a change.** Julia M. Smith\*, *University of Vermont, Burlington, VT.*

If a veterinary practitioner has tried to persuade a client to take action and nothing happens, is the problem that the client did not listen well or that the practitioner did not communicate well? An understanding of a few communication and behavior change models can enhance one's ability to make change a reality. Key principles of risk communication and social marketing of behavior will be discussed in the context of controlling mycobacterial diseases of animals (MDA). Risk communication can be understood as the process of sharing information and opinion about something hazardous, where uncertainty is a characteristic feature of the risk. When addressing Johne's disease or bovine tuberculosis, the disease is the hazard and uncertainty is associated with disease progression, detection, and success of control strategies. An approach known as message mapping can be a useful tool when developing risk communication messages. According to Prochaska's stages of change model, the process of converting a decision-maker into a change-maker takes time and involves distinct changes in attitude toward doing something new or different. To have the most effective conversations, it is important to identify at what stage of change the decision maker is and to tailor one's key points accordingly. Selling someone on behavior change to prevent or control a disease is a lot like selling someone on buying the latest gadget. The Academy for Educational Development has developed a process for marketing behavior change using the BEHAVE framework. This framework involves asking a few more questions before developing a strategy designed to achieve a behavior change. The strategy guides the development of a marketing plan. An essential element of a BEHAVE-based marketing plan is prototyping and pre-testing messages and aspects of the plan. When practicing risk communication and social marketing of behavior, it is essential to actively listen to the client or potential change-maker. Converting a decision-maker into a change-maker makes all the difference in controlling diseases such as *Mycobacterium avium* ssp. *paratuberculosis* and *Mycobacterium bovis*.

**Key Words:** risk communication, stages of change, social marketing of behavior

**258 Zoonotic potential of bTB and MAP—Nothing to worry about...right?** Tim Bull\*, *Institute of Infection and Immunity, St George's University of London, London, UK.*

The threat of mycobacterial zoonotic transmission (particularly bTB) has driven expensive eradication efforts and large culling projects. In the absence of reliable vaccines however, the cost and complexity of programs needed to secure eradication from domestic herds and wildlife maintenance reservoirs has had limited progress. Few have succeeded in eradication and some arguably increased transmission rates. As a result bTB and MAP remain prevalent in domestic animals and wildlife reservoirs. Given the opportunity and a favorable host susceptibility these mycobacteria can cause disease in humans. Acute bTB infection is a potentially life threatening disease clinically indistinguishable from tuberculosis. Pasteurization remains an effective defense reflected by the fact that most human bTB cases are either a result of direct animal contact or re-activation from an older latent infection. For MAP, the issue is less clear cut. Despite significant indications that MAP invades, is present and persistent in many humans and has the capacity in susceptible individuals to trigger inflammatory bowel conditions such as Crohn's disease, the role of MAP as a direct human pathogen is still not widely accepted. Worryingly MAP survives pasteurization and prevalence is such that viable MAP is now significantly present in retail and dried infant formula milk. If MAP is truly a human pathogen, current ineffectual MAP control methods are permitting an extensive, unchecked, worldwide, chronic exposure to humans, some of whom are susceptible to developing disease. Latency and persistence are important characteristics of mycobacterial disease evoking long term immunological influences promoting disease. These states have been difficult to quantitate due to dormant unculturable phenotypes. Better tests are needed but the likelihood is that the extent of bTB and MAP disease burden in humans is not yet fully evident. The threat from bTB and MAP to human health thus remains tangible. Only vigilance currently prevents many potential animal reservoirs becoming re-seeded. Failure to improve screening and eradication tools will continue to allow global spread, increase the risk of human exposure and ensure more trouble is in store.

**Key Words:** bovine tuberculosis, paratuberculosis, zoonotic potential



## Breeding and Genetics: EAAP Genetics Symposium: Breeding for environmental sustainability

**259 Breeding goals to deal with climate change and food security.** Eileen Wall\*, *Scotland's Rural College (SRUC), Edinburgh, UK.*

In today's world, livestock production systems need to deliver a secure supply of food (locally and globally) with ever increasing competition for resources (land, water, feed/food, energy). As the competition for resources increases, the costs of production can increase and therefore farmers are challenged to improve efficiencies within their systems with both short and long-term sustainability in mind, trying to balance the profitability of the farming system while minimizing any negativity externalities of that production system (e.g., greenhouse gas emissions (GHG), animal welfare, land capability). As with the rest of society, livestock systems also need to consider potential climate change impacts in their longer term management options. Climate change is expected to have detrimental effects on milk yield and quality, fertility and health, putting further pressure on the sustainability of the sector and introducing new challenges for breeding goals and animal welfare. As climate change advances, indirect effects of weather on wider agricultural systems will become more important. An animal's ability to 'cope' with extreme weather could be further compromised if climate change decreases the availability of these resources, as predicted. This would result in increased competition for the water and nutrients needed to maintain cows' production and fitness. Therefore, incorporating longer-term challenges in our livestock breeding goals (e.g., climate resilience, optimizing production within farming system) is a crucial and timely goal. This paper takes an interdisciplinary approach to quantifying the potential output from livestock systems (dairy and beef) under projected climate change. This will be extended to breeding goals that optimize productivity and climate resilience while meeting GHG targets and wider goals for animal health and welfare. The results could be used to provide solutions to key challenges limiting the efficiency, productivity and sustainability of the livestock sectors.

**Key Words:** breeding goal, climate change, livestock efficiency

**260 Genomic selection for the high-hanging fruit in livestock breeding programs.** Donagh P. Berry\*<sup>1</sup>, Yvette de Haas<sup>2</sup>, Roel F. Veerkamp<sup>2</sup>, Mike Coffey<sup>3</sup>, and Mario P. L. Calus<sup>2</sup>, <sup>1</sup>*Animal & Grassland Research and Innovation Centre, Teagasc, Moorepark, Co. Cork, Ireland,* <sup>2</sup>*Animal Breeding and Genomics Centre, Wageningen UR Livestock Research, Wageningen, the Netherlands,* <sup>3</sup>*Animal and Veterinary Sciences, SRUC, Easter Bush Campus, Easter Bush, Edinburgh, UK.*

Genomic selection has by now been adopted for most of the "low-hanging" traditionally measured traits in developed breeding programs. The benefit of incorporating genomic information into genetic evaluations, however, is greatest for economically important traits not routinely recorded and thus where only low accuracy of selection is being achieved using traditional approaches. Irrespective of species, difficult to measure traits (i.e., the high-hanging fruit) include feed intake and efficiency, environmental footprint, product quality, and animal health and disease. Options to develop genomic predictions for these high hanging fruits include (1) collation of (inter)national databases which in themselves are too small to achieve high accuracy of predictions, (2) exploitation of low-cost, easy to measure predictors, (3) development of an optimal reference population based on phenotypic and genomic

diversity (and possible financial incentives or investment in collection of same), and (4) detection and exploitation of the causal mutations. A successful international initiative (gDMI) based on the collation of feed intake data from 9 countries in dairy cows concluded that exchange of phenotypic and genomic information can augment the accuracy of genetic/genomic evaluations. Such an approach should also be embarked on for other breeds and species. Steps should, however, be taken now in anticipation of such an initiative including the exchange of germ-plasm between research centers or the use of a pan-global list of sires. Maximizing phenotypic and genomic diversity (within the constraints of relatedness to the candidate population) could improve the accuracy of genomic predictions; whole populations could be screened using low-cost predictor traits (e.g., sensors, mid-infrared spectroscopy) and divergent animals collated to a centralized unit for deep phenotyping. Combining transcriptomic and genomic data could aid in the detection of causal mutations. Successful genomic selection will also require a re-evaluation of current phenotyping strategies and may include measurements on the parents themselves (e.g., feed intake on bulls) as currently undertaken in other species or half-/full-sibs (e.g., deliberate infection with pathogens).

**Key Words:** genomic selection, phenotype, breeding scheme

**261 Statistical approaches to increase resilience of animals towards environmental challenges and to increase homogeneity of animal products.** Han A. Mulder\*<sup>1</sup>, Ewa Sell-Kubiak<sup>1</sup>, Juanma Herrero-Medrano<sup>1,2</sup>, Pramod K. Mathur<sup>2</sup>, and Egbert F. Knol<sup>2</sup>, <sup>1</sup>*Animal Breeding and Genomics Centre, Wageningen University, Wageningen, the Netherlands,* <sup>2</sup>*TOPIGS Norsvin BV, Beuningen, the Netherlands.*

In animal husbandry, there is a growing demand for animals that need less labor and are capable to handle diseases or other environmental challenges. For various markets, homogeneity of animal products is desired and uniform animals would ease management, e.g., less fluctuation in production in spite of challenges. Furthermore, due to globalization of breeding programs animals need to be capable to perform in a wide range of environments. Here we show 2 statistical approaches that can be used to breed for resilience and uniformity: a reaction norm model to breed for resilience and a double hierarchical generalized linear model (DHGLM) to breed for uniformity. Both were applied to reproduction traits such as total number of born piglets and number of piglets born alive in sow lines. For the reaction norm model, we first developed a challenge load indicator to estimate the level of challenge, based on drops in production. Subsequently, we used this challenge load indicator as a covariate in the reaction norm analysis. We found genetic correlations of 0.5–0.85 between healthy and diseased periods indicating substantial reranking of animals, or in other words genetic variation in resilience. We applied a DHGLM to total number born and found substantial genetic variation in residual variance of litter size with a genetic coefficient of variation at variance level of 0.17. Using deregressed variance EBV, we found a few highly significant genomic regions affecting the variance of litter size. These genomic regions could be utilized in genomic selection. Both statistical approaches can yield breeding values that could be used to select for increased resilience and uniformity of animal production. Due to its low heritability, accuracies of breeding values for resilience and uniformity are low, though substantial genetic variation

is present. Accuracy of breeding values for these traits can be enhanced by genomic selection.

**Key Words:** statistical modeling, resilience, uniformity

**262 The role of sustainable commercial pig and poultry breeding for food security.** Pieter W. Knap\*<sup>1</sup>, Anne-Marie Neeteson-Van Nieuwenhoven<sup>2</sup>, and Santiago Avendaño<sup>2</sup>, <sup>1</sup>*Genus-PIC, Schleswig, Germany*, <sup>2</sup>*Aviagen, Newbridge, UK*.

The worldwide demand for animal products is increasing: global meat consumption is projected to double in the coming 35 years. At the same time, availability of resources such as land and water are decreasing. This requires livestock production to increase its productivity and its reproductive efficiency, and to reduce its environmental impact. Animal breeding must support this. Livestock breeding goals should then broaden in a balanced way, focusing on productivity and efficiency, subject to certain external constraints and internal restrictions. External constraints are due to feed availability and environmental load (which calls for genetic improvement of livestock feed efficiency, reproductive efficiency, gross production levels, and liveability), and to animal welfare (which calls

for genetic improvement of liveability, adaptability and health). Internal restrictions are due to genotype by environment interaction (which calls for data recording at the producer level and using those data for genetic evaluation at the nucleus level), antagonisms between traits (which calls for wide data recording and balanced breeding goals incorporating all possibly antagonistic traits), and selection limits (which calls for proper strategies for restriction of inbreeding rate, and optimization of genetic contributions and mate allocation within populations). Commercial poultry and pig breeding goals have been evolving in that direction since the 1950s, and this logically extrapolates toward a shift of focus on animal robustness, feed efficiency, and product quality traits. At the same time, selection technology is becoming more powerful at a very fast rate, mainly due to molecular genetics tools. As a result of both these developments, animal breeding can make an increasing contribution to sustainable food security.

**Key Words:** food security, pig breeding, poultry breeding

## Forages and Pastures: Grasses and silages

**263 Maturity is a larger driver of fatty acid content in summer annuals than nitrogen fertility.** Caleb P. Goossen\*, Sidney C. Bosworth, Heather M. Darby, and Jana Kraft, *University of Vermont, Burlington, VT.*

Summer annual forages are a potentially important supplement for summer grazing. The C<sub>4</sub> photosynthetic pathway allows increased mid-season growth relative to C<sub>3</sub> perennial pasture. Maximizing grazing availability is important to I) producers seeking to optimize the fatty acid content of their dairy products, and II) organic dairy producers that must satisfy the National Organic Program 30% minimum pasture rule. Although total fatty acid (FA) and polyunsaturated fatty acid (PUFA) content responses to nitrogen (N) fertility and harvest timing have been considered in C<sub>3</sub> forage species, to our knowledge no studies have enumerated the FA profiles of summer annual forage species. This experiment was conducted over two years (2013 and 2014) at two Vermont locations (loamy sand and silt loam, respectively) to assess the effects of N fertility and harvest timing on total FA and PUFA content of sudangrass (*Sorghum × drumondii* ‘Hayking’) and pearl millet (*Pennisetum glaucum* ‘Wonderleaf’). The experiment was a split-plot design, with harvest height whole plot treatments (60 cm vs. 90+ cm) and N fertility sub-plot treatments (39, 79, 118, and 157 kg N/ha). A measure of leafiness—the proportion of leaf blades relative to the entire plant as dry matter (DM)—was recorded, as most FA from vegetative forages are found in chloroplast thylakoid membranes. At both locations in 2013, harvests at 60 cm instead of 90+ cm resulted in higher total FA concentration (17.2 vs. 12.6 g/kg DM,  $P < 0.001$ ) and a greater proportion of PUFA (62.3 vs. 57.7 g/100g total FA identified,  $P < 0.001$ ). At the loamy sand location however, both total FA and PUFA effects of harvest height were present only in sudangrass (interaction effect,  $P < 0.0001$ ). The loamy sand location was the only to show a sub-plot effect of increasing N fertility, with a total FA gain of only 1.9 g/kg DM from the lowest N level to the highest ( $P = 0.0478$ ). Leafiness was positively correlated with PUFA content in sudangrass and pearl millet ( $r = 0.645$  and  $0.503$ , respectively). PUFA and total FA content of summer annual forages are primarily driven by maturity. Nitrogen fertility effects were confined to the low fertility soil location.

**Key Words:** summer annual forage, PUFA, n-3 fatty acid

**264 Effect of grazing seedhead-suppressed tall fescue pasture on the vasoactivity of serotonin receptors.** James L. Klotz\*<sup>1</sup>, Glen E. Aiken<sup>1</sup>, Amanda M. Egert<sup>2</sup>, and David L. Harmon<sup>2</sup>, <sup>1</sup>USDA-ARS, Forage-Animal Production Research Unit, Lexington, KY, <sup>2</sup>Department of Animal and Food Sciences, University of Kentucky, Lexington, KY.

Previous research has demonstrated that exposure to ergot alkaloids reduces vasoactivity of serotonin (5HT) receptors. Chemical suppression of tall fescue seedhead production is a tool to reduce the level of exposure to ergot alkaloids by a grazing animal. Therefore, the objective was to evaluate contractility of lateral saphenous veins biopsied from mixed breed steers following a 87–101 d grazing period on 3-ha pastures of bermudagrass (BG;  $n = 5$ ;  $340 \pm 9$  kg), or standard endophyte-infected tall fescue that was not sprayed (UNSUP;  $n = 5$ ;  $300 \pm 6$ ; 0.56 ppm ergovaline) or sprayed (SUP;  $n = 5$ ;  $294 \pm 9$  kg; 0.24 ppm ergovaline) with herbicide containing aminopyralid and metsulfuron-methyl (Chaparral; Dow AgroSciences). To evaluate contractility, biopsied veins were mounted in a multimyograph and exposed to increasing concentrations

of a tall fescue seed extract (EXT; ergovaline source); and 5HT<sub>1B</sub> (CP9), 5HT<sub>1D</sub> (L), and 5HT<sub>2A</sub> (TCB) agonists. All contractility data were normalized to a maximal response of  $1 \times 10^{-4}$  M norepinephrine and were analyzed as a split plot treatment design using SAS for effects of pasture treatment, agonist concentration, and the interaction. There was no contractile response to any concentration of CP9 in any of the pasture treatments. There were pasture  $\times$  concentration interactions for contractile responses to TCB ( $P < 0.01$ ) and EXT ( $P < 0.01$ ). For both EXT and TCB, BG veins were more vasoactive to the higher concentrations ( $P < 0.05$ ) and there were no differences between UNSUP and SUP veins. There was also a pasture  $\times$  concentration interaction for the contractile responses to L ( $P < 0.01$ ). However, these responses were not sigmoidal and reached a zenith at  $5 \times 10^{-7}$  and  $1 \times 10^{-6}$  M. At these concentrations, the response was greatest for UNSUP ( $P < 0.05$ ) and did not differ between SUP and BG veins. Although lower levels of ergovaline in SUP pastures did not alter vasoactivity of 5HT<sub>2A</sub> or 5HT<sub>1B</sub> receptors in the lateral saphenous vein, elevated vasoactivity of 5HT<sub>1D</sub> in UNSUP veins suggests that lower ergovaline levels in SUP pastures reduces the vascular effects of ergot alkaloids at this receptor.

**Key Words:** ergot alkaloid, cattle, serotonin receptor

**265 Microbial count, fermentation, and aerobic stability of regular- and brown midrib-corn hybrids ensiled with and without a combo inoculant.** Juan J. Romero\*<sup>1</sup>, Yuchen Zhao<sup>2</sup>, Youngho Joo<sup>3</sup>, Jinwoo Park<sup>3</sup>, Marco A. Balseca-Paredes<sup>1</sup>, Adam J. Heitman<sup>1</sup>, and Miguel S. Castillo<sup>1</sup>, <sup>1</sup>Department of Crop Science, North Carolina State University, Raleigh, NC, <sup>2</sup>Department of Animal Nutrition and Feed Science, China Agricultural University, Beijing, China, <sup>3</sup>Department of Animal Science, Gyeongsang National University, Jinju, Korea.

The objective was to evaluate the effects of a combo inoculant on microbial count, fermentation, and aerobic stability of 4 hybrids of corn. Treatment design was the factorial combination of 4 corn-types ensiled with (INO) and without (CON) inoculant. Corn-types were TMF2R737, F2F817 (A and B, respectively; from Mycogen), P2089YHR, and P1449XR (C and D, respectively; from Pioneer). The NDF digestibility of A and C conventional hybrids was ~56.5% compared with 65.5% for B and D brown midrib hybrids. The inoculant added contained *Lactobacillus buchneri* and *Pediococcus pentosaceus* ( $4 \times 10^5$  and  $1 \times 10^5$  cfu/g of fresh corn). Experimental design was a complete randomized block design (3 field blocks) with treatments replicated 2 times (2 plots) within blocks, for a total of 16 experimental units within block. Corn from each plot was chopped, treated or not with inoculant, packed into 7.6L bucket silos, and stored for 100 d. At d 0 there were differences due to hybrids solely. The %DM was 44.0, 38.3, 42.1, and 41.3 for A, B, C, and D, respectively; lactic acid bacteria (LAB) count (log cfu/g of fresh corn) was greatest and similar for B and D (7.7) compared with A (6.7) and C ( $7.1 \pm 0.07$ ); yeast count was greatest in D ( $6.8$  vs.  $6.6 \pm 0.02$ ) and mold count was lowest in C compared with the others ( $5.7$  vs.  $5.9 \pm 0.05$ ); and pH was higher for A (5.81) vs. C (5.60) and D (5.70), but B ( $5.79 \pm 0.04$ ) was only higher than C ( $P \leq 0.05$ ). At opening, INO increased LAB (from 6.2 to  $6.6 \pm 0.08$ ) and pH (from 3.80 to  $4.02 \pm 0.01$ ), and decreased yeast (from 5.10 to  $3.74 \pm 0.18$ ,  $P \leq 0.05$ ) compared with CON. Aerobic stability (h) was extended by INO compared with CON (181 vs.  $35 \pm 1.1$ ) but to a lesser extent for B (73 vs.  $36 \pm 1.2$ ;  $P \leq 0.05$ ). The DM recovery (%) was higher for A (95.8) vs. B (93.4) and D (93.1), but C ( $95.0 \pm 0.6$ ) was only higher than D ( $P$



≤ 0.05). The results indicate that corn-hybrid had an effect on %DM and DM recovery, and that the combo inoculant used improved silage conservation to different extents depending on hybrid type.

**Key Words:** corn, microbe, inoculant

**266 In vitro digestion kinetics of NDF from stockpiled Tifton 85 bermudagrass as influenced by N fertilization.** Samantha R. Sechler\*, Courtney M. Holland, Carla J. Weissend, Mary K. Mullenix, and Russell B. Muntiferling, *Auburn University, Auburn, AL.*

Productivity and nutritive value of stockpiled bermudagrass are influenced by N fertilization and environmental conditions. Intake of forage DM and DE are highly correlated with kinetics of NDF digestion; yet, NDF digestion kinetics of stockpiled bermudagrass have not been extensively investigated. Therefore, a study was conducted to evaluate effects of N fertilization and seasonal changes in nutritive value on NDF digestion kinetics from stockpiled bermudagrass. Replicate (n = 2) pastures of Tifton 85 bermudagrass were cut on Aug 1 and fertilized with 56 (56N), 112 (112N) or 168 (168N) kg N/ha. Stockpiled forage was sampled monthly beginning on Oct 24, 2012 and analyzed for kinetic parameters of NDF digestion using in vitro fermentation. Undigested NDF residues were recovered at 3, 6, 9, 12, 18, 24, 48 and 72 h of incubation. Potential extent of NDF digestion was assumed complete at 72 h. Rates of NDF digestion were determined by least-squares regression of ln % potentially digestible NDF remaining vs. time; solution of the first-order kinetic equation to equal 100% potentially digestible NDF remaining yielded estimates of lag time. Data were analyzed as a completely randomized design using the PROC MIXED procedure of SAS 9.2. No N fertilization × sampling date interactions ( $P > 0.05$ ) were observed for any kinetic parameters measured. Potential extent of NDF digestion was greatest ( $P < 0.05$ ) in Oct (75.6%), least ( $P < 0.05$ ) in Jan (47.8%), and intermediate but not different ( $P > 0.05$ ) between Nov (61.1%) and Dec (59.0%). Potential extent of NDF digestion was greatest ( $P < 0.05$ ) for 168N (63.2%) and least ( $P > 0.05$ ) for 56N (58.8%). Rates of NDF digestion were not affected ( $P > 0.05$ ) by N fertilization, but were greater ( $P < 0.05$ ) in Oct ( $0.069 \text{ h}^{-1}$ ) and Nov ( $0.068 \text{ h}^{-1}$ ) than in Jan ( $0.063 \text{ h}^{-1}$ ). Lag time was less ( $P < 0.05$ ) for the 168N (7.3 h) than 56N (9.1 h) and 112N (8.8 h) treatments. Lag time was least ( $P < 0.05$ ) in Oct (6.0 h), intermediate ( $P < 0.05$ ) in Jan (7.8 h), and greatest but not different ( $P > 0.05$ ) between Nov (10.5 h) and Dec (9.4 h). Potential extent of NDF digestion was the kinetic parameter most affected by N fertilization and seasonal changes in stockpiled Tifton 85 bermudagrass.

**Key Words:** bermudagrass, digestion kinetics, neutral detergent fiber

**267 Variation in aerobic stability of maize silage with and without microbial inoculant.** Ida K. Hindrichsen\* and Nina Milora, *Chr. Hansen A/S, Hørsholm, Denmark.*

Forages to be ensiled for subsequent feeding to dairy cows are often contaminated with spoilage microorganisms that negatively affect feed quality and feed safety. Aerobic instability due to these spoilage organisms can cause heating and significant DM loss. *L. buchneri* improves aerobic stability, but because it grows slowly it is often combined with homofermentative strains to achieve a fast reduction in pH early in fermentation. Efficacy of silage inoculants can vary depending on forage type, harvest time, management etc. In the current study it was aimed at keeping the experimental conditions equal between experiments and to test the efficacy of a dual-purpose silage inoculant. Three samples of chopped whole maize were collected from farms in 2012 and 2013 on the day of harvest. Aerobic stability, microbial population and dry matter

loss of the maize was measured using the same methodology. Briefly, maize was inoculated with a combination of *L. buchneri* and *L. lactis* (150,000 cfu/g) or pure tap water and allocated into 1 kg bags, vacuum packed and stored for 15 weeks at 25°C. Then silage was placed in a polystyrene container (1.2 L; 274 kg/m<sup>3</sup>) with 1 cm diameter hole in top and bottom. The change in temperature was recorded continuously for 6.5 d using a data logger placed in the middle of each sample. Results showed that the pooled freshly harvested maize was very similar in microbial composition between the farms and years. All samples were highly contaminated with yeast and mold, while epiphytic total lactic acid bacteria were lower than the inoculated. The effect of the silage inoculant varied considerably among the 3 maize samples. Acetate concentration after 15 weeks of anaerobic fermentation was significant ( $P < 0.05$ ) for all inoculated samples compared with control. The aerobic stability varied considerably between the different control samples (46 h, 75 h, >156 h). Inoculation significantly increased ( $P < 0.05$ ) aerobic stability of the unstable samples (72 h, >156 h, >156 h). This study showed that maize with similar quantities of spoilage organisms in the fresh maize can vary widely in their aerobic stability.

**Key Words:** aerobic stability, maize silage, silage inoculant

**268 A comparative study of lignin assays and relationship with grass digestibility.** Alejandro Vargas Velásquez\* and Romualdo Shiguelo Fukushima, *Department of Animal Nutrition and Production (VNP), Faculty of Veterinary and Animal Science (FMVZ), University of São Paulo, Pirassununga, São Paulo, Brazil.*

Accurate determination of lignin is essential to understand the mechanism by which it inhibits carbohydrate digestion. Available analytical procedures have repeatedly produced conflicting results. The gravimetric methods ADL, potassium permanganate lignin (PL) and KL were compared against the spectroscopic acetyl bromide lignin method (ABL). Grass species *Brachiaria brizantha* 'Marandu' and 'Xaraés', *Panicum maximum* 'Mombaça' and *Pennisetum purpureum* 'Cameroon' and 'Napier' were harvested at 7 maturity stages. A completely randomized experimental design with duplicate analysis for the lignin assays was used. A randomized block design was used for the in vitro experiment. Highly significant effects for maturity, lignin method and their interaction on lignin content were observed. The ADL yielded the lowest values (28.05 to 103.05 g/kg DM) in all species. Values for PL were in accordance to previous observations from our laboratory and other authors (65.45 to 160.65 g/kg DM). The KL values observed (61.35 to 136.20 g/kg DM) were approximately double the values observed for ADL. Values for ABL were higher than all corresponding values from the other methods. Strong negative correlations between lignin contents and IVDMD or IVNDFD were observed for all methods. Higher correlations were observed for IVDMD than for IVNDFD, contrary to what could be expected, because lignin affects the CW digestibility but not the cellular contents. Regression analysis of ADL, KL and ABL produced negative slopes when plotted against IVDMD and IVNDFD. The PL method failed to reliably estimate the digestibility of tropical grasses, possibly because of other substances (pectin, tannins or flavonoids) solubilized by the KMnO<sub>4</sub> solution. Although ADL and KL had similar correlations with digestibility and therefore, estimate digestibility of forages with similar accuracy, these methods use strong acids and require determination of ash in the lignin residues, increasing time and cost of analysis. The ABL method has no need for corrections and is a fast and a convenient method for determination of total lignin content in plants, thus, a good option for routine laboratory analysis.

**Key Words:** lignin, acetyl bromide, digestibility

**269 Using Bayesian inference to delineate diet composition of mixed forages.** Napoleón Vargas Jurado<sup>\*1</sup>, Amy E. Tanner<sup>2</sup>, and Ronald M. Lewis<sup>1</sup>, <sup>1</sup>University of Nebraska, Lincoln, NE, <sup>2</sup>Virginia Tech, Blacksburg, VA.

Diet preferences in grazing animals are important for range management and ecology. However, estimating the composition of diet mixtures is challenging. The plant-wax marker technique, combined with nonnegative least squares (NNLS), has traditionally been used to estimate diet composition across herbivore species. More flexible methods, such as the normal compositional model (NCM) under Bayesian inference, offers an alternative approach for predicting diet composition. The efficiency of NCM was assessed by simulation. Mean *n*-alkane (C<sub>27</sub>, C<sub>29</sub>, C<sub>31</sub>, C<sub>33</sub>) and long-chain alcohol (LCOH; C<sub>26</sub>OH, C<sub>28</sub>OH, C<sub>30</sub>OH) concentrations (mg/kg) were obtained for 11 subsamples for each of 2 forages – tall fescue and red clover – by gas chromatography. The CV of those measures was 10.7%, which was used to derive a common SD (20 mg/kg). Forage mixtures ranging from 0.10 to 0.90 fescue, in 0.10 increments, were simulated from a Gaussian distribution. Values for each *n*-alkane and LCOH, using their respective mean and the common SD, were drawn independently. For each mixture, 100 sets of observations were generated. Data were analyzed using NNLS and NCM. For the *n*-alkanes and LCOH alone and in combination, efficiency was assessed by normalized mean squared error (NMSE), mean average difference (MAD), and coverage of 95% CI. For both types of markers, both methods accurately predicted the forage mixtures, although more so with NCM. The NMSE for the NNLS were 0.032%, 0.019% and 0.012% for *n*-alkanes, LCOH and their combination, respectively; for NCM, those respective values were 0.028%, 0.011% and 0.008%. Similarly, MAD for NNLS were 1.29%, 0.92% and 0.75% for *n*-alkanes, LCOH and their combination, respectively; for NCM, the respective values were 1.22%, 0.74% and 0.63%. Coverage was high for the combined markers: 97.1% for NCM and 91.2% for NNLS. Given the scenario used, the NCM more accurately predicted forage mixtures. In the current study, a common SD was assumed. Such is doubtfully the case in practice, and the NCM methodology can accommodate non-trivial covariance structures among markers and better model differences in variation for individual markers. Such extensions are underway. The NCM approach provides a robust tool for estimating forage mixtures.

**Key Words:** plant-wax markers, Bayesian inference, diet composition

**270 Interaction of isoflavones and endophyte-infected tall fescue seed extract on vasoactivity of bovine mesenteric vasculature.** Yang Jia<sup>\*1</sup>, David L. Harmon<sup>1</sup>, and James L. Klotz<sup>2</sup>, <sup>1</sup>Department of Animal and Food Sciences, University of Kentucky, Lexington, KY, <sup>2</sup>USDA-ARS, Forage-Animal Production Research Unit, Lexington, KY.

It is hypothesized that isoflavones may attenuate ergot alkaloid-induced vasoconstriction and possibly alleviate diminished contractility of vasculature after exposure to ergot alkaloids. The objective of this study was to determine if incubation of bovine mesenteric vasculature with formononetin (F), biochanin A (B), or an ergovaline-containing tall fescue seed extract (EXT) and their combinations affect ergotamine (ERT) induced contractility. Multiple segments of mesenteric artery and vein supporting the ileal flange of the small intestine were collected from Angus heifers at slaughter (n = 5, BW = 639 ± 39 kg). After cleaning and sectioning, duplicates of each vessel type were incubated in tissue culture flasks at 37°C with a 50-mL volume of Krebs-Henseleit buffer containing: only buffer (Control); or 1 × 10<sup>-6</sup> M EXT; F; or B; and combinations of 1 × 10<sup>-6</sup> M EXT+F; 1 × 10<sup>-6</sup> M EXT+B; 1 × 10<sup>-6</sup> M

F+B; or 1 × 10<sup>-6</sup> M EXT+F+B. After incubation for 2 h, sections were mounted in a multimyograph chamber. The ERT dose responses were normalized to 0.12 M KCl. Pretreatment with F, B, and F+B without EXT resulted in similar contractile responses to ERT in mesenteric artery and the incubations containing EXT resulted in a complete loss of vasoactivity to ERT. In mesenteric artery pretreated with EXT, treatments that contained B had higher contractile responses (P < 0.05) at ERT concentrations of 1 × 10<sup>-7</sup> M and 5 × 10<sup>-7</sup> M. Also, treatments containing B tended (P < 0.1) to have greater responses than treatments without B at ERT concentrations of 1 × 10<sup>-6</sup> M, 5 × 10<sup>-6</sup> M, and 5 × 10<sup>-5</sup> M. In mesenteric vein pretreated with EXT, treatments containing F had greater contractile responses to ERT at 1 × 10<sup>-5</sup> M, 5 × 10<sup>-5</sup> M, and 1 × 10<sup>-4</sup> M (P < 0.05). These data indicated that F and B at 1 × 10<sup>-6</sup> M and their combination did not affect the overall contractile response to ERT in mesenteric vasculature. However, F and B may alleviate the reduction of vasoconstriction caused by prior exposure to alkaloids in endophyte-infected tall fescue seed extract.

**Key Words:** ergot alkaloid, isoflavone, mesenteric vasoconstriction

**271 Proximate and fiber composition of leaves and stems of *Pennisetum purpureum* varieties fertilized with animal manure.** Victoria O. Ojo<sup>\*</sup>, Sarafadeen T. Adewuyi, Alaba O. Jolaosho, Adebayo O. Oni, and Oludotun O. Adelusì, Federal University of Agriculture, Abeokuta, Ogun State, Nigeria.

An experiment was conducted at the Organic Research Farm, Federal University of Agriculture, Abeokuta between September 2013 and March 2014 to evaluate the influence of manure type and season on the chemical composition of plant fractions of *Pennisetum purpureum* varieties. The experiment was a split-split plot design with 4 manure types: swine (10.16 kg), cattle (22 kg), poultry (7.92 kg)/plot and control) were applied in one application as the main plot, 4 varieties of *P. purpureum* (green, purple, S13 and S15) as the sub-plot and 2 seasons (early and late dry season) as the sub-sub plot with 3 replicates. An area of land of 53m × 11m was divided into individual plot size of 2m<sup>2</sup> after land clearing and was sown with *P. purpureum* stems. The level of manure application was determined according to the nitrogen need of the soil which contained: total N (0.15%), organic C (1.31%), and available P (32.87 mg kg<sup>-1</sup>). Forage subsamples were harvested after 12 weeks of growth, weighed and oven-dried. Analysis of proximate and fiber contents of leaves and stem fractions was carried out and data obtained were analyzed using GLM procedure of SPSS Statistics 20. Results from this experiment showed (P < 0.05) CP content of *P. purpureum* leaves was highest in early dry and late dry seasons for swine manure (25.94%, 21.94%) compared with control (24.86%, 21.83%), cattle (25.53%, 18.49%) and poultry (24.19%, 18.04%) respectively. *P. purpureum* variety Local green had the highest (P < 0.05) CP content (23.32%) in leaf fractions while *P. purpureum* variety purple had the highest (P < 0.05) CP (4.73%) in stem fractions. Cellulose contents of leaf (42%) of the grasses harvested in the early dry season was higher (P < 0.05) than those harvested in the late dry season. The ADL content were lower (P < 0.05) in the leaves than in the stem fractions in all the parameters measured. Feeding values of all the varieties (DDM, DMI, RFV and FI) were higher (P < 0.05) in the leaf fractions than stem fractions. It could therefore be concluded that swine manure is a good source of nutrient for sown pasture production and the use of animal manures maintained the quality of the grasses throughout the dry season.

**Key Words:** manure, leaf, stem

**272 Effects of inoculated lactic acid bacteria on aflatoxin B<sub>1</sub> in corn silage.** Zhengxin Ma\*, Felipe X. Amaro, Juan J. Romero, and Adegbola T. Adesogan, *Department of Animal Sciences, University of Florida, Gainesville, FL.*

The objective was to determine the effect of adding viable and unviable silage inoculant bacteria on binding of aflatoxin B<sub>1</sub> (AFB<sub>1</sub>) and silage quality. A corn hybrid was harvested at 35% DM, chopped to achieve a theoretical length of 1.9 cm and treated in quadruplicate with deionized water (Con) or 30 µg/kg of AFB<sub>1</sub>, or with a mixture of AFB<sub>1</sub> and viable, heated or acid-treated forms ( $1 \times 10^9$  cfu/g) of *Lactobacillus plantarum* R2014 (Lp), *L. buchneri* R1102 (Lb), or *Pediococcus acidilactici* EQ01 (Pa). Each of the inoculants was incubated for 1 h in 30 mL of phosphate-buffered saline (pH 7.3) at 37°C (viable) or 85°C (heated) or in 30 mL of 0.003 M HCl (pH 2.5) at 37°C (acid-treated) before application. Silages (3 kg) were stored for 21 d in polythene bags. Concentrations of AFB<sub>1</sub> were measured after 0, 24, 48, and 72 h of ensiling and silage samples from d 21 were analyzed for chemical composition, pH, volatile fatty acids and lactate. The experiment had a completely randomized design. Data were analyzed using the GLIMMIX procedure of SAS (SAS Institute Inc., Cary, NC). The AFB<sub>1</sub> concentration decreased linearly ( $P < 0.05$ ) to 0.35 µg/kg or less within 3 d of ensiling across treatments. Bacterial inoculation did not ( $P > 0.05$ ) increase the AFB<sub>1</sub> reduction. Treatment with AFB<sub>1</sub> reduced ( $P = 0.03$ ) the CP concentration (8.83 vs. 8.25%) and increased ( $P < 0.001$ ) butyric acid concentration (0.37 vs. 0.62%) but inoculation prevented ( $P < 0.05$ ) these effects. Treatment with viable Lp increased ( $P < 0.05$ ) the acidification of the forage in the first 3 d and the lactic acid concentration on d 21 (1.52 vs. 3.97%). However, inoculation with viable Lb or Pa did not ( $P > 0.05$ ) affect the chemical composition, suggesting that typical beneficial effects of these bacteria were attenuated by AFB<sub>1</sub>. The spiked aflatoxin was reduced to a safe level within 3 d of addition. Inoculation with bacteria did not increase AFB<sub>1</sub> removal but viable Lp prevented the increase in butyric acid and decrease in CP caused by the toxin.

**Key Words:** aflatoxin B<sub>1</sub>, lactic acid bacteria, silage

**273 In vitro digestibility of dried cassava peel, mushroom degraded cassava peel and silage from guinea grass.** Bolanle T. Akinyemi\*, Moronfolu Ige, Alaba O. Jolaosho, Moses O. Arigbede, And Shamsideen O. Iposu, *Federal University of Agriculture, Abeokuta, Abeokuta, Ogun State, Nigeria.*

This study was carried out to determine the quality and in vitro digestibility of dried cassava peel, mushroom degraded cassava peel when mixed with 40% of silage from guinea grass and the grass at different levels. Samples from dried cassava peel, mushroom degraded cassava peel (SMS), silage and grass was collected and the nutritional potential was investigated by determining the chemical composition and the level of digestibility. The result shows that there was a significant difference ( $P < 0.05$ ) on the in vitro gas production of dried cassava peel, mushroom-degraded cassava peel and when mixed with 40% of silage from guinea grass and the grass at different levels. The highest gas production of whole samples was from 100%CP having the ( $P < 0.05$ ) value of 56.25 mL/200 mg of DM and the lowest was from which had the ( $P < 0.05$ ) value of 26.50 mL/200 mg of DM. The highest gas production of diets was from Diet 6 (40% grass + 50% cassava peel + 10% PKC) having the ( $P < 0.05$ ) value of 59.00mL/200mgDM and the lowest was from Diet 2 (40% silage + 50% cassava peel + 10% PKC), which had the ( $P < 0.05$ ) value of 17.50 mL/200 mg of DM. Diet 6 had the highest values for metabolizable energy (ME), organic matter digestibility (OMD) and short-chain fatty acids (SCFAs; 7.26 MJ·kg<sup>-1</sup>, 59.93% and 0.82 µmol respectively). Diet 3 has the lowest value for ME, OMD and SCFAs (3.84 MJ·kg<sup>-1</sup>, 35.79% and 0.22 µmol respectively). The highest methane gas production of whole samples was from 100%CP having the ( $P < 0.05$ ) value of 26.33 mL and the lowest was from 100%SMS which had the ( $P < 0.05$ ) value of 12.88 mL. The highest gas production of Diets was from Diet 1 having the ( $P < 0.05$ ) value of 25.00 mL and the lowest was from Diet 3 which had the ( $P < 0.05$ ) value of 11.25 mL. The diet having 40% grass + 50% cassava peel + 10% PKC had the highest nutritional qualities. It is therefore concluded from this study that the use of cassava peel as substrate for producing edible mushroom improved the potential feeding value of the resultant substrate.

**Key Words:** silage, mushroom degraded cassava peel, in vitro digestibility



# Graduate Student Competition: ADSA Production Division

## Graduate Student Oral Competition, PhD

**274 Carry-over effect of an individual feeding strategy on milk production of Holstein cows managed for extended lactation.** Charlotte Gaillard\*<sup>1</sup>, Nicolas C. Friggens<sup>2</sup>, Martin R. Weisbjerg<sup>1</sup>, and Jakob Sehested<sup>1</sup>, <sup>1</sup>Aarhus University, Foulum, Tjele, Denmark, <sup>2</sup>AgroParisTech, INRA UMR 0791 MoSAR, Paris, France.

Feeding can induce significant individual variation in cows' milk production. Previous studies showed that an increase of energy density of the ration for a short period in early lactation induced greater total milk yield and persistency, defined by the shape of the lactation curve. The objective of our feeding strategy was to reduce the intensity of mobilization and thereby to sustain the mobilization for a longer period. We hypothesized that a supply of energy during the mobilization period of each individual cow will have a positive carry-over effect on milk production. A total of 53 Holstein cows performed extended lactations (461 ± 7 d). They were housed in free stalls with access to a milking robot. The group 1 (G1) were fed a control partially mixed ration, with a 60:40 forage:concentrate ratio (energy density 11.8 ± 0.1 MJ of ME/kg of DM), for the full lactation. The group 2 (G2) received a diet enriched in energy (ratio 50:50, energy density 12.1 ± 0.1 MJ of ME/kg of DM) followed by the control diet. The diet shift of G2 was defined individually when DIM ≥ 42, and live weight gain ≥ 0 kg/d on a 5 d average. The production data were smoothed using the *fda* package in R software. The daily smoothed data and the associated slopes were analyzed using a linear mixed effects model with parity and treatment as fixed effects. From calving to 42 DIM, an interaction indicated that the ECM of multiparous G2 was higher than for G1 (42.8 vs. 40.1 ± 1.5 kg ECM/d), while the primiparous presented opposite effects (29.0 vs. 32.5 ± 2.2 kg ECM/d) ( $P = 0.02$ ). The 300 d following the shift in diet, G1 and G2 had similar ECM (35.0 ± 1.0 kg ECM/d,  $P = 0.76$ ) but the curves had different slopes. From 0 to 100 d from shift, the ECM increased for the primiparous G2 while it decreased for the primiparous G1 (+0.008 vs. -0.01 ± 0.007 kg ECM/d,  $P = 0.04$ ). From 201 to 300 d from shift, the ECM decreased faster for G2 than for G1 (-0.06 vs. -0.04 ± 0.008 kg ECM/d,  $P = 0.02$ ). To conclude, the treatment had a 100-d positive carry-over effect only on primiparous persistency, as well as a 300 d negative carry-over effect on the ECM persistency for all the cows.

**Key Words:** extended lactation, persistency, individual feeding strategy

**275 Moisture, temperature, cow health, and bedding bacteria relationships in compost bedded pack barns.** Elizabeth A. Eckelkamp\*, Joseph L. Taraba, Robert J. Harmon, Katherine A. Akers, and Jeffrey M. Bewley, *University of Kentucky, Lexington, KY.*

Attracting global interest, the compost bedded pack barn (CB) is a loose housing system bedded with shavings or sawdust, without the stalls and partitions found in freestall and tie-stall housing. The objective of this study was to assess relationships among compost bedded pack (CB) moisture and temperature, ambient weather conditions, cow hygiene, mastitis indicators, and bedding bacterial counts. The CB moisture and temperature are affected by ambient conditions, which in turn alter the conditions experienced by the cow and the bacteria in the bedding. The study was conducted using data from 8 CB farms in Kentucky from May 2013 to May 2014. Biweekly, one observer hygiene scored 50 cows per farm and collected CB internal temperature at 20 cm depth

(CIT), moisture, nutrient profile, and bedding samples for bacteriological culture from 9 areas in each barn. Somatic cell count and high SCC prevalence (HSP, percent of cows with SCC ≥ 200,000 cells/mL) were collected from DHIA. The MIXED procedure of SAS 9.3 (SAS Institute, Inc., Cary, NC) was used to assess fixed effects for all models. Stepwise backward elimination was used to remove non-significant interactions ( $P \geq 0.05$ ) with all main effects remaining in the models regardless of significance. Internal temperature increased with increasing BTHI ( $P < 0.01$ ) and decreased with increasing milk yield ( $P < 0.01$ ). Compost moisture content decreased with increasing BTHI ( $P < 0.01$ ). Herd hygiene score decreased with increasing BTHI ( $P < 0.01$ ) and increased with increasing CB moisture ( $P = 0.02$ ). Herd SCC and HSP both increased with increasing BTHI ( $P < 0.01$ ), but did not change with compost factors. Staphylococcus ( $P = 0.01$ ), streptococcus ( $P = 0.01$ ), and bacillus ( $P = 0.03$ ) species growth in the bedding decreased with increasing CIT while coliform species growth ( $P = 0.02$ ) increased with increasing CIT. Maintaining higher internal temperature did not reduce all bedding bacteria levels. In CB farms, BTHI affected cow hygiene and udder health indicators more than CB moisture and CIT.

**Key Words:** compost bedded pack barn, bedding bacteria, somatic cell count

**276 Effect of dry period length and dietary energy source in dairy cows on natural antibody titers and somatic cell count in milk.** Novi Mayasari\*<sup>1,2</sup>, Wilke Rijks<sup>1</sup>, Ger de Vries Reilingh<sup>1</sup>, Gerrit Remmelink<sup>3</sup>, Bas Kemp<sup>1</sup>, Henk Parmentier<sup>1</sup>, and Ariette van Knegsel<sup>1</sup>, <sup>1</sup>Adaptation Physiology Group, Department of Animal Science, Wageningen University, Wageningen, Gelderland, the Netherlands, <sup>2</sup>Faculty of Animal Husbandry, Universitas Padjadjaran, Bandung, West Java, Indonesia, <sup>3</sup>Livestock Research, Wageningen University and Research Centre, Wageningen, Gelderland, the Netherlands.

Omission of the dry period of cows improved energy balance (EB) and showed variable effects on somatic cell counts (SCC) and natural antibodies (NAb) in milk. A glucogenic diet compared with lipogenic diet enhanced plasma NAb binding keyhole limpet hemocyanin (KLH). NAb in milk were associated with SCC. It was hypothesized that during negative energy balance, NAb either have a role or reflect in inflammatory processes and are associated with SCC. The objective was to study effects of dry period length and dietary energy source on titers of NAb binding KLH and lipopolysaccharide (LPS) in milk, SCC and mastitis. In total, 167 Holstein-Friesian dairy cows were randomly assigned to treatments. Treatments consisted of 3 dry period lengths: 0-, 30- or 60-d and 2 early lactation diets (glucogenic or lipogenic), in a 3 × 2 factorial design. Cows enrolled in this study were clinically healthy and had SCC in milk < 250,000 cell/mL. Milk samples for NAb and SCC measurement were sampled weekly and 4 times per week, respectively, from wk 1 until 14 postpartum. The data collected were statistically analyzed using ANOVA and logistic regression. Cows with a 0-d dry period had higher titers of IgG and IgM binding KLH and LPS and higher SCC in milk compared with cows with a 30- or 60-d dry period ( $P < 0.01$ ). Mastitis incidence was 17% and did not differ between dry period lengths or lactation diets. A glucogenic diet showed higher titers of IgM binding LPS and tended to have higher titers of IgG binding LPS in milk compared with a lipogenic diet ( $P < 0.01$  and  $P = 0.08$ , respectively).

Higher titers of IgG and IgM binding KLH and IgG binding LPS were associated with increased risk of high SCC ( $P < 0.05$ ). Higher IgG and IgM binding KLH and LPS were also associated with increased risk of mastitis ( $P < 0.05$ ). The results demonstrate that cows with a 0-d dry period and fed a glucogenic diet showed high titers of IgG binding LPS in milk. Moreover, we can conclude that IgG and IgM binding KLH or LPS in milk might be additional valuable tools to detect increased risks for mastitis in dairy cows.

**Key Words:** dry period, somatic cell count, antibodies

**277 Towards a better understanding of the effect of genetic merit for milk production on post-partum cyclicity of first lactation dairy cows.** Nicolas Bedere\*<sup>1</sup>, Luc Delaby<sup>1</sup>, Vincent Ducrocq<sup>2</sup>, Ségolène Leurent-Colette<sup>3</sup>, and Catherine Disenhaus<sup>1</sup>, <sup>1</sup>INRA-Agrocampus-Ouest UMR 1348 PEGASE, Saint-Gilles, France, <sup>2</sup>INRA UMR 1313 GABI, Jouy-en-Josas, France, <sup>3</sup>INRA UE 326 Domaine Expérimental du Pin-au-Haras, Exmes, France.

Milk genetic merit is known to affect commencement of luteal activity (CLA) in dairy cows. This effect is considered to be due to energy exported in milk production. We hypothesize that cows with genetic predisposition to export milk energy through milk yield would have degraded cyclicity compared with those exporting energy through fat and protein contents. This study aimed to identify and quantify the effect of breed and genetic groups on postpartum cyclicity of primiparous dairy cows. From 2006 to 2013, an experiment was conducted on 194 primiparous dairy (Holstein) and dual purpose (Normande). Within breeds, cows were classified into 2 groups with regards to their estimated breeding value (EBV). Cows with high EBV for milk yield were included in a "Milk" group (MG) and those with high EBV for fat and protein content were included in a "Content" group (CG). Within breeds, exported milk energy and weight loss were similar for cows in both MG and CG groups. Interval from calving to CLA was studied performing survival analyses (Weibull regression). Progesterone profile, milk yield and body condition were analyzed using Chi<sup>2</sup>-test and ANCOVA. Holstein cows produced more milk (+1450 kg,  $P < 0.001$ ) and lost more body weight (BW: -1.4kg/wk,  $P < 0.01$ ) than Normande ones. Normande and Holstein cows had different baseline hazard functions for CLA, Normande cows having earlier CLA than Holstein ones. Within breeds, cows in the CG group had earlier CLA (associated HR = 2.0,  $P = 0.001$ ) than cows in the MG group. For Holstein only, BW loss from wk 1 to 14 of lactation tended to be associated with later CLA ( $P < 0.1$ ). Within breeds, no effect of milk yield on CLA was observed. Prolonged luteal phases were frequent (18% of cows) and neither associated with breed nor with differences between MG and CG. Interovulatory intervals were longer for Holstein than for Normande cows (+1.7 d,  $P < 0.001$ ) because of a longer luteal phase and a longer interluteal interval. To conclude, this study showed that cows with genetic predisposition to export milk energy through fat and protein contents had earlier CLA than those exporting milk energy through yield.

**Key Words:** genetic merit, cyclicity, primiparous cows

**278 Forage yield, quality, and digestibility when intercropping vining soybean with MasterGraze seed corn at different seeding rates.** Ishwary Acharya\*<sup>1</sup>, David Casper<sup>1</sup>, Xingyou Gu<sup>2</sup>, and Ahamed Charif<sup>2</sup>, <sup>1</sup>Dairy Science Department, South Dakota State University, Brookings, SD, <sup>2</sup>Department of Plant Science, South Dakota State University, Brookings, SD.

The production of forage resulting from the intercropping of corn and soybean at planting has the potential to yield greater quantities of digestible nutrients to meet the nutrient requirements of lactating dairy cows. A field plot experiment was conducted to measure forage yield, nutrient concentration and digestibility when intercropping MasterGraze (MG) seed corn and vining (V) soybean lines at different seeding rates. A randomized complete block design with 5 different seeding rates [100:0 (T1); 67:33 (T2); 50:50 (T3); 33:67 (T4), and 0:100 (T5) of V and MG] with 3 replicates was used to determine the optimal intercropping seeding rates. Forage was hand harvested 97 d after planting during the 2014 growing season, inoculated, packed into plastic buckets, weighed, and ensiled for 60 or 90 d. Buckets were then re-weighed, opened, and samples of forage collected. Fresh, 60- and 90-d ensiled forage samples were submitted to a commercial laboratory (Analab Inc., Fulton, IL). Fresh forage yield was greatest ( $P < 0.05$ ) for T5 (all MG) compared with other ratios of MG and V (40.7, 78.0, 75.6, 75.5 and 80.9 T/ha for T1, T2, T3, T4, and T5, respectively). Fresh DM yield (16.5, 22.0, 21.1, 20.2, and 18.7 T/ha) and digestible DM (12.3, 15.2, 14.7, 14.1 and 13.9 T/ha) were similar ( $P > 0.05$ ) across all V:MG ratios. Fresh CP yield (3.74, 2.29, 2.14, 1.97, and 1.18 T/ha) was greatest ( $P < 0.05$ ) for T1 compared with other treatments and T2 greater ( $P < 0.05$ ) than T5 with remaining ratios being intermediate and similar. Fresh digestible fiber yield was similar ( $P > 0.10$ ) for all V:MG ratios (3.78, 5.55, 6.02, 5.22, and 6.45 T/ha). The 60 d DM ensiling loss (5.23, 1.34, 1.22, 1.71 and 1.97%) was greatest ( $P < 0.05$ ) for T1 compared with the other V:MG ratios. The 60 d ensiling digestible DM yield (12.3, 15.6, 14.9, 14.0 and 12.9 T/ha) was greatest ( $P < 0.05$ ) for T2 and lowest for T1 with other ratios being intermediate. In conclusion, the intercropping of V and MG holds great potential for increasing the production of forages to meet the nutrient requirements of lactating dairy cows.

**Key Words:** corn, soybean, yield

**279 Intravaginal administration of prostaglandin F<sub>2α</sub> induces luteolysis in lactating dairy cows.** Robert Wijma\*, Matias L. Stangaferro, and Julio O. Giordano, Department of Animal Science, Cornell University, Ithaca, NY.

Our objectives were to test the efficacy of PGF<sub>2α</sub> (PGF) to induce luteolysis and evaluate progesterone (P4) dynamics after intravaginal (IVG) administration in lactating dairy cows. Our hypothesis is that PGF given IVG will induce luteolysis. In 2 experiments, cows were synchronized using Ovsynch (GnRH-7 d-PGF-56 h-GnRH) to induce ovulation and the formation of a corpus luteum (CL). Cows with at least 1 CL  $\geq 15$  mm 8 d after Ovsynch remained in the study. In Exp 1, cows (n = 31) stratified by parity and number of CL received: 5 mL of saline solution IVG (SAL-IVG, n = 6), 25 mg of PGF im (PGF25-im, n = 7), 25 mg of PGF IVG (PGF25-IVG, n = 6), 50 mg of PGF IVG (PGF50-IVG, n = 6) and 125 mg of PGF IVG (PGF125-IVG, n = 6). In Exp 2, cows (n = 32) stratified as in Exp 1 received: SAL-IVG (n = 7), PGF25-im (n = 7), PGF25-IVG (n = 6), PGF50-IVG (n = 6) as in Exp 1 whereas another group received 2 IVG boluses of 25 mg of PGF 12 h apart (PGF25-2X-IVG, n = 6). Blood was collected at -1 h, 0 h, every 6 h up to 24 h, and then every 12 h up to 96 h after treatment (trt). Concentrations of P4 after treatment were analyzed by ANOVA with repeated measures using PROC MIXED of SAS. Mean separation was conducted with LSD method. In Exp 1, P4 concentrations from -1 to 96 h were affected by trt ( $P < 0.01$ ), time ( $P < 0.001$ ) and trt by time ( $P < 0.001$ ). From 12 to 96 h, all PGF groups had less P4 ( $P < 0.05$ ) than SAL-IVG cows. In Exp 2, from -1 to 96 h P4 concentrations were affected by trt ( $P < 0.001$ ), time ( $P < 0.001$ ) and trt by time ( $P < 0.001$ ). From 12 to 96 h all PGF groups had less ( $P < 0.05$ ) P4 than SAL-IVG cows. From 60 to 96 h,

PGF25–2X-IVG and PGF25-im tended ( $P < 0.1$ ) to or had less ( $P < 0.05$ ) P4 than PGF25-IVG. When data from Exp 1 and 2 were combined, P4 concentrations from –1 to 96 h were affected by trt ( $P < 0.001$ ), time ( $P < 0.001$ ) and trt by time ( $P < 0.001$ ). Concentrations of P4 in all the PGF groups were less ( $P < 0.05$ ) than in the SAL-IVG group from 6 to 96 h. At 60 h, the PGF25–2X-IVG group had less ( $P < 0.05$ ) P4 than the rest of the groups. We conclude that it is possible to induce luteolysis in lactating dairy cows by intravaginal administration of PGF. Two 25-mg boluses 12 h apart were more effective than one bolus of 25, 50 or 125 mg of PGF and as effective as a 25-mg bolus of PGF im.

**Key Words:** intravaginal, prostaglandin, dairy cow

**280 Dry period plane of energy: Effects on glucose tolerance in peripartum dairy cows.** Sabine Mann\*<sup>1</sup>, Francisco A. Leal Yepes<sup>2</sup>, Thomas R. Overton<sup>2</sup>, Joseph J. Wakshlag<sup>3</sup>, Bethany P. Cummings<sup>4</sup>, and Daryl V. Nydam<sup>1</sup>, <sup>1</sup>Department of Population Medicine and Diagnostic Sciences, College of Veterinary Medicine, Cornell University, Ithaca, NY, <sup>2</sup>Department of Animal Science, College of Agriculture and Life Sciences, Cornell University, Ithaca, NY, <sup>3</sup>Department of Clinical Sciences, College of Veterinary Medicine, Cornell University, Ithaca, NY, <sup>4</sup>Department of Biomedical Sciences, College of Veterinary Medicine, Cornell University, Ithaca, NY.

Overfeeding energy during the dry period may affect peripartum insulin sensitivity of dairy cows. The objectives were to describe the effect of different dry period dietary energy strategies on glucose tolerance and on concentrations of insulin, glucagon, glucose,  $\beta$ -hydroxybutyrate (BHBA) and nonesterified fatty acids (NEFA) in the peripartum period. To test our hypothesis that dry period plane of energy leads to changes in glucose tolerance, cows ( $n = 84$ ) were blocked by expected calving date and randomized into 3 treatment groups 57 d before expected parturition: a TMR formulated to supply 100% of energy requirements (C), or 150% of requirements (H), or 100% for the first 28 d of the dry period followed by a TMR supplying 125% of energy requirements until calving (I). At 28 and 10 d pre-, as well as 4 and 21 d postpartum, intravenous glucose tolerance tests (IVGTT; 0.25 g glucose/kg) were performed and the area under the curve (AUC) of glucose and insulin were estimated. Baseline samples were analyzed for concentration of insulin, glucose, NEFA, glucagon and BHBA. Mixed-effects ANOVA were carried out (SAS, v. 9.3). The AUC for glucose and insulin was not different among groups at any time point ( $P > 0.10$ ). Differences in baseline values were most notable on d 4 postpartum (Table 1) and indicated increased postpartum lipolysis and ketogenesis in group H whereas we detected a tendency for lower glucose concentration despite higher glucagon concentration in this group. In conclusion, these findings suggest that prepartum plane of energy had an effect on the hormonal regulation of gluconeogenesis and ketogenesis (as reflected by the concentrations of insulin and glucagon), but did not lead to a detectable effect on peripartum glucose tolerance as assessed by IVGTT.

*Contd.*

**Table 1 (Abstr. 280).** Mean (95% CI) baseline concentrations on day 4 postpartum

Item	C	I	H	P-value
Glucose (mg/dL)	60 (57–62) <sup>a</sup>	58 (56–61) <sup>ab</sup>	56 (53–58) <sup>b</sup>	0.09
Insulin ( $\mu$ IU/mL)	2.1 (1.2–3.6) <sup>a</sup>	2.3 (1.3–4.1) <sup>a</sup>	1.1 (0.5–1.7) <sup>b</sup>	0.08
Glucagon (pg/mL)	120 (108–132) <sup>A</sup>	117 (105–129) <sup>A</sup>	137 (127–150) <sup>B</sup>	0.01
NEFA (mEq/L)	1.37 (1.26–1.48) <sup>A</sup>	1.38 (1.27–1.50) <sup>A</sup>	1.63 (1.52–1.74) <sup>B</sup>	0.002
BHBA (mmol/L)	0.75 (0.64–0.88) <sup>A</sup>	0.83 (0.71–0.97) <sup>AB</sup>	1.0 (0.85–1.2) <sup>B</sup>	0.06

Row values with different superscript letters differ (<sup>A,B</sup> $P < 0.05$ ; <sup>a,b</sup> $P < 0.10$ ) in Tukey's HSD.

**Key Words:** transition, energy, dairy

**281 Infusion of 5-hydroxytryptophan increases serum calcium and mammary gland calcium pump activity during the transition period.** Samantha R. Weaver\*<sup>1</sup>, Austin P. Prichard<sup>1</sup>, Elizabeth L. Endres<sup>1</sup>, Stefanie A. Newhouse<sup>2</sup>, Rupert M. Bruckmaier<sup>3</sup>, Matt S. Akins<sup>1</sup>, and Laura L. Hernandez<sup>1</sup>, <sup>1</sup>University of Wisconsin-Madison, Madison, WI, <sup>2</sup>University of Wisconsin-Platteville, Platteville, WI, <sup>3</sup>University of Bern, Bern, Switzerland.

Hypocalcemia during the transition period in dairy cows has detrimental effects on animal health, welfare, and production. While clinical hypocalcemia affects 2 to 5% of cows in the US, approximately 50% of cows succumb to subclinical hypocalcemia. Serotonin (5-HT) has been suggested as a therapeutic target for prevention of hypocalcemia. Our objective was to determine the effects of pre-partum intravenous (IV) administration of a 5-HT precursor on calcium homeostasis postpartum in multiparous dairy cows. We hypothesized that the treatment would increase serum calcium and calcium transport into the mammary gland. Twelve (avg. lactation number  $3.67 \pm 0.43$ ) Holstein cows were IV infused for  $5.75 \pm 0.82$  d pre-partum, beginning approximately 7d before their predicted calving date until calving, with saline (CTL;  $n = 6$ ) or 1.0 mg/kg 5-hydroxytryptophan (5-HTP;  $n = 6$ ), the immediate precursor for 5-HT synthesis. Mammary gland biopsies were performed approximately 2 weeks pre-partum, and d1 and d7 postpartum. Blood and urine were collected daily from the first biopsy through d14 and on d30 of lactation. Colorimetric assays were performed for total calcium in serum and relative mammary mRNA expression was evaluated by RT-PCR. All statistical analysis was performed in SAS using a mixed model ANOVA. Cows infused with 5-HTP had decreased feed intake postpartum compared with CTL ( $P = 0.0004$ ;  $34.75 \pm 1.6$  kg CTL vs.  $30.25 \pm 2.8$  kg 5-HTP) and overall decreased milk yield ( $P = 0.0054$ ;  $18.35 \pm 1.07$  kg CTL vs.  $17.10 \pm 1.04$  kg 5-HTP), although colostrum milk yield was not different ( $P = 0.88$ ). Serum total calcium tended to increase in 5-HTP cows for 14d postpartum ( $P = 0.07$ ;  $2.89 \pm 0.09$  mM 5-HTP vs.  $2.66 \pm 0.09$  mM CTL). Basolateral mammary epithelial cell calcium sensing receptor (CaSR) mRNA was increased in 5-HTP compared with CTL cows ( $P = 0.035$ ), as was apical calcium pump plasma membrane calcium ATPase2 (PMCA2) ( $P = 0.018$ ) on d 1 and d 7 of lactation. These results suggest that 5-HTP treatment prepartum increases postpartum circulating calcium concentrations and calcium transport in the mammary gland.

**Key Words:** serotonin, calcium, hypocalcemia



**282 The use of gene expression in milk fat as an indicator of trace mineral status in dairy cows.** M. J. Faulkner<sup>1</sup>, E. H. Wall<sup>2</sup>, and W. P. Weiss<sup>1</sup>, <sup>1</sup>OARDC, The Ohio State University, Wooster, OH, <sup>2</sup>Pancosma, Geneva, Switzerland.

No non-invasive methods exist to accurately evaluate Cu, Zn, and Mn status in dairy cows. Thirty lactating Holstein cows were used to determine whether intake and source of Cu, Zn, and Mn affected erythrocyte Cu/Zn SOD activity and expression of genes in milk fat that are related to Cu and Zn transport. Cows were fed a diet void of supplemental Cu, Zn, and Mn (9, 41, and 41 mg/kg, respectively) for 30 d and then fed 1 of 3 diets for 30 d. One diet (UNSUP) contained no supplemental Cu, Zn, and Mn (9, 41, and 41 mg/kg); one diet (SUL) contained Cu, Zn, and Mn from sulfates (total concentrations = 17, 59, and 54 mg/kg, respectively); one diet (GLY) contained Cu, Zn, and Mn in the glycinate form (B-Traxim 2C, Pancosma; total concentrations = 20, 66, and 58 mg/kg). Data were analyzed using a mixed model with treatment and parity as fixed effects. Using the NRC (2001) model and absorption coefficients (AC), UNSUP provided about 82% of requirements for Cu and Zn for primiparous cows and 95% for multiparous cows. Assuming an AC of 0.05 and 0.20 for Cu and Zn from supplements, supplemented diets provided 1.3 to 2.3 times more absorbed Cu and Zn than requirements. Total RNA was extracted using Tri Reagent from milkfat samples collected on d 60. Expression of 1 Zn transport gene (ZnT4), 2 Zn and Mn transport genes (Zip3 and Zip8), 2 Cu transport genes (ATP7B and CCS), 1 heavy-metal binding gene (MT), and 2 reference genes (KEAP1 and ADSL) were analyzed using qPCR. Activity of SOD was higher ( $P = 0.04$ ) in primiparous than multiparous cows, but was not affected by diet. Expression of CCS, a protein that transports Cu to SOD, and SOD were negatively correlated ( $P < 0.06$ ). Expression of most genes was not affected by treatment; however, MT expression was greater ( $P = 0.04$ ) in primiparous compared with multiparous cows, and Zip8 expression tended ( $P = 0.10$ ) to be greater in cows fed supplemental Cu, Zn, and Mn regardless of source. When supplemental Cu, Zn, and Mn were fed, cow requirements were exceeded and no differences were observed between mineral sources. Zip8 has potential to detect cows with deficient trace mineral status.

**Key Words:** trace mineral, dairy cow, gene expression

**283 Feed efficiency and performance of lactating Holstein dairy cows fed two different concentrations of dried distillers grains with solubles.** Eric D. Testroet\*, Stephanie Clark, and Donald C. Beitz, Iowa State University, Ames, IA.

Dried distillers grains with solubles (DDGS) has become more nutritionally consistent as the industry has evolved. The objective of this study, therefore, was to evaluate the feed efficiency of lactating Holstein dairy cattle fed 2 different concentrations of DDGS. Thirty cows were fed 0, 10, and 20% DDGS dietary dry matter (DM) as a total mixed ration. Cows were stratified by parity and days in milk into 3 treatment groups of 10 cows in a 3 × 3 Latin square with repeated measures. We hypothesized that feeding 20% DDGS by dietary DM would negatively influence production and efficiency. Feeding DDGS had no consistent effect on dry matter intake (DMI), with the control diet being intermediate to the 10 and 20% diets (25.22, 24.03, and 26.37 kg/day, respectively;  $P < 0.0001$ ). Milk yield was unaffected by feeding DDGS; feeding DDGS, however, caused milk fat depression (3.45, 2.94, and 2.68%; 0, 10, and 20%, respectively;  $P < 0.0001$ ) and reduction of daily fat yield (1.35, 1.15, 1.04 kg/day;  $P < 0.0001$ ) that resulted in decreases in 3.5% fat-corrected milk (FCM) yield (36.20, 32.97, and 31.32 kg/day;  $P < 0.0001$ ) and energy-corrected milk (ECM) yield (40.95, 38.12, and 36.72 kg/day;  $P < 0.0001$ ). Both protein (3.58, 3.62, and 3.65%;  $P =$

0.0409) and lactose percentage (5.07, 5.15, and 5.17%;  $P = 0.0067$ ) were increased when cows were fed 20% DDGS; neither protein nor lactose yield, however, was affected. Protein efficiency decreased for cows fed 20% DDGS (35.42, 36.21, and 32.76%;  $P < 0.0001$ ), likely resulting from heat-damaged protein. All measures of energetic efficiency [ECM/DMI, kg ECM/net energy for lactation (NE<sub>L</sub>) intake (mcal), and gross energy (GE) of milk produced (mcal)/NE<sub>L</sub> caloric intake (mcal)] were decreased when cows were fed 20% DDGS, but not when cows were fed 10% DDGS (ECM/DMI: 1.66, 1.64, and 1.41; kg ECM/NE<sub>L</sub>: 1.02, 1.03, 0.89; GE of milk/NE<sub>L</sub> caloric intake: 0.65, 0.66, 0.56;  $P < 0.0001$ ). These results indicate that, with the exception of an approximate 0.5% loss of milk fat, the DDGS used in this study can be effectively fed at 10%, but not at 20%, of dietary DM without a loss in feed efficiency.

**Key Words:** corn, feed intake, milk

**284 Including sunflower seed in prepartum diet positively influenced postpartum ovarian function without affecting uterine health.** R. Salehi<sup>1</sup>, M. G. Colazo<sup>2</sup>, U. Basu<sup>1</sup>, A. Ruiz-Sanchez<sup>1</sup>, and D. J. Ambrose<sup>1,2</sup>, <sup>1</sup>University of Alberta, Edmonton, Alberta, Canada, <sup>2</sup>Alberta Agriculture and Rural Development, Edmonton, Alberta, Canada.

We investigated the influence of prepartum dietary oilseed supplementation on postpartum (pp) uterine inflammatory status (UIS) based on polymorphonuclear cells (PMN): normal ( $\leq 8\%$  PMN) or subclinical endometritis (SCE;  $> 8\%$  PMN), and associations among SCE, pro/anti-inflammatory cytokine gene expression and resumption of ovarian cyclicity. We hypothesized that feeding a diet containing sunflower seed (SUN; high linoleic acid) will induce pro-inflammatory effects in the early pp uterus facilitating early resumption of cyclicity. During late gestation (last 5 wk), Holstein cows received 1 of 3 diets supplemented with 8% DM rolled sunflower seed (SUN;  $n = 10$ ) or canola seed (CAN; high oleic acid;  $n = 9$ ), or no oilseed (CON;  $n = 9$ ). Ovaries were scanned 2×/wk until 35 d pp to record first appearance of a 10 (DF) or 16 mm (PreOVF) follicle, and ovulation. Endometrial cell sampling was done  $25 \pm 1$  d pp for cytology and gene expression. Data were analyzed using PROC MIXED, CORR and REG of SAS. Prepartum diets did not influence SCE incidence (SUN: 4/10; CAN: 4/9; CON: 3/9). The mRNA expressions of interleukin (IL)-1 $\beta$ , IL-6, IL-8, IL-10, tumor necrosis factor (TNF)- $\alpha$  and interferon (INF)- $\gamma$  were not affected ( $P > 0.05$ ) by either prepartum diet or diet × UIS interaction. Feeding SUN prepartum hastened ( $P < 0.05$ ) the establishment of PreOVF and increased the proportion of cows ovulating within 35 d pp. Regardless of prepartum diet, cows with SCE had higher ( $P < 0.05$ ) mRNA expression of IL-1 $\beta$ , IL-8, IL-10 and TNF- $\alpha$  than normal cows. The intervals from calving to DF formation and first ovulation were not affected by UIS; however, PreOVF formed earlier ( $P < 0.01$ ) in normal ( $13.2 \pm 0.9$ ) than in SCE ( $18.7 \pm 1.4$  d) cows. The mRNA expression of IL-1 $\beta$ , IL-8, IL-10 and TNF- $\alpha$  were positively correlated with each other ( $P < 0.01$ ) and with PMN% ( $P < 0.07$ ). The interval to PreOVF was determined by IL-1 $\beta$ , IL-8, IL-10, INF- $\gamma$  expression, PMN%, and the interval to DF. In summary, feeding SUN prepartum positively influenced pp ovarian function without affecting UIS. Increased endometrial expression of pro-inflammatory cytokines is likely linked to delayed establishment of PreOVF.

**Key Words:** endometritis, ovarian function, cytokine

**285 Pretreatment with saturated and unsaturated fatty acids regulates [1-<sup>14</sup>C] C16:0 metabolism in Madin-Darby bovine kidney cells.** Katherine E. Boesche\*, Stephanie L. Koser, and Shawn S. Donkin, *Purdue University, West Lafayette, IN.*

Metabolic fates of fatty acids (FA) may be influenced by circulating FA concentration. Previous work in our lab demonstrated an ability of C18:3n-3 *cis* to ameliorate gene expression of pyruvate carboxylase (PC) after depression by either C16:0 or C18:0. PC catalyzes oxaloacetate (OAA) synthesis and ostensibly links gluconeogenesis and FA metabolism. Our objective was to determine effects of co-presence of saturated and unsaturated FA pretreatments on cellular partitioning of [1-<sup>14</sup>C] C16:0 metabolism to CO<sub>2</sub> or acid-soluble products (ASP) in Madin-Darby bovine kidney (MDBK) cells. We hypothesized that the ratio of saturated to unsaturated FA pretreatments regulates [1-<sup>14</sup>C] C16:0 partitioning to CO<sub>2</sub> or ASP. Cells at 80% confluence were exposed for 21h to either individual FA bound to BSA (C16:0, C18:0, C18:1n-9 *cis* or C18:3n-3 *cis*) or FA cocktails in 10:90, 25:75, 50:50, 75:25 or 90:10 ratios for combinations of C16:0: C18:3n-3 *cis* or C18:0: C18:3n-3 *cis* or C18:1n-9 *cis*: C18:3n-3 *cis*. Total pretreatment FA concentration was 1.0 mM and was applied in triplicate to 3 cell replicates. Following pretreatment, cells were incubated in the presence of 1.0 mM [1-<sup>14</sup>C] C16:0 for 3h before CO<sub>2</sub> and ASP collection. Data were analyzed using PROC MIXED of SAS. The model accounted for fixed effects of pretreatment and random effects of replicate. Pretreatments with either C16:0 or C18:0 alone significantly ( $P < 0.01$ ) depressed subsequent oxidation of [1-<sup>14</sup>C] C16:0 to ASP by 62.7% and 41.2%, respectively, compared with C18:3n-3 *cis* pretreatments. Similar patterns were seen with [1-<sup>14</sup>C] C16:0 oxidation to CO<sub>2</sub>. ASP production from [1-<sup>14</sup>C] C16:0 positively correlated ( $r = 0.68$ ,  $P < 0.01$ ) with PC gene expression from previous experiments. CO<sub>2</sub> production from [1-<sup>14</sup>C] C16:0 did not correlate ( $r = 0.30$ ,  $P > 0.10$ ) with PC expression. In conclusion, activation of PC gene expression by C18:3n-3 *cis* may play a critical role in setting the capacity for OAA synthesis and determining metabolic fates of FA. Results show a regulation of ketone production by MDBK cells in response to saturated and unsaturated FA pretreatments.

**Key Words:** fatty acid oxidation, ketogenesis, pyruvate carboxylase

**286 A novel method to determine rumen biohydrogenation kinetics of alpha-linolenic acid (18:3 n-3).** Michel Baldin\*<sup>1</sup>, Natalie L. Urrutia<sup>1</sup>, Daniel E. Rico<sup>2</sup>, Kelsie Baxter<sup>1</sup>, Yun Ying<sup>1</sup>, and Kevin J. Harvatine<sup>1</sup>, <sup>1</sup>*Penn State University, University Park, PA*, <sup>2</sup>*Université Laval, Québec, QC, Canada.*

Biohydrogenation (BH) of unsaturated fatty acids (FA) has been extensively studied in vitro but BH rates and intermediates formed in vitro may not parallel BH pathways in vivo. The objective was to develop an in vivo method to determine the rate of  $\alpha$ -linolenic acid (18:3 n-3) BH and identify intermediates formed. Eleven rumen cannulated high-producing Holstein cows [40  $\pm$  6 kg milk/d (Mean  $\pm$  SD)] were fed at a rate of 6%/h of expected total DMI a diet balanced to 29% NDF and 5.9% EE (1.5% soybean oil). A single bolus consisting of 200 g of flaxseed oil (53% 18:3) and 15 g of tridecanoic acid (13:0) was mixed with rumen contents and rumen digesta was collected at -1, 0.1, 0.5, 1, 2, 3, 4, 6 and 8 h relative to the bolus. Samples were immediately placed in dry ice, stored at -20°C, freeze-dried, methylated and analyzed by gas chromatography. Data were first analyzed using PROC Mixed with repeated measures for time point comparison. Second, the disappearance of 13:0 and 18:3 was fit to a single exponential decay model using the nonlinear procedure of JMP Pro. The bolus increased total fat in the rumen from 4.3 to 6.0% and enriched 13:0 from 0.04 to 2.2% of FA and 18:3 from 2.0 to 11.3% of FA. The fractional rate of

disappearance of 13:0 was 0.4%/min ( $r^2 = 0.98$ ) and of 18:3 was 2.5%/min ( $r^2 = 0.99$ ), with 18:3 reaching pre-bolus concentration within 4 h. Assuming that 13:0 disappeared only by passage, 18:3 disappeared by passage and biohydrogenation, and the rate of passage of 13:0 and 18:3 are the same, the extent of bolused 18:3 BH was 85%. The concentration of *cis*-9,*trans*-11,*cis*-15 18:3 peaked at 1.2% of FA at 1 h (8-fold increase), *trans*-11, *cis*-15 18:2 peaked at 3.9% of FA at 2 h (13-fold increase), and *trans*-11 18:1 peaked at 6.6% FA at 3 h (43% increase). In conclusion, the in vivo method resulted in the expected extent of biohydrogenation and biohydrogenation intermediates, but the rate of ruminal biohydrogenation of 18:3 was much higher than that commonly observed in vitro. The method developed provides an in vivo assay of ruminal biohydrogenation for use in future experiments.

**Key Words:** biohydrogenation, fatty acid

**287 Effects of feeding algae rich in docosahexaenoic acid (DHA) on lactation and reproductive performance of dairy cows.** Leticia D. P. Sinedino\*<sup>1</sup>, Thiago F. Fabris<sup>1</sup>, Cesar D. Narciso<sup>2</sup>, Leticia R. Lucena<sup>1</sup>, Paula M. Honda<sup>1</sup>, Gilson G. Maia<sup>1</sup>, Maurice P. Boland<sup>3</sup>, Charles R. Staples<sup>1</sup>, William W. Thatcher<sup>1</sup>, and Jose E. P. Santos<sup>1</sup>, <sup>1</sup>*University of Florida, Gainesville, FL*, <sup>2</sup>*Sequoia Veterinary Services, Tulare, CA*, <sup>3</sup>*Alltech Animal Nutrition and Health, Nicholasville, KY.*

Previous work by our group showed that supplementing diets with Ca salts containing fish oil fatty acids improved pregnancy per AI (P/AI) in dairy cows. Objectives were to determine the effects of supplementing DHA by feeding algae on lactation and reproductive performance of Holstein cows. The hypothesis was that feeding DHA improves P/AI in dairy cattle. Weekly cohorts of cows were blocked by parity and randomly assigned at 27 DIM to a control [CON; n = 372, 115 primiparous and 258 multiparous] or an algae supplemented (ALG; n = 366, 105 primiparous and 261 multiparous) diet. Cows were fed the same TMR; however, ALG received 100 g/cow/d of a 17% DHA supplement top-dressed and hand-mixed onto the TMR for 120 d. Milk yield was recorded daily and sampled every 3 wk for analysis. Cows were subjected to the Presynch-Ovsynch protocol starting at 44  $\pm$  3 DIM and those in estrus after 58  $\pm$  3 DIM were inseminated on the same day, otherwise, they received timed AI at 80  $\pm$  3 DIM. Pregnancy was diagnosed on d 60 after AI. Data were analyzed by ANOVA, logistic regression or the Cox's proportional hazard model using the GLIMMIX and PHREG procedures of SAS. Intake of DM did not differ between treatments (ALG = 24.8 vs. CON = 25.5  $\pm$  0.6 kg/d). Milk yield was greater ( $P = 0.01$ ) in ALG than CON (43.6 vs. 42.5  $\pm$  0.3), but yield of energy-corrected milk was similar between treatments and averaged 39.2  $\pm$  0.2 kg/d. Feeding ALG reduced ( $P = 0.03$ ) content and yield of milk fat (3.08 vs. 3.23  $\pm$  0.03% and 1.30 vs. 1.34  $\pm$  0.01 kg/d); however, ALG increased ( $P < 0.01$ ) yields of protein (1.23 vs. 1.20  $\pm$  0.01 kg/d) and lactose (2.07 vs. 2.02  $\pm$  0.01 kg/d). Feeding ALG increased ( $P = 0.04$ ) the proportion of primiparous cows that resumed estrous cyclicity by 58 DIM (77.6 vs. 65.9%) and P/AI at first AI (47.6 vs. 32.8%). Feeding ALG increased ( $P < 0.01$ ) P/AI at all AI in all cows (41.4 vs. 30.8%). Cows fed ALG became pregnant 21 d (103 vs. 124 d) sooner ( $P < 0.01$ ) than those fed Control (adjusted HR = 1.38; 95% CI = 1.13 to 1.69). Supplementing DHA by feeding ALG did not affect intake of DM, increased yields of milk and protein, but reduced milk fat content and yield. Cows fed ALG had improved estrous cyclicity, P/AI, and reduced interval to pregnancy.

**Key Words:** dairy cow, docosahexaenoic acid, reproduction

# Graduate Student Competition: ADSA-ASAS Northeast Section

## Graduate Student Oral Competition

### 288 Assessment of acute pain during and after knife and band castration of beef calves at three different industry-relevant ages.

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The aim of this study was to identify which age and method of castration causes less acute pain and distress in beef calves. One hundred five Angus bull calves were blocked by age and body weight, and randomly assigned to 1 of 3 treatments: control (C; n = 12); band castration (B; n = 12); and surgical castration (S; n = 12); at 1 wk, 2 and 4 mo of age. Physiological and behavioral parameters were collected before, during and after castration to assess acute pain. Physiological measures included blood count, salivary cortisol, haptoglobin, substance P and infrared thermography. Behavioral measures consisted of visual analog score, stride length, hobo data loggers and behavioral scoring of walking, standing, lying, tail flick, foot stamping and head turning. Overall, no physiological or behavioral parameters differed significantly for calves castrated at 1 wk of age. Salivary cortisol tended ( $P = 0.07$ ) to differ at 1 wk of age when S calves had greater concentrations than B calves. Conversely, both physiological and behavioral indicators of pain/distress were clearly observed when calves were castrated at 2 and 4 mo of age regardless of the method used. A time  $\times$  treatment interaction ( $P < 0.0001$ ) was observed for salivary cortisol in 4 mo old calves, with B and S calves having greater concentrations than C calves 60 min after castration, while B had greater concentrations than S and C calves 120 min after castration. Based on behavioral data, S calves at 2 and 4 mo of age, stood ( $P < 0.0001$ ) and walked ( $P = 0.04$ ) more but lie down and ate less ( $P = 0.01$ ;  $P = 0.002$ ) compared with B and C only at 2 mo of age. At 4 mo of age, S calves tail flicked more ( $P = 0.0006$ ) and had shorter stride length ( $P = 0.036$ ) than B and C calves. The indicators of acute pain/discomfort assessed in this study suggest that the most welfare-friendly age and method of castration is band castration at 1 wk of age.

**Key Words:** castration, pain, beef

### 289 The influence of dietary strong ions on rumen ion concentrations.

Tracy L. Catterton\* and Richard A. Erdman, *Department of Animal and Avian Sciences, University of Maryland, College Park, MD*.

Dietary cation-anion difference (DCAD) is being used as a basis for diet formulation in the dairy industry. While extensive research has been conducted on the effects of DCAD concentration on animal performance and rumen characteristics, the effects of the individual strong ions that contribute to DCAD have yet to be determined. In addition to DCAD, there is evidence that cation source (Na vs. K) affects animal performance (Iwaniuk et al., 2015). We hypothesized that dietary strong ion concentrations (Na, K, and Cl) would affect the rumen ion concentrations. Our objective was to determine if rumen Na, K, and Cl concentrations were affected by dietary mineral content. Literature data were collected from 2 dairy cattle studies (Bailey, 1961; Bennink et al., 1978) that included 11 different diets with (mean  $\pm$  SE) concentrations of  $47 \pm 35$ ,  $358 \pm 312$ , and  $108 \pm 188$  mEq/kg dietary Na, K, and Cl, respectively. Rumen samples, collected by rumen cannula, were taken between 0 to 9 h (Bennink et al., 1978) and 1 to 14 h post-feeding (Bailey, 1961). Principal components analysis (PCA) suggested that rumen Na

and K were PCA negative, while rumen Cl and diet K clustered together. Subsequent multiple regression analysis (PROC GLMSELECT) showed that dietary K and Cl were significant factors ( $P < 0.01$ ) associated with changes in rumen Na, K, and Cl, and diet Na was associated ( $P \leq 0.06$ ) with changes in rumen Na and K. As rumen K increased, rumen Na decreased ( $P < 0.001$ ). We concluded that manipulation of diet strong ion concentrations could be used to alter rumen Na, K, and Cl concentrations and, thus, alter the rumen environment.

**Table 1 (Abstr. 289).**

Item	Rumen ion		
	Na	K	Cl
Regression statistics			
Mean, mEq/L	122.0	46.9	18.7
Root MSE	17.4	11.8	4.04
Adj. R <sup>2</sup>	0.61	0.60	0.63
Regression coefficients			
Intercept, mEq/L	151.9	63.4	9.35
Diet Na, mEq/kg	0.254	-0.83	NS
Diet K, mEq/kg	-0.07	0.042	0.251
Diet Cl, mEq/kg	0.064	-0.08	-0.193
Rumen Na, mEq/L	—	-0.21	NS
Rumen K, mEq/L	-0.47	—	NS
Rumen Cl, mEq/L	NS	0.22	—
Time post-feeding, h	NS	NS	-0.38

**Key Words:** rumen environment, strong ions, dairy cattle

### 290 Disbudding and dehorning practices in dairy calves among Ontario bovine veterinarians.

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An online survey was conducted in the fall of 2014 to explore current practices, including analgesic use, in the disbudding or dehorning (D/D) of dairy calves by veterinarians in Ontario. Members of the Ontario Association of Bovine Practitioners (n = 238) were invited to participate. Ninety-three veterinarians (39%) from 51 clinics (63%) responded, with 94% of clinics reporting veterinarians or veterinary technicians (VT) performing D/D for an estimated mean of 29% (SD = 20) of their dairy clients. Twenty-five percent of clinics employed VT to perform this service. Of these clinics, VT accounted for an estimated mean of 69% (SD = 29) of the clinic's total D/D. Veterinarians who perform D/D were asked to report calf age at time of D/D. Seventy-five percent of veterinarians disbudded calves < 4 weeks of age. Of these veterinarians, 99% used local anesthetic, 56% used a sedative, and 50% used an NSAID. Ninety-one percent of veterinarians disbudded calves 4–8 weeks of age. Of these veterinarians, 99% used a local anesthetic, 61% used a sedative, and 54% used an NSAID. Finally, 76% of vets performed D/D in calves > 8 weeks of age. Of these veterinarians, 97% used a local anesthetic, 66% used a sedative, and 59% used an NSAID. Injectable meloxicam accounted for 89% of all NSAIDs given in all age groups. Common reasons for NSAID use were: pain control, known withdrawal time, and reasonable cost. Common reasons for the lack of NSAID use



were: objections to cost, and client requests excluding cost. Nearly all (98%) veterinarians practicing for at least 10 years reported changing D/D practices over this time period. Common changes included: use or increased use of an NSAID (60%), use or increased use of sedation (34%), and use or increased use of local anesthetic (29%). Reasons for changes included: concern for the welfare of the calf, information from continuing education, and improved calf handling. Use of NSAID in this survey is higher than previously reported, as was the proportion of calves disbudded < 4 weeks of age. This indicates a trend toward improved analgesia and adoption of best practices for welfare in the D/D of dairy calves.

**Key Words:** calf, disbud, welfare

**291 Identification of early pregnancy and fetal landmarks via transabdominal ultrasound in sheep.** Amanda K. Jones\*<sup>1</sup>, Rachael E. Gately<sup>2</sup>, Katelyn K. McFadden<sup>1</sup>, Steven A. Zinn<sup>1</sup>, Kristen E. Govoni<sup>1</sup>, and Sarah A. Reed<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Connecticut, Storrs, CT, <sup>2</sup>Department of Environmental and Population Health, Tufts Cummings School of Veterinary Medicine, North Grafton, MA.

Field application of small ruminant ultrasound is primarily used for pregnancy detection and fetal number beginning at mid-gestation. Efforts to estimate fetal age and number early in small ruminant pregnancies are limited. We hypothesized that detection of pregnancy and fetal landmarks before d 45 via transabdominal ultrasound is reliable in the sheep, and may be used to determine fetal age during early in sheep. To test this hypothesis, 106 Western Whiteface ewes were exposed to 1 of 4 rams. The day a ewe was marked by a ram was considered d 0 of pregnancy. Transabdominal ultrasound (Easi-Scan, BCF Technology, Rochester, MN) was performed 3 times/wk with a 5 MHz rectal transducer in the right non-haired abdominal region of the ewe starting at d 26.0 ± 0.3 (range d 22 to d 30). Pregnancy was confirmed in 88 ewes, with the remaining identified as non-pregnant. Fluid-filled uterine cross-sections provided first evidence of pregnancy from d 26.0 ± 2.9 onward. Pregnancy was confirmed by the presence of a fetus with a heartbeat on d 28.5 ± 0.4. Singleton pregnancies were detected later than multiple pregnancies (singletons: d 31.2 ± 0.9, twins: d 27.6 ± 0.9, triplets: d 26.3 ± 1.0,  $P < 0.01$ ). The uterine horn of fetal origin had no effect on identifying singleton pregnancies ( $P = 0.52$ ). Placentome evagination was first observed at d 33.8 ± 0.4, separation of limb buds at d 35.2 ± 0.7, fetal genitalia spots at d 37.9 ± 0.7, mature placentomes at d 40.6 ± 0.4, and ribs at d 42.9 ± 1.4. Additionally, 3 fetal losses were identified by d 40.0 ± 0.7 of pregnancy (3.4% early embryonic loss rate). Accuracy of counting fetuses (77.6%) increased with decreasing number of offspring. That is, identification of singletons, twins and triplets was 95, 80 and 41%, respectively. Pregnancy detection via transabdominal ultrasound was accurate as early as d 28 in sheep and sensitive enough to consistently identify early embryonic developments before d 45. Distinguishing these fetal landmarks may allow for estimating fetal age to improve early detection of pregnancy and breeding management of sheep.

**Key Words:** ultrasound, pregnancy, sheep

**292 Isolation and characterization of chemical components of *Leucaena leucocephala* with anti-methanogenic properties by using in vitro gas production technique.** D. Dineshkumar\*<sup>1</sup>, A. L. Abdalla<sup>1</sup>, C. L. Linander<sup>1</sup>, A. P. Massarioli<sup>2</sup>, A. L. Abdalla Filho<sup>1</sup>, P. P. Santos<sup>1</sup>, A. S. Natel<sup>1</sup>, S. M. Alencar<sup>2</sup>, and H. Louvandini<sup>1</sup>, <sup>1</sup>Centre for Nuclear Energy in Agriculture, University of Sao Paulo,

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Studies emphasized that group of plant secondary metabolites (saponins, flavonoids and tannins) seems to present the ability to manipulate rumen fermentation lessening the CH<sub>4</sub> formation. The purposes of this research were to find out individual bioactive compound with anti-methanogenic activity. However, we anticipate that advances technology such as GC-MS will provide unprecedented data on the distribution of component existing in plant extracts. *Leucaena leucocephala* plant samples were extracted with methanol solvent using ultra sonication. Crude solvent free extract (8.58 g) was then extracted with different organic solvent with increasing polarity provided extracts of hexane (1.03 g), chloroform (0.34 g), ethyl acetate (0.48 g), butanol (0.77 g) and residual crude fractions (1.99 g) respectively. Assessment of phytoconstituents in such organic extracts was subjected to find out individual bioactive compound with the modified GC-MS (Shimadzu gas chromatography-mass spectrometer (GCMS-QP2010) method. 38 components were identified from the chromatograms of the different organic solvent extracts. Dried 0.5g of ground leucena plant, alfafa plant (positive control) and the different crude methanolic extracts with 3 different levels (125, 250, and 500 µg/mL) were tested for anti-methanogenic properties in terms of in vitro gas production and nutrient degradability. We found significant organic solvent effects for CH<sub>4</sub> production. Hexane extract reduces Net CH<sub>4</sub>/OMD ( $P < 0.001$ ) compared with other solvent extracts tested. Nutrient degradability, ruminal parameters and VFA production were non-significantly differed between the treatment groups. But, there was an improvement on nutrient degradability compared with *Leucaena leucocephala* and control plants. However, we found no doses and interaction between solvent and dose effects among the treatment groups. This study explained hexane extract from whole plant methanolic extract is effective against anti-methanogenic activity in modifying ruminal degradation of nutrients. The most active components still have to be identifying by fractionation of hexane extract.

**Key Words:** *Leucaena leucocephala*, anti-methanogenic properties, GC-MS

**293 Immune cells populate mesenteric adipose tissues of Holstein Friesian cows.** Bridget A. Aylward\*<sup>1</sup>, Megan Clark<sup>1</sup>, Amanda Barnard<sup>1</sup>, Jen Wilson<sup>1</sup>, Candice Gittens<sup>1</sup>, Tanya Gressley<sup>1</sup>, Erin Branick<sup>1</sup>, Marie Fecteau<sup>2</sup>, and Robert Dyer<sup>1</sup>, <sup>1</sup>Department of Animal and Food Sciences, College of Agricultural and Natural Resources, University of Delaware, Newark, DE, <sup>2</sup>Department of Clinical Studies, New Bolton Center, University of Pennsylvania, School of Veterinary Medicine, Kennet Square, PA.

In many animal species, anti-inflammatory immune cells are normal residents of lean adipose depots and produce a barrier of related cytokines that protect against metabolic syndrome. Our objective was to determine if similar immune cells reside in mid-jejunum mesenteric adipose tissue (MAT) of randomly selected lean Holstein Friesian cows from an abattoir. Body condition scores were determined before slaughter (mean BCS = 2.74). Stromal cell fractions (SCF) were prepared from washed, minced MAT digested with type I collagenase. Following digestion and cleaning of the sample, cells were stained with bovine immune cell marker specific monoclonal antibodies expressed on macrophages, dendritic cells, T lymphocytes and T regulatory lymphocytes (T regs). Background controls consisted of cells stained with irrelevant, isotype-matched control antibodies. SCF composition was analyzed across 10,000 cells/sample using a Becton Dickinson FACS Calibur Flow Cytometer. Marker specific fluorescence was compared with background fluorescence in controls by ANOVA. Marker expression is summarized

in the table below. Results from this work indicate that populations of innate (macrophage and dendritic cells) and adaptive (lymphocytes and T regs) immune response cells do in fact reside in MAT of lean cows (Table 1). Furthermore, expression of MHC class II in the context of dendritic cells or macrophages and T lymphocytes suggests adaptive immune responses do occur in MAT. Subpopulations of FoxP3<sup>+</sup> regulatory T lymphocytes were the dominant effector lymphocyte, implying that anti-inflammatory functions could contribute to MAT homeostasis in lean cows and potentially protect against development of metabolic disease.

**Table 1 (Abstr. 293).** Presence of immune cells mesenteric adipose tissue (MAT) of Holstein Friesian cows

Presumptive cell type	Marker	Percent of SCF
Macrophages	CD11b (n=9)	8.71 ± 0.75*
	CD172 (n=8)	12.57 ± 1.20*
	CD11b/CD172 (n=8)	6.74 ± 0.84*
	CD3/CD11b (n=4)	2.48 ± 0.62
Dendritic cells	MHC II (n=5)	10.62 ± 0.86*
	CD11c (n=9)	9.43 ± 0.57*
	MHC II/CD11c (n=5)	2.59 ± 0.52
	CD11c/CD3 (n=4)	1.76 ± 0.44
T lymphocytes	CD3 (n=17)	8.99 ± 0.33*
	CD8 (n=4)	3.58 ± 0.89
	CD3/CD8 (n=4)	1.07 ± 0.27
T regulatory lymphocytes	FoxP3 (n=6)	8.00 ± 0.56*
	CD4/FOXP3(n=6)	0.04 ± 0.12

\*Expression greater than background ( $P \leq 0.05$ ).

**Key Words:** immunology, adipose

#### 294 Effects of under- and over-feeding during gestation on organ development of offspring at days 45 and 90 of gestation.

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Poor maternal nutrition during gestation can lead to intrauterine growth restriction resulting in offspring with low birth weight and altered organ development, negatively affecting production. We hypothesized that poor maternal nutrition during gestation would alter fetal body weight and organ mass of lambs during gestation. Western Whiteface ewes (n = 82) were bred to 1 of 4 rams and confirmed pregnant by ultrasound. Ewes were fed 100%, 60%, or 140% of NRC requirements for TDN beginning at d 30.2 ± 0.2 of gestation and offspring from these ewes will be referred to as CON, RES, and OVER, respectively. Fetal weights and organs were collected at d 45, 90, and 135 of gestation and within 24 h of birth. To date, organ weights have been collected from d 45 and 90. Data are expressed as percent of fetal weight and were analyzed using PROC MIXED in SAS. At d 45, fetal weight tended to be different ( $P = 0.067$ ) between treatments with OVER offspring smaller than CON ( $P = 0.021$ ) and RES intermediate (CON = 11.0 ± 0.6 g; RES = 10.1 ± 0.5 g; OVER = 9.2 ± 0.5 g). Liver weight at d 45 tended to be different ( $P = 0.055$ ) with RES offspring larger than OVER ( $P = 0.017$ ) and CON intermediate (CON = 6.7 ± 0.5%; RES = 7.6 ± 0.5%; OVER 6.1 ± 0.4%). We did not observe a difference in the weight of heart and kidney at d 45 ( $P \geq 0.148$ ). Although fetal weights were not different at d 90 ( $P = 0.329$ ) liver weights were greater in OVER offspring vs CON and RES ( $P \leq 0.05$ ; CON = 5.5 ± 0.2%; RES = 5.7 ± 0.2%; OVER = 6.3 ± 0.2%). At 90 d an effect of maternal diet was not observed for kidney, heart, pancreas, adrenal, or renal fat weights ( $P \geq 0.153$ ). In conclusion, maternal under- and over-feeding affects liver development during early gestation, a period of rapid liver growth, which may lead to altered health and growth of the offspring later in life.

**Key Words:** maternal nutrition, sheep, organ development

# Lactation Biology I

## 295 CLOCK regulation of mammary epithelial cell growth.

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The circadian timing system (CTS) influences virtually all physiological processes, including lactation. However, the manner that the multitude of molecular clocks that make up the CTS both centrally and peripherally affect lactation performance is currently not understood. Preliminary studies from our lab led us to hypothesize clocks in the mammary play an important role in regulation of gland development. The objective of this study was to determine if decreasing abundance of CLOCK, a core component of the circadian clock mechanism, affects growth of mammary epithelial cells. For this study a mouse mammary epithelial cell line, HC11, was transfected with shRNA that targeted *Clock* or a negative control scramble sequence. Cells transfected with sh*Clock* expressed 70% less *Clock* mRNA than wild-type (WT) HC11 cultures, which resulted in significantly depressed levels of CLOCK protein ( $P < 0.05$ ). Scramble had no effect on mRNA or CLOCK protein levels. Six-day growth curve analysis revealed HC11 lines carrying sh*Clock* had 4-fold higher growth rates ( $P < 0.05$ ) and reached at least 2-fold higher cell density than scramble transfected or WT HC11 cultures. To understand how CLOCK regulates growth, sh*Clock* transfected and WT HC11 cells were plated in growth medium, serum starved for 24 h, returned to growth medium and then collected every 4 h over a 48-h period. Cell cycle analysis of propidium iodide stained cells showed sh*Clock* significantly increased response to serum starvation ( $90.1 \pm 1.1\%$  of sh*Clock* versus  $71.3 \pm 3.6\%$  of WT-HC11 in G1 phase), and affected progression through cell cycle. Two-way ANOVA showed time and cell line had significant effects ( $P < 0.0001$ ) on relative expression levels of the cell cycle regulators, *Ccnd1* (regulates transition from G1 to S phase), *Wee1* (regulates transition from G1 to S and G2 to M phase) and *Tp63* (a tumor suppressor gene). Moreover, basal mean expression of *Ccnd1* was 43% greater ( $P = 0.002$ ) in sh*Clock* versus WT cell lines, and *Tp63* expression was depressed by ~3-fold in sh*Clock* cultures ( $P < 0.0001$ ). These data support circadian clocks play a role in regulation of epithelial cell growth in the mammary gland.

**Key Words:** circadian, clock, mammary

## 296 CLOCK regulates mammary differentiation and output.

Aridany Suárez Trujillo\*<sup>2</sup>, Jennifer Crodian<sup>1</sup>, Emily Erickson<sup>1</sup>, Avi Shamay<sup>3</sup>, Sameer Majbeesh<sup>4</sup>, Karen Plaut<sup>1</sup>, and Theresa Casey<sup>1</sup>, <sup>1</sup>Purdue University, West Lafayette, IN, <sup>2</sup>Universidad de Las Palmas de Gran Canaria, Arucas, Canary Islands, Spain, <sup>3</sup>Agriculture Research Organization, Volcani Center, Bet Dagan, Israel, <sup>4</sup>The Hebrew University of Jerusalem, Rehovot, Israel.

Circadian clocks synchronize internal physiology to the environment, thus understanding role in regulation of lactation may lead to development of noninvasive approaches to improve production efficiency of dairy animals. Circadian clocks are present in all mammalian cells. The *BMAL1* and *CLOCK* genes are core components of clocks, functioning together as a transcription factor. In the mammary gland, abundance of *BMAL1*-*CLOCK* increases during the transition from pregnancy to

lactation and upon lactogen-induced differentiation of mammary cells in culture. We hypothesize that in the mammary *BMAL1*-*CLOCK* regulates differentiation and milk synthesis. Our objective was to elucidate the effect of decreasing *CLOCK* abundance on expression of markers of differentiation (e-cadherin- *CDH1*) and metabolic output (fatty acid synthase- *FASN*;  $\beta$ -casein- *CSN2*) in a mouse mammary epithelial cell line, HC11. To decrease *CLOCK* abundance, HC11 cells were transfected with shRNA specific for *Clock* or a scramble sequence (negative control), and clonal lines were established. Cells transfected with sh*Clock* had a 70% reduction in *Clock* mRNA, which resulted in significantly reduced ( $P < 0.05$ ) *CLOCK* protein abundance relative to wild-type cultures. Abundance of *CLOCK* in scramble-line was not different from wild-type. RNA and protein were collected from undifferentiated cultures, and cultures treated 96 h with dexamethasone, insulin and prolactin (differentiated). Gene expression was analyzed using RT-qPCR. Two-way ANOVA showed cell line (wild-type, scramble, sh*Clock*) and state of differentiation had a significant effect ( $P < 0.05$ ) on relative expression of *Fasn*, *Csn2* and *Cdh1*. Post-hoc analysis revealed *Fasn* was significantly reduced by 2-fold ( $P < 0.05$ ) in sh*Clock* treatments relative to wild-type cultures. *Cdh1* was also reduced more than 3-fold in sh*Clock* lines relative to wild-type cultures ( $P < 0.05$ ). Moreover, Western blot analysis showed abundance of *CDH1* protein was significantly reduced in cultures transfected with sh*Clock*. sh*Clock* sequence did not have a significant effect on *Csn2* expression. In conclusion *CLOCK* regulates differentiation markers and *Fasn*, future studies are needed to understand how factors affect mammary clocks and the relationship to dairy performance.

**Key Words:** mammary, circadian, differentiation

## 297 Expression of putative stem cell marker, hepatocyte nuclear factor 4 alpha, in mammary gland of water buffalo.

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Buffaloes account for more than 56% of total milk production in India. Cyclic remodeling of mammary glands of human, mice, cow, sheep and goat is determined by mammary stem cells. It is logical to assume that buffalo mammary gland will have mammary stem/progenitor cells. Thus far, no report exists on identification of buffalo mammary stem cells. Hepatocyte nuclear factor 4  $\alpha$  (HNF4A) is a candidate marker for hepatic progenitor cells and has recently been suggested as a marker of bovine mammary stem/progenitor cells. We hypothesized that (1) HNF4A identifies putative buffalo mammary stem/progenitor cells and (2) the number of mammary stem/progenitor cells increases during mastitis. Thirteen buffalo mammary samples were collected from a local slaughterhouse. Hematoxylin and eosin staining were performed on 5- $\mu$ m-thick sections. Based on histomorphology of mammary glands, physiological stages were estimated to be nonlactating ( $n = 4$  animals) and mastitis ( $n = 9$  animals). In total, ~22,000 cells were counted (10 microscopic fields/animal;  $n = 13$  animals), of which 40% cells were mammary epithelial cells (MEC) and 60% cells were the stromal cells. The percentage of MEC in nonlactating animals was higher compared with that in mastitic animals (47.3 vs. 37.3%), which was likely due to loss of MEC caused by infection. HNF4A staining was observed in



nuclei of MEC of ducts, alveoli and stromal cells. Basal location and low frequency of basally located HNF4A positive cells (ranges from 0.4 to 4.5%) was consistent with characteristics of mammary stem cells. HNF4A-positive MEC (basal and luminal; light and dark stained) tended to be higher during nonlactating stage than mastitis ( $8.73 \pm 1.71$  vs.  $4.29 \pm 1.19\%$ ;  $P = 0.07$ ). MEC proliferation (assessed by immunohistochemical expression of Ki67) was higher in mastitic glands in comparison to nonlactating glands ( $15.3 \pm 5.7\%$  vs.  $0.53 \pm 0.1\%$ ;  $P = 0.03$ ). This is the first report outlining the expression of HNF4A as a putative mammary stem/progenitor cells of buffalo mammary gland; and evaluation of MEC proliferation in naturally infected mastitic buffaloes.

**Key Words:** buffalo, mammary stem/progenitor cell, HNF4A

**298 Optimization of transfection and real-time monitoring of fluorescent proteins in bovine cells: An untapped molecular biology approach for dairy sciences.** J. S. Osorio\* and M. Bionaz, Oregon State University, Corvallis, OR.

The study of nuclear receptor activation by specific dietary compounds via gene reporter technology (GRT) is essential in nutrigenomics research. The GRT requires inserting into cells an artificial plasmid containing a promoter region with the response element for the nuclear receptor of interest and DNA coding for luciferase or a fluorescent protein. The main challenge of using GRT is the low efficiency and high variability of plasmid transfection; thus limiting the sensitivity and increasing intra-assay variability. To investigate the efficiency of transfection in immortalized (MacT) and primary mammary bovine (BMEC) cells we have tested several concentrations of transfection reagents in combination with several concentrations of a plasmid for the constitutively expressed enhanced green fluorescent protein (EGFP). Cells were seeded 24 h prior transfection at 30,000 cells/well in a 96-well plate and treated with a nuclear staining (NucBlue Live). The transfection reagents Lipofectamine 2000, Lipofectamine LTX, and TransIT-X2 were used at 0.2, 0.3, 0.4, or 0.5 mL/well with 50, 100, 200, or 300 ng/well of EGFP. Using a robotic inverted fluorescent microscope for live imaging (Leica DMI6000B), 2 pictures/well were taken every hour for 30 h post-transfection. Cell number, viability, and quantification of transfection efficiency were assessed using the CellProfiler software. Data were analyzed using GLIMMIX of SAS. EGFP protein was visualized as early as 4 h after transfection and plateau expression was observed >7 h post-transfection. We observed high variability in transfection efficiency ranging from 1 to 30%. Transfection efficiency was best using Lipofectamine 2000 in BMEC and Lipofectamine LTX in MacT. In general we observed that high dose of transfection reagent and plasmid provided best results; however, high dose of transfection reagent tend also to kill the cells. Overall, our data confirmed the large intra- and inter-cells variation in transfection efficiency and prompt for a more precise approach to obtain high reliable data for nutrigenomics studies in dairy cows.

**Key Words:** mammary epithelial cell, nutrigenomics, transfection

**299 Palmitate and peroxisome proliferator-activated receptor (PPAR) $\gamma$  synthetic agonists, but not *trans*-10,*cis*-12 CLA, activate PPAR in MacT and primary goat mammary cells.** J. S. Osorio\* and M. Bionaz, Oregon State University, Corvallis, OR.

The peroxisome proliferator-activated receptors (PPAR) can play pivotal nutrigenomics roles in ruminants owing their capacity to be activated by long-chain fatty acids (LCFA). Data on expression of putative PPARs target genes suggested that palmitate (PA) is a potent PPARs activator in

cows and goats whereas *trans*-10,*cis*-12 conjugated linoleic acid (CLA) is not or is a very weak PPAR agonist. Furthermore, previous in vivo data in goats and cows suggested that 2,4-thiazolidinedione (TZD) is a weak activator of PPAR $\gamma$ . To test if palmitate, *trans*-10,*cis*-12-CLA, and TZD are PPAR agonists we have transfected immortalized MacT and primary mammary goat (PMG) cells with a PPRE-X3-TK-Luc plasmid. Prior transfection cells were seeded at  $10^4$  cells/well in a 96-well plate. We have used as transfection reagents Lipofectamine 3000 with PMG and TransIT-X2 with MacT at 0.3  $\mu$ L/well with 200 ng/well of plasmid in OptiMEM deprived of fetal bovine serum (FBS). Transfected cells were treated for 24h in quadruplicates with 10% FBS or 0.1 mM of ethanol (control), rosiglitazone (ROSI), PA, and CLA as well as 2 doses of TZD (0.1 mM and 1 mM) in medium without FBS. All were supplemented with 10  $\mu$ M of 9-*cis*-retinoic acid. Cell number and transfection efficiency was obtained via CellProfiler software and luciferase was measured using luminometer and normalized by number of viable cells. Data were analyzed using GLIMMIX of SAS and significance was declared with  $P < 0.05$ . Transfection was higher with MacT ( $24 \pm 3\%$ ) compared with PMG ( $6 \pm 4\%$ ). In MacT cells, all treatments with exception of CLA activated PPAR compared with control with the highest activation observed with TZD followed by PA, ROSI, and FBS. In PMG the PPAR activation was observed with FBS, PA, and TZD, while CLA and ROSI did not affect luciferase. Overall our data confirmed that TZD is a PPAR $\gamma$  synthetic agonist in ruminant mammary cells. In addition, the data confirmed that PA but not CLA is a PPAR activator and that blood serum in ruminants contains potent natural PPAR agonists, likely LCFA. These findings underscore the nutrigenomics importance of PA and confirmed the PPAR $\gamma$  responsiveness of mammary cells in ruminants.

**Key Words:** nutrigenomics, PPAR, transfection

**300 Protection of bovine mammary epithelial cells from hydrogen peroxide-induced oxidative cell damage by resveratrol.**

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Knowledge in the effects of oxidative stress and antioxidants on bovine mammary epithelial cells (bMEC) is limited. The objectives of this study were to investigate the oxidative damaging effects of hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) and the cytoprotective effects of resveratrol, a well-known natural product rich in grape seeds, against oxidative stress in cultured bMEC (MAC-T). To establish an oxidative stress model in bMECs, 500  $\mu$ M H<sub>2</sub>O<sub>2</sub> was added to MAC-T cells. The CCK-8 assay was applied to detect cell viability and flow cytometry method was used to detect intracellular production of reactive oxygen species. Pretreatment of MAC-T cells with resveratrol could rescue the decrease in cell viability and resulted in lower intracellular accumulation of reactive oxygen species after H<sub>2</sub>O<sub>2</sub> exposure. Using qRT-PCR, we found that resveratrol helped MAC-T cells to prevent H<sub>2</sub>O<sub>2</sub>-induced endoplasmic reticulum stress, indicated by significantly decreased abundance of endoplasmic reticulum stress marker GRP78 and CHOP mRNA ( $P < 0.01$ ). Resveratrol also inhibited mitochondria-related cell apoptosis by downregulating the expression of pro-apoptotic Bax gene ( $P < 0.01$ ) and upregulating expression of anti-apoptotic Bcl-2 gene ( $P < 0.01$ ) compared with H<sub>2</sub>O<sub>2</sub> group. Moreover, resveratrol increased the

abundance of multiple antioxidant defense genes (HO-1, xCT, Txnrd1, and NQO-1) in MAC-T cells under normal/oxidative conditions. It is confirmed that Nrf2 was required for the cytoprotective effects on MAC-T cells by resveratrol, because knockdown of Nrf2 abolished resveratrol-induced cytoprotective effects against oxidative stress, accompanied by no significant differences in gene expression of Nrf2, HO-1, Txnrd, and CHOP between resveratrol+H<sub>2</sub>O<sub>2</sub> and H<sub>2</sub>O<sub>2</sub> groups in Nrf2 knockdown MAC-T cells. Finally, by using selective inhibitors, we confirmed that the induction of Nrf2 by resveratrol was mediated through the activation of the PI3K/Akt and ERK/MAPK pathways, but negatively regulated by p38/MAPK pathway. In conclusion, our study provided evidence that resveratrol may be potentially used as a therapeutic agent for cytoprotection of bMEC against oxidative stress.

**Key Words:** resveratrol, oxidative stress, mammary epithelial cell

### 301 Stabilization of Nrf2 by tBHQ attenuates heat shock-induced cell damage in bovine mammary epithelial cells. X.L.

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Nrf2-ARE signaling plays a key role in cellular antioxidant-defense system, but whether Nrf2 activation has protective effects against heat shock (HS) stress remains unclear. The objective of the present study was to determine whether the Nrf2 activator, tBHQ, could attenuate the heat

stress-induced responses in bovine mammary epithelial cells (bMECs). MAC-T cells were exposed to HS (42.5°C for 1 h) followed by recovery at 37°C to mimic HS stress. Compared with cells consistently cultured at normothermia (37°C), cell viability was dropped after HS treatment ( $P < 0.01$ ), and arrived at the lowest ( $64.5 \pm 7.68\%$ ) after 12 h recovery. Accordingly, mRNA abundance of cell-apoptosis marker genes (Bax and CHOP,  $P < 0.01$ ) and cellular antioxidant defense genes (HO-1 and Txnrd1,  $P < 0.01$ ) increased time-dependently in HS challenged cells and reached to the highest level after 12 h recovery, compared with normothermic cultured MAC-T cells. When the MAC-T was pretreated with tBHQ (10  $\mu$ M) for 2 h and performed HS following 12 h recovery, pre-incubation of tBHQ significantly prevented loss of cell viability and downregulated mRNA expression of Bax and CHOP ( $P < 0.01$ ) than HS treated MAC-T cells in the absence of tBHQ. The presence of tBHQ also significantly blocked accumulation of reactive oxygen species induced by HS in MAC-T cells after recovering for 2 h ( $P < 0.01$ ). More importantly, tBHQ pre-treated cells showed stronger activation of Nrf2-ARE signaling compared with the HS group, including more nuclei-accumulations of Nrf2 and higher upregulations of Nrf2-ARE driven gene expressions (including Nrf2, HO-1 and Txnrd1,  $P < 0.01$ ). RNA silencing of Nrf2 in HS-treated MAC-T cells almost abolished the cytoprotective effects of tBHQ. These results indicated that HS could cause oxidative stress in bMECs, and stabilization of Nrf2 by tBHQ could attenuate HS-induced bMECs damage.

**Key Words:** heat shock, Nrf2, tBHQ

## Nonruminant Nutrition: Amino acids and minerals

**302 Long-term leucine and branched-chain amino acid supplementation in a protein and energy deficient diet increases muscle mTORC1 activation in neonatal pigs.** Rodrigo Manjarín\*, Daniel A. Columbus, Agus Suryawan, Hanh V. Nguyen, Adriana Hernández-García, Rosemarie D. Parada, Marta L. Forotto, and Teresa Davis, *USDA-ARS, Children's Nutrition Research Center, Houston, TX.*

Suboptimal nutrient intake represents a limiting factor for growth and long-term survival of low-birth weight infants and runt pigs. The objective of this study was to determine if enteral leucine or branched chain amino acid (BCAA) supplementation will upregulate the mammalian target of rapamycin complex 1 (mTORC1) pathway in skeletal muscle and lead to an increase in protein synthesis in neonates who consume only 70% of their protein and energy requirements. Twenty-two 4-d-old piglets were fed by gastric tube 1 of 4 diets, containing (kg body weight<sup>-1</sup>·d<sup>-1</sup>) 15 g protein and 215 kcal (CON; n = 5), 10.5 g protein and 156 kcal (R; n = 5), 10.5 g protein + 1.19 g leucine and 156 kcal (RL; n = 6), or 10.5 g protein + 1.67 g BCAA (n = 6) at 4-h intervals for 21 d. On d 21, plasma AA and insulin levels were measured during 6 post-feeding intervals, whereas muscle protein synthesis rate and mTORC1 activation were determined at 120 min post-feeding. Data were analyzed by ANOVA using a linear mixed model with repeated measures. *P*-values for pre-planned pairwise comparisons were calculated using Student's *t*-tests. Compared with CON, the RL and BCAA diets increased plasma leucine levels (*P* ≤ 0.01), whereas RL decreased isoleucine and valine (*P* ≤ 0.01) between 0 and 120 min post-feeding. At 120 min insulin was higher in CON compared with R (*P* ≤ 0.01). Compared with CON, BCAA, RL and R decreased (*P* < 0.01) body weight gain (3.5, 2.9, 3.0 and 2.9 ± 0.01 kg, respectively), protein synthesis (19.3, 14.2, 15.2 and 14.6 ± 1.3%·d<sup>-1</sup>), phosphorylation of S6 kinase (p-S6K1; 0.6, 0.4, 0.4 and 0.2 ± 0.01 AU) and 4E-binding protein (p-4EBP1; 0.4, 0.2, 0.2 and 0.1 ± 0.03 AU), and activation of eukaryotic initiation factor 4 complex (eIF4E·eIF4G; 1.5, 1.1, 1.1, and 0.4 ± 0.02 AU). BCAA and RL increased (*P* ≤ 0.01) p-S6K1, p-4EBP1 and eIF4E·eIF4G compared with R. In conclusion, when protein and energy intakes are restricted, leucine or BCAA supplementation increases muscle mTORC1 activation but does not improve body weight gain or skeletal muscle protein synthesis in neonatal pigs.

**Key Words:** leucine, neonate, muscle

**303 Effect of fermentation on ileal digestibility of nutrients and amino acids in soybean meal with low solubility in growing pigs.** S. D. Upadhyaya<sup>1</sup>, J. H. Ryu<sup>2</sup>, K. I. Kang<sup>2</sup>, S. J. Cho<sup>2</sup>, and I. H. Kim<sup>1</sup>, <sup>1</sup>*Department of Animal Resource & Science, Dankook University, Cheonan, Chungnam, South Korea,* <sup>2</sup>*CJ Food Ingredients R&D Center, Seoul, South Korea.*

Fermentation of soybean meal with microbes has been reported to increase digestibility of nutrients. The objective of this study was to evaluate the effects of fermentation on digestibility of soybean meal with protein solubility below optimal range. Apparent ileal digestibility (AID) and standardized ileal digestibility (SID) of GE, DM, N and amino acids were evaluated in 5 cannulated [(Landrace × Yorkshire) × Duroc] barrows with an average initial BW of 27.05 kg. The pigs were used in a 5 × 5 Latin square design with treatments of soybean meal (SBM) with protein solubility of 84% and fermented soybean meal (FSM) with protein solubility of 52%, 62% and 72% and designated as

FSMA, FSMB and FSMC, respectively. A purified diet without protein was used to determine basal endogenous losses of CP and amino acids. Data were analyzed using the Mixed procedure of SAS with pig as the experimental unit for all analyses. Tukey's test was used to determine significant differences among treatments. The AID of DM and N was greater (*P* < 0.05) in FMSC (75.9% and 73.7%) and tended to improve in FSMA (74.4% and 71.3%) and FSMB (73.7% and 71.8%) compared with SBM (71.1% and 69.1%). The AID of Ile, Phe and Val was greater in all fermented soybean meal whereas that of Lys was greater (*P* < 0.05) in FSMC (76.19%) compared with SBM (69.05%). Likewise, the AID of Asp and Pro was greater (*P* < 0.05) in FSMA and FSMC than SBM. The SID of CP and Lys was higher (*P* < 0.05) in FSMC (85.03% and 83.86%) and tended to improve in FSMA (82.4% and 81.08%) and FSMB (83.11% and 82.23%) compared with SBM (80.68% and 77.72%) whereas the SID of Ile was higher (*P* < 0.05) in FSMA (87.47%), FSMB (84.86%) and FSMC (86.75%) compared with SBM (80.29%). In conclusion, fermentation of soybean meal showed better digestibility compared with SBM regardless of its protein solubility values.

**Key Words:** amino acid, ileal digestibility, fermented soybean meal

**304 Requirement of valine and optimal valine:lysine ratio in diets for 8- to 18-kg pigs.** John K. Htoo<sup>\*1</sup> and Georg Dusel<sup>2</sup>, <sup>1</sup>*Evonik Industries AG, Hanau-Wolfgang, Germany,* <sup>2</sup>*University of Applied Sciences Bingen, Bingen am Rhein, Germany.*

A 28-d experiment was conducted to estimate the optimal SID Val:Lys in diets for starter pigs. A total of 192 weaned pigs (Topigs; initial BW of 8.5 ± 0.12 kg) were randomly assigned to 8 dietary treatments with 6 pen replicates (2 barrows and 2 gilts/pen) per treatment. A corn-wheat-soybean meal-based basal diet was formulated using analyzed ingredient AA contents and published SID coefficients to meet requirements of AA other than Val and Lys. Valine was first limiting (0.69% SID Val) and Lys was second limiting (1.15% SID Lys). Graded levels of L-Val were added to the basal diet to obtain 5 SID Val:Lys ratios (60, 64, 68, 72 and 76%). In addition, diets 6, 7 and 8 were produced to be adequate in SID Lys (1.34%) but varied in SID Val (0.86, 0.91 and 0.97%). All diets contained 2,474 kcal/kg NE. Based on analyzed AA contents, the corrected SID Val:Lys ratios in diets 1 to 5 were 63, 66, 72, 76 and 78%, and corrected SID Val contents in diets 6 to 8 were 0.86, 0.94 and 1.01%, respectively. Data were analyzed by ANOVA using the GLM procedure of SAS. Orthogonal-polynomial contrasts were used to determine linear and quadratic effects of SID Val:Lys ratios (diets 1–5) on response criteria. Increasing dietary SID Val:Lys from 63 to 78% (Diet 1 to 5) increased (*P* = 0.046) the final BW quadratically, and tended to increase (*P* = 0.075) the ADG (d 0–28) quadratically. The greatest final BW (16.47 vs. 15.55 kg) and ADG (284 vs. 258 g/d) in comparison to the basal diet were achieved at a SID Val:Lys ratio of 66%. The FCR (d 0–28) tended to improve (*P* = 0.078; quadratic) and the best FCR (1.377 vs. 1.545) in comparison to the basal diet was achieved at a SID Val:Lys ratio of 72%. Feed intake was not affected by dietary treatments. A 2-slope broken-line regression estimated the SID Val:Lys ratio of 67% to optimize both the ADG and FCR. When containing adequate Lys (diets 6 to 8), increasing the SID Val from 0.86 to 0.94% (SID Val:Lys from 66 to 71%) improved FCR (*P* = 0.011) but did not affect ADG (*P* = 0.148). Based on these results an optimal SID Val:Lys ratio of at least 67% is recommended in diets for 8 to 18 kg pigs.

**Key Words:** lysine, pig, valine



**305 Effects of dietary fiber on the optimum threonine:lysine ratio for 25- to 50-kg gilts.** John K. Mathai<sup>1</sup>, John K. Htoo<sup>2</sup>, John Thomson<sup>3</sup>, Kevin J. Touchette<sup>4</sup>, and Hans H. Stein<sup>1</sup>, <sup>1</sup>University of Illinois, Urbana, IL, <sup>2</sup>Evonik Industries AG, Hanau-Wolfgang, Hesse, Germany, <sup>3</sup>Evonik Degussa Corporation, Kennesaw, GA, <sup>4</sup>Ajinomoto Heartland Inc., Chicago, IL.

Two experiments were conducted to determine the effect of dietary fiber on the ideal Thr:Lys ratio for 25 to 50 kg gilts. In Exp. 1, 192 gilts (26.3 ± 4.64 kg) were used with 2 pigs/pen and 8 pens/trt. A 2 × 6 factorial arrangement was used to determine the effect of dietary fiber (low and high) and standardized ileal digestible (SID) Thr:Lys ratios 45, 54, 63, 72, 81, and 90:100. At both fiber levels, ADG and G:F increased quadratically ( $P < 0.05$ ), as the concentration of Thr increased in the diets. For pigs fed low-fiber diets, broken-line analyses estimated the optimum SID Thr:Lys ratio for ADG and G:F, respectively, as 0.60 and 0.59, quadratic analyses as 0.76 and 0.73, and combined linear-quadratic analyses as 0.66 and 0.63. For pigs fed high fiber diets, broken-line analyses estimated the optimum SID Thr:Lys ratio for ADG and G:F, respectively, as 0.66 and 0.55, quadratic analyses as 0.80 and 0.75, and combined linear-quadratic analyses as 0.71 and 0.63. In Exp. 2, pigs were fed either low-fiber or high-fiber diets that were very deficient (0.45 SID Thr:Lys) or marginally deficient (0.60 SID Thr:Lys) in Thr. Thirty-six gilts (29.0 ± 0.74 kg) were housed in metabolism crates with 9 replicate pigs per diet. Output of N in feces was greater ( $P < 0.05$ ) from pigs fed high-fiber diets, whereas output of N in urine was greater ( $P < 0.05$ ) from pigs fed low-fiber diets. The ATTD of N and retention of N were greater ( $P < 0.05$ ) in pigs fed low-fiber diets. There was a lower ( $P < 0.05$ ) N output in urine and a greater ( $P < 0.05$ ) N retention in pigs fed high-Thr diets with 0.60 SID Thr:Lys compared with pigs fed low-Thr diets containing 0.45 SID Thr:Lys. There was also an interaction ( $P < 0.05$ ) between fiber level and Thr for output of N in feces with N output increasing ( $P < 0.05$ ) as Thr in the high-fiber diet increased. Results of these experiments indicate that increased fiber levels in diets fed to growing gilts increase the requirement for Thr, and diets with higher fiber levels should, therefore, include a greater concentration of Thr.

**Key Words:** pig, fiber, threonine

**306 Evaluation of L-methionine bioavailability in piglet diets.** Helvio C. Ferreira Júnior<sup>1</sup>, Melissa I. Hannas<sup>1</sup>, Luiz F. T. Albino<sup>1</sup>, Horácio S. Rostagno<sup>1</sup>, Lyssa Otani<sup>2</sup>, and Lília T. Lopes<sup>1</sup>, <sup>1</sup>Federal University of Viçosa, Viçosa, Minas Gerais, Brazil, <sup>2</sup>CJ of Brazil Ltda, São Paulo, São Paulo, Brazil.

The objective of this trial was to evaluate the bioavailability of L-Met considering DL-Met as the standard source (100% availability) based on average daily gain (ADG) and feed conversion (FC) of piglets. A total of 135 barrows (14.77 ± 1.46 kg) 24 d old were distributed in a completely randomized block design into 5 treatments with 9 replicates and 3 animals per pen. The treatments were a basal diet (BD) formulated to meet or exceed the nutritional recommendations of all nutrients (except for methionine), BD + 0.041% DL-Met; BD + 0.082% DL-Met; BD + 0.041% L-Met and BD + 0.082% L-Met. The calculated content of Met and Met+Cys in basal diet were 0.206% and 0.441% respectively. The experimental period lasted 28 d and individual pig body weight and feed intake were recorded to calculate performance: ADG, average daily feed intake (ADFI) and FC. Multiple regression model was obtained from linear regression analysis using SAS PROC MIXED procedure at probability level of  $P < 0.05$  was regarded as statistically significant. The regression coefficient ratios were utilized to determine bioavailability of L-Met considering that both equations had the same intercept. A linear

increase was observed in ADG ( $P < 0.05$ ) with the supplementation of both DL-Met and L-Met. DL-Met supplementation levels increased ADFI linearly ( $P < 0.05$ ). However there was a linear decrease ( $P < 0.05$ ) in ADFI with supplemental sources of L-Met. The L-Met bioavailabilities estimated by ADG and FC were 105.48 and 130.94%, respectively.

**Table 1 (Abstr. 306).** Performance and bioavailability (BV) of L-Met for piglets from 15 to 30 kg<sup>1</sup>

Item	DL-Met			P-value	L-Met			s
	BD	0.041%	0.082%		0.041%	0.082%	P-value	
ADG, g	581	648	714	0.001	664	715	0.001	0.246
ADFI, g	1121	1275	1368	0.003	1320	1312	0.052	0.487
FC, g/g	2.085	1.975	1.916	0.001	1.990	1.839	0.001	0.427
L-Met BV	Intercept	b <sup>D</sup>	b <sup>L</sup>	Bioavailability				
ADG	0.584	1.581	1.668	105.48				
FC	2.086	-2.196	-2.876	130.94				

<sup>1</sup>Linear regression analysis; b<sup>D</sup> = DL-methionine regression coefficients; b<sup>L</sup> = L-Met regression coefficients

**Key Words:** L-isomer amino acid, swine, L-methionine

**307 Oral tryptophan supplementation to nursing piglets increases serotonin synthesis and improves performance after weaning under social mixing stress.** T. J. Pasquetti<sup>1,2</sup>, I. Park<sup>1</sup>, J. Y. Guo<sup>1</sup>, P. C. Pozza<sup>2</sup>, and S. W. Kim<sup>1</sup>, <sup>1</sup>Department of Animal Science, North Carolina State University, Raleigh, NC, <sup>2</sup>Departamento de Zootecnia, Universidade Estadual de Maringá, Bolsista do Ciências sem Fronteiras / CNPQ, Maringá, PR, Brazil.

This study was to evaluate the effect of oral Trp supplementation to nursing piglets on serotonin (5-HT) metabolism, growth performance, and behavior during nursing (from 14 to 21 d of age) and nursery (from 21 to 56 d of age) periods under social mixing stress at weaning (from 21 to 22 d of age). Eighty piglets from 10 sows (4 males and 4 females per litter) at 14 d of age were randomly allotted to 2 treatments based on sex. Treatments (Trp, 0.46 g/kg BW/d; or Ala + glucose, 0.38 + 0.42 g/kg BW/d, respectively) were orally given to piglets during nursing period. At weaning on d 21, 6 piglets from each treatment were euthanized to collect hypothalamus to analyze 5-HT and 5-hydroxyindoleacetic acid (5-HIAA) concentrations. Remaining 68 piglets were weaned and moved to nursery pens. Pigs within a treatment, same sex, but from different litters were paired and housed together (2 pigs/pen, 17 pens/treatment). Normal and aggressive behaviors were recorded from 21 to 22 d of age immediately after weaning and mixing. All pigs were fed the same diet during nursery period. Growth performance was measured during nursing and nursery periods. Data were analyzed using Proc Mixed of SAS with treatment and sex as fixed effects. Oral Trp feeding during nursing period increased ( $P \leq 0.05$ ) hypothalamic concentration of 5-HT and 5-HIAA (2.84 to 5.94 and 59.1 to 104.6 ng/mg of protein, respectively), tended to increase the frequency of visits to the feeders ( $P = 0.057$ , 0.42 to 0.83%) during mixing period and G:F ratio ( $P = 0.094$ , 0.440 to 0.514) during the first week of nursery period. Collectively, oral Trp feeding during nursing period increased hypothalamic serotonin affecting eating behavior and potentially enhancing feed efficiency immediately after weaning.

**Key Words:** mixing stress, pig, tryptophan

**308 Concentrations of nitrogen-corrected apparent metabolizable energy and amino acid digestibility in soybean meal from Argentina, Brazil, China, Thailand, and the United States fed to broilers.** Kelly M. Sotak-Peper\*<sup>1</sup>, Rommel C. Sulabo<sup>2</sup>, Carl M. Parsons<sup>1</sup>, and Hans H. Stein<sup>1</sup>, <sup>1</sup>University of Illinois at Urbana-Champaign, Urbana, IL, <sup>2</sup>University of the Philippines Los Banos, Los Banos, Luzon, Philippines.

The Philippines import soybean meal (SBM) from many different countries, but the quality of the SBM from the different sources is not known. Therefore, 2 experiments were conducted using SBM from Argentina (ARG 1 and ARG 2), Brazil, China, Thailand, and the U.S (US1, US2, and US3) that was collected at feed mills in the Philippines. The objective was to determine concentrations of nitrogen-corrected apparent metabolizable energy (AMEn) and standardized ileal digestibility (SID) of AA in each source of SBM. In both experiments, ANOVA was used with the PROC MIXED function in SAS. In Exp. 1, 72 d-old male broilers were allotted to 1 of 9 treatments using a completely randomized design with 8 broilers per treatment. Treatments included a corn-SBM based basal diet and 8 diets that were formulated by mixing 70% basal diet and 30% of each SBM source. Broilers were placed in individual metabolism crates equipped with an aluminum tray for total collection of excreta. Excreta were collected for 3 d after a 10 d adaptation period. The AMEn (2,762 kcal/kg) of Chinese SBM was greater ( $P < 0.05$ ) than in ARG 1 (2,699 kcal/kg) and ARG2 (2,737 kcal/kg), but not different from the other sources. No differences in BW gain were observed, but there was reduced ( $P < 0.05$ ) feed intake and improved ( $P < 0.05$ ) G:F for birds fed US1 SBM compared with birds fed Argentinian or US2 SBM. In Exp. 2, 48 cecectomized roosters were allotted to 1 of 8 treatments using a completely randomized design with 6 roosters per treatment. The cecectomized roosters were tube fed 30 g of SBM and excreta were collected for 48 h following feeding. There was a tendency for greater ( $P < 0.10$ ) SID of indispensable AA for US3 SBM than for the other sources of SBM. The SBM from US3 had greater ( $P < 0.05$ ) SID of Lys compared with Brazilian SBM. The SBM from US3 also had greater ( $P < 0.05$ ) SID of dispensable AA and total AA than SBM from China, Thailand, US1 and US2. These studies demonstrated that differences in energy and AA digestibility exist among different SBM sources.

**Key Words:** amino acid, energy, soybean meal

**309 Effect of dietary cation-anion difference during late gestation and lactation on blood and urine parameters of sow.** J. Y. Guo\*<sup>1</sup>, D. E. Axe<sup>2</sup>, and S. W. Kim<sup>1</sup>, <sup>1</sup>Department of Animal Science, North Carolina State University, Raleigh, NC, <sup>2</sup>Granco Minerals, Disputanta, VA.

The experiment was conducted to evaluate the effect of dietary cation-anion difference (DCAD) on pH of blood and urine, mineral concentrations in serum, and hypocalcemia for sows during late gestation and lactation. A total of 22 pre-parturient sows (average parity of  $4.5 \pm 2.9$  and BW of  $224.1 \pm 38.7$  kg) were initially allotted to 2 dietary treatments in a completely randomized design: a control diet with positive DCAD (100 mEq/kg) and a diet with negative DCAD (-100 mEq/kg) with supplemental anionic salt (Cad-mate, Granco Minerals, Disputanta, VA) which were fed from d 93 of gestation to d 18 of lactation. Each sow had a daily access to 2 kg feed during gestation and had ad libitum access to feed during lactation. Feed with positive DCAD was provided during the subsequent parity. The pH of blood and urine were determined at d 93, 108 of gestation, d 1, 9 and 18 of lactation. At d 1 and 18 of lactation, Ca concentration in urine and milk and serum concentrations of Na, K, Mg, Ca, P, and Cl were measured. Data were analyzed using Proc Mixed of SAS with treatment as a fixed effect. Feed intake and

BW loss in current lactation, and litter performance in current as well as subsequent lactations were not affected by changing DCAD. At d 1 of lactation, decreasing DCAD reduced ( $P < 0.05$ ) blood pH (7.83 to 7.72). Urinary pH was reduced ( $P < 0.05$ ) by decreasing DCAD at d 108 of gestation, d 1, 9, and 18 of lactation (6.18 to 5.28; 6.38 to 5.47; 6.47 to 5.10; 6.23 to 5.04). Reducing DCAD increased ( $P < 0.05$ ) Ca concentrations in serum (9.23 to 9.71 mg/dL) and colostrum (66.11 to 79.34 mg/dL) at d 1 of lactation and increased ( $P < 0.05$ ) serum Ca concentration (9.74 to 10.14 mg/dL) at d 18 of lactation. Reducing DCAD tended to increase Mg concentration in serum at d 1 of lactation ( $P = 0.097$ , 18.73 to 19.84 mg/L) and at d 18 of lactation ( $P = 0.087$ , 22.11 to 23.97 mg/L). At d 1 of lactation, reducing DCAD increased ( $P < 0.05$ ) Cl concentration in serum (101.8 to 104.7 mEq/L). Collectively, negative DCAD increased Ca mobilization and decreased potential risk of urine tract infection without affecting litter performance.

**Key Words:** sow, dietary cation-anion difference, Ca

**310 Effects of copper sources and levels on growth performance and carcass traits in wean-finishing pigs.** Ferdinando N. Almeida\*<sup>1</sup>, Jeffery Escobar<sup>1</sup>, Gary Allee<sup>2</sup>, Junmei Zhao<sup>1</sup>, Yulin L. Ma<sup>1</sup>, and Mercedes Vazquez-Anon<sup>1</sup>, <sup>1</sup>Novus International Inc., St Charles, MO, <sup>2</sup>Porktech LLC, Columbia, MO.

A study was conducted under commercial conditions to compare the effects of 2 different Cu sources [tri-basic copper chloride ( $\text{Cu}_2[\text{OH}]_3\text{Cl}$ ) or  $\text{Cu}(\text{HMTBa})_2$ ] on growth performance and carcass traits in pigs and to evaluate a step-down feeding strategy when supplementing pigs with  $\text{Cu}(\text{HMTBa})_2$ .  $\text{Cu}(\text{HMTBa})_2$  is a chelated Cu with 2-hydroxy 4-methylthio butanoic acid (HMTBa) at 1:2 ratio (Novus International Inc., St. Charles MO). A total of 2,300 weaned pigs (initial BW =  $6.60 \pm 0.34$  kg, 21 d of age, mixed sex) were randomly allotted to 1 of 4 feeding programs with 23 replications and 25 pig/pen. Treatments included 1) control (16 mg/kg Cu in the premix); 2) ( $\text{Cu}_2[\text{OH}]_3\text{Cl}$ ) at 150 mg/kg (wean-finishing); 3)  $\text{Cu}(\text{HMTBa})_2$  at 150, 80, and 60 mg/kg (Nursery-Phase-I, Nursery-Phases-II-III, and grow-finishing phases, respectively) and 4)  $\text{Cu}(\text{HMTBa})_2$  at 150 mg/kg in Nursery-Phase-I and 80 mg/kg from Nursery-Phase-II to finishing phase. Copper levels were chosen based on current industry practices. Data were analyzed using the Mixed Procedure of SAS. The model included treatment as fixed effect and replication as random effect. From d 7 to 21 post-weaning, the ADG and GF was greater ( $P = 0.03$ ) for pigs fed  $\text{Cu}(\text{HMTBa})_2$  than that of the control pigs. Pigs fed ( $\text{Cu}_2[\text{OH}]_3\text{Cl}$ ) were intermediate. From d 0-42, ADG tended ( $P = 0.11$ ) to be greater for pigs fed  $\text{Cu}(\text{HMTBa})_2$  than control pigs. In the grow-finishing phase (d 42-153), by d 111, the BW of pigs fed  $\text{Cu}(\text{HMTBa})_2$  at 80 mg/kg was heavier ( $P < 0.05$ ) than control pigs and pigs fed ( $\text{Cu}_2[\text{OH}]_3\text{Cl}$ ) or  $\text{Cu}(\text{HMTBa})_2$  at 60 mg/kg were intermediate. Overall (d 42-153) the GF was greater ( $P < 0.05$ ) for both (60 and 80 mg/kg)  $\text{Cu}(\text{HMTBa})_2$  treatments compared with control (0.377 and 0.378 vs. 0.370, respectively). Pigs fed ( $\text{Cu}_2[\text{OH}]_3\text{Cl}$ ) were intermediate. On average, the hot carcass weight (HCW) of pigs fed both levels of  $\text{Cu}(\text{HMTBa})_2$  was 101.48 and 101.49 (60 and 80 mg/kg, respectively) vs. 100.44 kg in pigs fed control, whereas the HCW of pigs fed ( $\text{Cu}_2[\text{OH}]_3\text{Cl}$ ) was 101.34 kg. In conclusion, growth performance was improved by  $\text{Cu}(\text{HMTBa})_2$  supplementation to a greater extent than ( $\text{Cu}_2[\text{OH}]_3\text{Cl}$ ). Supplementing  $\text{Cu}(\text{HMTBa})_2$  either at 60 or 80 mg/kg resulted in similar performance in terms of feed efficiency.

**Key Words:** chelated trace mineral, copper, pig growth performance

## Physiology and Endocrinology: Nutrition, reproduction and metabolism

**311 Dietary omega-3 supplementation alters gene expression in equine endometrial and embryonic tissues.** Robert D. Jacobs\*<sup>1</sup>, Alan D. Ealy<sup>1</sup>, Parker M. Pennington<sup>2,3</sup>, Budhan Pukazhenth<sup>2</sup>, Lori K. Warren<sup>4</sup>, Ashley L. Wagner<sup>5</sup>, Tanja M. Hess<sup>6</sup>, and Rebecca K. Splan<sup>1,7</sup>, <sup>1</sup>Virginia Polytechnic Institute and State University, Blacksburg, VA, <sup>2</sup>Smithsonian Conservation Biology Institute, Front Royal, VA, <sup>3</sup>George Mason University, Fairfax, VA, <sup>4</sup>University of Florida, Gainesville, FL, <sup>5</sup>Cooperative Research Farms, Richmond, VA, <sup>6</sup>Colorado State University, Fort Collins, CO, <sup>7</sup>Virginia Tech MARE Center, Middleburg, VA.

Adverse maternal events around the time of conception influence embryonic development. Thus, aberrations in the uterine environment during early pregnancy, such as maternal metabolic or nutritional disruption, can alter gene expression in the developing embryo, leading to alteration in its developmental trajectory. Dietary supplementation of long-chain omega-3 polyunsaturated fatty acids (LCPUFA) improves metabolic and reproductive health across species. The objective of this study was to evaluate effects of peri-conceptual LCPUFA supplementation on endometrial and embryonic gene expression in overweight horses. Light horse mares (n = 13; mean age = 13.56 ± 0.11 yr; mean BCS = 7.07 ± 0.21) were supplemented with concentrate (n = 6) or an isocaloric, isonitrogenous diet containing 0.06 mg/kg BW marine-derived omega-3 LCPUFA (n = 7) 60 d before first sample collection. Four consecutive ovulatory cycles were monitored and uterine endometrial samples were obtained 12 d post-ovulation 1, 3 and 4. Mares were bred to one stallion on ovulatory cycles 2, 3 and 4, and embryos were flushed 12 d post ovulation. Candidate genes essential to inflammation, prostaglandin synthesis and embryonic development were evaluated by quantitative PCR. Data were log-transformed and analyzed using the GLM procedure in SAS (v9.3). When examining the data independent of breeding and pregnancy status, samples from LCPUFA supplemented mares contained reduced ( $P = 0.04$ ) *IL6* mRNA abundance and tended to have increased transcript abundance for Uterocalin ( $P = 0.09$ ), *SAA* ( $P = 0.06$ ) and *IL10* ( $P = 0.06$ ). Mares fed LCPUFA pregnant in cycle 3 contained greater *IL10* ( $P < 0.001$ ) and *PLA2G3* mRNA ( $P = 0.009$ ) and had a tendency for increased *SAA* abundance ( $P = 0.08$ ). Supplemented mares bred but not pregnant in cycle 3 had a tendency for reduced expression of *PTGER2* ( $P = 0.100$ ). In the conceptus, relative transcript abundance of *GATA4* and *GATA6*, markers of endoderm differentiation, along with *GATA3* and *ELF3*, markers of trophectoderm differentiation were greater ( $P < 0.05$ ) in embryos from LCPUFA supplemented mares (n = 5), than controls (n = 5). These results indicate that LCPUFA supplementation during the peri-conceptual period may alter the post-ovulatory uterine environment and early embryonic development in the horse.

**Key Words:** fatty acid, pregnancy, fetal programming

**312 Omega-6 fatty acid-rich sunflower oil supplements in diet affect uterine health, ovarian function and oocyte characteristic in heat-stressed dairy cows.** Chainarong Navanukraw\*<sup>1,2</sup>, Aree Kraison<sup>1</sup>, Vilaivan Khanthusaeng<sup>1</sup>, Suthipong Uriyapongson<sup>1</sup>, and Chuchart Kamollirt<sup>3</sup>, <sup>1</sup>Department of Animal Science, Khon Kaen University, Khon Kaen, Thailand, <sup>2</sup>Agricultural Biotechnology Research Center for Sustainable Economy, Khon Kaen University, Khon Kaen, Thailand, <sup>3</sup>Department of Anatomy, Faculty of Veterinary Medicine, Khon Kaen, Thailand.

The objective was to examine effects of plant oil supplement on uterine health, ovarian function, and oocyte characteristic in heat-stressed dairy cows. Parturient cows (n = 36) were randomly allocated to one of 3 dietary treatments; control, 4% refined palm oil (PO), and 4% refined sunflower oil (SO). ANOVA using the general linear model procedure was performed to determine treatments effect. All variables were analyzed with a nested ANOVA with treatment, animal (treatment), and time included in the model. Differences between specific means were evaluated by least significance difference. Placental expulsion and uterine involution in cows fed oil supplements occurred earlier than control cows (3.4 h and 33.2 d for cows fed SO, 4.9 h and 42.0 d for cows fed PO, and 21.0 h and 50.4 d for control cows;  $P < 0.05$ ). Numbers of class 1 (2–5 mm) and class 3 (≥10 mm) follicles of cows fed oil supplements were greater than those of control cows ( $P < 0.05$ ). In the second experiment, lactating cows (n = 42) were hormonally synchronized and were randomly offered diets supplemented either with 4% PO or 4% SO for 42 d. Follicle and oocyte numbers and corpus luteum (CL) volume were determined on d 5, 9, and 13 of the estrous cycle and morphologic quality assessed. Neither follicle nor oocyte characteristics were affected by the dietary treatment, however volume of CL and progesterone concentrations on d 13 in cows fed SO were greater ( $P < 0.05$ ) than those fed PO diet (8,290 vs. 7,875 mm<sup>3</sup> and 5.0 vs. 3.9 ng/mL). The data support the beneficial effects of SO supplement in diet on reproductive function in dairy cows

**Key Words:** plant oil, ovarian function, dairy cow

**313 Effects of excess dietary MP from corn gluten meal or soybean meal on ovarian function of beef cows consuming low quality forage.** Taylor C. Geppert\*<sup>1</sup>, George A. Perry<sup>2</sup>, and Patrick J. Gunn<sup>1</sup>, <sup>1</sup>Department of Animal Science, Iowa State University, Ames, IA, <sup>2</sup>Department of Animal Sciences, South Dakota State University, Brookings, SD.

The objective of this study was to determine the effects of feeding excess MP from feedstuffs differing in rumen degradability on ovarian function of beef cows. Non-pregnant, nonlactating beef cows (n = 18) were stratified by age, BCS, and BW to 1 of 2 isocaloric, isonitrogenous diets: (1) corn gluten meal (CGM) or (2) soybean meal (SBM), supplemented at 150% of MP requirements with ad libitum access to corn stalks for 58d. After a 20d supplement adaptation period, cows were synchronized for ovulation using the 5-d CO-Synch + CIDR protocol. Ten d after synchronization completion, 100 µg of GnRH was administered to reset follicular growth. Starting at GnRH administration and daily thereafter until spontaneous ovulation, transrectal ultrasonography was performed to diagram ovarian follicular growth, and blood samples were collected for hormone and metabolite analyses. Seven d after visual detection of estrus, corpus luteum (CL) size was determined and supplements were terminated. Data were analyzed using the MIXED procedures of SAS. Body weight and BCS, ovulatory follicular wavelength, antral follicle count, size of ovulatory follicle at dominance and duration of dominance were not different ( $P = 0.13$ ) between treatments. Cows supplemented with CGM had greater ( $P < 0.01$ ) post-dominance dominant follicle growth, larger ( $P = 0.03$ ) dominant follicles at spontaneous luteolysis, shorter proestrus (36 vs. 68 h;  $P < 0.01$ ) and larger (15.4 vs. 13.5 mm;  $P = 0.03$ ) ovulatory follicles than SBM cows. No differences ( $P = 0.11$ ) in estradiol, ratio of estradiol to ovulatory follicle volume, or plasma urea nitrogen concentrations were observed between treatments. Although CL volume and the ratio of progesterone to CL volume were not affected



by treatment ( $P = 0.24$ ), CGM treated cows tended to have lesser (4.7 vs. 5.7 ng/mL;  $P = 0.07$ ) concentrations of circulating progesterone 7d post-estrus than SBM. In summary, these data illustrate that excess MP when supplemented to cows consuming a low quality forage may differentially affect ovarian function depending on rumen degradability of the protein source.

**Key Words:** follicle, ovary, RUP

**314 Effects of supplementing excess amounts of MP from a moderately abundant RUP source on ovarian function of beef cows consuming low quality forage.** Taylor C. Geppert<sup>\*1</sup>, George A. Perry<sup>2</sup>, and Patrick J. Gunn<sup>1</sup>, <sup>1</sup>*Department of Animal Science, Iowa State University, Ames, IA*, <sup>2</sup>*Department of Animal Sciences, South Dakota State University, Brookings, SD*.

The objective of this experiment was to determine the effects of excess MP supplementation from a moderately abundant RUP source on ovarian function in beef cows consuming low quality forage. Non-pregnant, nonlactating mature beef cows ( $n = 16$ ) were allocated by age, BW and BCS to 1 of 2 isocaloric supplements designed to maintain BW for 60 d. All cows were offered ad-libitum access to corn stalks and individually offered a corn gluten meal-based supplement at (1) 125% (MP125) or (2) 150% (MP150) of NRC MP requirements. After a 20-d supplement adaptation period, cows were synchronized for ovulation using the 5-d CO-Synch + CIDR protocol. Ten days after synchronization, 100  $\mu$ L of GnRH was administered to reset follicular growth. Starting at GnRH administration and daily thereafter, transrectal ultrasonography was performed to diagram follicular waves, and blood samples were collected for hormone and metabolite analyses. Seven days after observation of estrus, corpus luteum (CL) size was determined via ultrasound and supplementation was terminated. Data were analyzed using the MIXED procedures of SAS. There were no differences ( $P \geq 0.21$ ) between BW and BCS. Preovulatory ovarian follicle characteristics including size at dominance, duration of dominance, post-dominance growth, size at spontaneous luteolysis, post-luteolysis growth, length of proestrus, and wavelength were not different between treatments ( $P \geq 0.11$ ). However, ovulatory follicles were larger ( $P = 0.04$ ) and average antral follicle count was greater ( $P = 0.01$ ) in the MP150 than MP125 treatment. Estradiol concentrations and the ratio of estradiol to ovulatory follicle volume was not affected ( $P \geq 0.51$ ) by treatment. Although CL volume 7 d post-estrus was greater ( $P < 0.01$ ) in MP150 than MP125, circulating progesterone 7 d post-estrus and the ratio of progesterone to CL volume was not different ( $P \geq 0.21$ ). In conclusion, supplementation of CP at 150% of NRC MP requirements from a moderately undegradable protein source may enhance growth of the ovulatory follicle and subsequent CL compared with MP supplementation at 125% of NRC requirements.

**Key Words:** crude protein, follicle, ovulation

**315 Effect of top-dressing rumen-protected methionine in lactating Holstein cows II: Fertility and embryo development.** Mateus Z. Toledo<sup>\*1</sup>, Giovanni M. Baez<sup>1</sup>, Eduardo Trevisol<sup>1</sup>, Nelson E. Lobos<sup>1</sup>, Alvaro Garcia-Guerra<sup>1</sup>, Jerry N. Guenther<sup>1</sup>, Daniel Luchini<sup>2</sup>, Randy D. Shaver<sup>1</sup>, and Milo C. Wiltbank<sup>1</sup>, <sup>1</sup>*University of Wisconsin-Madison, Madison, WI*, <sup>2</sup>*Adisseo, Alpharetta, GA*.

Experimental objectives were to evaluate the effects of supplementation with rumen-protected methionine (RPM) from 31  $\pm$  2 to 127  $\pm$  2 DIM (61 d after timed AI; TAI) on fertility and embryo development of dairy cows. Holstein cows ( $n = 309$ ) were housed in a free-stall barn, milked twice daily, fed a basal diet formulated to 16.7% CP to

deliver 2521 g of metabolizable protein (MP) with 6.93 lysine as % of MP and randomly assigned to once daily top-dressing with either (1) RPM, 21.2 g of Smartamine M mixed with 38.8 g of dry distillers grains (2.34 methionine as % of MP) or (2) Control (CON), 60 g of dry distillers grain (1.87 methionine as % of MP). All cows were synchronized using a Double Ovsynch protocol (GnRH-7d-PGF-3d-GnRH-7d-GnRH-7d-PGF-56h-GnRH-16h-TAI) to receive first TAI at 66  $\pm$  3 DIM. Pregnancy diagnosis was done at 28 d (pregnancy-specific protein B) and at 32 and 61 d (ultrasound) after TAI. Embryo size and amniotic vesicle volume were determined by ultrasound on d 33 of pregnancy. Binomial data were analyzed by parity using Chi-squared or Fisher's exact test. Continuous outcomes were analyzed by *t*-test or Wilcoxon sum-rank test. Only synchronized cows (92.1%, determined by progesterone concentrations) were used in the final analysis ( $n = 285$ ). There was no effect of treatment on pregnancies per AI at 28 (65.5 vs. 66.7%; CON vs. RPM), 32 (58.6 vs. 61.4%), or 61 (54.4 vs. 58.3%) d after TAI. However, pregnancy loss was greater in multiparous cows for CON compared with RPM cows from 28 to 61 d (19.6 vs. 6.1%;  $P = 0.04$ ) or from 32 to 61 d (8.9 vs. 0.0%,  $P = 0.03$ ). However, there was no effect of treatment on pregnancy loss in primiparous cows (28 to 61 d,  $P = 0.50$ ; 32 to 61 d,  $P = 0.50$ ). Consistent with data on pregnancy loss, RPM increased embryonic size in multiparous cows (RPM vs. CON; amniotic vesicle volume, 592 vs. 472 mm<sup>3</sup>,  $P = 0.03$ ; embryo abdominal diameter, 11.0 vs. 10.5 mm,  $P = 0.01$ ; crown-rump length, 5.8 vs. 5.4 mm,  $P = 0.13$ ), but had little effect on embryonic size in primiparous cows. Thus, top-dressing RPM increased embryo size and pregnancy maintenance in multiparous cows.

**Key Words:** methionine, dairy cattle, reproduction

**316 Intravenous glucose infusion in early postpartum dairy cows: Effects on plasma metabolites, milk production, and interval to first ovulation.** Stephen Butler<sup>\*1</sup>, Shane Leane<sup>1</sup>, Francis Curran<sup>1</sup>, Stephen Moore<sup>1</sup>, Mark Crowe<sup>2</sup>, and Matthew Lucy<sup>3</sup>, <sup>1</sup>*Teagasc Moorepark-Animal & Grassland Research and Innovation Centre, Fermoy, Ireland*, <sup>2</sup>*School of Veterinary Medicine, University College Dublin, Dublin, Ireland*, <sup>3</sup>*Department of Animal Sciences, University of Missouri, Columbia, MO*.

Glucose supply is less than required during early lactation. The glucose deficit affects the homeostatic mechanisms that coordinate milk production and also impinges upon reproduction. The effect of glucose infusion during early lactation was tested. Postpartum dairy cows ( $n = 22$ ) that calved in either the autumn ( $n = 11$ ) or spring ( $n = 11$ ) were treated with either glucose (GLUC;  $n = 11$ ) or saline (SAL;  $n = 11$ ). The GLUC cows received 750 g of glucose per day via continuous intravenous infusion of 40% dextrose solution (0.52 g glucose/min). The SAL cows received an equal volume of 0.9% saline. Infusions began during the second week postpartum (7.9  $\pm$  1.9 d) and continued for 14 d. Blood was collected 4 times daily. Data were analyzed by using PROC MIXED of SAS. Compared with SAL, the GLUC cows had greater blood glucose concentrations (69.7  $\pm$  1.5 vs. 61.7  $\pm$  1.5 mg/dL;  $P < 0.01$ ). There was a tendency ( $P < 0.11$ ) for a treatment by season interaction for plasma glucose (spring: 71.6  $\pm$  2.3 vs. 60.0  $\pm$  2.5 ng/mL,  $P < 0.01$ ; autumn: 67.7  $\pm$  2.0 vs. 63.4  $\pm$  1.8 mg/mL,  $P > 0.10$ ; GLUC vs. SAL) and there was a treatment by season interaction ( $P < 0.01$ ) for plasma insulin (spring: 4.4  $\pm$  0.4 vs. 3.1  $\pm$  0.4 ng/mL,  $P < 0.05$ ; autumn: 3.4  $\pm$  0.5 vs. 3.1  $\pm$  0.5 ng/mL,  $P > 0.10$ ; GLUC vs. SAL). GLUC cows had lesser plasma nonesterified fatty acids (NEFA; 448  $\pm$  39 vs. 580  $\pm$  39  $\mu$ Eq/L;  $P < 0.03$ ) and lesser plasma  $\beta$  hydroxybutyrate (BHBA; 0.63  $\pm$  0.10 vs. 1.15  $\pm$  0.10 mM;  $P < 0.01$ ). Insulin sensitivity (RQUICKI) was less in spring cows compared with autumn cows (0.50  $\pm$  0.01 vs 0.53

± 0.01;  $P < 0.05$ ). There was no effect of treatment on milk produced (27.8 ± 1.1 vs. 27.8 ± 1.1 kg/d; GLUC vs. SAL) or as-fed consumption of total mixed ration (44.1 ± 1.6 vs. 45.1 ± 1.7 kg/d; GLUC vs. SAL). The number of cows with first ovulation during the infusion was not affected by treatment [5/11 (45%) for GLUC and SAL] and interval to first ovulation was similar (13.7 ± 0.9 d postpartum). There was a tendency ( $P < 0.10$ ) for more autumn cows [7/11 (64%)] compared with spring cows [3/11(27%)] to ovulate during infusion. Glucose infusion affected plasma hormone and metabolite concentrations but did not affect milk production or interval to first ovulation.

**Key Words:** glucose, dairy, reproduction

**317 Intravenous glucose infusion during pregnancy in dairy cows: Effects on plasma hormones, metabolites, milk production, and conceptus growth.** Matthew Lucy<sup>\*1</sup>, Shane Leane<sup>2</sup>, Francis Curran<sup>2</sup>, Stephen Moore<sup>2</sup>, Mark Crowe<sup>3</sup>, and Stephen Butler<sup>2</sup>, <sup>1</sup>Department of Animal Sciences, University of Missouri, Columbia, MO, <sup>2</sup>Teagasc Moorepark-Animal & Grassland Research and Innovation Centre, Fermoy, Ireland, <sup>3</sup>School of Veterinary Medicine, University College Dublin, Dublin, Ireland.

Glucose supply in postpartum dairy cows may impinge upon early pregnancy by affecting maternal and placental endocrinology as well as substrate availability to the developing conceptus. The objective was to test the effect of glucose infusion from d 32 to 45 of pregnancy on plasma hormones and metabolites, milk production and conceptus growth. Dairy cows (n = 10) were assigned to glucose (GLUC; n = 5) or saline (SAL; n = 5). The GLUC cows received 750 g of glucose per d via continuous intravenous infusion of 40% dextrose (0.52 g glucose/min). The SAL cows were infused with an equal volume of 0.9% saline. Infusions began on d 32 of pregnancy (98 ± 15 d postpartum) and ended on d 45 of pregnancy. Blood was collected twice daily during the infusion. Fetal and amniotic vesicle length and width were measured on d 31 (1 d before infusion) and d 33, 35, 37, 39, 41, 43 and 45. Data were analyzed by using PROC MIXED of SAS. Compared with SAL, the GLUC cows had greater blood glucose (81.7 ± 2.1 vs. 74.2 ± 2.1 mg/dL;  $P < 0.05$ ). Plasma progesterone (P4; 9.5 ± 1.3 vs. 9.5 ± 1.3 ng/mL), insulin (7.7 ± 1.7 vs. 7.0 ± 1.7 ng/mL), IGF1 (159 ± 27 vs. 143 ± 27 ng/mL), β hydroxybutyrate (0.93 ± 0.08 vs. 1.02 ± 0.08 mM), nonesterified fatty acids (280 ± 10 vs. 264 ± 10 μEq/L) and pregnancy-associated glycoproteins (PAG; 64 ± 21 vs. 73.2 ± 21% of assay control) were not affected ( $P > 0.10$ ; GLUC vs. SAL). There was no effect of treatment on milk yield (26.8 ± 2.4 vs. 24.6 ± 2.2 kg/d; GLUC vs. SAL) or as-fed consumption of total mixed ration (63 ± 4 vs. 67 ± 3 kg/d; GLUC vs. SAL). There was an effect of day of pregnancy ( $P < 0.001$ ) but no effect of treatment on length, width, or volume of the fetus or amniotic vesicle. All hormone and metabolic data were provided as independent variables to explain variation in conceptus measurements using backward elimination (GLMSELECT of SAS). The only significant effects remaining were day ( $P < 0.001$ ) and plasma progesterone (P4;  $P < 0.001$ ; positive association for P4 with all conceptus measurements). Glucose infusion from d 32 to 45 of pregnancy increased plasma glucose but did not affect growth of the conceptus. Greater plasma P4 was associated with larger conceptuses.

**Key Words:** glucose, pregnancy, dairy

**318 Rumen-protected methyl donors during late pregnancy: 1. Maternal Smartamine M and its association with neonatal Holstein calf blood immunometabolic biomarkers.** Carolina Besspahlak Jacometo<sup>\*1</sup>, Zheng Zhou<sup>2</sup>, Erminio Trevisi<sup>3</sup>, Daniel Luchini<sup>4</sup>,

Marcio Nunes Corrêa<sup>1</sup>, and Juan J. Looor<sup>2</sup>, <sup>1</sup>Universidade Federal de Pelotas, Pelotas, Rio Grande do Sul, Brazil, <sup>2</sup>University of Illinois, Urbana, IL, <sup>3</sup>Università Cattolica del Sacro Cuore, Piacenza, Italy, <sup>4</sup>Adisseo NA, Alpharetta, GA.

The aim was to evaluate the effect of supplementing pregnant cows with rumen-protected methionine (MET) on growth and blood biomarkers of hepatic and energy metabolism, inflammation and oxidative stress. Forty Holstein calves born to cows receiving during the last ~4 wk of pregnancy MET (Smartamine M, Adisseo NA.; ~2.9:1 Lys:Met; n = 20) or control (CON, ~3.35:1 Lys:Met, n = 20) were used. Immediately after birth calves were separated from the dam, fed first colostrum (3.8 L with minimum IgG concentration of 50 g/L), housed individually and fed a common milk replacer (25% CP, 17% fat) twice daily. Calves were bled at birth (before colostrum), 24 h after first colostrum, at 14, 28 and 50 (~1 wk post-weaning) d of age. Data were analyzed as repeated measures using the MIXED procedure of SAS. No maternal diet effect ( $P > 0.05$ ) was observed in calf growth (body weight and withers height) from birth through weaning. MET calves had lower glucose at birth (4.05 vs. 4.73 mmol/L,  $P > 0.01$ ), but there was no overall maternal diet effect ( $P = 0.18$ ). Regardless of maternal diet, glucose, AST and GGT increased markedly ( $P < 0.01$ ) from birth to 24 h after colostrum intake, then decreased ( $P < 0.01$ ) at 14 d and remained unchanged until 50 d. NEFA and creatinine concentrations had a sharp decrease after birth ( $P < 0.01$ ) while BHBA concentrations increased ( $P < 0.01$ ) over time. Paraoxonase, albumin and ceruloplasmin concentration increased ( $P < 0.01$ ) over time. MET calves had lower albumin (30.1 vs. 30.9 g/L,  $P = 0.09$ ) and ceruloplasmin tended to be lower (1.58 vs. 1.85 μmol/L,  $P = 0.11$ ). IL1-B and IL-6 had a marked decrease ( $P < 0.01$ ) from birth to 24 h after colostrum intake. Tocopherol (1.31 vs. 2.19 μg/mL), myeloperoxidase (466 vs. 544 U/L) and ROMt (12.4 vs. 15.5 mg H<sub>2</sub>O<sub>2</sub>/100 mL) were lower ( $P < 0.05$ ) in MET calves at 14 d of age. Retinol increased over time ( $P < 0.01$ ). Overall, data suggest that maternal supplementation with MET during the last ~4 wk of gestation affected some biomarkers of metabolism and oxidative stress, hence, seemed to elicit a beneficial effect on the neonatal calf.

**Key Words:** dairy cattle, fetal programming, nutrition

**319 Rumen-protected methyl donors during late pregnancy: 2. Maternal Smartamine M and its association with hepatic gene expression in neonatal Holstein calves.** Carolina Besspahlak Jacometo<sup>\*1</sup>, Zheng Zhou<sup>2</sup>, Daniel Luchini<sup>3</sup>, Marcio Nunes Corrêa<sup>1</sup>, and Juan J. Looor<sup>2</sup>, <sup>1</sup>Universidade Federal de Pelotas, Pelotas, Rio Grande do Sul, Brazil, <sup>2</sup>University of Illinois, Urbana, IL, <sup>3</sup>Adisseo NA, Alpharetta, GA.

The aim was to evaluate the effect of supplementing pregnant cows with rumen-protected methionine (MET) on neonatal calf liver expression of genes related to energy/lipid metabolism, insulin signaling, growth hormone signaling and inflammation. Forty Holstein calves born to cows receiving during the last ~4 wk of pregnancy MET (Smartamine M, Adisseo NA.; ~2.9:1 Lys:Met; n = 20) or control (CON, ~3.35:1 Lys:Met, n = 20) were used. Immediately after birth calves were separated from the dam, fed first colostrum within 6 h (3.8 L with minimum IgG concentration of 50 g/L), housed individually and fed a common milk replacer (25% CP, 17% fat) twice daily. Liver biopsies were harvested (n = 8/group) at 4, 14, 28 and 50 (~1 wk post-weaning) d of age. Data were analyzed as repeated measures using the MIXED procedure of SAS. No maternal diet effect ( $P > 0.05$ ) was observed on calf growth (body weight and withers height) from birth through weaning. Expression of genes related to lipoprotein metabolism (*APOB*, *MTTP*) and growth hormone signaling (*IGFI*, *GHR1A*) were not ( $P > 0.05$ ) affected by

maternal diet, but increased in expression over time ( $P < 0.05$ ). *PCK1* and *FBP1* expression was greater ( $P = 0.05$  and  $0.02$ ) in MET calves and increased ( $P < 0.001$ ) over time in both groups. *PC* expression, however, was lower ( $P = 0.007$ ) in MET calves and decreased ( $P < 0.001$ ) over time in both groups. Lower ( $P = 0.001$ ) *ACOX1* expression was observed in MET, while *CPT1A* was greater ( $P < 0.001$ ). The insulin-signaling related genes *AKT2* and *SLC2A2* had greater ( $P < 0.01$ ) expression in MET calves. Except for *FOXO1* and *SLC2A2*, all other genes evaluated in this pathway (*INSR*, *IRS1*, *AKT2*, *SREBF1*) increased ( $P < 0.05$ ) expression over time regardless of maternal diet. MET calves had higher *NFKB* ( $P = 0.009$ ) and *SOD2* ( $P < 0.001$ ) expression, and also a trend ( $P = 0.08$ ) for higher *SOD1*. Overall, the data suggest that maternal supplementation with MET during the last ~4 wk of gestation elicited changes in calf hepatic gene expression and, as such, might have led to functional differences in improving neonatal energy metabolism.

**Key Words:** fetal programming, nutrition, nutrigenomics

### 320 Rumen-protected methyl donors during late pregnancy:

**3. Maternal Smartamine M and its association with neonatal Holstein calf neutrophil gene network expression.** Carolina Bespalhok Jacometo<sup>\*1</sup>, Zheng Zhou<sup>2</sup>, Erminio Trevisi<sup>3</sup>, Daniel Luchini<sup>4</sup>, Marcio Nunes Corrêa<sup>1</sup>, and Juan J. Looz<sup>2</sup>, <sup>1</sup>Universidade Federal de Pelotas, Pelotas, Rio Grande do Sul, Brazil, <sup>2</sup>University of Illinois, Urbana, IL, <sup>3</sup>Università Cattolica del Sacro Cuore, Piacenza, Italy, <sup>4</sup>Adisseeo NA, Alpharetta, GA.

The aim was to evaluate the effect of supplementing pregnant cows with rumen-protected methionine (MET) on calf neutrophil expression of genes related to cell adhesion and chemotaxis, oxidative stress and inflammation. Forty Holstein calves born to cows receiving during the last ~4 wk of pregnancy MET (Smartamine M, Adisseeo NA; ~2.9:1 Lys:Met;  $n = 20$ ) or control (CON, ~3.35:1 Lys:Met,  $n = 20$ ) were used. Immediately after birth calves were separated from the dam, fed first colostrum within 6 h (3.8 L with minimum IgG concentration of 50 g/L), housed individually and fed a common milk replacer (25% CP, 17% fat) twice daily. Blood neutrophils were isolated at birth (before receiving colostrum), 24 h after first colostrum and at 14, 28 and 50 (~1 wk post-weaning) d of age. Data were analyzed as repeated measures using the MIXED procedure of SAS. Neutrophil phagocytosis was not affected ( $P > 0.05$ ) by maternal MET supplementation, but increased ( $P < 0.01$ ) over time in both groups. Regardless of maternal diet *SELL*, *CADMI*, *LCP1* and *CYBA* expression increased ( $P < 0.05$ ) from birth to 24 h after colostrum intake, then decreased ( $P < 0.05$ ) until 28 d. *ZBPI* increased ( $P < 0.01$ ) from birth to 28 d. *SELL* expression was overall greater ( $P = 0.04$ ) in MET than CON calves. Expression of genes related to oxidative stress (*MPO*, *NOS2*, *SOD1*, *SOD2*, *NFE2L2*) was not affected ( $P > 0.05$ ) by maternal diet. Similarly, blood biomarkers related to oxidative stress (ROMt, myeloperoxidase, retinol, tocopherol) were not affected ( $P > 0.05$ ) by diet. *TLR2* had lower ( $P = 0.04$ ) expression in MET calves, but other inflammatory mediators (*TLR4*, *MYD88*, *IRAK1*, *TRAF6*, *NFKB*, *TNF*, *IL1B*, *SLAMF7*) and blood IL-1B and IL-6 concentrations were not affected ( $P > 0.05$ ). A marked decrease ( $P < 0.01$ ) in both cytokines from birth to 24 h after colostrum intake was observed regardless of diet. Overall, the data suggest that maternal supplementation with MET during the last ~4 wk of gestation had a minor effect on calf neutrophil gene network expression and blood biomarkers of oxidative stress and inflammation.

**Key Words:** fetal programming, nutrition, nutrigenomics

**321 Chromium supplementation alleviates heat stress in growing pigs.** Fan Liu<sup>\*1</sup>, Jeremy J Cottrell<sup>1</sup>, Danni Wijesiriwardana<sup>1</sup>, Fletcher W. Kelly<sup>1</sup>, Pietro Celi<sup>2,1</sup>, Brian J. Leury<sup>1</sup>, and Frank R. Dunshea<sup>1</sup>, <sup>1</sup>Faculty of Veterinary and Agricultural Sciences, the University of Melbourne, Parkville, VIC, Australia, <sup>2</sup>Faculty of Veterinary Science, the University of Sydney, Camden, NSW, Australia.

Reduced insulin sensitivity is a characteristic of heat stress (HS) in pigs. Therefore the aim of the experiment was to investigate the effect of chromium (Cr) supplementation in ameliorating HS due to its properties augmenting insulin sensitivity. Thirty-six gilts (Large White × Landrace,  $29 \pm 4$  kg) were randomly assigned to 2 diets containing 0 (control) or 3200 ppb Cr picolinate (400 ppb Cr). After 14 d supplementation pigs were allocated to 8 d thermoneutral (20°C; TN) or cyclic HS (8h/d 35°C) ( $n = 9$ /group). Production performance was recorded in the thermal exposure period. Respiration rate (RR) and rectal temperature (RT) were measured at 0900, 1300 and 1600h daily, and blood gas was measured on d 7. Area under the curve (AUC) of glucose and NEFA was studied in an intravenous glucose tolerance test ( $n = 6$ /group) on d 8. Data were analyzed by ANOVA in Genstat. In TN pigs fed Cr diet had higher ADFI (2.0 vs. 2.3 kg,  $P < 0.05$ ), but ADG was not improved (0.63 vs. 0.69 kg,  $P = 0.26$ ). Heat stress decreased ADFI by 35% and ADG by 84% (both  $P < 0.001$ ) and no effect of Cr was observed. Heat stress increased RT (38.8 vs. 40.0°C,  $P < 0.001$ ) and RR (34 vs. 155 breaths/min,  $P < 0.001$ ). The increased RR led to reductions in blood CO<sub>2</sub>, bicarbonate and base excess in HS (all  $P < 0.05$ ). Collectively the results indicate that heat treatment resulted in a “heat stressed” state. Besides, HS increased glucose and decreased NEFA AUC (both  $P = 0.05$ ), suggesting reduced insulin sensitivity. Compared with control diet, Cr pigs had lower RT (40.2 vs. 39.9,  $P < 0.05$ ) and RR (173 vs. 136,  $P < 0.01$ ) under HS, indicating an amelioration in the level of HS experienced in Cr pigs. Chromium did not alter the glucose AUC in HS, although Cr reduced the glucose “basal to peak” increment in TN (4.15 vs. 2.55 mM,  $P < 0.05$ ). Besides, Cr tended to increase NEFA recovery (20 to 90 min) rate and AUC in HS (both  $P = 0.09$ ), indicating that Cr facilitated lipid mobilization in HS. In summary, pig growth performance was not improved by Cr during HS, possibly because of the severe reduction in ADFI. However, dietary Cr mitigated the physiological responses to HS, including lipid mobilization. Therefore, an inclusion of 400 ppb Cr may reduce HS in growing pigs.

**Key Words:** chromium, pig, insulin

**322 Metabolome-based relationships of four biofluids from dairy cows.** H. Z. Sun<sup>\*1,2</sup>, L. L. Guan<sup>3</sup>, and J. X. Liu<sup>1,2</sup>, <sup>1</sup>Institute of Dairy Science, College of Animal Sciences, Hangzhou, China, <sup>2</sup>MoE Key Laboratory of Molecular Animal Nutrition, Zhejiang University, Hangzhou, China, <sup>3</sup>Department of Agricultural, Food & Nutritional Science, University of Alberta, Edmonton, Canada.

To date, most studies have focused on the improvement of dairy nutritional management strategies to enhance milk production and quality, whereas the physiological and metabolic mechanisms involved have not been well examined. This study was conducted to investigate the relationships of 4 biofluids from lactating dairy cow to identify mechanisms and potential biomarkers as well as key pathways involved in potential regulation of lactation. Eight multiparous Holstein dairy cows with similar lactation stage ( $164 \pm 19.6$  d, mean  $\pm$  SD) and milk yield ( $30.4 \pm 2.29$  kg, mean  $\pm$  SD) were offered a diet with 16.7% (DM basis) of crude protein and 1.57 Mcal/kg of net energy for lactation.



Rumen fluid, serum, milk, and urine were collected after 65-d feeding to identify the metabolome using gas chromatography–time of flight/mass spectrometry. A total of 165, 195, 218 and 156 metabolites were identified from rumen fluid, milk, serum, and urine, respectively, with 29 metabolites detected in all 4 biofluids. The TIC chromatograms showed a clear discrimination among 4 biofluids; and principal component analysis of the relative concentration of mutual metabolites revealed 4 separated metabolite profile clusters of 4 biofluids. The clusters derived from the rumen fluid, milk, and serum partly overlapped with each other, whereas the cluster from the urine was separated from the other 3 biofluids. The dendrogram of hierarchical clustering analysis revealed different subclusters containing varying numbers of metabolites within each biofluid; and the subclusters from rumen fluid and serum

were grouped together and highly correlated with each other, but were separated from milk. Based on metabolomic profiles, urine is the most different biofluid, compared with other 3 biofluids. When the mutual metabolites were used for pathway analysis, the impact values of the pathway were 0.29, 0.28, and 0.11 for glycine, serine and threonine metabolism, glycerolipid metabolism, and tyrosine metabolism, respectively. These 3 pathways may play important roles in improvement of lactation performance. Our results suggest that all the 4 biofluids represent the comprehensive metabolism of dairy cow that can be further used for metabolic pathway analysis.

**Key Words:** biofluid, metabolomics, dairy cow

# Physiology and Endocrinology Symposium: Progesterone as an endocrine regulator of fertility in cattle

## 323 The role of progesterone in uterine biology of ruminants.

Thomas E. Spencer\*, *University of Missouri, Columbia, MO.*

This review integrates established and new information on the role of progesterone in uterine biology of ruminants. Establishment of pregnancy in ruminants occurs during the peri-implantation period and involves growth of the blastocyst, elongation of the conceptus (embryo and extraembryonic membranes), and suppression of the endometrial luteolytic mechanism to maintain progesterone production by the ovary. Conceptus elongation involves exponential increases in length and weight of the trophoblast and onset of extraembryonic membrane differentiation, including gastrulation of the embryo and formation of the yolk sac and allantois that are vital for embryonic survival and formation of a functional placenta. Antiluteolytic effects of the elongating conceptus are due to production of interferon tau (IFNT) by the trophoblast that has a paracrine effect to inhibit upregulation of oxytocin receptors in the endometrial epithelia, thereby disrupting uterine release of luteolytic prostaglandin  $F_{2\alpha}$  (PGF) pulses. Survival of the blastocyst and elongation of the conceptus requires embryotrophic factors (ions, amino acids, carbohydrates, proteins, lipids, and other substances) from the epithelia of the uterus, and those embryotrophic factors are primarily regulated by progesterone. Available results from studies in sheep support the idea that the individual, interactive, and coordinated actions of progesterone, IFNT, prostaglandins, and cortisol regulate expression of elongation- and implantation-related genes in the endometrial epithelia and are essential regulators of conceptus elongation. The outcome of these gene expression changes is alterations in endometrial epithelial secretions that govern conceptus elongation. Elevated concentrations of circulating progesterone immediately after conception have been associated with an advancement of conceptus elongation, an increase in IFNT production and, in some studies, higher pregnancy rates in cattle. An increased knowledge of progesterone biology and conceptus-endometrial interactions is necessary to understand and elucidate the causes of recurrent pregnancy loss and to provide a basis for new strategies to improve pregnancy outcome and reproductive efficiency in ruminants.

**Key Words:** uterus, conceptus, progesterone

## 324 Cellular and molecular mechanisms controlling corpus luteum function and progesterone concentrations in cattle.

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Steady-state, circulating progesterone (P4) concentrations are determined primarily by 2 factors: the rate of P4 production, principally by the corpus luteum (CL), and the rate of P4 metabolism, mainly by the liver. Rate of P4 production is primarily related to number of large luteal cells (LLC), differentiation state of LLC, and provision of substrate for P4 production. Steroid production increases 1000-fold from estrus to d 7 with growth of the follicular granulosa cells from 10 mm diameter to 38 mm LLC, a 50-fold increase in cellular volume. The LLC of ruminants are distinguished by a large number of mitochondria and high constitutive P4 production meaning that luteotrophic hormones are not required for P4 production. Nevertheless, cholesterol substrate, primarily in the form of high-density lipoprotein (HDL) is essential for high P4 production by ruminant LLC. Circulating HDL is not limiting

in well-fed cattle but may be manipulated by diet. One limiting factor is number of granulosa cells in the follicle and therefore number of LLC. Ovulation of larger follicles or ovulation of multiple follicles can produce greater quantity of luteal tissue and therefore increased luteal P4 production. Regression of the CL is marked by decrease P4 production due to decreased transport of cholesterol to the inner mitochondrial membrane of the LLC. The rate-limiting step in cholesterol transport is STAR protein. Other steroidogenic enzymes remain active as P4 production by the CL plummets. Thus, luteal P4 production can be increased or decreased by hormonal, dietary, and management manipulations. Metabolic clearance rate (MCR) of P4 has been found to vary substantially between cows in different physiological conditions. For example, P4 MCR more than doubles in cows during high milk production as compared with similar size and age nonlactating cows. The P4 metabolism enzymes are concentrated in the liver. Thus, MCR of P4 and other steroids is primarily related to the rate of liver blood flow, which has been related to dry matter intake in dairy cattle. In conclusion, manipulation of P4 production and metabolism appear to be fertile areas for future research aimed at improving fertility in cattle.

**Key Words:** P4, corpus luteum, fertility

## 325 Novel concepts on the role of prostaglandins on luteal maintenance and maternal recognition of pregnancy in ruminants.

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In ruminants, the corpus luteum (CL) of early pregnancy is resistant to luteolysis. PGE<sub>2</sub> is considered as a luteoprotective mediator. Early studies indicate that during the establishment of pregnancy in sheep, a factor(s) from the conceptus or gravid uterus reaches the ovary locally through the utero-ovarian plexus (UOP) and protects the CL from luteolysis. The local nature of the embryonic antiluteolytic or luteoprotective effect precludes any direct effect of a protein transported or acting between the gravid uterus and CL in ruminants. At the time of establishment of pregnancy, interferon tau (IFNT) secreted by the trophoblast of the conceptus inhibits endometrial pulsatile release of PGF<sub>2α</sub> and increases endometrial PGE<sub>2</sub>. Our recent studies indicate that (1) luteal PG biosynthesis is selectively directed toward PGF<sub>2α</sub> at the time of luteolysis and toward PGE<sub>2</sub> during establishment of pregnancy; (2) the ability of CL of early pregnancy to resist luteolysis is likely due to increased intraluteal biosynthesis of PGE<sub>2</sub> and signaling; (3) IFNT protein is not transported from the uterus to the CL through the UOP vascular route; and (4) a large proportion of endometrial PGE<sub>2</sub> is transported from the uterus to the CL through the UOP vascular route during the establishment of pregnancy in sheep. Our ongoing studies indicate that intrauterine co-administration of IFNT and PGES-1 inhibitor inhibits IFNT action and IFNT fails to rescue the CL in cyclic sheep. By contrast, intrauterine co-administration of IFNT and PGES-1 inhibitor along with intraovarian administration of PGE<sub>2</sub> rescues the CL in cyclic sheep. These results suggest that IFNT may prolong the lifespan of the CL by increasing endometrial secretion of PGE<sub>2</sub> which in turn increases luteal PGE<sub>2</sub> biosynthesis and signaling and promotes luteal resistance. Together, our studies provide compelling evidence

that PGE2 produced by the CL in response to endometrial PGE2 may protect the CL, as a component of luteoprotective mechanisms, at the time of maternal recognition of pregnancy in ruminants. These novel findings may offer therapeutic strategies to target PGE2 biosynthesis and signaling selectively in the endometrium and/or CL to improve reproductive efficiency in ruminants.

**326 The role of progesterone receptor on control of ovulation and oviductal transport in mammals.** Rebecca Robker\*, Darryl Russell, and Lisa Akison, *University of Adelaide, Adelaide, South Australia, Australia.*

Progesterone is critical for many aspects of female reproduction such as ovulation, oviductal transport, implantation, and maintenance of pregnancy. Its effects are mediated by the progesterone receptor (PGR), which occurs in both PGR-A and PGR-B isoforms, acting as nuclear hormone transcription factors. In the ovary, PGR is transiently expressed, specifically in granulosa cells of preovulatory follicles in response to the luteinizing hormone (LH) surge that triggers ovulation. To identify PGR regulated genes that are responsible for these key reproductive events we conducted microarray analyses of mRNA from granulosa cells and oviducts comparing progesterone receptor null mice (PRKOs) and heterozygous littermates. Microarray results confirmed that proteases Adamts1 and Cathepsin L are expressed at greatly reduced levels in PRKO granulosa cells, and identified novel gene products involved in cellular migration and invasion processes that are likely to mediate the dynamic tissue remodeling that facilitates ovulation. In the oviduct, PGR is expressed predominantly in luminal epithelial cells but also muscle cells. A large number of oviductal genes were also dysregulated in PRKO mouse oviducts during the peri-ovulatory period and we identified and subsequently validated several that have potential roles in cumulus-oocyte capture and transport following ovulation. The majority were genes associated with adhesion and muscular contractility including integrin  $\alpha$  8, endothelin 3, myocardin and angiotensin II receptor. Thus, the actions of PGR play key roles in coordinating the functions of multiple tissues, including the important peri-ovulatory events of oocyte release and acquisition of oocyte developmental competence, as well as subsequent oviductal transport of the newly formed embryo and immunological events at implantation.

**Key Words:** progesterone receptor, ovulation, oviduct

**327 Contrasting effects of progesterone on fertility of dairy and beef cows.** Jeffrey S. Stevenson\*<sup>1</sup> and G. Cliff Lamb<sup>2</sup>, <sup>1</sup>*Kansas State University, Manhattan, KS,* <sup>2</sup>*University of Florida, Marianna, FL.*

Role of progesterone in maintaining pregnancy is well known in the bovine. Subtle differences exist between dairy (milked) and beef (suckled) cows because of differing concentrations of progesterone during recrudescence of postpartum estrous cycles, rate of follicular growth and maturation, proportions of 2- and 3-follicular wave cycles, and other effects on pregnancy outcomes per AI (P/AI). Because proportions of anovulatory cows at the onset of the AI period are greater and more variable in beef (usually ranging from 30 to 70%) than dairy (25%) cows, AI programs were developed to accommodate anovulatory and cycling beef cows enrolled therein. Incorporating a progestin as part of an AI program in beef cows improved P/AI by reducing the proportion of cows having premature luteal regression and short post-AI luteal phases. Two- vs. 3-follicle wave cycles are more common in beef cows (86 vs. 14%) than in dairy cows (68 vs. 32%), respectively, even though P/AI may not differ in 2- vs. 3-wave beef cows (82 vs. 100%), but dif-

ferred in dairy cows (63 vs. 81%). When dominant follicles matured in subluteal-phase progesterone concentrations compared with those matured in luteal-phase concentrations, P/AI increased in beef cows, but were reduced in dairy cows when the first dominant follicle (matured in sub-luteal milieu) was induced to ovulate compared with cows ovulating a second-wave dominant follicle. Further, supplementing progesterone during growth of the first-wave dominant follicle improved fertility in dairy cows. Initiating timed AI programs in dairy cows in a greater progesterone environment and during cycle d 5 through 12 enhanced fertility. In contrast, progesterone status in beef cows at the onset of synchronization does not seem to be related to P/AI in multiparous cows, whereas P/AI were suppressed in primiparous cows that began a timed AI program in a low-progesterone environment. Pregnancy losses after AI between 35 and 60 to 70 d are less than 5% in beef cows and are not associated with pre-AI progesterone or cycling status, whereas losses in dairy cows (6 to 20%) are inversely related to progesterone and adversely affected in anovular dairy cows.

**Key Words:** beef and dairy cattle, progesterone, fertility

**328 The influence of progesterone (P4) during follicle development on endometrial and conceptus biology and fertility in dairy cows.** Rafael S. Bisinotto\*, Eduardo S. Ribeiro, Leandro F. Greco, Natalia Martinez, Leticia D. P. Sinedino, Fabio S. Lima, William W. Thatcher, and Jose E. P. Santos, *Department of Animal Sciences, University of Florida, Gainesville, FL.*

The effects of P4 concentrations during follicle growth on fertility responses in dairy cows were evaluated. In study 1, pregnancy per artificial insemination (P/AI) increased ( $P < 0.01$ ) in cows initiating the timed AI program in diestrus compared with anovular and cyclic cows without a corpus luteum (CL; 36.9, 25.1, 28.1%); however, pregnancy loss was smaller ( $P = 0.08$ ) in cyclic cows without CL, followed by cows in diestrus, and anovular cows (10.0, 13.5, 15.0%). In study 2, concepti on d 15 after AI were longer ( $P < 0.05$ ) in anovular compared with cyclic cows ( $47.8 \pm 8.8$  vs.  $9.4 \pm 5.8$  mm). Concepti transcriptome analysis depicted 417 genes differentially expressed in response to cyclic status, mostly involved with embryonic development. In study 3, cyclic cows were induced to ovulate the dominant follicle from the first wave (FW; P4 =  $1.2 \pm 0.3$  ng/mL), first wave supplemented with P4 (FWP4; P4 =  $4.3 \pm 0.3$  ng/mL), or second wave (SW; P4 =  $5.4 \pm 0.2$  ng/mL). Cows in FW had ( $P < 0.05$ ) larger ovulatory follicles ( $19.6 \pm 0.6$ ,  $15.6 \pm 0.6$ ,  $15.2 \pm 0.5$  mm), greater estradiol concentrations during proestrus ( $8.0 \pm 0.6$ ,  $7.0 \pm 0.7$ ,  $5.9 \pm 0.6$  pg/mL), and a faster P4 rise after AI. Concepti on d 17 after AI were longer ( $P = 0.05$ ) for FW compared with FWP4 and SW ( $16.6 \pm 2.3$ ,  $9.8 \pm 2.2$ ,  $9.6 \pm 2.0$  cm); however, major changes on transcriptome were not observed. Treatment did not affect any biological process in the endometrium on d 17 of gestation. In studies 4 and 5, a single ultrasound evaluation identified a low-fertility cohort based on the absence of CL at the initiation of the timed AI program. Increasing P4 concentrations to at least 2.0 ng/mL improved ( $P \leq 0.01$ ) P/AI in cows lacking CL similar to those in diestrus in the 5-d timed AI (No CL = 28.6, 2CIDR = 43.7, Diestrus = 47.3%) and the Ovsynch protocols (No CL = 28.9, 2CIDR = 37.2, Diestrus = 33.9%). Low concentrations of P4 during the development of the ovulatory follicle impair fertility and a minimum of 2.0 ng/mL seems to be needed to restore P/AI. Lack of differences on conceptus and endometrium on d 17 suggest that effects occur early in gestation.

**Key Words:** anovulation, embryo, progesterone



**329 Effect of manipulating progesterone before timed artificial insemination on reproductive and endocrine parameters in Irish Holstein-Friesian dairy cows.** Paul M. Fricke\*<sup>1</sup>, Paulo D. Carvalho<sup>1</sup>, Matthew C. Lucy<sup>2</sup>, Francis Curran<sup>3</sup>, Mary M. Herlihy<sup>3</sup>, and Stephen T. Butler<sup>3</sup>, <sup>1</sup>University of Wisconsin - Madison, Madison, WI, <sup>2</sup>University of Missouri, Columbia, MO, <sup>3</sup>Teagasc Moorepark-Animal & Grassland Research and Innovation Centre, Fermoy, Ireland.

Irish Holstein-Friesian dairy cows managed in a seasonal calving, grazing-based dairy system were randomly assigned to 2 treatments to manipulate progesterone (P4) before timed AI (TAI). Cows in the first treatment (High P4; n = 30) were submitted to a Double Ovsynch protocol [Pre-Ovsynch protocol (GnRH; 7 d, PGF<sub>2α</sub>; 3 d, GnRH) followed 7 d later by an Ovsynch-56 protocol (GnRH (G1); 7 d PGF<sub>2α</sub> (PGF); 24 h, PGF<sub>2α</sub>; 32 h, GnRH (G2); 16 h, TAI)]. Cows in the second treatment (n = 30; Low P4) received the same protocol with an additional PGF injection 24 h after G1. Data were analyzed using PROC MIXED and PROC GLIMMIX of SAS. Overall, synchronization rate was 92% (55/60) and did not differ between treatments. As expected, High P4 cows had more ( $P < 0.001$ ) CL at PGF ( $1.93 \pm 0.09$  vs.  $1.04 \pm 0.04$ ),

and P4 (ng/mL) did not differ between treatments at G1 ( $3.8 \pm 0.3$  vs.  $4.1 \pm 0.4$ ). Circulating P4 was greater ( $P < 0.001$ ) for High vs. Low P4 cows at PGF ( $8.5 \pm 0.9$  vs.  $1.6 \pm 0.4$ ) and G2 ( $0.30 \pm 0.04$  vs.  $0.08 \pm 0.02$ ). Although diameter (mm) of the preovulatory follicle was greater ( $P < 0.001$ ) for Low vs. High P4 cows at G2 ( $17.1 \pm 0.4$  vs.  $15.6 \pm 0.4$ ), neither CL diameter nor volume differed between treatments 15 d after TAI. The proportion of pregnant cows 39 and 60 d after TAI did not differ between treatments (63%; 17/27 vs. 61%; 17/28 for Low P4 vs. High P4 cows), and no pregnancy loss occurred. Both treatment ( $P = 0.006$ ) and time ( $P < 0.001$ ) affected P4 concentrations in pregnant cows from 1 to 39 d after TAI with Low P4 cows having greater P4 than High P4 cows. Treatment did not affect plasma pregnancy-associated glycoprotein (PAG) concentrations for cows diagnosed pregnant 39 d after TAI; however, pregnant primiparous cows had greater ( $P = 0.02$ ) PAGs from 20 to 39 d after TAI than pregnant multiparous cows. We conclude that although low P4 before TAI increased follicle growth before TAI and P4 concentrations in pregnant cows after TAI compared with high P4, low P4 before TAI did not negatively affect fertility, pregnancy loss, or PAGs after TAI in Irish Holstein-Friesian dairy cows.

**Key Words:** progesterone, timed AI, dairy cow

## Ruminant Nutrition: General

**330 Effect of trace minerals and different levels of starch on digestibility and ruminal fermentation in diets for dairy heifers.** Felipe Pino\* and Jud Heinrichs, *The Pennsylvania State University, State College, PA.*

Eight rumen cannulated dairy heifers ( $15.4 \pm 0.8$  mo of age and  $438.31 \pm 18.08$  kg of BW) were fed a high forage diet with 4 different levels of starch (3.5, 12.9, 22.3, and 31.7%) and 2 types of trace minerals (TM; organic and inorganic; OTM and ITM). Inorganic TM were provided as sulfates and organic TM as proteinate (Bioplex; Alltech, Inc.). The TM analyzed were Cu, Mn, Se, and Zn. Heifers were subject to a split-plot,  $4 \times 4$  Latin square design with 19-d periods; 15 d adaptation and 4 d sampling. The whole-plot factor was TM, and the subplot was starch concentration. Total collection of feces and urine were completed on d 15 to 19 to determine digestibility and TM excretion. Rumen contents were sampled (d 18 to 19) from 5 different locations in the rumen at 0, 1, 2, 4, 8, 12, 16, 20, and 22 h after feeding to measure pH and VFA concentrations. Plasma samples were collected to evaluate TM concentrations. Data were analyzed with the mixed procedure of SAS. Trace mineral intake was lower for OTM compared with ITM. No effect of TM on digestibility was detected, but as level of starch increased, DM digestibility increased ( $P = 0.08$ ). Rumen pH was lower for diets with OTM ( $P = 0.01$ ), which is consistent with higher total VFA production and butyrate proportion ( $P = 0.03$ ) observed for OTM diets. These variables may be explained by the higher bioavailability of OTM and faster utilization and fermentation by rumen microorganisms. Heifers that consumed ITM had higher moisture in feces and urine excretion, which increased total manure production ( $P < 0.01$ ). Excretion of TM was not different by treatment. Blood plasma was not different between treatments except for Mn that was higher for OTM. Because mineral intake was reduced and blood mineral levels were not different, these results suggest enhanced absorption of OTM compared with ITM. In conclusion, based on pH, VFA production and plasma TM concentration, OTM may be more ruminally bioavailable and absorbed in a greater extent than ITM. Also, the form of TM affects fecal moisture and urine excretion suggesting that ITM might stimulate water intake.

**Key Words:** trace mineral, starch digestibility, heifer

**331 Small intestinal starch digestion in cattle is linearly increased by duodenal glutamic acid flow.** Ethan J. Blom\*<sup>1</sup>, Derek W. Brake<sup>1</sup>, and David E. Anderson<sup>2</sup>, <sup>1</sup>South Dakota State University, Brookings, SD, <sup>2</sup>University of Tennessee Knoxville, Knoxville, TN.

Postruminal infusion of glutamic acid has been shown to increase small intestinal starch digestion in cattle. Our objective was to determine response among small intestinal starch digestion in cattle provided increasing amounts of postruminal glutamic acid. Five ruminally, duodenally, and ileally cannulated steers ( $351 \pm 11$  kg BW) were fed ( $5.1 \pm 0.06$  kg DM/d) a soybean hull-based diet that contained only slight amounts of starch (0.8% DM) and placed in a  $5 \times 5$  Latin square with 12-d periods. Cattle received (DM basis) continuous duodenal infusion of raw cornstarch ( $1.5 \pm 0.08$  kg/d), and either 0 (control),  $30.9 \pm 0.6$ ,  $62.4 \pm 1.2$ ,  $120.4 \pm 3.4$  g/d glutamic acid or  $407 \pm 18.3$  g/d casein (a positive control). Ileal and fecal nutrient flows were measured during the last 4 d of each period and CrEDTA served as an indigestible marker. Amounts of starch infused did not differ ( $P = 0.46$ ). Casein increased ( $P = 0.05$ ) small intestinal starch digestion. Similarly, small intestinal starch digestion increased linearly ( $P = 0.02$ ) with increasing amounts

of glutamic acid. Ileal flow of ethanol soluble starch tended to decrease (linear = 0.16) with greater amounts of glutamic acid, but ileal flow of ethanol soluble starch was not affected by casein ( $P = 0.42$ ). Ileal flow of glucose was small ( $8.5 \pm 3.0$  g/d); however, greater glutamic acid flow tended (quadratic = 0.08) to decrease ileal glucose flow. Fecal starch flow decreased in response to casein ( $P = 0.01$ ) and glutamic acid (linear = 0.04; cubic = 0.04); however, fecal flow of ethanol soluble starch and glucose were not affected by casein or glutamic acid. Postruminal starch digestion was increased by both casein ( $P = 0.02$ ) and glutamic acid (linear = 0.05; cubic = 0.03). Our data indicate that small intestinal starch digestion is linearly increased by postruminal glutamic acid flow.

**Key Words:** cattle, small intestinal starch digestion, glutamic acid

**332 Protein chemical profile, energy values, protein fractions, and rumen degradation characteristics of the newly developed yellow and brown carinata lines for dairy cattle compared with commercial canola seeds.** Yajing Ban\*, David A. Christensen, John J. McKinnon, and Peiqiang Yu, *Department of Animal and Poultry Science, College of Agricultural and Bioresources, University of Saskatchewan, Saskatoon, SK, Canada.*

*Brassica carinata* (Yellow: AAC A110; Brown: 110915EM) are newly developed oil crop lines for bio-fuel industry. However, there is little research on chemical profile, nutritive value and biodegradation characteristics of seeds as a feed source for animal diets. The objectives of this study were to systematically determine (1) chemical composition, (2) energy values, (3) protein fractions, (4) rumen protein degradation kinetics of the new yellow and brown carinata lines in comparison with brown commercial canola seeds, and to provide detailed information for feed registration in Canada. The 2 carinata lines were bred by Agriculture and Agri-Food Canada, and 2 sources of seeds were combined for each line. Four cannulated lactating dairy cows were used for rumen degradation trial. Protein fractions were partitioned based on Cornell Net Carbohydrate and Protein System (CNCPS). Statistical analyses were performed using PROC MIXED procedure of SAS 9.3 with significance declared at  $P \leq 0.05$ . The results showed that yellow carinata seeds were higher than canola seeds in crude protein, non-protein nitrogen but lower in neutral detergent insoluble crude protein and acid detergent insoluble crude protein ( $P \leq 0.05$ ). Yellow carinata seeds were not significantly different from canola seeds in total digestible nutrient (TDN) and energy values, while brown carinata seeds had the lowest TDN and energy values. Results from CNCPS indicated that yellow carinata seeds were lower in unavailable protein C fraction and higher in true protein ( $P \leq 0.05$ ), while brown carinata seeds had no significant difference with canola seeds in protein subfractions. Yellow carinata seeds had more rumen undegraded feed crude protein (RUP) and effective degradability of feed crude protein (EDCP) than canola seeds. In conclusion, *Brassica carinata* have potential to be utilized as a good feed protein source compared with commercial canola seeds, and the yellow carinata line had a higher protein supply than the brown line.

**Key Words:** carinata seed, protein fractions, rumen degradation

**333 Effect of an increased ruminal ammonia supply on lysine utilization by growing steers.** A. H. Hussein<sup>\*1</sup>, E. D. Batista<sup>1,2</sup>, M. D. Miesner<sup>3</sup>, and E. C. Titgemeyer<sup>1</sup>, <sup>1</sup>Department of Animal Sciences and Industry, Kansas State University, Manhattan, KS, <sup>2</sup>Departamento de Zootecnia, Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil, <sup>3</sup>Department of Clinical Sciences, Kansas State University, Manhattan, KS.

Six ruminally cannulated Holstein steers (202 ± 15 kg) were used to study effects of ruminal ammonia loading on whole-body lysine utilization. Steers were housed in metabolism crates and used in a 6 × 6 Latin square design. All steers received 2.52 kg of DM/d of a diet (10.1% CP) containing 82% soybean hulls, 8% wheat straw, and 5% cane molasses, and 10 g/d of urea was ruminally infused continuously to all steers to ensure adequate ruminal ammonia concentrations. All steers were ruminally infused continuously with 200 g/d acetic acid, 200 g/d propionic acid, and 50 g/d of butyric acid and abomasally infused with 300 g/d of glucose continuously to increase energy supply without increasing microbial protein supply. Steers were also abomasally infused continuously with an excess of all essential amino acids except lysine to ensure that lysine was the only limiting amino acid. Treatments were arranged as a 3 × 2 factorial with 3 levels of urea (0, 40, or 80 g/d) continuously infused ruminally to induce ammonia loading and 2 levels of lysine (0 or 6 g/d) continuously infused abomasally. Treatments did not affect fecal N output ( $P \geq 0.37$ ). Lysine supplementation decreased ( $P < 0.01$ ) urinary N excretion from 51.9 g/d to 44.3 g/d (SEM = 1.3), increased ( $P < 0.01$ ) retained N from 24.8 to 33.8 g/d (SEM = 1.2), and tended ( $P = 0.09$ ) to reduce plasma urea. Urea infusions linearly increased retained N (27.1, 29.3, and 31.5 g/d; SEM = 1.5;  $P = 0.05$ ) and also linearly increased ( $P < 0.01$ ) urinary N excretion (31.8, 48.1, and 64.4 g/d; SEM = 1.6), urinary urea-N (21.9, 37.7, and 54.3 g/d; SEM = 1.5), urinary ammonia-N (1.1, 1.4, and 1.9 g/d; SEM = 0.1), and plasma urea (2.7, 4.0, and 5.1 mM; SEM = 0.5). Assuming that retained protein is 6.25 × retained N and contains 6.4% lysine, the incremental efficiencies of infused lysine utilization were 51, 59, and 69% for steers receiving 0, 40, and 80 g/d of urea, respectively, suggesting that ruminal ammonia loads might improve efficiency of lysine utilization; this is supported by the observed increases in whole body protein deposition in response to ammonia loading of our steers that were, by design, lysine deficient.

**Key Words:** ammonia, lysine, cattle

**334 Relationship between antioxidant capacity, oxidative stress, and feed efficiency in beef steers.** J. R. Russell<sup>\*1</sup>, W. J. Sexten<sup>2</sup>, M. S. Kerley<sup>2</sup>, and S. L. Hansen<sup>1</sup>, <sup>1</sup>Iowa State University, Ames, IA, <sup>2</sup>University of Missouri, Columbia, MO.

The objective was to evaluate the relationship between feed efficiency (FE), antioxidant activity (ANTI) and oxidative stress (OXI) in feedlot steers representing phenotypic extremes for FE. Steers (n = 182) were fed a 70 d growing phase (GP) diet of whole shell corn (GCorn) or roughage (GRough) in GrowSafe bunks at University of Missouri, then shipped to Iowa State University where the 12 greatest (HFE) and 12 least (LFE) efficient steers from each diet (n = 48; 467 ± 48 kg) were selected for evaluation. Steers received diets similar to GP diets and 3 d after arrival, blood was sampled to evaluate ANTI and OXI markers for GP. Steers were transitioned to finishing phase (FP) corn (FCorn) or byproduct-based diets (FByp) and on FP d 97, blood samples for FP were collected. Data for GP were analyzed as a 2 × 2, and data for FP as a 2x2x2 using PROC MIXED of SAS. Looking at GP diet effects, GRough had greater ( $P = 0.04$ ) GP plasma protein carbonyl concentrations (PC) and a tendency for greater ( $P = 0.06$ ) FP total blood lysate superoxide dismutase activity (SOD) than GCorn. The FByp had

greater ( $P < 0.01$ ) PC during FP than FCorn and no other FP diet effects were noted ( $P > 0.2$ ). Analyzing FE group effects, during GP the HFE had greater ( $P < 0.04$ ) PC and ratio of oxidized:reduced blood lysate glutathione concentrations than LFE. There were GP diet × FE group interactions, as LFE-GRough had greater ( $P \leq 0.03$ ) GP total SOD and greater ( $P \leq 0.05$ ) FP glutathione peroxidase activity (GPX) compared with LFE-GCorn and HFE-GRough; HFE-GCorn was intermediate. During GP, the LFE-GRough had greater ( $P < 0.01$ ) GPX than other treatments and had greater ( $P = 0.03$ ) plasma malondialdehyde concentrations than LFE-GCorn. No GP diet × FP diet, FP diet × FE group, or 3-way interactions were noted ( $P > 0.3$ ). The GP diet and FE groups had stronger relationships with ANTI and OXI markers measured during the GP than the FP. Antioxidant activity may play a role in FE as LFE, driven largely by LFE-GRough, had greater SOD and GPX than HFE, potentially using a greater proportion of energy otherwise utilized for tissue accretion.

**Key Words:** antioxidant, feed efficiency, oxidative stress

**335 Efficiency of lysine utilization by growing steers.** Erick D. Batista<sup>\*1,2</sup>, Ali H. Hussein<sup>1</sup>, Matt Miesner<sup>1</sup>, Edenio Detmann<sup>2</sup>, and Evan C. Titgemeyer<sup>1</sup>, <sup>1</sup>Kansas State University, Manhattan, KS, <sup>2</sup>Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.

This study evaluated the efficiency of lysine utilization by growing steers. Five ruminally cannulated Holstein steers (165 kg ± 8 kg) housed in metabolism crates were used in a 6 × 6 Latin square design. Data from a sixth steer was excluded due to erratic feed intake. All steers were limit-fed (2.46 kg DM/d) twice daily diets low in RUP (81% soybean hulls, 8% wheat straw, 6% cane molasses, and 5% vitamins and minerals). Treatments were: 0, 3, 6, 9, 12, and 15 g/d of L-lysine abomasally infused continuously. To prevent amino acids other than lysine from limiting performance, a mixture providing all essential amino acids in excess was continuously infused abomasally. Additional continuous infusions included 10 g urea/d, 200 g acetate/d, 200 g propionate/d, and 50 g butyrate/d to the rumen and 300 g glucose/d to the abomasum. These infusions provided adequate ruminal ammonia and increased energy supply without increasing microbial protein supply. Each 6-d period included 2 d for adaptation and 4 d for total fecal and urinary collections for measuring N balance. Blood was collected on d 6 (10 h after feeding). Plasma urea-N was decreased ( $P = 0.01$ ) by lysine supplementation, but plasma glucose was unaffected ( $P = 0.73$ ). Diet DM digestibility was not altered ( $P = 0.94$ ) by treatment and averaged 72.2%. Increasing lysine supplementation from 0 to 9 g/d decreased urinary N excretion ( $P < 0.01$ ) from 32.3 to 24.3 g/d with no further reduction when more than 9 g/d of lysine was supplied. Changes in total urinary N excretion were due to changes in urinary urea-N, which decreased ( $P < 0.01$ ) from 21.6 to 14.8 g/d as supplemental lysine increased from 0 to 9 g/d. Urinary ammonia-N was not affected by treatment ( $P = 0.48$ ). Increasing lysine supply from 0 to 9 g/d increased ( $P < 0.01$ ) N retention from 20.7 to 29.9 g/d with no further increase observed beyond 9 g/d of lysine. Break-point analysis estimated maximal N retention at 9.0 g/d supplemental lysine. Over the linear response surface of 0 to 9 g/d lysine, assuming that retained protein is 6.25 × retained N and contains 6.4% lysine, the efficiency of lysine utilization for protein deposition was 40%.

**Key Words:** cattle, lysine, efficiency



### 336 Effects of preweaning nutrient supply on growth and pre and post-weaning glucose tolerance test in male Holstein calves.

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Perinatal nutrient supply can affect metabolism of mammals with effects that persist later in life. We hypothesized that pre-weaning calf milk replacer (CMR) supply can alter energy homeostasis in later growth phases. One hundred twenty male Holstein calves ( $20 \pm 3.2$  d old and  $48.4 \pm 2.2$  kg BW) were assigned to a low (LP) or high (HP) plane of nutrition, in a randomized block design with 12 blocks of 2 pens. LP received 25 kg and HP 50 kg of the same CMR (18% fat, 23% protein) for 7 weeks before weaning. Throughout the 12 weeks of experiment, calves were offered ad libitum calf starter and straw. Body weight (BW) was recorded weekly. A glucose tolerance test (GTT) was conducted before (d 42, PW) and after (d 84, AW) weaning in 24 calves (1 calf randomly selected per pen). After a 12h fasting period, glucose was infused at 180 mg/kg of BW through a venous catheter. Blood was collected at -15, -5, 0, 5, 10, 15, 20, 25, 30, 35, 45, 60, and 90 min relative to glucose infusion. Data were analyzed using PROC MIXED in SAS. BW and average daily gain (ADG) were affected ( $P < 0.01$ ) by treatment, week and treatment  $\times$  week interactions. HP calves were heavier from wk 2 leading to a final BW of  $131.3 \pm 1.88$  and  $140.0 \pm 1.88$  for LP and HP at wk 12, respectively. ADG was greater for HP on wk 1 ( $P < 0.01$ ), 2 ( $P < 0.01$ ), 3 ( $P < 0.01$ ), 10 ( $P = 0.05$ ) and 12 ( $P = 0.04$ ) although on wk 7, LP presented a higher ( $P = 0.01$ ) ADG. Consequently, overall ADG was lower ( $P < 0.01$ ) for LP ( $0.969 \pm 0.029$  vs.  $1.089 \pm 0.029$  kg/d). Glucose responses PW or AW did not differ ( $P = 0.78$  and  $P = 0.32$ , respectively) between the treatments. Serum insulin presented no treatment  $\times$  time interactions ( $P = 0.24$  and  $P = 0.06$ , for PW and AW, respectively). In the PW test, insulin did not differ ( $P = 0.89$ ) between treatments. In the AW test, however, plasma insulin was greater ( $P < 0.01$ ) in HP calves 5, 10, 15, 20, 30 35 and 45 min following glucose infusion. Nutrient supply to calves in early life affects post weaning insulin response, supporting the importance of perinatal nutritional stimuli for later life metabolism.

**Key Words:** calf milk replacer, insulin, metabolic programming

### 337 Effects of hemp seed oil on serum antioxidant indicators in dairy water buffalo.

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The aim of the present study was to investigate the effects of hemp seed oil supplementation on serum antioxidant indicators in dairy water buffalo. Eighteen dairy water buffaloes were randomly assigned to 3 treatments: (1) 0 g/d hemp seed oil supplement, (2) 100 g/d hemp seed oil supplement, and (3) 200 g/d hemp seed oil supplement. The whole feeding experiment duration was 6 weeks with 2 weeks for adaptation and measurements were made at the end of whole feeding experiment. The results showed that, compared with group 1, hemp seed oil supplementation tended to increase the activity of total antioxidative capacity (T-AOC), glutathione peroxidase (GSH-PX) and catalase (CAT), and group 3 reached significance level ( $P < 0.05$ ). Group 2 resulted in significantly decreased activity of peroxidase (MDA) compared with group 1, while group 3 had no significant effect. On the other hand, no significant differences were found between group 1 and the other treatments in

the activity of peroxidase (POD) and superoxide dismutase (SOD). In conclusion, these results of this study indicated that supplement hemp seed oil would increase the antioxidant capacity of serum and enhance the immune function in organism of dairy water buffalo.

**Key Words:** dairy water buffalo, hemp seed oil, antioxidant indicator

### 338 Effects of maternal metabolizable protein supply during late gestation on maternal and fetal intestinal development in sheep.

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We hypothesized that metabolizable protein (MP) supply during late gestation would affect maternal and fetal intestinal development in sheep. In Exp. 1, multiparous ewes ( $n = 45$ ) were allotted randomly to 1 of 3 treatments, 60% (MP60), 80% (MP80), or 100% (MP100) of MP requirements fed from d 100 to d 130 of gestation. In Exp. 2, multiparous singleton pregnant ewes ( $n = 18$ ) were randomized to receive 1 of 3 diets that were similar in energy and formulated to supply 60% (MP60), 100% (MP100), or 140% (MP140) of MP requirements during late gestation (d 100 to 130). Pregnant ewes and fetuses were euthanized and necropsied on d  $130 \pm 1$  of gestation, and maternal and fetal intestinal tissues were collected. Data were analyzed with GLM and means separated using PDIF of SAS. In Exp. 1, as previously reported, maternal BW of MP80 was greater ( $P = 0.05$ ) than MP60 and MP100 which were not different, whereas fetal BW was not altered by treatment. Neither maternal nor fetal small intestinal mass or % proliferating jejunal crypt cells (labeling index) were affected by dietary MP supply during late gestation. Fetal large intestinal mass in MP80-fed ewes tended ( $P = 0.08$ ) to be greater than MP60 and MP100 fetuses when examined as a proportion of eviscerated BW. Likewise, in Exp. 2, crypt cell proliferation in fetal jejunal samples was unaltered by maternal MP supply. Thus, maternal MP supply did not affect maternal and fetal intestinal development during late gestation; however, fetal large intestinal weight appeared to be increased when dams were supplied MP80 compared with MP100. Additional research is needed to confirm and extend these results. We hypothesized that metabolizable protein (MP) supply during late gestation would affect maternal and fetal intestinal development in sheep. In Exp. 1, multiparous ewes ( $n = 45$ ) were allotted randomly to 1 of 3 treatments, 60% (MP60), 80% (MP80), or 100% (MP100) of MP requirements fed from d 100 to d 130 of gestation. In Exp. 2, multiparous singleton pregnant ewes ( $n = 18$ ) were randomized to receive 1 of 3 diets that were isocaloric and formulated to supply 60% (MP60), 100% (MP100), or 140% (MP140) of MP requirements during late gestation (d 100 to 130). Pregnant ewes and fetuses were euthanized and necropsied on d  $130 \pm 1$  of gestation, and maternal and fetal intestinal tissues were collected. In Exp. 1, as previously reported, maternal BW of MP80 was greater ( $P = 0.05$ ) than MP60 and MP100 which were not different, whereas fetal BW was not altered by treatment. Neither maternal nor fetal small intestinal mass nor % proliferating jejunal crypt cells (labeling index) were affected by dietary MP supply during late gestation. Fetal large intestinal mass in MP80-fed ewes tended ( $P = 0.08$ ) to be greater than MP60 and MP100 fetuses when examined as a proportion of eviscerated BW. Likewise, in Exp. 2, crypt cell proliferation in fetal jejunal samples was unaltered by maternal MP supply. Thus, maternal MP supply did not affect maternal and fetal intestinal development during late gestation; however, fetal large intestinal weight appeared to be increased when dams were supplied MP80 compared with MP100. Additional research is needed to confirm and extend these results.

**Key Words:** gestation, intestine, metabolizable protein

### 339 Effects of supplementation to steers consuming green chopped wheat pasture on energy losses and nitrogen balance.

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British cross steers (n = 10; initial BW: 206 ± 10.7 kg) were studied using respiration calorimetry chambers to evaluate energy losses, N balance, and nutrient digestibility of greenchop wheat forage as affected by energy supplementation. The study was designed as a 2 × 2 factorial arrangement of treatments. Eight steers were assigned to one of 2 BW blocks (4 steers per block) with factors consisting of either dietary treatment [no supplementation (CON) vs. supplemented with a corn-based energy supplement that contained 150 mg monensin sodium per steer/daily) at 0.5% of BW daily (SUP)] and NE<sub>m</sub> intake level [(1-times (1×) vs. 1.5-times (1.5×) maintenance]. Wheat forage was harvested daily and fed as green chop to steers continuously during the 56-d study. No difference (P ≥ 0.29) was observed between CON and SUP for apparent digestibility of OM (79.6% vs. 79.6%), NDF (68.7% vs. 64.7%), or N (82.2% vs. 80.4%). No difference was observed for DE (2.90 Mcal/kg), heat production (8.54 vs. 8.29 Mcal), or retained energy (4.98 vs. 4.06 Mcal) between dietary treatments when fed at 1.5× maintenance. CON steers had greater (P = 0.01) O<sub>2</sub> consumption (1,790 vs. 1,711 L/d), tended to have (P = 0.06) greater CH<sub>4</sub> production (78 vs. 65 L/d), and had similar CO<sub>2</sub> production (1,704 vs. 1,627 L/d; P = 0.62) compared with SUP. Methane, as a proportion of GE intake, was 6.50% and 6.00%, for CON and SUP, respectively, but was not significantly different (P = 0.60) between treatments. No difference (P = 0.69) was noted in retained energy as a proportion of GE intake (24.8 vs. 23.0%) between treatments. We determined that the CON treatment (wheat forage alone) contained 1.61 Mcal/kg NE<sub>m</sub> and 0.98 Mcal/kg NE<sub>g</sub> and the SUP treatment contained 1.78 Mcal/kg NE<sub>m</sub> and 0.95 Mcal/kg NE<sub>g</sub>. By difference, supplementation improved the calculated NE<sub>m</sub> of wheat forage by 10.6% but decreased NE<sub>g</sub> of wheat forage by 3.1%. Under the conditions of this study, supplementation of wheat forage with a corn-based supplement did not affect losses of energy and resulted in similar NE values.

**Key Words:** methane, net energy, wheat forage

### 340 Supplement and undegradable protein influence on beef calf performance grazing stockpiled tall fescue.

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**ABSTRACT:** Ninety crossbred beef calves (277.7 ± 7.1 kg) were utilized in a completely randomized design to determine supplement and rumen undegraded protein (RUP) influence on calf performance while grazing stockpiled tall fescue. Calves were supplemented at 1.15% of BW DM with dried distillers grains w/solubles (DDGS); 80.7% DDGS, 19.3% AminoPlus (DDG19); 80.7% soybean hulls (SBH), 19.3% AminoPlus (SBH19); 61.7% SBH, 38.3% AminoPlus (SBH38), or unsupplemented control (CON). We hypothesized supplement would increase ADG, and supplements with increased RUP would improve performance. Calves were stratified by BW, source, color, and sex, and then randomly assigned to a pasture. Pasture replicates (n = 15) were fertilized with nitrogen at 24.4 kg/ha on 25 August 2014 and at experiment initiation were blocked by percent endophyte infection. Weekly forage allocations were based on 3.25% BW DM hd<sup>-1</sup>d<sup>-1</sup>. Supplement and forage DM allocations were adjusted every 21 d using interim BW. Forage DM substitution was set at 0.5 times average supplemental DMI. Forage allocations were adjusted to maintain 509 kg DM/ha residual. Proc Glimmix procedure of

SAS was used for ANOVA. Supplement increased (P < 0.01) ADG and final BW compared with CON (0.68 vs. -0.03 kg/d; 336.0 vs. 276.7 kg). ADG did not differ (P > 0.05) between DDG19 and SBH38 (0.78 vs. 0.73 kg/d), however DDG19 increased (P < 0.05) ADG compared with SBH19 (0.62 kg/d) and DDGS (0.59 kg/d). ADG was not different (P > 0.05) between SBH38, SBH19, and DDGS. Forage DM disappearance was not different (P > 0.05) between CON (6.7 kg/d), DDG19 (6.5 kg/d) and SBH38 (6.2 kg/d). SBH19 (5.5 kg/d) forage DM disappearance did not differ (P > 0.05) from DDG19, SBH38 whereas DDGS (4.3 kg/d) decreased (P < 0.01) forage DM disappearance compared with CON, DDG19, and SBH38. DDG19 improved (P < 0.05) supplemental G:F compared with DDGS, (0.23 vs. 0.18 kg/kg). SBH38 (0.22 kg/kg) and SBH19 (0.19 kg/kg) supplemental G:F did not differ (P > 0.05) compared with DDG19 or DDGS. DMI did not differ (P > 0.05) between DDG19 (10.0 kg/d), SBH38 (9.7 kg/d), and SBH19 (8.9 kg/d), and was least for DDGS (7.8 kg/d). Supplementation increased ADG, while additional RUP improved ADG and supplemental G:F.

**Key Words:** supplementation, grazing, DDGS

### 341 Performance and economics of supplementing calves with distillers grains or fertilization of smooth brome grass pastures.

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Ten years of performance data were summarized to evaluate distillers grains supplementation and smooth brome grass fertilization on cattle performance and economics. Each year, 45 steer calves (321 kg, SD = 21 kg) were assigned to 1 of 3 treatments with 3 replications per treatment. Treatments included non-fertilized control pasture (CON), fertilized pasture (89.75 kg N/ha; FERT), or non-fertilized and supplemented with distillers grains plus solubles (DGS) at 0.6% of BW daily for an average of 152 d (SUPP). Cattle on FERT and SUPP were stocked at 9.88 AUM (308 kg of forage)/ha while CON cattle were stocked at 6.82 AUM/ha. Pastures had 5 test animals and put and take animals were used to maintain similar grazing pressure among treatments. Cattle on CON and FERT treatments gained the same through the grazing period (0.77 kg/d; P = 0.67); however, fertilized pastures had greater gain per hectare. Supplemented cattle gained 0.31 kg/d more than FERT or CON (P < 0.01), resulting in 45.4 kg more BW at the end of the grazing season. Utilizing performance data from this 10 year period, 3 corn prices (\$3, \$4, and \$5/25.4 kg) and 3 DGS prices (95, 105, and 115% price of corn) were used in a partial budget to compare profit potential of these treatments. To account for the greater BW of SUPP cattle at the end of grazing, a \$10/45.4 kg price slide was used to calculate SUPP cattle revenue. At all prices, CON cattle were set to breakeven for profit comparisons. Across all scenarios, fertilizer costs were offset by savings in land rent, resulting in a \$16.71/animal profit for FERT relative to CON (P < 0.01). As price of corn increased, profit of SUPP relative to CON decreased; however, SUPP cattle had greater profit than CON and FERT (P < 0.01) at all prices evaluated. For every 10% increase in DGS price relative to corn, SUPP profit decreased \$5.32, \$7.09, and \$8.86 per animal for \$3, \$4, and \$5/25.4 kg corn, respectively. Using an average DGS price of 105% the value of corn, SUPP cattle were \$80.76, \$62.14, and \$43.52/animal more profitable than CON (P < 0.01) at corn prices of \$3, \$4, and \$5/25.4 kg, respectively. In these scenarios, supplementing cattle grazing smooth brome grass pasture with DGS increased ADG and profitability.

**Key Words:** beef cattle, grazing, supplementation

# Ruminant Nutrition Symposium: Time required for diet adaptation and minimization of carry-over effect in ruminants: Evidence-based decisions

**342 Time required for adaptation of rumen fermentation and the rumen microbiome.** Timothy Hackmann\*, *University of Florida, Gainesville, FL.*

Dietary shifts disturb microbiota and fermentation of the rumen, and varying amounts of time are required for them to re-stabilize. Shifts from high-forage to high-grain diets have been studied for over 60 years. During these shifts, abundance of total and cultured bacteria generally increases and then stabilizes. For some shifts in some studies, stabilization was complete within 24 h, but in others, it was incomplete even after 3 wk. Specific groups of bacteria (cellulolytics, amylolytics, lactate-utilizers) and protozoa also require varying amounts of time to stabilize. The time required for methanogens and fungi to stabilize has seldom been studied. Stabilization of fermentation (pH, short-chain fatty acid concentration) does not necessarily coincide with stabilization of the microbiota. For diets with very high (>70%) grain, abundance of protozoa and cultured bacteria vacillates greatly and stabilizes slowly, if at all. More recent studies have employed culture-independent methods, such as qPCR and 16S rDNA sequencing, to investigate taxonomic composition of bacteria and methanogens during dietary shifts. These studies confirm that taxonomic composition does change during dietary shifts, but more frequent sampling is required to resolve at what exact point it stabilizes. Other culture-independent methods, such as whole-genome or transcriptome sequencing, need to be deployed to resolve changes in microbial function, not just taxonomic composition. Changes in microbial communities, whether assessed by culture-independent or dependent methods, need to be better related to variables of interest in feeding trials (e.g., diet intake and digestibility).

**Key Words:** dietary adaptation, microbiota, rumen

**343 Time required for adaptation of behavior, feed intake, and dietary digestibility in cattle.** Richard J. Grant\*, Heather M. Dann, and Melissa E. Woolpert, *William H. Miner Agricultural Research Institute, Chazy, NY.*

Experimental designs such as Latin square, crossover, and switchback have been used extensively in nutritional studies to assess behavioral, intake, and digestibility responses to diet. A critical consideration when using these designs is the necessary length of an adaptation period to accurately measure treatment effect and to minimize potential confounding influence of carry-over from previous treatment. The objective of this review was to determine typical time required for adaptation of these responses based on research evidence primarily with dairy cattle. We focused on (1) eating, ruminating, and resting behavior, (2) dry matter intake (DMI), and (3) total-tract nutrient digestibility. Animal response to dietary treatment integrates response to physicochemical properties of the diet being studied as well as the feeding environment, and so the environment must be similar among treatments. For example, competition for feed can profoundly affect eating behavior and DMI. Response to diet for eating, ruminating, and resting behaviors usually stabilizes within 1–7 d. Consequently, DMI response to diet reflects the pattern of change in eating behavior. This time course is affected by physical and chemical attributes of the diet such as particle size, carbohydrate fermentability, and fat or protein content and characteristics. Most research indicates that ruminant total-tract digestibility ordinarily adjusts to diet within 10 to 14 d. Meta-analysis reveals that change-over designs are as accurate

as continuous designs in assessing DMI. However, diets of extremely high or low digestibility may cause lingering effects on digestive and metabolic processes related to ruminal function or dynamics of body fat and protein that may confound interpretation of response to diet in subsequent experimental periods. Examples of expected time to adaptation and potential carryover in lactating dairy cattle will be illustrated using Latin square studies that evaluated diets with considerable range in forage amount and carbohydrate fermentability. In summary, adaptation periods of approximately 7–14 d are usually adequate for measuring response in eating behavior and DMI, but potential consequences of carryover effects in change-over designs are less clear.

**Key Words:** adaptation, carryover

**344 Time required for adaptation of protein metabolism.** G. I. Zanton\*, *USDA, Agricultural Research Service, US Dairy Forage Research Center, Madison, WI.*

Animals that can appropriately adjust to varying environmental and nutritional conditions possess a survival advantage. Maintaining homeostasis and homeorhesis in response to changing nutritional conditions requires flexibility in nutrient partitioning and use efficiencies. This is especially the case for protein metabolism because there is no dedicated pool of amino acids held in reserve for times of changing dietary protein availability. Research on the adaptation in protein metabolism to changing dietary conditions could have potential implications on experimental design and on basic and applied ruminant nutrition. However, experiments directly evaluating the metabolic characteristics and appropriate timing of dietary adaptation are very limited. This is especially the case when compared with the number of experiments that have evaluated a response to a nutritional change after the period of adaptation is presumed to be complete, although rarely verified. Factors affecting the adaptive responses in protein metabolism in the ruminant are multiple and likely interacting, although the time required for adaptation appears to depend most on factors such as the duration and level of the previous nutritional condition, the difference from the current nutritional condition, the timing of these dietary changes with respect to the physiological state of the animal, the priority of the metabolic demand, and the productive state of the animal (i.e., the respective combinations of maintenance, growth, and/or lactation). As a consequence of these varied factors, from an experimental perspective, the time required for adaptation may depend on the response variables of interest, productive state of the animal, and the treatments under investigation. Existing literature will be reviewed to highlight time-related adaptive responses to diet changes on protein digestion, post-absorptive metabolism, and production and to emphasize areas where this understanding is incomplete.

**Key Words:** protein nutrition, adaptation, ruminant

**345 Time required for diet adaptation and minimization of carry-over effect in ruminants: The perspective of an experimental researcher.** Kevin J. Harvatine\*, *Penn State University, University Park, PA.*

The ideal experimental design depends on the biological mechanisms and physiology under investigation. Classical metabolic data provides



insight into clearance of specific metabolic pools, but recent discoveries in epigenetic provide the opportunity for long-term responses to diet. Arguably, the most complete experimental design would determine the effect of a dietary treatment over entire lactations, multiple lactations, and possibly over multiple generations. However, these approaches are not practical and arguably other interactions are of greater importance. Experimentally the time course of response to a treatment is very important to provide mechanistic insight into the primary mechanism and secondary adaptations. Both primary mechanisms and secondary adaptations are biologically relevant and application of time-series analysis in experiments may provide key insight in understanding these mechanisms. In our own work with milk fat depression we have determined the time course of rumen response to diet changes and mammary response to bioactive fatty acids. This information has been invaluable to efficient experimental design and increasing confidence in mechanisms. Lastly, a common goal is to determine the expected response to a treatment. Temporal adaptation is one factor to consider, but many other interactions such as composition of the basal diet (e.g., high vs. low fiber) and physiological state of the cow (including level of intake and production) are also extremely important factors. Arguably, testing treatments over several conditions with adequate, but not excessive, treatment durations would optimize the accuracy of the predicted response when a treatment is applied in practice.

**Key Words:** time-course, milk fat depression

**346 The perspectives of a beef cattle nutritionist.** Shawn L. Archibeque\*<sup>1</sup> and Gerald B. Huntington<sup>2</sup>, <sup>1</sup>*Department of Animal Sciences, Colorado State University, Fort Collins, CO*, <sup>2</sup>*Department of Animal Science, North Carolina State University, Raleigh, NC*.

There are numerous changes in beef cattle in both their symbiotic and physiological aspects as they are adjusted to various diets associated with varying production states and research needs. Within the beef industry, there is a need to address changes within diet types as well as across diet types (i.e., high roughage vs. high concentrate). These changes will influence our understanding of how nutrients are used during varying production states. These variations in treatments that deal with source more than daily supply will allow for minimal variation in measurements within a shorter period of time than those studies with large changes in particularly nutrients or overall plane of nutrition. In particular, these adaptations are greatly varied in the literature. This is of particular interest in studies that use a study design that minimizes animal-to-animal variation through the repeated use of animals in the study, such as a Latin square or crossover design. To highlight the significance of this issue, over 35% of the articles in the ruminant nutrition section of the *Journal of Animal Science* used these research techniques and had a range in adaptation periods from 9 to 27 d, with an average of 14.6 d. There is a need for greater understanding and homogeneity in these adaptation periods for future understanding of data, particularly those that will be happening over a fixed period of time.

**Key Words:** homeorhetic adaptation, ruminal adaptation, variance

**347 Algae meal for ruminants: I. Nutrient digestibility in finishing lambs.** Rebecca S. Stokes\*, Megan L. Van Emon, Daniel D. Loy, and Stephanie L. Hansen, *Department of Animal Science, Iowa State University, Ames, IA*.

Heterotrophic microalgae combined with soyhulls forms an algae meal (ALG) which contains partially deoiled microalgae (DMA; 57% DM

basis) and soyhulls (43%). Eight whiteface wethers ( $23.02 \pm 0.54$  kg) were used in a  $4 \times 4$  Latin square design to determine the effect of the DMA portion of ALG on total-tract nutrient digestibility. Lambs received 1 of 4 isonitrogenous dietary treatments (2 sheep·diet<sup>-1</sup>·period<sup>-1</sup>) where ALG was added at the expense of soyhulls: a soyhulls-based control (CON; 53% soyhulls), 10% DMA from ALG (DMA10), 20% DMA from ALG (DMA20), and 30% DMA from ALG (DMA30). There were 4 periods, with 10 d of adaptation and 5 d of total fecal and urine collection. Prior to each collection period was a 14 d washout period where all lambs were fed a common diet. Data were analyzed using Proc Mixed of SAS, and pooled LSMEANS and SEM are reported. Intake of OM and fecal OM output were similar ( $P \geq 0.11$ ) between CON and ALG-fed lambs. Urine output linearly increased ( $P = 0.02$ ) as DMA increased in diets ( $0.57, 0.72, 0.77$ , and  $0.87 \pm 0.08$  L/d for CON, DMA10, DMA20, and DMA30, respectively). Digestibility of OM (73.6, 72.9, 71.0, and  $69.1 \pm 1.01\%$  for CON, DMA10, DMA20, and DMA30, respectively) linearly decreased ( $P < 0.01$ ) as DMA increased in diets. Also, NDF ( $65.5, 61.3, 53.6$ , and  $39.0 \pm 2.36\%$  for CON, DMA10, DMA20, and DMA30, respectively) and ADF ( $65.0, 60.7, 50.9$ , and  $30.2 \pm 1.94\%$  for CON, DMA10, DMA20, and DMA30, respectively) digestibility linearly decreased ( $P < 0.01$ ) as DMA increased in diets. Ether extract digestibility did not differ ( $P = 0.24$ ) between CON and DMA-fed lambs. Nitrogen digestibility linearly decreased ( $P = 0.05$ ) as DMA increased in the diet ( $60.2, 58.9, 58.0$ , and  $57.2 \pm 1.10\%$  for CON, DMA10, DMA20, and DMA30, respectively). Also, N balance linearly decreased ( $P < 0.01$ ) as DMA increased in the diet ( $10.1, 8.4, 8.9$ , and  $6.8 \pm 0.64$  g/d for CON, DMA10, DMA20, and DMA30, respectively). These results suggest that the DMA portion of ALG may be less digestible than soyhulls, and even though the ALG had minimal effects on OM digestibility the differences in N balance may suggest an effect on growth in sheep, under the conditions of this study.

**Key Words:** algae, digestibility, sheep

**348 Algae meal for ruminants: II. Growth and carcass characteristics of finishing steers.** Rebecca S. Stokes\*, Daniel D. Loy, and Stephanie L. Hansen, *Department of Animal Science, Iowa State University, Ames, IA*.

De-oiled microalgae from large-scale production of heterotrophic microalgae can be combined with soyhulls to form a novel feedstuff called algae meal (ALG). To determine the effects of replacing corn in a finishing diet with ALG on growth and carcass characteristics, crossbred steers (168) were blocked by BW ( $432.2 \pm 30.8$  kg) into pens of 6 steers (7 pens per treatment) and assigned to 1 of 4 diets: a corn-based control (CON), 14% ALG (ALG14), 28% ALG (ALG28), and 42% ALG (ALG42). Corn was replaced by ALG on a DM basis. Steer BW were taken on d 0, 1, 28, 56, 74, 101, and 102, and steers were harvested on d 103. Pen was the experimental unit and DMI, ADG, and G:F data were analyzed as repeated measures. Two steers per pen were selected for sampling of blood and liver (d -1 and 96), and collection of rib facings at harvest. Overall DMI linearly increased ( $P < 0.01$ ) as ALG increased in the diet ( $12.7, 13.4, 13.8$ , and  $14.4 \pm 0.15$  kg/d for CON, ALG14, ALG28, and ALG42, respectively). There was a treatment by time effect for ADG ( $P < 0.01$ ), with ADG linearly decreasing ( $P \leq 0.03$ ) in the first and third month, not differing ( $P = 0.95$ ) in the second month, and linearly increasing ( $P < 0.01$ ) in the fourth month as ALG increased in the diet. Final BW did not differ ( $P = 0.74$ ) between CON and ALG-fed cattle. There was a treatment by time effect for G:F ( $P < 0.01$ ), with G:F linearly decreasing ( $P < 0.01$ ) in the first 3 mo as ALG increased in the diet, while G:F linearly increased ( $P < 0.01$ ) in

the fourth month. Yield grade linearly decreased ( $P = 0.02$ ), and there was a tendency for dressing percent and 12th rib backfat to linearly decrease ( $P \leq 0.10$ ) as ALG increased in the diet. Plasma Cu, Fe, and Mg concentrations were not different ( $P \geq 0.31$ ) in CON vs. ALG cattle; however, plasma Zn concentrations linearly increased ( $P = 0.03$ ) as ALG increased in the diet. Total lipid, SFA, MUFA, and PUFA concentrations in the longissimus thoracis did not differ ( $P \geq 0.13$ ) between CON and

ALG-fed cattle. It appears ALG has less energy than corn; however, minimal effect on carcass performance suggests ALG may serve as a potential replacement for corn in feedlot diets.

**Key Words:** algae, performance, cattle

# Animal Health: Lactating cows

**T1 Health and productive responses of dairy cows treated with reduced doses of recombinant bovine somatotropin during the periparturient period.** Paula R. B. Silva<sup>\*1</sup>, Henrique F. Soares<sup>1</sup>, Gabriel D. Bombardelli<sup>1</sup>, Wagner D. Braz<sup>1</sup>, Daniela N. Liboreiro<sup>1</sup>, and Ricardo C. Chebel<sup>1,2</sup>, <sup>1</sup>University of Minnesota, St Paul, MN, <sup>2</sup>University of Florida, Gainesville, FL.

The objectives were to evaluate the effects of treatment of dairy cows with recombinant bovine somatotropin (rbST) during the periparturient period on health and production. Holstein (HO, n = 302) and Jersey (JS, n = 522) cows were enrolled in the experiment 21 d before calving. Cows were assigned randomly to the control and rbST (125 mg of rbST every 7 d from -21 to 21 d relative to calving) treatments. At -21, 0, 30, and 60 d relative to calving, BCS was recorded. Occurrence of retained placenta (RP) was recorded and cows were examined for diagnosis of metritis 4, 7, 10, and 14 d postpartum (DIM). Acute metritis (metritis and fever and/or anorexia) was recorded in the JS herd. Blood was sampled 7 and 14 d postpartum to determine ketosis incidence. Cows were milked thrice daily. In the HO herd, milk yield and composition were determined weekly until 21 d postpartum; thereafter, monthly milk yield was recorded. In the JS herd, milk yield and composition were recorded monthly. Dichotomous data were analyzed by logistic regression and continuous data were analyzed by ANOVA. Incidence of RP tended to be affected by treatment (HO-control = 14.4%, rbST = 6.1%; JS-control = 1.5%, rbST = 1.2%;  $P = 0.10$ ). Incidence of metritis was reduced by rbST treatment (HO-control = 26.2%, rbST = 16.8%; JS-control = 19.9%, rbST = 13.3%;  $P < 0.01$ ). Treatment tended to affect the incidence of ketosis (HO-control = 8.2%, rbST = 10.0%; JS-control = 8.4%, rbST = 13.1%;  $P = 0.09$ ). Postpartum BCS was ( $P < 0.05$ ) lower for rbST treated cows (HO-control = 3.08 ± 0.03, rbST = 2.99 ± 0.03; JS-control = 2.83 ± 0.03, rbST = 2.80 ± 0.03). Yield of energy corrected milk (ECM) in the first 3 weeks postpartum in the HO herd was greater for rbST cows (control = 33.0 ± 1.3, rbST = 36.3 ± 1.3 kg/d;  $P = 0.05$ ) but yield of ECM in the first month postpartum was not affected by treatment in the JS herd (control = 26.6 ± 0.6, rbST = 27.7 ± 0.6 kg/d;  $P = 0.15$ ). Milk yield in the first month postpartum was ( $P = 0.05$ ) increased by rbST treatment (HO-control = 35.0 ± 0.9, rbST = 37.9 ± 0.8 kg/d; JS-control = 26.9 ± 0.8, rbST = 28.1 ± 0.8 kg/d). Treatment of periparturient cows with small doses of rbST improved postpartum health and increased milk yield.

**Key Words:** transition cow, recombinant bovine somatotropin, health

**T2 Reduction in hepatic functionality can delay resumption of ovarian activity postpartum in dairy cows.** Paula Montagner<sup>\*1,2</sup>, Rubens A. Pereira<sup>1,2</sup>, Ana Rita T. Krause<sup>1,2</sup>, Marina M. Weschenfelder<sup>1,2</sup>, Elizabeth Schwegler<sup>1,2</sup>, Fernanda M. Gonçalves<sup>1,2</sup>, Carolina B. Jacometo<sup>1,2</sup>, Cássio C. Brauner<sup>1,2</sup>, and Marcio N. Corrêa<sup>1,2</sup>, <sup>1</sup>Federal University of Pelotas, Pelotas, RS, Brazil, <sup>2</sup>Center for Research, Teaching and Extension in Animal Science (NUPEEC), Pelotas, RS, Brazil.

After calving, the liver undergoes a metabolic overload due to high energetic and nutritional demand caused by milk synthesis and these challenges can induce inflammation and impair liver function. Several studies demonstrate that inflammation around calving is responsible for decreased productive efficiency and fertility. For this a composite index based on multiple variables associated with inflammation, the liver functionality index (LFI), can be promising for aid in the diagno-

sis of diseases and reproductive problems on dairy farms. The LFI is determined utilizing 3 biomarkers of hepatic function: albumin, total cholesterol and bilirubin, measured on 3 and 28 d after calving. The aim of this study was to evaluate the resumption of postpartum ovarian activity of dairy cows with low LFI. Twenty cows Holstein were evaluated from d 21 prepartum until 30 d postpartum. The cows were divided into 2 groups: Low LFI (LLFI:  $\leq -7$  to  $-12$ ; n: 10) and High LFI (HLFI:  $> -7$  to  $-4$ ; n: 10). Serum concentration of progesterone (P4) was analyzed on d 16, 23, 30, 37 and 44 postpartum to predict the resumption of ovarian activity. The cows which had P4 concentration higher than 1ng/mL in 2 consecutive assays were considered ovulatory, while cows with P4 concentration below 1ng/ml during same period were considered anovulatory. The statistical analyses were performed using SAS 9.0 software. The statistical model CATMOD (Categorical Data Analysis Procedures) was used for analysis of categorical data of ovulation.  $P < 0.05$  was considered significant. The proportion cows that resumed ovarian activity to 44 d postpartum was lower (29%; 3/10;  $P < 0.05$ ) in the LLFI, compared with HLFI group (86%, 9/10). In conclusion, dairy cows with reduced hepatic functionality after calving have delayed resumption of ovarian activity, which demonstrates that the LFI can be a useful index to predict problems in resumption of ovulation in dairy cows.

**Key Words:** liver functionality index, ovarian activity, progesterone

**T3 Organic trace minerals during the transition period. 4. Corium gene expression profiling reveals a beneficial effect of supplementing Zn, Mn, and Cu from Availa Mins and Co from CoPro on hoof health of peripartal dairy cows.** J. S. Osorio<sup>\*1</sup>, E. F. Garrett<sup>1</sup>, M. M. Elhanafy<sup>1</sup>, E. Trevisi<sup>2</sup>, J. K. Drackley<sup>1</sup>, M. T. Socha<sup>3</sup>, and J. J. Looor<sup>1</sup>, <sup>1</sup>University of Illinois at Urbana-Champaign, Urbana, IL, <sup>2</sup>Università Cattolica del Sacro Cuore, Piacenza, Italy, <sup>3</sup>Zinpro Corporation, Eden Prairie, MN.

Lameness remains a major health problem and cause of death, in dairy cows. The negative effects of the transition period may make cows prone to become lame. Positive effects of supplementation of organic trace minerals (AAC) on hoof health have been reported. A hoof biopsy method to study the transcriptome of corium tissue has been published (Osorio et al., J. Dairy Sci. 95:6388–6396). Objective was to evaluate corium mRNA expression of genes related to claw conformation, oxidative stress, chemotaxis, inflammation, and transcription regulation in peripartal cows supplemented with AAC or inorganic (INO) trace minerals. Twenty Holstein dairy cows received a common prepartal (1.5 Mcal/kg DM, 15% CP) and postpartal (1.76 Mcal/kg DM, 18% CP) diet. Both diets were partially supplemented with an INO mix of Zn, Mn, and Cu to supply 35, 45, and 6 ppm, respectively, of the total diet DM. Cows were assigned to treatments in a randomized complete block design, receiving an oral bolus daily with a mix of INO (n = 10) or AAC (n = 9) containing Zn, Mn, Cu, and Co to achieve 75, 65, 11, and 1 ppm, respectively, in total diet DM. Treatments began on -30 d and continued until 30 d postpartum. Inorganic trace minerals were provided in sulfate form and AAC were supplied via Availa Zn, Availa Mn, Availa Cu, and CoPro (Zinpro Corp., Eden Prairie, MN). Hoof biopsies were harvested at 30 d postpartum from the sole of the rear right lateral claw in the right rear limb. Data on 31 genes were analyzed using the MIXED procedure of SAS. Expression of keratin 5 (*KRT5*) was lower ( $P < 0.01$ ) and biotinidase (*BTD*) was greater ( $P < 0.01$ ) in AAC cows, suggesting that AAC cows had a lower requirement of keratins for hoof



tissue repair, while having additional biotin for claw conformation. In contrast, a concomitantly greater ( $P < 0.06$ ) expression of toll-like receptor 2 (*TLR2*), tumor necrosis factor (*TNF*), and interleukin 1  $\beta$  (*IL1B*) in INO cows suggested a greater inflammatory status in hoof tissue. Overall, data suggest that periparturient AAC supplementation ameliorates the negative effect of transition period on hoof health.

**Key Words:** hoof biopsy, trace mineral, transition cow

**T4 Effects of postpartum uterine diseases on milk yield, milk components, and culling in dairy cows under certified organic management.** J. M. Piñeiro<sup>\*1</sup>, M. G. Maquivar<sup>2</sup>, A. A. Barragan<sup>1</sup>, J. S. Velez<sup>3</sup>, H. Bothe<sup>3</sup>, and G. M. Schuenemann<sup>1</sup>, <sup>1</sup>The Ohio State University, Columbus, OH, <sup>2</sup>Washington State University, Pullman, WA, <sup>3</sup>Aurora Organic Farms, Boulder, CO.

The objective was to assess the effect of postpartum uterine diseases on milk yield (kg), milk components (SCC and percent fat and protein), and culling. Cows ( $n = 3,227$ ) from 2 dairy herds were screened for retained placenta (RP;  $> 24$  h after parturition), metritis (within 20 d in milk [DIM]), and purulent vaginal discharge (PVD) at  $26 \pm 3$  DIM. Milk yield and components from the DHIA test-days and cows culled from farm records up to 300 DIM were collected. Weekly, a list of cows by DIM was obtained using on-farm computer records and screened for RP (presence of fetal membranes outside the vulva), metritis (fetid brown-red watery vaginal discharge and fever), and PVD (gloved hand technique). PVD was defined as any cow presenting a score of 2 or 3 (0–3 scale; mucopurulent or worse vaginal discharge) at the time of exam. Parity (lactations 1, 2 and  $\geq 3$ ) of cows was considered for milk yield, milk components, and culling. The statistical analyses were performed using MIXED (milk yield and components) and GLIMMIX (culling) procedures of SAS. Cows with metritis, RP or PVD had an additive effect on milk yield, milk components, and culling. Regardless of parity, lactating cows diagnosed with uterine diseases (all combined) had significantly reduced milk yield (by 2–3.9 kg/cow/d) for at least one of the first 4 DHIA test-days ( $P < 0.05$ ), but was not different at later tests. For the first 2 DHIA test-days, lactating cows diagnosed with uterine disease (all combined) had significantly higher SCC ( $232 \times 10^3$  cells/mL) and fat content (3.7%) compared with cows without uterine diseases ( $164 \times 10^3$  cells/mL and 3.5%, respectively;  $P < 0.05$ ). Milk protein content (%) was not different between cows with or without uterine diseases. Cows with uterine diseases had higher ( $P < 0.05$ ) culling within 60 DIM and significantly lower ( $P < 0.05$ ) pregnancy hazard up to 300 DIM compared with cows without uterine diseases, regardless of parity. Uterine diseases decreased milk yield and changed milk components (SCC and fat) early in lactation; and these diseases were a substantial risk factor within 60 DIM for culling.

**Key Words:** organic, dairy cattle, uterine disease

**T5 Cows diagnosed with metritis showed blood alterations related to innate immunity and carbohydrate and lipid metabolism during early dry off period.** Guanshi Zhang, Dagnachew W. Hailemariam, Elda Dervishi, Qilan Deng, Tran H. Lam, Seyed A. Goldansaz, Suzanna M. Dunn, and Burim N. Ametaj<sup>\*</sup>, *Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB, Canada.*

The objective of this study was to search for screening biomarkers of metritis in transition dairy cows. Blood samples were collected from the coccygeal vein once per week before morning feeding from 100 multiparous Holstein dairy cows during –8, –4, disease diagnosis, and

+4 and +8 wk relative to parturition. Six healthy cows (CON) and 6 cows that showed clinical signs of disease were selected for intensive serum analyses. Concentrations of interleukin-1 (IL-1), interleukin-6 (IL-6), and tumor necrosis factor (TNF); haptoglobin (Hp), serum amyloid A (SAA), and lipopolysaccharide binding protein (LBP); and nonesterified fatty acids (NEFA) and  $\beta$ -hydroxybutyrate (BHBA) were measured in serum by ELISA or colorimetric methods. Health status, feed intake, and milk yield were monitored for each cow during the whole experimental period. Data were processed statistically by MIXED procedure of SAS 9.2. Results showed that cows affected by metritis had greater concentrations of lactate ( $2,901$  vs  $2,251 \pm 183$   $\mu\text{mol/L}$ ,  $P = 0.04$ ), IL-6 ( $111$  vs  $27 \pm 13$   $\text{pg/mL}$ ,  $P = 0.01$ ), TNF ( $0.72$  vs  $0.19 \pm 0.11$   $\text{ng/mL}$ ,  $P = 0.01$ ), and SAA ( $13,441$  vs  $8,517 \pm 1,517$   $\mu\text{g/mL}$ ,  $P = 0.03$ ) in the serum vs CON. Most interestingly, elevated concentrations of all 4 variables were observed at –8 (lactate:  $3,902$  vs  $2,455$   $\mu\text{mol/L}$ ,  $P = 0.03$ ; IL-6:  $87$  vs  $19$   $\text{pg/mL}$ ,  $P = 0.05$ ; TNF:  $0.77$  vs  $0.34$   $\text{ng/mL}$ ,  $P = 0.09$ ; SAA:  $17,960$  vs  $8,448$   $\mu\text{g/mL}$ ,  $P = 0.02$ ) and –4 (lactate:  $3,311$  vs  $2,162$   $\mu\text{mol/L}$ ,  $P = 0.03$ ; IL-6:  $166$  vs  $48$   $\text{pg/mL}$ ,  $P = 0.02$ ; TNF:  $1.02$  vs  $0.27$   $\text{ng/mL}$ ,  $P = 0.03$ ; SAA:  $6,003$  vs  $3,461$   $\mu\text{g/mL}$ ,  $P = 0.03$ ) wk before the occurrence of metritis compared with those of CON cows. The disease also lowered the overall milk production ( $38.18$  vs  $42.16 \pm 1.48$   $\text{kg/d}$ ,  $P = 0.01$ ) and feed intake ( $36.87$  vs  $39.81 \pm 1.30$   $\text{kg/d}$ ) as well as milk fat ( $3.56$  vs  $3.88$   $\text{g/kg}$ ,  $P = 0.10$ ) and fat:protein ratio ( $1.10$  vs  $1.38$ ,  $P = 0.05$ ) and was associated with greater SCC ( $44,700$  vs  $29,250 \pm 4,880$  cells/mL,  $P = 0.05$ ). In conclusion, metritis affected serum concentrations of several variables related to innate immunity and carbohydrate metabolism that might serve to monitor health status of transition dairy cows. More research is warranted to validate these data.

**Key Words:** dairy cow, metritis, innate immunity

**T6 Activation of innate immunity ahead of occurrence of ketosis.** Guanshi Zhang, Dagnachew W. Hailemariam, Elda Dervishi, Qilan Deng, Tran H. Lam, Seyed A. Goldansaz, Suzanna M. Dunn, and Burim N. Ametaj<sup>\*</sup>, *Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB, Canada.*

The objective of this study was to evaluate alterations in blood variables related to innate immunity, carbohydrate and lipid metabolism in transition dairy cows with ketosis. Multiparous Holstein dairy cows ( $n = 100$ ) were involved in the study. Five blood samples were collected from the coccygeal vein during the –8 to +8 wk around parturition, once per week before the morning feeding. Serum samples collected at –8, –4, time of diagnosis of disease, +4 and +8 wk relative to parturition from 6 healthy control cows (CON) and 6 cows with ketosis were selected for analyses. Samples were analyzed for  $\beta$ -hydroxybutyrate (BHBA), lactate, nonesterified fatty acids (NEFA), interleukin-1 (IL-1), interleukin-6 (IL-6), tumor necrosis factor (TNF), haptoglobin (Hp), and serum amyloid A (SAA). Health status, feed intake, and milk yield data were monitored for each cow during the whole experimental period. Data were processed statistically by MIXED procedure of SAS 9.2. Results revealed that cows with ketosis had greater concentrations of serum BHBA ( $1,014$  vs  $504 \pm 140$   $\mu\text{mol/L}$ ,  $P = 0.04$ ), lactate ( $4,236$  vs  $2,240 \pm 351$   $\mu\text{mol/L}$ ,  $P < 0.01$ ), IL-6 ( $255$  vs  $27 \pm 55$   $\text{pg/mL}$ ,  $P = 0.03$ ), TNF ( $0.47$  vs  $0.19 \pm 0.07$   $\text{ng/mL}$ ,  $P = 0.03$ ), and SAA ( $24,107$  vs  $8,550 \pm 3,457$   $\mu\text{g/mL}$ ,  $P = 0.01$ ) in comparison with the CON animals. Enhanced serum concentrations of BHBA ( $483$  vs  $312 \pm 19$   $\mu\text{mol/L}$  at –4 wk,  $P = 0.02$ ), lactate ( $5,795$  vs  $2,455 \pm 349$   $\mu\text{mol/L}$  at –8 wk,  $P = 0.01$ ; and  $4,478$  vs  $2,162 \pm 185$   $\mu\text{mol/L}$  at –4 wk,  $P = 0.01$ ), IL-6 ( $183$  vs  $19 \pm 6$   $\text{pg/mL}$  at –8 wk,  $P < 0.01$ ; and  $330$  vs  $48 \pm 18$   $\text{pg/mL}$  at –4 wk,  $P < 0.01$ ) and TNF ( $0.64$  vs  $0.27 \pm 0.05$   $\text{ng/mL}$  at –4 wk,  $P = 0.03$ ) at –4 or –8 wks before parturition were identified in cows with ketosis compared with the CON group.

Cows with ketosis also had overall lower feed intake ( $35.70$  vs  $39.81 \pm 1.74$  kg/d,  $P = 0.03$ ) and milk production ( $32.25$  vs  $42.16 \pm 2.53$  kg/d,  $P < 0.05$ ) vs CON animals. Overall results indicate that cows affected by ketosis displayed an activated innate immunity and altered carbohydrate and lipid metabolism several weeks before diagnosis of disease. More research is warranted to better understand the agent(s) that contribute(s) to ketosis in transition dairy cows and to validate utilization of these blood variables to predict disease state in cows.

**Key Words:** dairy cow, ketosis, blood variable

**T7 Effects of repeated oral administration of lipopolysaccharide and lipoteichoic acid either alone or in combination with subcutaneous exposure on metabolite responses in periparturient dairy cows.** Emily F. Eckel\*, Dagnachew W. Hailemariam, Grzegorz Zwierzchowski, Guanshi Zhang, Suzanna M. Dunn, and Burim N. Ametaj, *University of Alberta, Edmonton, AB, Canada.*

Recent investigations implicate bacterial lipopolysaccharide (LPS) and lipoteichoic acid (LTA) in the pathogenesis of multiple diseases currently affecting transition dairy cows. The objective of this investigation was to evaluate the effect of repeated oral administration of LPS and LTA alone or with subcutaneous injections on carbohydrate and lipid metabolism which are associated with the immune response to LPS and LTA. Two hundred dairy cows were randomly assigned to one of 4 treatments ( $n = 50$  per group) which they received on d -28, -25, -21, -18, and -14 before the expected calving date: 1) 2 mL oral sterile saline (CON), 2) Flat doses of oral LPS and LTA (TRT1), 3) Flat doses oral LPS and LTA plus subcutaneous (sc) injection on d -18 and -14 (TRT2), 4) Oral increasing doses of LPS at  $6.5\mu\text{g}$  (d -28 and -25),  $32.5\mu\text{g}$  (d -21 and -18) and  $65\mu\text{g}$  (d -14), all with flat doses of LTA (TRT3). Flat doses of LPS and LTA were 100 and  $120\mu\text{g}/\text{cow}$ , respectively, in 2 mL sterile saline. Blood samples from the coccygeal vein were collected during d -28, -14, 0, +7, +14, and +28 and analyzed for insulin, glucose, cholesterol, nonesterified fatty acid (NEFA), lactate, and  $\beta$ -hydroxybutyric acid (BHBA). Preliminary statistical analysis showed a significant effect of treatment on serum insulin ( $P = 0.02$ ). In general, TRT2 had lower serum insulin concentrations than TRT1 and TRT3 while TRT1 had higher concentrations than CTR. A tendency for treatment to affect serum cholesterol ( $P = 0.10$ ) was observed with TRT3 lowering cholesterol compared CTR and TRT1, but not TRT2. Results also showed a tendency for TRT2 to increase NEFA ( $P = 0.06$ ) compared with all other treatments. No effect of treatment was observed for serum glucose ( $P = 0.11$ ), lactate ( $P = 0.13$ ), or BHBA ( $P = 0.21$ ). Overall, preliminary results of this study suggest that repeated oral administration of LPS and LTA alone or in combination with sc injections were associated with minimal changes to lipid and carbohydrate metabolism.

**Key Words:** transition dairy cow, vaccine, serum metabolite

**T8 Effect of repeated oral administration of lipopolysaccharide and lipoteichoic acid either alone or in combination with subcutaneous exposure on humoral immunity in periparturient dairy cows.** Emily F. Eckel\*, Dagnachew W. Hailemariam, Grzegorz Zwierzchowski, Guanshi Zhang, Suzanna M. Dunn, and Burim N. Ametaj, *University of Alberta, Edmonton, AB, Canada.*

Transition dairy cows experience a high incidence of metabolic and infectious diseases immediately after calving. Recent research implicates bacterial lipopolysaccharide (LPS) and lipoteichoic acid (LTA) in their pathogenesis. Minimal knowledge exists of the periparturient dairy cow immune response to oral endotoxin exposure while other routes have

been extensively explored. The objective of this study was to evaluate the humoral immune response of periparturient dairy cows to repeated oral administration of LPS and LTA alone or in combination with subcutaneous injections. Two hundred dairy cows were randomly assigned to one of 4 treatments ( $n = 50$  per group; administration d -28, -25, -21, -18, and -14 before expected calving date): 1) 2 mL oral sterile saline (CON), 2) Flat doses of oral LPS and LTA (TRT1), 3) Flat doses of oral LPS and LTA plus subcutaneous (sc) injection d -18 and -14 (TRT2), 4) Oral increasing doses of LPS at  $6.5\mu\text{g}$  (d -28 and -25),  $32.5\mu\text{g}$  (d -21 and -18) and  $65\mu\text{g}$  (d -14), all with flat doses of LTA (TRT3). Flat doses of LPS and LTA were 100 and  $120\mu\text{g}/\text{cow}$ , respectively, in 2 mL sterile saline. Blood samples collected from the coccygeal vein at d -28, -7, +7, and +28 were analyzed for lipopolysaccharide binding protein (LBP), interleukin 1 (IL-1), transforming growth factor- $\beta$  (TGF- $\beta$ ), immunoglobulin M (IgM), and immunoglobulin G (IgG). Statistical results showed there was no effect of treatment on serum concentrations of LBP ( $P = 0.82$ ), IL-1 ( $P = 0.50$ ), TGF- $\beta$  ( $P = 0.75$ ), IgG ( $P = 0.25$ ), or IgM ( $P = 0.40$ ). Overall, preliminary results suggest that repeated oral administration of LPS and LTA or in combination with sc injections did not modulate humoral immunity in periparturient dairy cows.

**Key Words:** transition dairy cow, vaccine, humoral immunity

**T9 Blood alterations indicate subclinical mastitis diagnosed postpartum might start during early dry-off period.** Guanshi Zhang, Dagnachew W. Hailemariam, Elda Dervishi, Qilan Deng, Seyed A. Goldansaz, Suzanna M. Dunn, and Burim N. Ametaj\*, *Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB, Canada.*

The objective of this study was to identify alterations in innate immunity reactants, carbohydrate and lipid metabolites in the blood of transition dairy cows with subclinical mastitis (SM). Multiparous Holstein cows ( $n = 100$ ) were involved in the study and the experimental period lasted 16 wk starting at -8 wk before until +8 wk postpartum. Health status, feed intake, and milk yield and composition were monitored for each cow during the whole experimental period. Blood samples were collected from the coccygeal vein once/week before the morning feeding. Six healthy cows (CON) and 6 cows with SM were selected for intensive blood analyses. Serum concentrations of lactate, nonesterified fatty acids (NEFA),  $\beta$ -hydroxybutyrate (BHBA), interleukin-1 (IL-1), interleukin-6 (IL-6), tumor necrosis factor (TNF), haptoglobin (Hp), and serum amyloid A (SAA) were determined. Feed intake, milk production and composition also were determined. Data were processed using the MIXED procedure of SAS 9.2. Results indicated that concentrations of lactate ( $3,043$  vs  $2,243 \pm 149\mu\text{mol}/\text{L}$ ,  $P < 0.01$ ), TNF ( $0.52$  vs  $0.19 \pm 0.04$  ng/mL,  $P < 0.01$ ) and SAA ( $23,915$  vs  $8,514 \pm 2,518\mu\text{g}/\text{mL}$ ,  $P < 0.01$ ) were greater in cows with SM than CTR cows. Moreover, serum lactate ( $3,478$  vs  $2,455\mu\text{mol}/\text{L}$  at -8 wk,  $P = 0.03$ ;  $3,467$  vs  $2,162\mu\text{mol}/\text{L}$  at -4 wk,  $P = 0.04$ ) and TNF ( $1.29$  vs  $0.27$  ng/mL at -4 wk,  $P < 0.01$ ) in cows with SM were different from CTR cows starting at about -8 and -4 wks before diagnosis of disease. Overall feed intake ( $36.18$  vs  $39.81 \pm 1.55$  kg/d,  $P = 0.01$ ) and milk production ( $33.76$  vs  $42.16 \pm 2.52$  kg/d,  $P = 0.04$ ) was lower in SM-affected cows. Additionally, milk fat ( $3.32$  vs  $5.08$  g/kg,  $P = 0.02$ ) and fat:protein ratio ( $1.11$  vs  $1.69$ ,  $P < 0.01$ ) were lower, whereas SCC ( $1,867,000$  vs  $28,330$  cells/mL,  $P = 0.02$ ) and milk urea N ( $18.70$  vs  $15.39$  mg/dL,  $P = 0.03$ ) were greater in SM cows at diagnosis of disease vs CON. In conclusion cows affected by SM showed enhanced concentrations of indicators of innate immunity and metabolites related to carbohydrate metabolism weeks before diagnosis of SM. More research is warranted to validate these

data and better understand the reasons for activation of innate immune responses to SM in transition dairy cows.

**Key Words:** dairy cow, subclinical mastitis, innate immunity

**T10 The effect of lipopolysaccharide (LPS) and phorbol 12-myristate 13-acetate (PMA) on whole blood oxidative response as assessed by luminol-amplified chemiluminescence in dairy cows.** Y. Qu<sup>\*1</sup>, S. Kahl<sup>2</sup>, T. H. Elsasser<sup>2</sup>, E. E. Connor<sup>2</sup>, and K. M. Moyes<sup>1</sup>, <sup>1</sup>University of Maryland, College Park, MD, <sup>2</sup>Agricultural Research Service, US Department of Agriculture, Beltsville, MD.

The differences between lipopolysaccharide (LPS) and phorbol 12-myristate 13-acetate (PMA) on whole blood oxidative response using luminol-amplified chemiluminescence (CL) are currently unknown in cattle. Luminol-dependent CL measures the amount of reactive oxygen species released from leukocytes after stimulation with LPS and/or PMA. The objective of this study was to compare in vitro the effect of PMA and LPS on oxidative response in whole blood of dairy cows during lactation as a rapid means of assaying the oxidative response of blood leukocytes. Jugular blood (20 mL) was collected from 6 healthy multiparous Holstein dairy cows in mid-lactation (>90 DIM) using EDTA Vacutainer tubes. For each cow, 500 µL of blood was incubated at final concentrations of either 0, 200, 800 or 1,600 ng/mL of PMA or LPS for 15 min at 37°C using a heating block. After incubation, oxidative response of whole blood was measured using a chemiluminometer. Data were analyzed by ANOVA using the PROC MIXED procedure of SAS. Overall, whole blood incubated with PMA resulted in higher ( $P < 0.001$ ) CL values (800 ng/mL; 2635 relative units) than LPS (800 ng/mL; 777 relative units). In PMA, a significant dose response relationship was observed where incubation with 200, 800 or 1,600 ng/mL resulted in progressively higher CL values than 0 ng/mL. In addition, incubation with PMA resulted in a higher CL values when compared with LPS. In conclusion, although both LPS and PMA both generated an oxidative response measurable by CL, PMA elicited a CL response greater than that of LPS. The data suggest that PMA stimulation of cells in whole blood may serve as a rapid test of oxidative burst responsiveness to assess a vital aspect of immune function in dairy cows.

**Key Words:** chemiluminescence, cow, endotoxin

**T11 Efficacy of a novel antimicrobial post-milking teat dip on rate of new intramammary infections with an experimental bacterial challenge against contagious mastitis organisms.** David M. Galton<sup>1</sup> and Leo L. Timms<sup>\*2</sup>, <sup>1</sup>Cornell University, Ithaca, NY, <sup>2</sup>Iowa State University, Ames, IA.

Study objective was to determine efficacy of a novel antimicrobial solution containing citrate ion, methylene blue, parabens and emollients (ZuraLac<sup>SD</sup>, Zurex PharmAgra) used as a post-milking teat dip against a positive and negative control in reducing the incidence of new quarter intramammary infections (QIMI) during a period of experimental exposure of teats to contagious mastitis organisms, *Streptococcus agalactiae* and *Staphylococcus aureus*. 120 Holstein cows were used in an 8-wk trial with 40 cows/ treatment. Cows were 66–122 DIM and free of IMI. Three treatments were experimental dip - ZuraLac<sup>SD</sup> containing 10.0% trisodium citrate with 5.0% emollients used as a post-milking teat dip; positive control – Theratec<sup>R</sup> (GEA Farm Technologies Inc.) containing 0.5% iodine with a 3% triple emollient system used as a post-milking teat dip; and a negative control where no post-milking teat dipping occurred. All teats were dipped immediately after machine removal at morning milking with broth suspensions of *Streptococcus agalactiae*

(ATCC 55194) and *Staphylococcus aureus* (ATCC 12600). Teats were post dipped with appropriate teat dip between 2 and 5 min after teats dipped with culture broth. Duplicate quarter samples were taken aseptically weekly (and when clinical mastitis occurred) to determine quarter bacteriological status (3rd sample when results differed). Infection data were analyzed using Student *t*-statistics based on percent eligible quarters becoming infected with respective mastitis pathogens. Novel germicide post milking teat dip (3.3% quarter IMI) significantly ( $P < 0.01\%$ ) reduced the number of new infections of *Streptococcus agalactiae* and *Staphylococcus aureus* compared with both the negative control (53.7% QIMI; 95.3% reduction) and positive control (13.1% QIMI; 80.9% reduction). Incidence of clinical mastitis with a bacteriological positive identification of one of the challenge organisms was 40, 4, and 0 for negative control, positive control and novel germicide dips, respectively. Novel germicide dip was significantly better than positive and negative controls.

**Key Words:** postmilking teat dipping, intramammary infection, germicide

**T12 Effects of a 6-week duodenal supplementation of quercetin on metabolic stress and liver health in periparturient dairy cows.** Ann-Kathrin Stoldt<sup>1</sup>, Manfred Mielenz<sup>1</sup>, Alexander Starke<sup>2</sup>, Siegfried Wolfram<sup>3</sup>, and Cornelia C. Metges<sup>\*1</sup>, <sup>1</sup>Institute of Nutritional Physiology, Leibniz Institute for Farm Animal Biology, Dummerstorf, Germany, <sup>2</sup>Faculty of Veterinary Medicine, University of Leipzig, Leipzig, Germany, <sup>3</sup>Institute of Animal Nutrition and Physiology, Christian-Albrechts University of Kiel, Kiel, Germany.

The periparturient period poses metabolic challenges for dairy cows resulting in negative energy balance often followed by fatty liver disease, a typical periparturient metabolic disorder, and further health problems. Quercetin (Q), a polyphenolic compound found in plants, has hepatoprotective and antioxidative potential, and can reduce hepatic lipid accumulation in rodents. In ruminants, knowledge on metabolic effects of Q is scarce. Thus, we investigated whether Q affects lipid metabolism, oxidative stress defense, and has hepatoprotective effects in periparturient dairy cows. Because Q is degraded in rumen, 5 cows were given 100 mg Q dihydrate per kg BW daily in 0.9% NaCl solution into a duodenal fistula while control (CTR; n = 5) cows received NaCl only, starting 3 wk antepartum (ap) to 3 wk postpartum (pp). Twice-weekly blood samples were collected and liver was biopsied twice ap and once pp. Selected hepatic transcript abundances were determined by quantitative real-time PCR. Effects of Q were analyzed using repeated-measure ANOVA (SAS PROC MIXED). Duodenal supplementation of Q resulted in higher ( $P < 0.05$ ) plasma flavonoid levels in Q than in CTR cows. In Q cows pp plasma values of aspartate aminotransferase (AST) were lower ( $P < 0.05$ ) whereas glutamate dehydrogenase (GLDH) and BHBA levels tended to be lower ( $P = 0.1$ ). Liver fat content tended ( $P = 0.1$ ) to be lower in Q cows pp, although groups did not differ ( $P = 0.7$ ) in fat mobilization indicated by plasma NEFA. We could not show group differences of hepatic mRNA abundance of genes related to lipid metabolism and oxidative stress defense (fatty acid synthase, carnitine palmitoyltransferase 1A, peroxisome proliferator-activated receptors  $\alpha$  and  $\gamma$ , paraoxonase, superoxide dismutase, catalase and glutathione peroxidase). In conclusion, when Q is systemically available during the periparturient period there is potential that metabolic stress and liver damage in dairy cows can be reduced. However, these results should be verified in a larger number of cows and mechanisms of action of Q in cows need to be clarified. Eventually a rumen-protected form of Q has to be developed.

**Key Words:** transition dairy cow, quercetin, liver health



**T13 Effects of manipulated insulin and glucose plasma concentrations on glucagon secretion during intramammary LPS challenge in dairy cows.** Mousa Zarrin\*<sup>1,2</sup>, Olga Wellnitz<sup>1</sup>, and Rupert Bruckmaier<sup>1</sup>, <sup>1</sup>*Veterinary Physiology Vetsuisse Faculty, University of Bern, Bern, Switzerland*, <sup>2</sup>*Department of Animal Science, Yasouj University, Yasouj, Iran.*

Insulin and glucagon are gluco-regulatory hormones that contribute to glucose homeostasis which is also critical during immune reaction. Intramammary LPS challenge causes an immune reaction, which is accompanied by metabolic and endocrine changes. The objective of the present study was to investigate effects of intramammary LPS challenge concomitantly with elevated insulin concentrations on glucagon concentration during simultaneous hypoglycemia or euglycemia in dairy cows. Animals were randomly assigned to 3 treatment groups: an intravenous insulin infusion (HypoG, n = 5) to decrease plasma glucose concentration (2.5 mmol/L), a hyperinsulinemic euglycemic clamp to maintain plasma glucose at pre-infusion level to study effects of insulin at simultaneously normal glucose concentration (EuG, n = 6), and a 0.9% saline solution infusion (Control, n = 8). Glucose was measured in blood at 5-min intervals to allow adjustments of glucose infusion rate. Plasma insulin and glucagon concentrations were analyzed hourly. Area under the curve was evaluated by ANOVA with treatment as fixed effect. Data are presented as means ± SEM. The mean of insulin infusion rate before LPS challenge (48 h), was 0.6 mU/kg BW /min in HypoG and EuG. In EuG the glucose infusion rate was 2.20 ± 0.04 mmol/kg/min to keep plasma glucose concentrations at a pre-infusion level and it unchanged during the LPS challenge. In response to LPS challenge, plasma insulin and glucose increased. Plasma glucose concentration increased in EuG (4.4 ± 0.1 mmol/L) compared with concentrations before the LPS challenge (3.8 ± 0.2 mmol/L; *P* < 0.01), and HypoG (2.6 ± 0.1 mmol/L; *P* < 0.01). Intramammary LPS challenge caused an increase of plasma glucagon in HypoG and control compared with basal level, 48 h infusions period, and EuG, but the increase of glucagon was more pronounced in control (219.7 ± 17.3 pg/mL; *P* < 0.001). In conclusion, intramammary LPS challenge induces increases of glucose, insulin, and glucagon concentrations. The results show that glucagon concentrations dramatically increased in the absence of insulin infusion. This is in agreement with previous reports that suggested a general inhibitory effect of insulin on glucagon secretion.

**Key Words:** glucagon, insulin, glucose

**T14 Total immunoglobulin concentration in colostrum produced by dairy cows in Costa Rica.** J. A. Elizondo-Salazar\*<sup>1</sup>, D. Benavides-Varela<sup>2</sup>, and A. Vargas-Ramírez<sup>1</sup>, <sup>1</sup>*Estación Experimental Alfredo Volio Mata. Facultad de Ciencias Agroalimentarias, Universidad de Costa Rica, San José, Costa Rica*, <sup>2</sup>*Alimentos del Norte S.A-DIPCR, Costa Rica.*

The objective of this study was to determine total immunoglobulin (Ig) concentration in colostrum produced by dairy cows and establish the effect that breed and parity can have on Ig concentration. The data presented correspond to determinations of total Ig concentration determined by a colostrometer in 537 colostrum samples obtained in 50 dairy farms in the provinces of San José, Alajuela, Heredia, and Cartago. Cow breeds were classified into Holstein, Jersey, Holstein × Jersey, and other. Farm size ranged from 10 to 300 milking cows. To determine breed and parity number effect on colostrum Ig concentration, MIXED procedure was used, where dam was considered as a random effect. For the purposes of this study, good quality colostrum was considered when Ig concentration was ≥50 mg/mL. Immunoglobulin concentration ranged from 10 to 140 mg/mL with an average of 85 mg/mL. Of all the

samples analyzed, 13.2% had an inadequate concentration of Ig. When considering breed effect on Ig concentration, no significant differences were found (*P* > 0.05). Parity number significantly (*P* < 0.05) influenced Ig concentration and it was found that Ig concentration increased with parity number. Based on the current study, Ig concentration was adequate for calf feeding in 87% of colostrum samples.

**Table 1 (Abstr. T14).** Effect of dam breed on total Ig concentration in 537 colostrum samples from 50 dairy farms in Costa Rica

Dam breed	Number	Ig (mg/mL)	SEM
Holstein	270	88.8	1.9
Holstein × Jersey	64	85.9	2.5
Jersey	146	85.5	2.6
Other	57	85.6	4.2

**Key Words:** passive immunity, colostrometer, immunoglobulin

**T15 Risk factors associated with milk fever occurrence in Costa Rican dairy cattle.** Alejandro Saborío-Montero\*<sup>2</sup>, Bernardo Vargas-Leitón<sup>1</sup>, Juan José Romero-Zúñiga<sup>1</sup>, and Jorge M. I. Sánchez<sup>2</sup>, <sup>1</sup>*Programa de Investigación en Medicina Poblacional, Escuela de Medicina Veterinaria, Universidad Nacional, Heredia, Costa Rica*, <sup>2</sup>*Centro de Investigaciones en Nutrición Animal y Escuela de Zootecnia, Universidad de Costa Rica, San José, Costa Rica.*

The aim of this study was to determine risk factors associated with milk fever (MF) occurrence in dairy cows in Costa Rica. A total of 69,870 cows from 127 dairy herds were included in the study. Data were collected in the VAMPP (Veterinary Automated Management and Production Control Program) software by the Population Medicine Research Program of the Veterinary Medicine School, National University of Costa Rica; from 1985 to 2014. To determine the risk factors, 2 logistic regression mixed models were evaluated. The first model used breed (B), month of calving (M), quinquennium of calving (Y) ecological life zones (Z) and calving number (N) as fixed effects. The second model excluded first lactation animals and cows without productive information, had the same fixed effects of the first model plus: previous MF case (C), previous lactation length (L), previous dry period length (D), previous corrected 305d milk yield (P), and calving interval length (I) as fixed effects. Both models used animal (a) and herd (h) as random effects. Of the 235,971 recorded lactations, 4,312 (1.83%) reported MF event. The significantly associated (*P* < 0.05) risk factors for MF occurrence, ranked by their highest odds ratio (OR) are listed on Table 1. The findings of this study are the first data reported for a population study on risk factors for MF in Costa Rica. Some of these results might be used to improve preventive management practices at the farms to reduce the incidence of this metabolic disease.

*Contd.*

**Table 1 (Abstr. T15).** Risk factors associated with milk fever (MF) occurrence ranked by odds ratio (OR) in Costa Rican dairy cattle<sup>1</sup>

Risk factor	Riskiest category	Reference category	OR	95% CI
Calving number	6th or more calving	1st calving	52.40	40.94–67.08
Breed	Jersey	Brown Swiss	2.88	1.66–4.98
Previous MF case	Previous case reported	No previous case reported	2.39	2.17–2.63
Quinquennium of calving	1990-1994	2005-2009	2.33	2.00–2.78
Month of calving	July	December	1.37	1.16–1.61
Previous 305-d milk yield	1,000 kg over $\mu$	$\mu$ for 305-d milk yield	1.17	1.14–1.21
Previous lactation length	30 d over $\mu$	$\mu$ for lactation length	1.06	1.03–1.09

<sup>1</sup> $\mu$  = population mean; 6,021 kg for previous 305-d milk yield, 316 d for previous lactation length.

**Key Words:** milk fever, risk factor, dairy cattle

**T16 Serum calcium concentration during the peripartum period in a Jersey herd grazing tropical pastures and supplemented with a low calcium grain mixture.** Jorge M. I. Sánchez\* and Alejandro Saborío-Montero, *Centro de Investigaciones en Nutrición Animal y Escuela de Zootecnia, Universidad de Costa Rica, San José, Costa Rica.*

The aim of this study was to analyze the serum Ca concentration in a Jersey herd during the peripartum period in a tropical production system, to compare Ca status in young ( $\leq 2$ nd parity) and adult ( $\geq 3$ rd parity) cows. The study was conducted on a dairy farm located in Cartago, Costa Rica, during a 6-mo period (January–June, 2014) and comprised 161 cows (62 young cows; 99 adult cows). During the close-up period cows grazed lush kikuyu grass (*Kikuyuocloa clandestina*) (14.8% DM, 23.4% CP, 0.35% Ca, 0.31% Mg and 3.5% K) and were supplemented with 4 kg/cow/d of a low Ca grain mixture (88% DM, 14% CP, 0.2% Ca, 0.42% Mg and 1.38% K) and 1 kg of hay (82.3% DM, 5.1% CP, 0.4% Ca, 0.35% Mg and 1.8% K)/d. After calving, cows were fed 1 kg of grain mixture (87.5% DM, 18.6% CP, 0.90% Ca, 0.42% Mg and 1.38% K)/2.5 to 3.0 kg of milk. A total of 752 blood samples were taken from 4d before calving until 5d postpartum and were analyzed for total calcium. Data were analyzed as a repeated measures with the fixed effect of parity class and cow nested within parity was the subject of the repeated measure of time. Statistical analysis showed significant effect for category, not so for day and day x category interaction. Means across the 2 categories were compared by Student's *t*-test (Table 1). Serum Ca concentrations in young cows were usually greater than concentrations in adult cows. Starting at 1 d before calving through 1 d post calving, serum Ca in adult cows was depressed reaching its nadir at 1 d in milk. Results show that adult cows were subclinically hypocalcemic (serum Ca <8.0 mg/dl) from d 2 before calving until 5 d in milk, requiring feeding practices designed to improve Ca metabolism in the periparturient adult cows.

*Contd.*

**Table 1 (Abstr. T16).** Serum Ca concentration (mg/dl) from 4 d before to 5 d after calving for young (1 and 2 parities) and adult (3 or more parities) Jersey cows grazing lush tropical pastures

Category	Day relative to calving									
	-4	-3	-2	-1	0	1	2	3	4	5
Young	8.41 <sup>a</sup>	8.18 <sup>a</sup>	8.19 <sup>a</sup>	8.37 <sup>a</sup>	7.57 <sup>a</sup>	7.85 <sup>a</sup>	7.90 <sup>a</sup>	8.15 <sup>a</sup>	7.97 <sup>a</sup>	8.09 <sup>a</sup>
Adult	8.66 <sup>a</sup>	8.09 <sup>a</sup>	7.74 <sup>a</sup>	7.49 <sup>b</sup>	6.98 <sup>b</sup>	6.72 <sup>b</sup>	7.42 <sup>a</sup>	7.67 <sup>a</sup>	7.68 <sup>a</sup>	7.86 <sup>a</sup>
<i>P</i> -value	0.781	0.901	0.280	0.046	0.027	0.009	0.127	0.092	0.445	0.438

<sup>ab</sup>Means in the same column with different superscripts are different ( $P < 0.05$ ).

**Key Words:** grazing cow, hypocalcemia, milk fever

**T17 Bacterial diversity and pathogen load in recovered dairy cows bedding materials following an aerobic composting of dairy manure.** Maral Rahmani\*, Hooman Derakhshani, Hein M. Tun, Jacqueline Donogh, Shadi Sepehri, and Ehsan Khafipour, *University of Manitoba, Winnipeg, MB, Canada.*

The FAN bedding recovery system consists of a solid separator and an in-vessel aerobic composting system, allowing the treatment of dairy manure into separate liquid and solid streams, with the solid stream later used within the barn as dairy cows bedding. The objective was to evaluate the effect of composting on the microbiota profile and pathogen load of recovered bedding material (RBM). Manure (MAN), liquid stream (LS), solid stream undigested (SSU) and solid stream digested (SSD) samples were collected on a weekly basis for a 2-mo period during summer and winter seasons ( $n = 36$ /category). Genomic DNA was extracted and subjected to bacterial 16S rRNA gene sequencing for community analysis and qPCR for absolute quantification of generic *Escherichia coli*, *E. coli* O157:H7, *Staphylococcus aureus*, *Mycobacterium avium* ssp. *paratuberculosis* (MAP); *Arcanobacterium pyogenes*, and *Klebsiella pneumoniae*. On average, 29,982 high-quality sequences were generated per sample. The  $\alpha$ -diversity of SSD microbial communities was found to be lower ( $P < 0.001$ ) than other treatment groups, with the MAN samples showing the most diverse microbiota followed by LS and SSU. The proportion of phylum Firmicutes (members of the family Bacillaceae) was greater ( $P < 0.05$ ) in SSD group, and the relative abundances of several members of the phyla Proteobacteria (family Moraxellaceae and genus *Acinetobacter*) and Bacteroidetes (family Sphingobacteriaceae) were found to be significantly ( $P < 0.05$ ) higher in SSU compared with other groups. The proportions of several members of the phylum Proteobacteria were also found to be higher in SSU compared with LS, suggesting that, in the absence of composting process, separation of the solid and liquid part of the RBM may increase the proportion of the opportunistic microorganism in the solid fraction. *E. coli* O157:H7, MAP and *S. aureus* were present only in a limited number of samples and only 2 SSD samples found to be MAP, O157:H7 or *S. aureus* positive. Aerobic composting had limited effect on the population of *E. coli*, *A. pyogenes*, and *K. pneumoniae*.

**Key Words:** aerobic composting, microbial community, pathogen load

**T18 Massive shotgun metagenomic sequencing reveals the potential mode of action of *Saccharomyces cerevisiae* fermentation product (SCFP) on rumen microbiome during subacute ruminal acidosis (SARA) in dairy cows.** H. M. Tun\*<sup>1</sup>, S. Li<sup>1</sup>, I. Yoon<sup>2</sup>, M. Scott<sup>2</sup>, J. C. Plaizier<sup>1</sup>, and E. Khafipour<sup>1</sup>, <sup>1</sup>*Department of Animal Science, University of Manitoba, Winnipeg, MB, Canada,* <sup>2</sup>*Diamond V, Cedar Rapids, IA.*

The effects of *Saccharomyces cerevisiae* fermentation product on rumen microbiome composition and function were studied in 8 rumen-cannulated lactating cows in a crossover study with two 5-wk periods. Each period consisted of a 4-wk normal feeding and a 1-wk grain-based SARA challenge. A 3-wk washout period separated the experimental periods. During each period, 4 cows received 14 g/d of SCFP (Original XPC, Diamond V) mixed with 126 g/d ground corn and the other 4 received 140 g/d ground corn as control. Rumen fluid was collected during wk 4 and 5 of each period. Genomic DNA was extracted and subjected to shotgun metagenomic sequencing using a MiSeq Illumina platform. Host genomic sequences were removed before analysis. Taxonomies were annotated against Greengenes database and functional genes were annotated against SEED subsystems in the MG-RAST pipeline. Both compositional and functional differences among treatments were analyzed by the linear discriminant analysis effect size. In total, 103.4 GB sequences were obtained from 32 shotgun metagenomic samples with an average of 10 millions sequences per sample. The SCFP supplement altered the  $\beta$ -diversity of rumen functional metagenome under both control and SARA conditions ( $P < 0.05$ ). The SCFP supplement restored several cellulolytic populations, including *Fibrobacteres*, *Paenibacillaceae* and *Spirochaetaceae*, as well as *Burkholderiales* that were suppressed during SARA ( $P < 0.05$ ). The SCFP supplement also tended to increase the population of *Eubacteriaceae*, *Coriobacteriaceae* and unclassified *Clostridiales* observed during SARA ( $P < 0.05$ ). Several enzymatic pathways were downregulated during SARA. The SCFP supplement increased the abundance of formate dehydrogenase and methylmalonyl-CoA mutase (MCM) that were suppressed during SARA ( $P < 0.05$ ). The MCM is a vitamin B12-dependent enzyme constantly found in methanol-utilizing bacteria, such as members of *Burkholderiales*. Data reveals potential mechanisms through which SCFP supplement contributes to the resilience of the rumen microbiome, especially during SARA.

**Key Words:** shotgun metagenomic, *Saccharomyces cerevisiae* fermentation product, subacute ruminal acidosis

#### **T19 Effects of *Saccharomyces cerevisiae* fermentation product (SCFP) on the predicted functional profiles of rumen microbiome in lactating dairy cows with subacute ruminal acidosis (SARA).**

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The effects of *Saccharomyces cerevisiae* fermentation product (SCFP) on the rumen bacterial function were studied in 8 rumen-cannulated lactating cows in a crossover study with 2 5-wk periods. Each period consisted of 4 wk of normal feeding and a 1-wk grain-based SARA challenge. A 3-wk washout period separated the experimental periods. During each period, 4 cows received 14 g/d of SCFP (Original XPC, Diamond V) mixed with 126 g/d ground corn and the other 4 received 140 g/d ground corn as control. Rumen fluid was collected during wk 4 and 5 of each period. DNA was extracted and V1-V3 region of 16S rRNA gene was amplified and subjected to pyrosequencing. Phylogenetic Investigation of Communities by Reconstruction of Unobserved States (PICRUSt) was used to impute metagenomic information based on 16S rRNA sequencing data. The SARA challenge and SCFP supplementation did not affect the  $\alpha$ -diversity of the predicted functional gene families of rumen bacterial communities. However, the  $\beta$ -diversity was affected by SARA ( $P < 0.01$ ), but not by SCFP. A total of 9 out of 36 level-2, and 39 out of 254 level-3 KEGG Orthology groups were found to be affected by SARA challenge ( $P < 0.01$ ). At the KEGG level-2, pathways upregulated during SARA included cell motility, membrane

transport, signal transduction and transcription, while pathways involved in the metabolism of terpenoids and polyketides, biosynthesis of other secondary metabolites, amino acid metabolism, signaling molecules and interaction, transport and catabolism were downregulated. However, SCFP supplementation and interaction between SARA challenge and SCFP supplementation were not significant. The relevance and accuracy of PICRUSt application to predict the function of microbiome in the rumen need further validation using shotgun sequencing approach.

**Key Words:** dairy cow, SARA, *Saccharomyces cerevisiae* fermentation product

#### **T20 Effect of milk yield genotype on response to repeated lipopolysaccharide (LPS) administration to lactating Holstein cows.**

Georgina Cousillas\*<sup>1</sup>, Wanda J. Weber<sup>1</sup>, Stanislaw Kahl<sup>2</sup>, Bruce Walcheck<sup>1</sup>, Ricardo Chebel<sup>1</sup>, David Kerr<sup>3</sup>, Theodore H. Elsasser<sup>2</sup>, and Brian A. Crooker<sup>1</sup>, <sup>1</sup>University of Minnesota, St. Paul, MN, <sup>2</sup>USDA-ARS, Beltsville, MD, <sup>3</sup>University of Vermont, Burlington, VT.

Cows (n = 12/genotype) from unselected (stable milk yield since 1964, UH) and contemporary (CH) Holsteins that differed by more than 4,500 kg milk/305 d were fed the same diet ad lib and housed together for more than 4 mo before being blocked (2/genotype) by DIM and randomly assigned within genotype to receive saline or 0.25  $\mu$ g LPS (*Escherichia coli* 055:B5) per kg BW. Cows were synchronized to be at d 8 of their estrous cycle for the first challenge (C1) at 70–84 d in milk. Jugular catheters were implanted 24 h before C1. Blood samples were collected at -1, -0.5, 0, 1, 2, 3, 4, 6, 8, 12, and 24 h relative to treatment administration and plasma harvested. Body temperatures (BT) were determined at these times and at 5 and 7 h. Liver biopsies and blood for flow cytometry and hemogram assays were obtained at 0, 4, and 24 h. A second identical challenge (C2) and sampling was conducted 4 d later. Data were analyzed by repeated measures using PROC MIXED (SAS). Means differed when  $P < 0.05$ . Pre-challenge glucose and IGF-1 were greater ( $P < 0.01$ ) and BT was less ( $P < 0.01$ ) in UH than CH. Glucose response to LPS was greater ( $P < 0.01$ ) in UH than CH, but IGF-1 and BT response was similar in both genotypes. TNF $\alpha$  and cortisol response to LPS was greater during C1 than C2 ( $P < 0.02$ ). TNF $\alpha$  response to LPS was greater ( $P < 0.05$ ) in UH than CH in C1 but similar in C2. Cortisol response to LPS increased in both genotypes but returned to baseline earlier in CH than UH ( $P < 0.05$ ). LPS decreased white blood cell count ( $P < 0.01$ ) but response did not differ between genotypes or challenge. Neutrophil oxidative burst was greater ( $P < 0.05$ ) and phagocytic capacity tended ( $P = 0.07$ ) to be greater in UH than CH. CD11b expression increased ( $P < 0.05$ ) in response to LPS at 4h, was less in CH than UH at 24h and did not differ between C1 and C2. L-selectin decreased in response to LPS at 4hr but did not differ between challenge or genotype. Results indicate that genotype affects bovine response to LPS and this effect differs among the response variables assessed.

**Key Words:** innate immunity, Holstein genotype, lipopolysaccharide

#### **T21 Use of chitosan microparticles to prevent metritis in lactating dairy cows.**

Rodolfo Daetz\*<sup>2</sup>, Federico Cunha<sup>2</sup>, Yosuke Maeda<sup>3</sup>, Carlos A. Risco<sup>2</sup>, Kwang C. Jeong<sup>1,4</sup>, Jose Eduardo P. Santos<sup>1</sup>, and Klibis N. Galvao<sup>2</sup>, <sup>1</sup>Department of Animal Sciences, University of Florida, Gainesville, FL, <sup>2</sup>Department of Large Animal Clinical Sciences, University of Florida, Gainesville, FL, <sup>3</sup>School of Veterinary Medicine, Kitasato University, Towada, Japan, <sup>4</sup>Emerging Pathogens Institute, University of Florida, Gainesville, FL.



The objective was to determine the efficacy of chitosan microparticles (CM) in preventing metritis in dairy cows. Holstein cows ( $n = 101$ ) from a 4,500-cow commercial herd that had risk factors for metritis (dystocia, twins, stillbirth, retained placenta) were randomly assigned to one of 2 treatments 1 d postpartum (DPP): CM ( $n = 51$ ) = intrauterine (i.u.) infusion of 8 g of CM dissolved in 40 mL of sterile water for 5 d; Control ( $n = 50$ ) = i.u. infusion of 40 mL of sterile saline solution for 5 d. Metritis prevalence was analyzed by logistic regression using the LOGISTIC procedure of SAS using a one-side test in accordance with sample size calculation for reduction in metritis prevalence using CM. Continuous outcomes were analyzed by ANOVA for repeated measures using the MIXED procedure of SAS. Models included the effects of treatment, parity, specific risk factor, body condition score at enrollment and interaction between treatment and other covariates. The effect of time and interaction between treatment and time was also included in repeated measures analyses. Treatment with CM resulted in decreased incidence of metritis at 7 DPP compared with Control (45.1 vs. 64.0%, respectively;  $P = 0.03$ ); however, there were only numerical differences at 4 (11.8 vs. 18%, respectively;  $P = 0.23$ ), 10 (60.1 vs. 72%, respectively;  $P = 0.12$ ), and 14 (62.7 vs. 72.0%, respectively;  $P = 0.16$ ). Treatment with CM resulted in decreased NEFA plasma concentrations at 10 DPP ( $464.2 \pm 57.2$  vs.  $639.5 \pm 57.2$   $\mu\text{Eq/L}$ ;  $P = 0.04$ ); however, there were no differences at 4 ( $813.8 \pm 56.7$  vs.  $780.4 \pm 56.7$   $\mu\text{Eq/L}$ ;  $P = 0.67$ ), 7 ( $669.9 \pm 56.7$  vs.  $692.9 \pm 56.7$   $\mu\text{Eq/L}$ ;  $P = 0.77$ ), and 14 ( $527.6 \pm 57.7$  vs.  $420.7 \pm 57.7$   $\mu\text{Eq/L}$ ;  $P = 0.18$ ). The uterine discharge pH was lower in Control than in CM cows ( $6.84 \pm 0.03$  vs.  $6.93 \pm 0.03$ ;  $P = 0.02$ ). BHBA ( $647.4 \pm 30.0$  vs.  $589.3 \pm 30.0$   $\mu\text{mol/L}$ ;  $P = 0.36$ ), temperature ( $39.2 \pm 0.04$  vs.  $39.1 \pm 0.04^\circ\text{C}$ ;  $P = 0.62$ ) and milk production ( $29.3 \pm 1.0$  vs.  $28.8 \pm 1.0$  L/d;  $P = 0.69$ ) were not different between CM and Control groups. In conclusion, CM may be a viable alternative for treatment of metritis; however, the duration of treatment may have to be extended to maintain differences in the incidence of metritis.

**Key Words:** chitosan microparticle, metritis, dairy cow

**T22 The role of Bacteroidetes and Bacteroides species in the development of metritis and fever in dairy cows.** Soo Jin Jeon<sup>\*2</sup>, Achilles Vieira-Neto<sup>1</sup>, and Klíbs N. Galvão<sup>2</sup>, <sup>1</sup>Department of Animal Sciences, University of Florida, Gainesville, FL, <sup>2</sup>Department of Large Animal Clinical Sciences, University of Florida, Gainesville, FL.

Objective was to evaluate if uterine microbiota was associated with the presence of fever in metritic cows. Uterine swabs were collected at  $6 \pm 2$  d postpartum (DPP) from 92 cows. Uterine swabs from 11 metritic cows that had a fever (rectal temperature  $\geq 39.5^\circ\text{C}$ ) at the time of metritis diagnosis (MF), 12 metritic cows that did not have a fever at the time of metritis diagnosis (MNF), and 11 cows that remained healthy (Healthy) were used for metagenomic sequencing of the 16S rRNA gene using the Illumina MiSeq platform. Data were analyzed by ANOVA using the GLM procedure of SAS. Rectal temperature was  $38.97 \pm 0.12$ ,  $38.96 \pm 0.08$ , and  $39.89^\circ\text{C} \pm 0.11$  for Healthy, MNF, and MF, respectively. At the phylum level, Bacteroidetes were found to have lower ( $P < 0.01$ ) relative abundance in Healthy than in MNF and MF (27.3 vs. 51.1 vs. 53.3%), whereas there was no difference ( $P > 0.25$ ) in Fusobacteria (23.9 vs. 28.6 vs. 24.6%), Actinobacteria (1.2 vs. 0.5 vs. 1.1%), and Firmicutes (18.6 vs. 12.4 vs. 15.6). Proteobacteria in Healthy was similar ( $P = 0.13$ ) to MNF but greater ( $P = 0.05$ ) than MF (16.6 vs. 6.5 vs. 2.9%). Tenericutes was lower ( $P < 0.04$ ) in Healthy than in MNF and MF (10.6 vs. 0.7 vs. 2.0%). At the species level, principal coordinates analysis showed that MNF were clustered together with MF, whereas Healthy were mostly separated from metritic

cows. Relative abundance of *Bacteroides heparinolyticus* in Healthy was lower ( $P = 0.01$ ) than in MNF and tended ( $P = 0.06$ ) to be lower than in MF (7.0 vs. 20.3 vs. 16.8%). *Bacteroides pyogenes* was lower ( $P \leq 0.05$ ) in Healthy and MNF than in MF (4.0 vs. 3.8 vs. 10.0%), but similar between Healthy and MNF. There was no difference in the relative abundance of *Fusobacterium necrophorum* ( $P = 0.68$ ) or other 10 most prevalent bacteria ( $P > 0.15$ ). In conclusion, Bacteroidetes is more abundant, whereas Firmicutes and Tenericutes are less abundant in metritic cows. Furthermore, *B. heparinolyticus* is an important species for the development of metritis, whereas *B. pyogenes* seem to be involved in the development of fever in metritic cows.

**Key Words:** Bacteroidetes, *Bacteroides* species, metritis

**T23 Incidence of health treatments among pure Holsteins of 8 high-performance dairies in Minnesota.** M. R. Donnelly<sup>\*1</sup>, A. R. Hazel<sup>1</sup>, B. J. Heins<sup>2</sup>, and L. B. Hansen<sup>1</sup>, <sup>1</sup>University of Minnesota, St. Paul, MN, <sup>2</sup>West-Central Research and Outreach Center, Morris, MN.

Health treatments of pure Holstein cows ( $n = 3,936$ ) were evaluated in 8 high-performance Minnesota dairies. Cows calved from March 2008 to February 2013, and 16 types of health treatments were defined uniformly across herds. For analysis, treatments were grouped into 5 categories: mastitis, reproduction (cystic ovary, retained placenta, and metritis), hoof, metabolic (milk fever, displaced abomasum, ketosis, and digestive), and miscellaneous (respiratory, injury, and other). Excluded from analysis were California Mastitis Test/culture, hoof trimming, palpation, reproductive aid, and abortion. Parity was coded as 1 ( $n = 2,285$ ), 2 ( $n = 2,529$ ), or 3 to 5 ( $n = 3,842$ ), and later lactations were deleted. Observations were recorded for entire lactations and coded as treated (1) or not treated (0) during a lactation. Independent variables for statistical analysis of all 5 treatment categories were the fixed effects of herd, year-season nested within herd, parity, and interaction of herd and parity. Cow was considered a repeated measure. For all 5 treatment categories, herd and year-season nested within herd were highly significant ( $P < 0.01$ ). Parity was significant ( $P < 0.05$ ) for all 5 categories of health treatments, except parity was not significant ( $P = 0.71$ ) for miscellaneous. Interaction of herd and parity was significant ( $P < 0.05$ ) for all categories. Least squares means increased with advancing parity for all 5 treatment categories except for the miscellaneous category. Treatment rates for parities 1, 2, and 3 to 5, respectively, for each health treatment category were 0.25, 0.34, and 0.42 (mastitis); 0.12, 0.13, and 0.16 (reproduction); 0.32, 0.39, and 0.40 (hoof); 0.07, 0.13, and 0.18 (metabolic); 0.14, 0.13, and 0.14 (miscellaneous). Across herds and parities, the largest treatment rate was for the hoof category followed by mastitis, and the other 3 treatment categories had similar treatment rates.

**Key Words:** health, treatment, mastitis

**T24 Rumination behavior alert indexes for detecting health disorders during early lactation.** Sushil Paudyal<sup>1</sup>, Fiona Maunsell<sup>2</sup>, Carlos Risco<sup>2</sup>, Arthur Donovan<sup>2</sup>, Albert De Vries<sup>3</sup>, John Richeson<sup>1</sup>, and Pablo Pinedo<sup>\*4,5</sup>, <sup>1</sup>West Texas A&M University, Canyon, TX, <sup>2</sup>Department of Large Animal Clinical Sciences, College of Veterinary Medicine, University of Florida, Gainesville, FL, <sup>3</sup>Department of Animal Sciences, University of Florida, Gainesville, FL, <sup>4</sup>Texas A&M AgriLife Research, Amarillo, TX, <sup>5</sup>Department of Veterinary Pathobiology, College of Veterinary Medicine & Biomedical Sciences, Texas A&M University System, College Station, TX.

Monitoring of health disorders during early lactation is a key component in the management of dairy systems. Electronic systems that allow for monitoring of rumination are now available. The objective was to evaluate the association between changes in rumination time (RT) and early stages of disease within the prepartum and early lactation. Two weeks before the due date, 198 cows at the University of Florida (UF) Dairy Unit were affixed with a neck collar containing rumination loggers (Hr-Tag rumination monitoring system, SCR Engineers Ltd., Netanya, Israel), providing rumination time (RT) for each 2-h interval. Occurrence of health disorders [mastitis (MAS), metritis (MET), clinical hypocalcemia (HYC), digestive disorders (DIG), lameness (LAM), and ketosis (KET),] were assessed until 60 DIM by UF veterinarians and farm personnel. Two indices were developed to explore the association between RT and disease: i) Cow Index (CI) = 24-h RT on the day of diagnosis (d0) minus average RT on d -3 to -5 relative to d0 (0to3-5RT), divided by the -3 to -5 d RT average; ii) Mates Index (MI) = (0to3-5RT - pen mates 0to3-5RT)/pen mates d0 RT. Subsequently an alert value (ACI and AMI) was determined for both indices when the value was lower than -0.1. Alert indices were evaluated by ROC curve analyses. Average CI in healthy cows ranged from 0.0491 to 0.0495 while CI in sick cows were -0.165, -0.029, -0.513, -0.022, -0.098, and -0.081 for MAS, MET, HYC, DIG, LAM, and KET, respectively. Average MI in healthy cows ranged from 0.0001 to 0.001 while MI in sick cows were -0.183, -0.101, -0.424, -0.101, -0.148, and -0.147 for MAS, MET, HYC, DIG, LAM, and KET, respectively. Sensitivity/specificity (%) of ACI were 56/72, 39/73, 100/72, 44/72, 67/72, and 61/72 for MAS, MET, HYC, DIG, LAM, and KET, respectively. Sensitivity/specificity (%) of AMI were 63/71, 42/72, 100/72, 48/72, 55/72, and 67/72 for MAS, MET, HYC, DIG, LAM, and KET, respectively. Area under the curve for our proposed cut-off value ranged from 0.56 (KET) to 0.87 (HYC) for CI and from 0.51 (MET) to 0.87 (HYC) for MI. Consistent negative changes in rumination activity, both within cow (CI) and compared with cohorts (MI), were observed on the day of diagnosis for each postpartum disease.

**Key Words:** rumination, disease, alert index

**T25 OmniGen-AF alters rectal temperature (RT) and leukocyte profiles in dairy cows exposed to heat stress (HS) following acute activation of the stress axis.** Nicole C. Burdick Sanchez<sup>1</sup>, Jeffery A. Carroll<sup>1</sup>, Paul R. Broadway<sup>1</sup>, Matthew L. McBride<sup>2</sup>, Xavier A. Ortiz<sup>2</sup>, Jayne L. Collier<sup>2</sup>, James D. Chapman<sup>3</sup>, Derek McLean<sup>3</sup>, and Robert J. Collier<sup>2</sup>, <sup>1</sup>USDA-ARS, Livestock Issues Research Unit, Lubbock, TX, <sup>2</sup>University of Arizona, Department of Animal Science, Tucson, AZ, <sup>3</sup>Phibro Animal Health Corp., Quincy, IL.

Differences in the response of OmniGen-AF (OG) supplemented dairy cows to a corticotropin releasing hormone (CRH) and vasopressin (VP) or an adrenocorticotrophic hormone (ACTH) challenge when housed at different temperature-humidity indices (THI) were studied. Holstein cows (n = 12; 162 ± 1 DIM) were balanced by milk yield, BW and DIM into 1 of 2 trts: 1) OminGen-AF, supplemented with OG at 4.54 g/kg BW for 70d; or 2) Control (CON), no supplement. Cows were moved to individual tie stalls in 1 of 2 temperature controlled chambers on d51 and fitted with indwelling RT devices and jugular catheters on d52. Initially THI was cycling at thermoneutrality (TN; THI < 72 for 24 h/d) for 7d, followed by HS (THI > 72 for 12h/d) for 10d. Cows were challenged with CRH (0.3 µg/kg) and VP (1 µg/kg) at 1000h on d4 of TN and d1 of HS, and with ACTH (0.1 IU/kg) at 1000h on d5 of TN and d2 of HS. Blood samples were collected from -2 to 8h at 30-min intervals relative to each challenge and analyzed for leukocyte profiles. There was a THI × time interaction ( $P \leq 0.01$ ) for RT such that RT was greater during

HS than TN (2 to 9h, and at 11h for CRHVP and 4 to 9h and 11 to 13h for ACTH). Also, RT was greater ( $P \leq 0.02$ ) in OG than CON cows regardless of challenge. Total white blood cells (WBC) and neutrophils (NT) increased ( $P < 0.01$ ) in response to CRHVP and ACTH. There was a trt × THI interaction ( $P < 0.01$ ) for WBC during both challenges. Specifically, WBC were decreased in CON during HS compared with TN in response to CRHVP, while WBC were greatest in CON during TN and least in OG during HS in response to ACTH. There was a trt × THI interaction ( $P = 0.02$ ) for NT in response to CRHVP challenge; NT decreased in CON cows during HS compared with TN. Also, NT were decreased during HS ( $P < 0.01$ ) compared with TN and decreased in OG compared with CON ( $P < 0.01$ ) following ACTH. Lymphocytes were decreased during HS compared with TN ( $P < 0.01$ ) and in OG compared with CON cows ( $P < 0.01$ ) regardless of challenge. These data suggest supplementing cows with OG can reduce the negative effects of HS on leukocyte profiles following activation of the stress axis.

**Key Words:** heat stress, immune, OmniGen-AF

**T26 Altered microbiomes in bovine digital dermatitis lesions, and the gut as a pathogen reservoir.** Martin Zinicola<sup>\*1</sup>, Fabio Lima<sup>1</sup>, Svetlana Lima<sup>1</sup>, Vinicius Machado<sup>1</sup>, Charles Guard<sup>1</sup>, Dörte Döpfer<sup>2</sup>, and Rodrigo Bicalho<sup>1</sup>, <sup>1</sup>Cornell University, Ithaca, NY, <sup>2</sup>University of Wisconsin, Madison, WI.

Bovine digital dermatitis (DD) is the most relevant infectious disease associated with lameness affecting cattle worldwide. Notwithstanding decades of research, the pathogenesis of this disease and the potential reservoir of pathogenic microbes involved in DD remain unclear. Here, we characterize the microbiome of healthy skin (HS) and lesions from dairy cows affected with different stages of DD and we also identified DD-causing *Treponema* spp. in rumen and fecal samples. A total of 140 biopsy samples (BS) (51 HS and 89 DD lesions) were collected from Holstein dairy cows housed in 3 different dairy farms. BS were trimmed in 2 different layers, resulting in 280 samples. Rumen fluid (n = 8) and fecal (n = 14) samples were also collected. DNA was extracted and the microbiome was determined by shotgun and 16S metagenomic techniques using Illumina MiSeq platform. Discriminant analysis revealed that microbiomes of HS, active and inactive DD lesions were completely distinct. The differences in microbiomes between the superficial and deep strata were found to be minor. *Treponema* spp. were found in greater ( $P < 0.05$ ) relative abundance in active DD lesions when compared with HS and inactive DD lesions and these *Treponema* spp. were nearly ubiquitously present in rumen and fecal microbiomes. *Candidatus amoebophilus asiaticus*, a bacterium not previously reported in DD lesions, was encountered in high ( $P < 0.05$ ) relative abundance in active and inactive lesions but not in HS. In conclusion, our data support the concept that DD is a polymicrobial disease, with active DD lesions having a markedly distinct microbiome dominated by *T. denticola*, *T. maltophilum*, *T. medium*, *T. putidum*, *T. phagedenis* and *T. paraluiscuniculi*. Furthermore, these *Treponema* species are nearly ubiquitously found in rumen and fecal microbiomes, suggesting that the gut is an important reservoir of microbes involved in DD pathogenesis. Further investigation into the potential role of the gut microbiome as a reservoir for pathogens leading to DD development and of prophylactic measures to control the potential environmental shedding of these pathogens is needed.

**Key Words:** bovine digital dermatitis, dairy cow, microbiome

### T27 Characterization of the leukocyte transcriptome in cows challenged with *Mycobacterium bovis* and healthy controls.

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*Mycobacterium bovis* (*M. bovis*) is a pathogenic bacteria that generates approximately \$3 billion worldwide production loss for the cattle industry, in terms of animal value and productivity. To help understand the response to this pathogen, the objective of this study was to characterize the transcriptome of cows infected with *M. bovis* relative to healthy controls. Cows positive for *M. bovis* were identified using the single intradermal comparative tuberculin test, and in vitro ELISA-based interferon-gamma. The transcriptome of peripheral blood leukocytes from Holstein cows detected positive was compared with healthy control cows (n = 8/group) using individual RNA-Seq libraries. Single-end reads were mapped to the *Bos taurus* reference genome (UCSC\_bosTau7) using Tophat v2.0.12. In total, 8,309 isoform transcripts pertaining to 8,127 genes were identified and 2,620 isoform transcripts pertaining to 2,608 genes were identified as differentially expressed (false discovery rate-adjusted *P*-value <0.05) using Cufflinks v2.2.1. Among the top 25 differentially abundant transcripts using log fold change, 76% were overexpressed in the infected group relative to control cows including polo-like kinase 3 (PLK3), atypical chemokine receptor 3 (CXCR7), and nuclear factor of kappa light polypeptide gene enhancer in B-cells inhibitor delta (NFKBID). PLK3 participates in the tuberculosis response pathway, activating the nuclear factor- $\kappa$ B, which in turn stimulates transcription of the inducible nitric oxide synthase as well as pro-inflammatory cytokines. CXCR7 is an innate immune gene reported in leukocytes infected with *M. bovis*. Also, NFKBID may regulate the expression of cytokines by regulating nuclear factor- $\kappa$ B activity. Functional analysis of the differentially expressed genes using DAVID identified 14 enriched functional category clusters (enrichment score >3) including leukocyte mediated immunity, leukocyte activation and proliferation, cellular apoptosis, together with cytokines and chemokines production. The majority of the genes in these clusters were overexpressed in the infected group relative to control cows. These results offer insights on leukocyte transcriptome changes in response to *M. bovis* infection.

**Key Words:** transcriptome, tuberculosis, production

**T28 Evaluation of antimicrobial activity of chitosan microparticles in different matrices from dairy cows.** Zhengxin Ma<sup>\*1,2</sup>, Lin Teng<sup>1,2</sup>, Donghyeon Kim<sup>1</sup>, Klibs N. Galvão<sup>3</sup>, Corwin D. Nelson<sup>1</sup>, Adegbola T. Adesogan<sup>1</sup>, and K. Casey Jeong<sup>1,2</sup>, <sup>1</sup>*Department of Animal Sciences, University of Florida, Gainesville, FL*, <sup>2</sup>*Emerging Pathogens Institute, University of Florida, Gainesville, FL*, <sup>3</sup>*Department of Large Animal Clinical Sciences, University of Florida, Gainesville, FL.*

The objective of this study was to evaluate antimicrobial activity of chitosan microparticles (CM) in different matrices from dairy cows. CM was prepared by cross-linking of chitosan solution to enhance antimicrobial activity. Chitosan is biocompatible, natural antimicrobial biopolymer and considered Generally Recognized As Safe in many countries. Although CM shows strong antimicrobial activity in vitro, antimicrobial activity of CM in different animal matrices is not understood. We assayed antimicrobial activity of CM in cow uterine fluids (n = 3), milk samples (n = 5) and rumen fluids (n = 4) that were collected from metritic, subclinical mastitic, and fistulated dairy cows, respectively. To evaluate antimicrobial activity in the rumen fluids, we inoculated  $5 \times 10^4$  cfu/mL *E. coli* O157:H7 into the fluids, but we used

endogenous microorganisms for milk and uterine samples. Concentrations of CM ranging from 0.1% to 0.8% were tested in these matrices. Efficacy of antimicrobial activity of CM was measured by direct plating of treatments to measure the concentration of pathogens. Milk and uterine fluid samples were plated on LB agar, but the rumen fluids were plated on CT-SMAC agar after 0, 2, 4 and 6 h incubation to select *E. coli* O157:H7. The experiment in each fluid was conducted as a completely randomized design independently and in triplicate. Data were analyzed using the GLIMMIX procedure of SAS (SAS Institute Inc., Cary, NC). Treatment with CM effectively reduced the concentration of pathogens in all of the tested samples, even though the antimicrobial activity varied depending on matrices and CM concentrations. In milk, naturally occurring pathogens were completely eliminated within 4 h with 0.1% CM (*P* < 0.001). Regarding the cow uterine fluid, although the growth of naturally occurring pathogens was inhibited at 0.1% (*P* < 0.001), higher concentration (0.6%) of CM was required to eradicate pathogens (*P* < 0.001). Finally, 0.2% CM reduced *E. coli* O157:H7 by 2 log cfu/mL in spiked rumen samples (*P* < 0.001). These data demonstrate that a natural antimicrobial agent, CM, retains antimicrobial activity in different matrices from dairy cows.

**Key Words:** chitosan microparticle, antimicrobial activity

**T29 Targeted oxylipid analyses of milk obtained from periparturient dairy cows.** Jeffery C. Gandy\*, Vengai Mavangira, and Lorraine M. Sordillo, *Michigan State University, East Lansing, MI.*

Dysfunctional mammary gland inflammatory responses around the time of parturition contribute to immunopathology associated with mastitis. Oxylipids are bioactive lipid mediators that restore tissue homeostasis following injury by orchestrating the initiation and resolution of inflammation. The objective of this study was to develop a method to detect milk oxylipid concentrations and profile changes with respect to lactation stage. Milk samples were obtained from 11 multiparous cows at the time of calving and again at approximately 80 and 220 d in milk. Analytes in milk were obtained using solid-phase column extraction and then measured using liquid chromatography with tandem mass spectrophotometry (LC/MS/MS). The amount of oxylipids in each sample were quantified relative to internal standard abundance and calibrated against standard curves. Using this newly developed extraction and analytical methodology, a total of 31 of the 63 total oxylipids species targeted for analyses could be detected in milk samples within the limits of detection. Linoleic acid (2,419  $\mu$ M) was the predominant polyunsaturated fatty acid detected in milk samples regardless of lactation state followed by arachidonic (743  $\mu$ M), docosahexaenoic (89.9  $\mu$ M), and eicosapentaenoic (82.14  $\mu$ M) acids. The linoleic acid-derived oxylipids were the most abundant found in milk with significant increases (*P* < 0.05) in several hydroxyl products including 9,10-dihydroxy-octadecenoic acid during late (109.6 nM) when compared with milk samples obtained in early lactation (0.01 nM). Lipoxin A4 is an arachidonic acid-derived oxylipids with known anti-inflammatory functions that was higher (*P* < 0.05) in milk samples obtained at the time of calving (0.01 nM) when compared with late lactation samples (0.002 nM). These are the first observations of how oxylipids profiles change with respect to lactation stage and in response to changing concentration of fatty acid substrates in milk. A better understanding of how individual milk oxylipids affect mammary gland immunity may provide new insight of how to better control dysfunctional inflammatory responses during disease.

**Key Words:** oxylipid, eicosanoid, inflammation



**T30 Prevalence and antimicrobial resistance of mastitis pathogens in cattle dairy in a region of Colombia.** Maria del P. Sanchez\*, Norma P. Gutierrez, and Ivan J. Posada, *Universidad Cooperativa de Colombia Sede Ibagué, Ibagué, Tolima, Colombia.*

Bovine mastitis is the most common and major economic problem in the dairy industry worldwide, with a wide variety of microorganisms involved. Identification of mastitis pathogens is important for the selection of appropriate antimicrobial therapies. Antimicrobial agents are used to treat these infections caused by bacteria in particular but in recent years some bacteria have demonstrated full or partial resistance to different antibiotics. This phenomenon called antimicrobial resistance is a rising concern in both public and animal health and in this case, dairy science. The objective of the research was to determine the prevalence and antimicrobial resistance of pathogens involved in bovine mastitis in a region of Colombia. 1392 quarters of 348 cows from Anaime Colombia region were tested, using the California mastitis test (CMT). The positive samples to CMT were cultured for bacteriological isolation and were tested for antibiotic susceptibility and resistance by disk diffusion method and performed according to CLSI guidelines in Mueller-Hinton agar. Analysis of the results was made through descriptive statistics and prevalence ratio. 190 (54,6%) cows were considered mastitis free. One hundred fifty-eight cows (45,4%) were positive for CMT and bacteriological culture. Clinical mastitis in 20 (5,74%) cows characterized by milk abnormalities and subclinical mastitis in 138 (39,65%) were found. Coagulase-negative staphylococci (CoNS) was the main pathogenic agent with 44,79% of the total isolations, followed by *Staphylococcus aureus* (CoNP) 30,73% and *Streptococcus* spp. 22,39%, both *Klebsiella pneumoniae* and *E. coli* with 1,1%. CoNS and *S. aureus* exhibited the highest degree of resistance to penicillin G, *S. aureus* and *Streptococcus* spp. showed a high resistance to streptomycin and erythromycin respectively. The most prevalent organism was CoNS considered currently a worldwide emerging mastitis and it was mainly evidenced as a cause of subclinical mastitis with high degree of resistance to penicillin.

**Key Words:** clinical mastitis, subclinical mastitis, coagulase-negative staphylococci

**T31 Variation in clinical mastitis detection frequency and etiology among milkers.** Paulo Cesar Duque-Madrid<sup>1,3</sup>, Cedric Blanc<sup>2</sup>, and Alfonso Lago<sup>\*3</sup>, <sup>1</sup>*Universidad de Caldas, Manizales, Colombia,* <sup>2</sup>*GTV Dairies, Tipton, CA,* <sup>3</sup>*DairyExperts Inc., Tulare, CA.*

Early detection of clinical mastitis cases is considered key for the success of therapy and to maintain milk quality. Our objectives were to describe variation in 1) clinical mastitis detection frequency among milkers; and, 2) etiology of clinical mastitis among low and high detection milkers. Clinical mastitis records were from 712 cases from one California Central Valley large dairy herd where cows were milked twice a day in 2 separate parlors. Milking procedures included forestripping with observation of milk appearance on the parlor floor. When abnormal milk was observed a milk sample was collected, labeled with the milker ID, and cultured on-farm following the Minnesota Easy Culture system. Culture results were classified as no-growths, gram-negatives, gram-positives, and, mixed culture when gram-negatives and gram-positives were isolated. The total number of cases detected among 10 milkers ranged from 21 to 113 and the number of working hours from 817 to 987. Milkers 1 to 5 detected 2 to 7 cases per 100 h and were classified as low detection milkers. Conversely, milkers 6 to 10 detected 9 to 12 cases per 100 h and were classified as high detection milkers. The percentage

of samples from which bacteria were isolated ranged from 38% to 71% among milkers. However, milker's clinical mastitis detection rate did not affect the percentage of samples with bacterial growth ( $P = 0.93$ ) - it was 58% for the 2 clinical mastitis detection rate groups. Gram-positives were the bacteria most commonly isolated from both groups with 32% of the samples; gram-negative bacteria represented 21% and 16% of isolates in groups low and high, respectively; and, mixed cultures 5% and 10% in groups low and high, respectively. In conclusion, there was a large variation among milkers in clinical mastitis detection rate and in the percentage of samples with bacterial growth. However, neither the percentage of samples with bacterial growth nor the etiology were influenced by the level of detection. Therefore, it appears that high clinical mastitis detection rates can be achieved without increasing the percentage of cases without bacterial growth.

**Key Words:** clinical mastitis, mastitis detection, milkers

**T32 The effect of concentrate allocation strategy on the metabolic and immune function of high genetic merit dairy cows offered a grass silage based diet.** Mark W. Little<sup>\*1,2</sup>, Niamh O'Connell<sup>2</sup>, Jason Barley<sup>3</sup>, Michael D. Welsh<sup>3</sup>, and Conrad P. Ferris<sup>1</sup>, <sup>1</sup>*Agri-Food and Biosciences Institute, Hillsborough, UK,* <sup>2</sup>*School of Biological Sciences, Queens University Belfast, Belfast, UK,* <sup>3</sup>*Agri-Food and Biosciences Institute, Veterinary Sciences Division, Belfast, UK.*

The impact of concentrate allocation strategy on metabolic and immune function has received little attention. This 140-d study (commencing at calving) examined the effect of either a group fed (GF) or individual cow fed (ICF) concentrate allocation strategy on hematology, biochemistry, inflammatory and immune competence of Holstein Friesian dairy cows ( $n = 72$ ). With GF, cows were offered a total mixed ration comprising grass silage and concentrates (50: 50 DM ratio) plus 0.35 kg chopped straw/cow/day throughout the study. With ICF, cows were offered a basal ration consisting of grass silage, concentrates (6 kg/cow/day) and chopped straw (0.35 kg/cow/day), with this diet designed to meet the cows energy requirements for maintenance plus 24 kg milk/cow/day. With this treatment additional concentrates were offered 'feed-to-yield' via an out-of-parlor feeding system (0.45 kg concentrate/kg milk) based on each individual cow's milk yield during the previous week. Blood samples were obtained from the coccygeal vein or artery of each cow at wk 2, 4, 6, 8, 10, 12, 16 and 20 ( $\pm 3$  d) post-calving. Data were analyzed using a residual maximum likelihood (REML) mixed model analysis using GenStat (Version 16.2). Cows on the GF treatment had a higher mean hemoglobin ( $P = 0.009$ ) and packed cell volume ( $P = 0.018$ ), higher lymphocyte ( $P = 0.020$ ) percentage and lower neutrophil percentage ( $P = 0.018$ ) than cows on the ICF treatment. Cows on the GF treatment had lower serum NEFA concentrations ( $P = 0.028$ ) and tended to have a higher serum albumin concentration ( $P = 0.055$ ) than cows on the ICF treatment. There was no effect of concentrate allocation strategy on serum haptoglobin ( $P = 0.356$ ) or interferon gamma production of whole blood incubated with pokeweed mitogen ( $P = 0.115$ ). Allocating concentrates on a group basis resulted in small physiological improvements to metabolic function, but had no effect on immunological function, compared with offering the same amount of concentrates on an individual cow basis.

**Key Words:** concentrate allocation, metabolic function, immune function

**T33 Evaluation of experimental novel germicide postmilking teat dips and a commercial iodine barrier postmilking teat dip on teat end and teat skin health and integrity.** Rae Sires, Kia Knutson, and Leo L. Timms\*, *Iowa State University, Ames, IA, 50011.*

Study objective was to evaluate prototype novel germicide post milking teat dips versus a control commercial iodine barrier post milking teat dip on overall teat end (TE) and teat skin (TS) health and integrity. Control dip was Bovi-Kote (1% iodine with 10% multiple emollient system (low drip): Boumatic, Inc.) and treatment dips were a novel germicide dip with 10% citrate, parabens, and varying methylene blue (MB) (0.1, 0.25, and 0.4%) and emollient (5, 10, and 11%) concentrations (Zurex Pharma Inc.). Trial used a split pen and split udder design. Twenty-four early- mid lactation Jersey cows in a single pen were used with 12 cows having a blue leg band (BLB group) to designate them as trial group 1 while the other 12 had no leg band (NLB group). Left teats of all 24 cows were post dipped with Bovi-Kote (control) while right teats were initially dipped with a 0.1% methylene blue dip (1/29) but differed in emollient % (5 v 11%). Dips changed to .25% MB (2/10) to enhance coloration. NLB teats switched to a 0.4% MB with 11% emollients (2/19), while BLB teats switched to a higher emollient (10 v 5%) no drip teat dip (2/20). Trial was 5 weeks in duration (Jan–Feb). TS and TE scoring were performed 1st 4 trial days, then 3×/week using Goldberg and Timms methods (1–5 scoring for both, 0.5 increments for TE scores). Mixed procedure of SAS with repeated measures (quarter within cow as a repeated measure) were used to analyze TS and TE data and GENMOD procedures of SAS with repeated measures was used to analyze % rough teat ends or dry/chapped teat skin, with  $P < 0.05$  considered significant. Results of experimental vs commercial control dip were similar across groups so data were combined. No significant differences in TS score were seen (99+% excellent). TE scores and % rough teats were not significantly different during weeks. One and 4, but were significantly higher [1.6, 13% (Exp) v 2.1, 27% (Con)  $P < 0.01$ ] and compromised in control teats during wks. 2–3, corresponding to temperature change (30°F (–1°C) to –4°F (–20°C)). Experimental dip showed significantly better TE during colder weather and temperature changes.

**Key Words:** postmilking teat dip, germicide, teat health

**T34 Study of the activity of soluble and nanostructured IFN $\gamma$  and metalloproteinases as a new tool for the optimization of the dry-off of dairy cows.** Francesc Fabregas<sup>1</sup>, Olivia Cano<sup>2,3</sup>, Sandra Genís<sup>\*1</sup>, Silvia Parés<sup>1</sup>, Joaquim Seras-Franzoso<sup>2,3</sup>, Alex Bach<sup>1,4</sup>, Antonio Villaverde<sup>2,3</sup>, Elena Garcia-Fruitos<sup>2,3</sup>, and Anna Aris<sup>1</sup>, <sup>1</sup>*Department of Ruminant Production, Institute of Research in Agriculture and Technology, Caldes de Montbui, Spain,* <sup>2</sup>*Department of Genetics and Microbiology, Institute of Biotechnology and Biomedics, Universitat Autònoma de Barcelona, Bellaterra, Spain,* <sup>3</sup>*CIBER de Bioingeniería, Biomateriales y Nanomedicina, Bellaterra, Spain,* <sup>4</sup>*ICREA, Barcelona, Spain.*

The use of bovine metalloproteinases and IFN $\gamma$  to hasten mammary gland involution and the stimulation of immune system response has been proposed to accelerate the dry off period and diminish the risk of intramammary infections. The protein nanoparticles technology has shown to be an economically viable production strategy of recombinant proteins. The objective of this work was to study the activity of bovine metalloproteinase 2 and 9 (MMP-2, MMP-9) and IFN $\gamma$  produced in *Lactococcus lactis* either as nanoparticles (Lu et al. Mol. Biotechnol. 2013 54:170–176) or soluble recombinant proteins. The activity of IFN $\gamma$  was tested using a macrophage activation test along with an assay of induction of nuclear factor kappa- $\beta$  (NF- $\kappa$ B) expression in mammary

epithelial cells. Monocytes were isolated from cow blood, differentiated to macrophages, and treated with IFN $\gamma$  for 24 h at 37°C, 5% CO<sub>2</sub> while primary culture of mammary gland epithelial cells were treated with IFN $\gamma$  for 4 h at 37°C, 5% CO<sub>2</sub>. Cell samples were taken to analyze the expression of NF- $\kappa$ B by qPCR. Activity of MMP-2 and MMP-9 soluble or as nanoparticles was tested by both zymography and ex vivo. For the ex vivo assay, mammary gland explants were incubated with MMP for 7 h at 37°C and 5% CO<sub>2</sub>. Then, apoptotic and proliferation gene expression was assessed by qPCR (PCNA, SPC25, CASP3 and BNIP). The sensitivity of NF- $\kappa$ B assay was greater than the macrophage activity test (detection limit of 19.6  $\mu$ g and 140  $\mu$ g IFN $\gamma$ , respectively;  $P < 0.05$ ). Soluble IFN $\gamma$  upregulated ( $P < 0.05$ ) NF- $\kappa$ B, whereas similar amounts of IFN $\gamma$  nanoparticles had no significant effect. Metalloproteinase activity was detected for MMP-2 and MMP-9 in both forms by zymography. As expected, the soluble form was more active than the nanostructured form ( $P < 0.05$ ). Only soluble MMP9 activity was detected ex vivo, stimulating ( $P < 0.001$ ) the expression of SPC25 and PCNA proliferative genes. In conclusion, the bovine recombinant MMP and IFN $\gamma$  proteins elicited the expected biological response and thus they can be used in further in vivo dry-off studies in dairy cows.

**Key Words:** dry period, nanoparticle, metalloproteinase

**T35 Metabolic parameters of cows with different status for bovine leukemia.** Irina V. Vinogradova<sup>\*1</sup>, Elena A. Gladyr<sup>1</sup>, Ludmila A. Ivanova<sup>2</sup>, Alexandr S. Kramarenko<sup>1</sup>, Igor V. Gusev<sup>1</sup>, Roman V. Rykov<sup>1</sup>, Michael I. Guljukin<sup>2</sup>, and Natalia A. Zinovieva<sup>1</sup>, <sup>1</sup>*L.K. Ernst Institute of Animal Husbandry, Dubrovitsy, Moscow, Russia,* <sup>2</sup>*Y.R. Kovalenko Institute of Experimental Veterinary Medicine, Moscow, Russia.*

Increased susceptibility of high-productive dairy cows to infectious diseases is associated with metabolic stress which is developed on the background of a negative energy balance. The objective of our study was to determine the differences in metabolic parameters and blood leukocyte profiles in cows infected by bovine leukemia virus (BLV+) compared with non-infected cows (BLV–). The blood samples were collected from 150 cows of the Russian Black-and-White breed. The cows were housed at identical environmental conditions in one free-stall facility and were fed by the same diets. BLV status was determined by immune diffusion reaction and confirmed by enzyme immune assay. The blood serum concentrations of carbohydrate, protein and lipid metabolites were measured and the blood leukocyte profile was evaluated. The experimental data were analyzed using logistic regression analysis of binary data, (<http://www.statistica-help.ru/node/28>) ( $p_1$ ) and linear model ANOVA ( $p_2$ ), where the BLV status and correlation coefficient were taken as a random factor. Student *t*-test was performed to evaluate the statistical significance of differences between the groups. The serum concentrations of albumin, creatinine and ALT did not differ significantly between the groups, but the concentrations of the total protein (+6.89 g/L,  $p_1 \leq 0.0001$ ,  $p_2 \leq 0.0141$ ), globulin (+6.95 g/L,  $p_1 \leq 0.0002$ ,  $p_2 \leq 0.0483$ ), urea (+1.02 mM/L,  $p_1 \leq 0.0020$ ,  $p_2 \leq 0.0006$ ) and bilirubin (+2.01 mM/L,  $p_1 \leq 0.0162$ ,  $p_2 \leq 0.0016$ ) were significantly higher among BLV+ cows compared with non-infected cows, but the concentrations of glucose (–0.50 mM/L,  $p_1 \leq 0.0007$ ,  $p_2 \leq 0.0940$ ), triglycerides (–0.04 mM/L,  $p_1 \leq 0.0384$ ,  $p_2 \leq 0.0065$ ), alkaline phosphatase (–63.1 IE/L,  $p_1 \leq 0.0384$ ,  $p_2 \leq 0.0065$ ) and AST enzyme (–1.56 IE/L,  $p_2 \leq 0.0713$ ) were significantly decreased. According to clinical indicators of blood (WBC, RBC, HGB, HCT, MCV, MCH, MCHC, RDW) significant differences between the groups were not found. The data obtained confirm the assessment that bovine leukemia has effect on the biochemical parameters of carbohydrate, lipid, protein metabolism.

The research was supported by the Russian Science Foundation, project number 14–16–00046.

**Key Words:** dairy cattle, bovine leukemia virus, biochemical paramete

**T36 High forage diet alters feeding behavior, health, and milk production in fresh Holstein dairy cows.** Juliana M. Huzzey<sup>\*1,2</sup>, Hesam A. Seifi<sup>1,3</sup>, Muhammad A. Khan<sup>1,4</sup>, Marina A. G. von Keyserlingk<sup>1</sup>, and Daniel M. Weary<sup>1</sup>, <sup>1</sup>*Animal Welfare Program, University of British Columbia, Vancouver, BC, Canada*, <sup>2</sup>*Animal Science Department, California Polytechnic State University, San Luis Obispo, CA*, <sup>3</sup>*Department of Clinical Sciences, School of Veterinary Medicine, Ferdowsi University of Mashhad, Iran*, <sup>4</sup>*AgResearch, Grasslands Research Centre, Palmerston North, New Zealand*.

High-forage diets for close-up dairy cows are gaining popularity and are increasingly perceived to provide health and production advantages. After calving cows are typically transitioned to a high-energy diet but it is not clear how this transition affects their health and behavior. Our objective was to test the effect of maintaining cows on a high-forage diet for 3 wk after calving, versus switching to the conventional high-energy diet. Sixty-eight Holstein dairy cows (21 primiparous and 47 multiparous cows) were all fed the same high-forage prepartum diet. After calving, cows were moved to a postpartum pen for 21 d and randomly assigned to feed bins according to their respective treatments (Straw TMR: STMR vs. Control TMR: CTMR). After 21 d, all cows were moved to a new pen and fed CTMR. An electronic feeding system continuously monitored individual feeding behavior and intake. Rumination time was recorded using data loggers. Blood was sampled twice per week and tested using the  $\beta$ -hydroxybutyrate (BHBA) Precision Xtra kit. Cows were considered subclinically ketotic (SCK) if BHBA was  $\geq 1.0$  mmol/L in the first wk postpartum or  $\geq 1.2$  mmol/L from d 7 to 35 postpartum. Milk yield was recorded daily. Data were analyzed using the Mixed and Genmod procedures of SAS; cow was treated as a random effect and period (wk or d) as the repeated measure. Fresh cows fed STMR had lower DMI, fewer meals, spent more time at the feed bunk and had a slower feeding rate ( $P \leq 0.02$ ). Rumination time of cows fed STMR was higher than cows fed CTMR ( $P = 0.04$ ). Cows fed STMR had higher BHBA concentrations during wk 3 ( $P = 0.03$ ) and were 4.9 times more likely than the CTMR group to develop SCK by wk 3 (odds ratio = 4.9, 95% confidence interval = 0.95 - 25.47,  $P = 0.06$ ). Over the first 35 d of lactation, cows fed CTMR produced more milk than those fed STMR ( $33.3 \pm 1.0$  vs.  $30.7 \pm 1.0$  kg/d;  $P = 0.03$ ). These results indicate that maintaining cows on a high-forage diet after calving can have detrimental effects on energy balance and cow health.

**Key Words:** transition cow, nutrition, welfare

**T37 Prepartum rumination patterns in dairy cows that develop health disorders in the early postpartum period.** Matias L. Stangaferro<sup>\*</sup>, Robert Wijma, Miranda M. Medrano, Mohammed A. Al Abri, and Julio O. Giordano, *Department of Animal Science, Cornell University, Ithaca, NY*.

The objective of this study was to compare prepartum rumination patterns of lactating dairy cows that developed health disorders (HD) versus cows that did not develop HD (NoHD) up to 30 DIM. A total of 559 Holstein cows (222 primiparous and 337 multiparous) were fitted with a rumination and activity monitoring tag (HR Tags, SCR Dairy) from -28 to 30 DIM. Rumination time (RT) was recorded in minutes per 2 h intervals and summarized in 24 h periods. After calving, farm

personnel examined cows daily for signs of clinical disease following farm standard operating procedures. From 1 to 10 DIM, personnel evaluated: appetite, rectal temperature, ketone bodies in urine, rumen fill and movements, retained placenta, vaginal discharge, daily milk weights, and milk conductivity. Milk culture was performed for all mastitis cases. Number of cows suffering HD was displaced abomasum (DA) 17, ketosis (KET) 16, indigestion (IND) 5, metritis (MET) 171, retained placenta (RP) 49, and mastitis (MAST) 36. Rumination time for the 7 d preceding calving was evaluated for cows with HD or NoHD within 30 DIM. Also, data were evaluated for all metabolic and digestive diseases (MDZ = DA, KET, IND) combined and RP, MET, and MAST individually. Data were analyzed by ANOVA with repeated measurements using PROC MIXED of SAS. For all HD combined, RT was less ( $P = 0.02$ ) for cows with HD ( $439.1 \pm 2.8$  min) than for cows with NoHD ( $455.9 \pm 2.0$  min). Rumination time was lowest ( $P < 0.01$ ) on the day of calving ( $391.3 \pm 4.3$  min) than the 6 d preceding calving (mean range =  $457.5 \pm 4.0 - 462.6 \pm 4.1$  min) for all cows. Cows with MDZ had reduced RT ( $P < 0.01$ ) than NoHD cows ( $409.6 \pm 7.3$  vs  $455.9 \pm 2.0$  min). Cows with RP had similar RT ( $P = 0.25$ ) than NoHD cows ( $440.2 \pm 5.0$  vs  $455.9 \pm 2.0$  min). Cows with MET tended to have reduced RT ( $P = 0.05$ ) than NoHD cows ( $440.5 \pm 3.1$  vs  $455.9 \pm 2.0$  min). Cows with MAST had similar RT ( $P = 0.82$ ) than NoHD cows ( $452.4 \pm 6.1$  vs  $455.9 \pm 2.0$  min). We conclude that starting 7 d prepartum, rumination patterns are altered in cows that suffer health disorders within 30 DIM. Rumination time is reduced in cows that suffer MDZ (DA, KET, IND) and MET but not in cows with RP and MAST

**Key Words:** rumination, dairy cow, disease

**T38 Plasma phosphatidylcholine and lysophosphatidylcholine profiling of heat-stressed lactating dairy cows.** He Tian<sup>1</sup>, Jianbo Cheng<sup>2</sup>, Yangdong Zhang<sup>1</sup>, Nan Zheng<sup>\*1</sup>, and Jiaqi Wang<sup>1</sup>, <sup>1</sup>*Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China*, <sup>2</sup>*College of Animal Science and Technology, Anhui Agricultural University, Hefei, China*.

Heat stress (HS) is a global challenge for the dairy industry; however, its abnormal physiological alterations still remain elusive. Here, a LC-MS-based study on lysophosphatidylcholine (lysoPC) and phosphatidylcholine (PC) in plasma of HS dairy cows was performed to explore HS-induced metabolic alterations. LysoPCs or PCs exist in 2 forms/isomers, with the fatty acyl groups at positions 1 (sn-1) or 2 (sn-2) on the glycerol backbone. First experiment, the LC-MS-based method of discriminating isomers of lysoPCs or PCs was constructed. The isomeric lysoPCs or PCs can be differentiated by chromatographic separations and relative intensities of their characteristic fragments. Second experiment, the enrolled 2 groups of Holstein cows were in second parity, mid-lactation stage, had similar milk yield (both around 40 kg/day), and fed the same diet. The HS-free group consisted of 22 cows, with plasma samples obtained in spring season, after temperature-humidity index (THI) was around 50–55 for 1 mo. The HS group consisted of another 22 cows, with samples obtained in summer season, after THI gradually increased from 68 to 80 over 1 mo. LC-MS in combination with multivariate analyses on plasma profiling of HS-free and HS groups was conducted to discover HS-induced discriminating metabolites ( $P < 0.05$ ), from which isomeric lysoPCs or PCs were identified by the constructed method in first experiment. The results revealed that concentrations of 14 lysoPCs and 17 PCs were significantly up- or downregulated in HS compared with in HS-free group, indicating HS-induced alterations of lysoPCs and PCs. Of these 31 discriminating metabolites, concentrations of sn-1 lysoPC 14:0, sn-2 lysoPC 15:0, sn-1 lysoPC 18:0, and sn-2 lysoPC 22:4 were upregulated in HS group ( $P < 0.05$ ), concentrations



of sn-1 PC 14:0/18:3, sn-1 PC 14:0/18:2, sn-2 PC 15:0/17:1, sn-1 PC 14:0/20:4, sn-1 PC 18:0/15:1, sn-1 PC 18:0/15:0, and sn-2 PC 15:0/20:4, were downregulated in HS group ( $P < 0.05$ ); whereas, concentrations of their respective isomers remained unchanged, suggesting isomers of these lysoPCs and PCs have different biological functions played in lactating dairy cows under HS states.

**Key Words:** lactating dairy cow, heat stress, phosphatidylcholine and lysophosphatidylcholine profiling

**T39 Assessment of an application to collect calving-related events in dairy herds.** A. A. Barragan\*, J. D. Workman, S. Bas, K. L. Proudfoot, and G. M. Schuenemann, *The Ohio State University, Columbus, OH.*

Calving-related losses (survival, health, and productivity of cows and calves) are known challenges for the dairy industry worldwide, and poor management practices can increase the risk of these losses. It is common to observe large variation within and between herds on the quality and quantity of calving-related records. The objective of the present study was to assess an application (APP) for hand-held smart devices to capture defined calving-related events in dairy herds. Calving events collected by personnel ( $n = 23$ ) from 6 large dairy operations (range: 900–5,000 cows) were recorded. Calving personnel received the same training on calving management and use of the APP before collecting events. Immediately after training, personnel recorded calving events ( $n = 448$ ) using the APP for 7 d. Personnel satisfaction with the APP and knowledge gained from the training were assessed. Calving personnel reported that the information provided during the training was relevant (agree = 14.3% and strongly agree = 85.7%) and of great immediate use (agree = 33.3% and strongly agree = 66.7%). The presented materials and demonstrations substantially increased ( $P < 0.05$ ) the knowledge level of the participants by 23.7 percentage points from pre- to post-test scores. The APP accurately captured (100%) calving events and integrated multiple metrics with personnel performance (accounting for the effect of shift change) such as the dam (e.g., date-time of calving), colostrum (e.g., timing, quality, and quantity) and newborn calf (e.g., presentation, vigor). The follow-up assessment with participants revealed that the APP was easy to use (91.3%) and that they would like to keep using it (100%). Dairywomen, consultants, and veterinarians often trouble-shoot calving related losses within-herd; however, the lack of meaningful records makes it difficult to implement effective corrective measures. It is important to note that the timing and accuracy of data is always dependent on the willingness of the individual recording the information. However, this effective and easy-to-use tool requires minimal personnel training. These findings showed that decision-makers can monitor calving events and losses (magnitude and time) at the farm level accounting for the effect of management.

**Key Words:** dairy cattle, calving, application

**T40 Difuctose anhydride III supplementation promotes passive calcium absorption in the small intestine immediately after calving in dairy cows.** Makoto Teramura\*<sup>1,2</sup>, Syaw Wynn<sup>1</sup>, Maimaiti Reshalaitihan<sup>3</sup>, Wakana Kyuuno<sup>2</sup>, Tadashi Sato<sup>2</sup>, Masayuki Ohtani<sup>2</sup>, Chiho Kawashima<sup>3</sup>, and Masaaki Hanada<sup>3</sup>, <sup>1</sup>United Graduate School of Agricultural Sciences, Iwate University, Morioka, Iwate, Japan, <sup>2</sup>Nippon Beet Sugar Manufacturing Co., Ltd., Obihiro, Hokkaido, Japan, <sup>3</sup>Obihiro University of Agriculture and Veterinary Medicine, Obihiro, Hokkaido, Japan.

Difuctose anhydride (DFA) III is an indigestible oligosaccharide. It has been shown to reach the duodenum without being degraded by ruminal bacteria and to promote the intestinal calcium (Ca) absorption via paracellular pathway in cattle, but these studies did not mention DFA III's effectiveness during early postpartum (pp) period. The objective of this study was to determine whether DFA III could promote intestinal paracellular Ca absorption during early pp period. Seventy-four multiparous Holstein cows were separated into the DFA and control groups based on their parity ( $3.7 \pm 0.3$  and  $3.7 \pm 0.3$ , respectively). The DFA group was fed 40 g/d of DFA III from -14 d to 6 d relative to calving and the control group was not fed DFA III. At calving (0 h pp), the serum Ca declined below 9 mg/dL in both groups. However, the serum Ca was significantly greater in the DFA group than in the control group at 6 h, 12 h, 24 h, and 48 h pp ( $P < 0.05$ ) and the time interval for serum Ca recovery to  $\geq 9$  mg/dL during the pp period was shorter in the high-parity cows of the DFA group than in those of the control group. The serum parathyroid hormone (PTH) increased at 0 h pp in both groups and was significantly greater in the control group than in the DFA group at 12 h and 24 h pp ( $P < 0.05$ ). The serum 1,25-dihydroxyvitamin D [ $1,25-(OH)_2D$ ] increased at 0 h and 12 h pp in both groups and was significantly greater in the control group than in the DFA group at 72 h pp ( $P < 0.01$ ). The serum bone-resorption marker cross-linked N-telopeptide of type I collagen (NTX) was not significantly different between the groups during peripartum period, and the serum NTX in all of the cows was lesser at 0 h, 6 h, 12 h, 24 h, 48 h, and 72 h pp than at -21 d, 4 d, and 5 d relative to calving ( $P < 0.05$ ). These results suggest that the DFA group had rapid recovery of serum Ca, although bone resorption was restrained and active intestinal Ca absorption via transcellular pathway was impaired. In conclusion, DFA III promotes intestinal passive Ca absorption via paracellular pathway during early pp period, which is unaffected by aging.

**Key Words:** hypocalcemia, dairy cow, DFA III

**T41 Supplementation with *Bacillus pumilus* 8G-134 enhances expression of T cell markers in dairy cows during early lactation.** Megan Duersteler\*<sup>1</sup>, Mike Brouk<sup>2</sup>, and Elizabeth Galbraith<sup>1</sup>, <sup>1</sup>DuPont, Waukesha, WI, <sup>2</sup>Kansas State University, Manhattan, KS.

Infectious disease during transition and early lactation periods has the potential to decrease production performance and increase morbidity and mortality in dairy cows. Enhanced adaptive immunity through nutritional supplementation may counter immunosuppression that occurs naturally during the periparturient and early lactation periods. The objective of this study was to evaluate the effect of a direct-fed microbial, *Bacillus pumilus* 8G-134, on systemic immunity in dairy cows through flow cytometric analysis of peripheral blood mononuclear cells. Thirty cows were fed a control diet or a diet including  $5 \times 10^9$  cfu/head/day *B. pumilus* 8G-134 from 3 weeks prepartum to 22 weeks postpartum. Peripheral blood mononuclear cells were collected at 60 d in milk and analyzed by flow cytometry. Leukocytes and lymphocytes were gated based on forward and side scatter, and relevant cell surface markers were examined in each population. *B. pumilus* 8G-134 supplementation increased ( $P < 0.05$ ) expression of T cell surface markers CD4, CD8, CD62L, CD25, and CD45RO, but did not affect proportions of T cell subsets. There was no effect on expression of cell surface markers on innate immune cells (Mono, CD14, CD45, CD172a, and CD62L) or B cells (Bcell, and CD21). Feeding *B. pumilus* 8G-134 induced expression of T cell surface markers that are naturally suppressed during parturition. By targeting the adaptive immune system, *B. pumilus* 8G-134 was able to enhance T cell marker expression without activating a systemic

immune response that would adversely affect energy balance. This may help dairy cows recover faster from periparturient immunosuppression.

**Key Words:** dairy, direct-fed microbial, immunology

**T42 A stochastic estimate of the economic impact of oral calcium supplementation in postparturient dairy cows.** J. A. A. McArt<sup>1</sup> and G. R. Oetzel\*<sup>2</sup>, <sup>1</sup>*Department of Population Medicine and Diagnostic Services, Cornell University, Ithaca, NY*, <sup>2</sup>*Department of Medical Sciences, University of Wisconsin-Madison, Madison, WI*.

Stochastic models were developed to estimate the economic impact in the first 30 d in milk of oral calcium supplementation (Bovikalc, Boehringer Ingelheim Vetmedica Inc., St. Joseph, MO) to multiparous postparturient dairy cows using 4 different strategies: 1) supplementation of cows with a high previous lactation mature equivalent milk yield, 2) supplementation of lame cows, 3) supplementation of cows that either have a high previous lactation mature equivalent milk yield or are lame, and 4) supplementation of all cows. Data from current literature was used to model input variables associated with the costs and risks related to milk production, postparturient disease, and culling. The mean net herd impact per 1,000 calvings for each of the 4 supplementation strategies was \$4,425, \$5,812, \$8,313, and \$3,065 respectively. Return on investment was 6.5 to 1 for supplementation of lame cows, 1.8 to 1 for high milk yield and lame cows, 1.1 to 1 for high milk yield cows, and 0.3 to 1 for supplementation of all multiparous postpartum cows. A herd's average milk yield at first test had the highest influence on the net impact of oral calcium supplementation to all multiparous cows and accounted for 30% of the variation in net herd financial impact of oral calcium administration. Other variables that explained at least 5% of the variation in financial impact included decreased risk of health events in lame cows, herd prevalence of lameness, and milk price. Whereas supplementation of all postpartum multiparous cows returned a positive net herd impact approximately 80% of the time, if a herd was willing to devote time to mature-equivalent milk yield calculations and locomotion scoring, supplementation of this subpopulation of postpartum cows with oral calcium was estimated to have a positive economic impact in all iterations. Depending on the supplementation strategy chosen and baseline milk yield and immediate postpartum lameness prevalence in a herd, a herd with 1,000 calvings per year can expect to see an average net impact ranging from approximately \$3,000 to \$8,000 after postpartum supplementation of oral calcium in multiparous animals.

**Key Words:** dairy cow, oral calcium, economic impact

**T43 Mastitic cows management practices on California dairies.** Pau Pallarés\*<sup>1</sup>, Arnau Espadamala<sup>1</sup>, Alfonso Lago<sup>2</sup>, and Noelia Silva-del-Río<sup>1</sup>, <sup>1</sup>*UC Davis School of Veterinary Medicine, VMTRC, Tulare, CA*, <sup>2</sup>*DairyExperts, Tulare, CA*.

Minimizing the risk of antibiotic resistant organisms and antibiotic residues in dairy and dairy beef products is a topic of nationwide interest. However, to design an effective outreach program on judicious use of antibiotics, it is imperative to describe the actual practices on dairies. Thus, our objective was to summarize management and treatment practices for mastitic cows on 15 California dairies as a first step in that outreach effort. Herds ranged in size from 600 to 9,500 cows. Data were collected by 2 bilingual veterinarians during the milking of mastitic cows, based on cow-side observation and responses from dairy employees. Cows identified with mastitis were kept in the same pen (n = 1) or moved to the hospital pen (n = 14). The hospital pen housed mastitic cows (n = 5); mastitic, sick and lame cows (n = 3); or mastitic,

sick, lame and fresh cows (n = 6). Four of the 10 herds housing mastitic and non-mastitic cows in the hospital pen did not clean milking units in between cows. Five dairies were fitted with a milking parlor just for fresh and mastitic cows. Dairies with a single parlor milked the mastitic cow pen last. Mastitic cows were identified during regular milking based on quarter inflammation (n = 2) and quarter inflammation and milk appearance (n = 13). Two dairies used California Mastitis Test (CMT) for confirmation. Cows with abnormal milk were sampled as soon as they were identified (n = 2) or after they were moved to the hospital pen (n = 8). Milk culture results were used for selective treatment of mastitis (n = 4). The first treatment options were intramammary cephalosporin [1 d (n = 1), 3 d (n = 2), 4 d (n = 1) or 5 d (n = 1)], ceftiofur [2 d (n = 1), 3 d (n = 4) or 4 d (n = 3)], hetacillin [3 d (n = 1)] or intravenous oxytetracycline and sulfamides [5 d (n = 1)]. If after treatment completion cows showed abnormal milk (n = 13) or positive CMT (n = 2) the antibiotic therapy was continued with the same (n = 10) or a different (n = 5) drug. Milk culture was used on some dairies as a treatment decision tool. Although the antibiotic drug of choice was similar across dairies, the length of treatment varied widely across dairies.

**Key Words:** dairy cattle, mastitis, intramammary treatment

**T44 Concentrations of saturated fatty acids in whole raw milk of dairy cows under different management systems and country of origin: A meta-analytical study.** Grzegorz Zwierchowski\* and Burim Ametaj, *Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB, Canada*.

The objective of this meta-analytical study was to investigate concentrations of milk saturated fatty acids (SFA) including C4, C6, C8, C10, C12, C14, C15, C16, C17, and C18 in whole raw dairy cow milk from different production systems (conventional [CVN] vs organic [ORG]) and several countries of origin. Data from 18 studies and 13 countries were used to create a data set, which was used for statistical analyses. Data are reported in milk fat percent. The average concentrations of C15 and C17 in the raw milk from ORG system were greater (1.49 vs 1.12 and 1.17 vs 0.61%, respectively) than those from CVN farms ( $P < 0.05$ ). However, no differences were found with regards to concentrations of C4 (3.21 vs 3.17%), C14 (10.70 vs 10.13%), and C16 (28.69 vs 26.82%), in the raw milk between ORG and CNV systems. Also no differences were obtained in the concentration of C6, C8, C10, C12, and C18. On the other hand, country of origin affected concentrations of SFA ( $P < 0.05$ ). For example, raw milk from Ireland had lower amounts of C4 compared with milk from Belgium (1.33 vs 6.26%;  $P < 0.05$ ). Additionally, the greatest concentrations of C6, C8, C10, and C14 were reported in Norway (3.11, 1.95, 4.43, and 13.88%, respectively), whereas the lowest levels of those fatty acids were reported in Germany (0.82%), Switzerland (0.95%), Mexico (0.91%), and United Kingdom (7.13%), respectively ( $P < 0.05$ ). Average C12 levels varied from 1.97% (Mexico) to 4.83% (Netherlands) ( $P < 0.05$ ). Likewise, milk originating from US had lower means of C15 (0.63 vs 2.53%) and C17 (0.33 vs 1.61%) compared with milk from Germany ( $P < 0.05$ ). Additionally milk from Germany had greater C16 levels compared with milk from UK (15.84 vs 41.31%) ( $P < 0.05$ ). Means of C18 varied from 5.49 to 13.12% (China,  $P < 0.05$ ). In conclusion, data from this meta-analytical study indicated that ORG farms were characterized by greater concentrations of some of the SFA compared with CVN system of management. This study also showed high variability in concentrations of SFA in the raw milk with regards to the country of origin.

**Key Words:** whole raw milk, saturated fatty acid, meta-analysis

**T45 Concentrations of unsaturated fatty acids in the whole raw milk of dairy cows under different management systems and country of origin: A meta-analytical study.** Grzegorz Zwi-  
erzchowski\* and Burim Ametaj, *Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, Alberta, Canada.*

The objective of this meta-analytical study was to investigate concentrations of milk unsaturated fatty acids (UFA), including C14:1, C18:1, C18:2, CLA, C18:3, C20:1, C20:2, C20:4, docosapentaenoic (DPA), and eicosapentaenoic acid (EPA) in whole raw bovine milk from different production systems (conventional (CNV) vs organic (ORG)) and countries of origin. Data from 18 studies and 13 countries were used to create a data set, which was used for statistical analyses. Data are reported in milk fat percent. The average concentrations of C18:2 and C18:3 in the raw milk from CNV system were greater (1.94, and 0.60 respectively) than those coming from ORG farms (1.90, and 0.54%, respectively), but difference reached no significance. In addition, there were greater concentrations of C20:4 (0.14%) in the raw milk from CNV vs ORG system (0.11%) ( $P < 0.05$ ). Lower ( $P < 0.05$ ) amounts of C14:1 (0.76%), C20:1 (0.17%), C20:2 (0.03%), and EPA (0.04%) were reported for milk originating from ORG vs CNV (0.97, 0.26, 0.13, and 0.10%, respectively). In addition, no differences with regards to C18:1 ( $P = 0.90$ ), CLA ( $P = 0.31$ ), and DPA ( $P = 0.07$ ), were reported for milk from the ORG vs CNV system. Country of origin had significant effect on concentrations of UFA ( $P < 0.05$ ). The lowermost concentrations of C18:1, C18:2, and C20:4 were reported in Germany ( $P < 0.04\%$ ), whereas the greatest levels of those UFA were reported in Belgium and US ( $P < 0.05$ ). Milk from Poland had lower amounts of C14:1 and C20:1 compared with milk from Germany and Norway, respectively ( $P < 0.05$ ). Average DPA levels varied from 0.01% (Norway) to 0.19% (Switzerland). Moreover, milk from UK had greater means of C18:3 and CLA compared with milk from Norway and Netherlands, respectively ( $P < 0.05$ ). Norwegian milk had greater EPA levels compared with milk from China ( $P < 0.05$ ). In conclusion, data from this meta-analytical study indicated that ORG farms were characterized by greater concentrations of n-3 FA (CLA, DPA, EPA) in the milk compared with milk from the CNV system. This study also showed high variability in concentrations of UFA in raw milk with regards to the country of origin.

**Key Words:** whole raw milk, unsaturated fatty acid, meta-analysis

**T46 Owner and veterinarian involvement on fresh cow health management on California dairies.** Pau Pallarés\*<sup>1</sup>, Arnau Espadamala<sup>1</sup>, Alfonso Lago<sup>2</sup>, and Noelia Silva-del-Río<sup>1</sup>, <sup>1</sup>*UC Davis School of Veterinary Medicine, VMTRC, Tulare, CA, 2**DairyExperts, Tulare, CA.*

Minimizing the risk of antibiotic resistant organisms and antibiotic residues in dairy and dairy beef products is a topic of nationwide interest. However, to design an effective outreach program on judicious use of antibiotics, it is imperative to describe the actual practices on dairies. Thus, as a first step in that outreach effort, our objective was to evaluate the involvement of owners and veterinarians on fresh cow (FC) evaluations and to identify who FC evaluators requested advice from. Herds ( $n = 15$ ) ranged in size from 600 to 9,500 cows. Cow-side observation and responses from FC evaluators were collected by 2 bilingual veterinarians during FC evaluations. Dairy owner conducted FC evaluations ( $n = 2$ ), supervised FC evaluations ( $n = 4$ ) or delegated FC evaluations ( $n = 9$ ). The dairy veterinarian identified and treated cows once a week ( $n = 2$ ) or every other week ( $n = 2$ ), provided occasional advice on treatments ( $n = 7$ ) or they were not involved on FC evaluations ( $n = 4$ ). Dairies

with minimum veterinarian involvement relied on pharmaceutical sales representatives (PSR) and other consultants for advice on herd health. During FC evaluations a single individual identified sick cows and decided on treatments [dairy owner ( $n = 2$ ), herdsman ( $n = 5$ ), herdsman assistant ( $n = 8$ )]. Evaluators had  $< 1$  yr ( $n = 2$ ), 1–5 yr ( $n = 3$ ),  $> 5$  yr ( $n = 10$ ) of experience. New knowledge was acquired based on formal training [ $< 1$  year ( $n = 4$ ) or 1–5 yr ( $n = 1$ ) ago], communication with other dairies workers ( $n = 7$ ), working with more experienced employees at the dairy ( $n = 7$ ) or through self-teaching ( $n = 3$ ). Based on FC evaluators, treatments were decided by the owner ( $n = 8$ ) or manager ( $n = 7$ ). And, when advice on treatments was needed, the owner ( $n = 6$ ), the veterinarian ( $n = 8$ ), or the PSR ( $n = 1$ ) were consulted. Information on new drugs and treatments came through veterinarians ( $n = 8$ ), and/or PSR ( $n = 13$ ). Veterinarians involvement on FC evaluations can be strengthened. Furthermore, many FC evaluators relied on their peers to learn and discuss work related issues. This information suggested that teaching tools that promote peers networking might be of relevance for fresh cow employees.

**Key Words:** dairy cattle, fresh-cow evaluation, veterinarian

**T47 Fresh cow evaluations and treatments on California dairies.** Arnau Espadamala\*<sup>1</sup>, Pau Pallarés<sup>1</sup>, Alfonso Lago<sup>2</sup>, and Noelia Silva-del-Río<sup>1</sup>, <sup>1</sup>*UC Davis School of Veterinary Medicine, VMTRC, Tulare, CA, 2**DairyExperts, Tulare, CA.*

Minimizing the risk of antibiotic resistant organisms and antibiotic residues in dairy and dairy beef products is a topic of nationwide interest. To design an effective outreach program on judicious use of antibiotics, it is imperative to describe the actual practices on dairies. Thus, our objective was to summarize management and treatment practices for fresh cows (FC) on 15 California dairies (600 to 9,500 cows) as a first step in that outreach effort. Data were collected by 2 bilingual veterinarians during the FC evaluations, based on cow-side observations and responses from dairy employees. Daily ( $n = 14$ ) or thrice a week ( $n = 1$ ) FC evaluations and treatments administrations lasted [median (range)] 13.8 (1.5 to 45) s/cow. To identify sick cows evaluators relied on thermometer ( $n = 1$ ), stethoscope ( $n = 7$ ) or both ( $n = 3$ ). All dairies visually inspected cows for abnormal uterine discharge. Twelve dairies evaluated 2 to 5 signs of diseases [rumen fill ( $n = 7$ ), eyes-ears ( $n = 7$ ), milk yield / udder fill ( $n = 7$ ), appetite ( $n = 7$ ), feces ( $n = 5$ ), temperature ( $n = 5$ )]. Antibiotic therapy was given systematically after eutotic ( $n = 2$ ), twinning ( $n = 7$ ) and dystotic calvings [all FC ( $n = 5$ ), primiparous FC ( $n = 1$ ), severe cases ( $n = 7$ )]. Cows with retained placenta were treated at 24 ( $n = 8$ ), 48 ( $n = 4$ ) and 72 ( $n = 3$ ) h postpartum with systemic ceftiofur ( $n = 9$ ), penicillin ( $n = 1$ ) or ampicillin ( $n = 3$ ), or intrauterine urea or essential oils ( $n = 2$ ). Cows with foul-smelling vaginal discharge were treated for metritis with NSAIDs ( $n = 3$ ) and systemic antibiotics [ceftiofur ( $n = 12$ ), penicillin ( $n = 2$ ) or ampicillin ( $n = 1$ )], as well as with antiseptic ( $n = 1$ ) or antibiotic ( $n = 2$ ) uterine flushing. Two dairies used antibiotic uterine flushings if systemic antibiotics were ineffective. Non-foul-smelling abnormal vaginal discharge was treated with uterine antiseptic flushings ( $n = 4$ ). On 2 dairies, antibiotics were the treatment of choice for sick cows with an unknown disease. In this study we have observed that some dairies used antibiotics either as prophylactic therapy or to treat sick cows with unspecific diagnose. Sick cows identification, disease classification, and treatments were not consistent across dairies.

**Key Words:** dairy cattle, fresh cow, metritis



**T48 Association among gestation length with health, reproduction, and production in Holstein cows.** Achilles Vieira-Neto\*, Klips N. Galvao, and Jose E. P. Santos, *University of Florida, Gainesville, FL.*

Objectives were to evaluate the association between gestation length (GL) and incidence of diseases, reproduction, and milk production. The data were screened to eliminate cows with GL longer or shorter than 3 SD from the population mean, resulting in 104 cows excluded from the analyses. Holstein cows ( $n = 6,254$ ) on a farm using only artificial insemination (AI) were evaluated. Responses measured included the incidences of stillbirth, retained placenta (RP), metritis, mastitis, and other diseases within 90 d in milk (DIM). Pregnancy at first AI and interval to pregnancy were evaluated. Milk yield and removal from the herd by death or culling were recorded for the first 300 DIM. Gestation length was categorized as short (S; at least 1 SD below the population mean; group mean = 266, range 256 to 269), normal (N; population mean  $\pm$  1 SD; group mean = 276, range 270 to 282 d), and long (L; > 1 SD above the population mean; group mean = 285, range 283 to 296 d). Data were analyzed by ANOVA, logistic regression, and the Cox's proportional hazard model using the GLIMMIX and PHREG procedures of SAS. Models included the fixed effects of GL category (S, N, L), gender of calf (female, male, twin), parity (1 or >1), season of calving (cool or hot), and all 2-way interactions. Gestation length affected ( $P < 0.01$ ) the incidences of stillbirth (S = 11.4,  $n = 7.3$ , L = 6.7%), RP (S = 32.7,  $n = 10.3$ , L = 9.4%), metritis (S = 51.1,  $n = 36.5$ , L = 33.9%), but not ( $P = 0.85$ ) that of mastitis (S = 4.7,  $n = 4.0$ , L = 5.2%). The rate of removal from the herd by culling or death was faster ( $P < 0.01$ ) for S than N (adjusted hazard ratio [HR] = 1.32; 95% CI = 1.10–1.58) and tended ( $P = 0.10$ ) to be faster for L than N (adjusted HR = 1.12; 95% CI = 0.98–1.29). Pregnancy at first AI did not differ ( $P = 0.94$ ) among groups (S = 33.2,  $n = 34.3$ , L = 33.5%). The rate of pregnancy was greater ( $P < 0.05$ ) for N than L (adjusted HR = 1.11; 95% CI = 1.02–1.21), but it did not differ between N and S or L and S. Daily milk yield was greater ( $P < 0.01$ ) for N than S or L (S = 35.2,  $n = 38.1$ , L = 35.8  $\pm$  0.4 kg/d). Cows with GL within 1 SD of the population mean (range 270 to 282 d) had improved health, reproduction, and production.

**Key Words:** dairy cow, gestation length, health

**T49 Hepatic mRNA expression of genes related to inflammatory and immune responses of dairy cows treated with recombinant bovine somatotropin during the periparturient period.** Paula R. B. Silva\*<sup>1</sup>, Wanda Weber<sup>1</sup>, Brian Crooker<sup>1</sup>, and Ricardo C. Chebel<sup>1,2</sup>, <sup>1</sup>University of Minnesota, St Paul, MN, <sup>2</sup>University of Florida, Gainesville, FL.

Objectives were to determine the effects recombinant bovine somatotropin (rbST) treatment during the peripartum period on hepatic mRNA expression of genes related to inflammation and immune response. Holstein cows were assigned randomly to receive no treatment (control;  $n = 10$ ), 87.5 mg (rbST87.5;  $n = 12$ ), or 125 mg (rbST125;  $n = 10$ ) of rbST every 7 d from -21 to 21 d relative to calving. Liver biopsies were collected -21, -7, and 7 d relative to calving. Twenty-four genes were assessed by direct molecular counts using NanoString technology. Continuous data were analyzed by ANOVA. Gene expression on d -21 was used as a covariate for analyses of mRNA expression on d -7 and 7. No differences in mRNA expression were observed among treatments on d -21 for any of the genes except SOCS3, which had lower ( $P \leq 0.05$ ) mRNA expression in control cows compared with rbST87.5 cows. On d -7, expression of mRNA for ANGPTL4 and SCARB1 was higher ( $P \leq 0.05$ ) in rbST87.5 and rbST125 than control cows. The rbST87.5

cows had higher ( $P \leq 0.05$ ) mRNA expression for HP, ICAM1, SOCS2 and XBP1 on d -7 than control and rbST125 cows. Control cows had higher ( $P \leq 0.05$ ) mRNA expression for HIF1A than rbST125 cows on d -7. On d 7, control cows had higher ( $P \leq 0.05$ ) mRNA expression for CXCL1, IL1RN, MYD88, NFKBIA, and SOCS3 compared with rbST87.5 and rbST125 cows. Control cows had higher ( $P \leq 0.05$ ) mRNA expression for ICAM1 and XBP1 than rbST125 cows and had higher mRNA expression for HIF1A than rbST87.5 cows. Expression of mRNA for NR3C1 and SOCS2 was lower ( $P \leq 0.05$ ) in control cows than rbST125 and rbST87.5 cows, respectively. Treatment did not affect hepatic expression of CEBPD, JUN, M-CSF1, NFKB1, PPARGC1A, STAT5B, TLR2, TNF, TNFRSF1 and TNFRSF5. Weekly treatment of periparturient cows with rbST regulates liver mRNA expression of genes related to inflammation and immune response during the prepartum and postpartum periods. These rbST-induced changes in gene expression have resulted in improved neutrophil function, antibody production, and reduced incidence of retained placenta and metritis.

**Key Words:** periparturient cow, recombinant bovine somatotropin, hepatic gene expression

**T50 Changes in serum triacylglycerols may indicate disease risk for retained placenta and mastitis in multiparous dairy cows.** Fereshteh Zandkarimi, Massimo Bionaz, Jan S. Stevens, Claudia S. Maier, and Gerd Bobe\*, *Oregon State University, Corvallis, OR.*

Compromised liver function has been proposed to be involved in the etiology of many diseases in early lactation dairy cows. Examining changes in individual serum triacylglycerols (TAG) may identify previously unknown metabolic pathways involved in the etiology of disease in early lactation. We used ultraperformance liquid chromatography (UPLC) in conjunction with high-resolution accurate mass spectrometry to comprehensively and quantitatively profile individual TAG in serum. The objective of this study was to examine whether individual TAG in serum may serve as disease risk indicators of mastitis, retained placenta, or both. Serum samples were collected from 161 multiparous cows 3, 2, and 1 week before calving and at calving. For this nested case-control study, serum samples of 3 groups of cows that either developed after calving retained placenta (RP;  $n = 8$ ), mastitis (MA;  $n = 8$ ), or remained healthy (Healthy;  $n = 9$ ) were selected and serum levels of individual TAG were measured. Using PROC MIXED, overall and individual TAG levels in serum were compared between groups. The mass spectrometry-driven strategy resulted in the identification and comparative quantification of 24 individual saturated and unsaturated TAG between C48:1 and C54:4. Total TAG levels decreased until calving ( $P < 0.0001$ ). Cows that subsequently developed RP had and tended to have lower TAG levels than cows that subsequently developed mastitis ( $P = 0.03$ ) or remained healthy ( $P = 0.12$ ). Similarly, RP cows had lower TAG with 4 double-bonds compared with Healthy cows ( $P = 0.04$ ) and MA cows ( $P = 0.0006$ ), which could serve as potential RP indicator. In contrast, both diseased groups had lower saturated TAG levels compared with Healthy cows (MA:  $P = 0.03$ ; RP:  $P = 0.05$ ). Consequently, the ratio between TAG with 4 double bonds to saturated TAG was higher in MA cows compared with Healthy ( $P = 0.007$ ) and RP cows ( $P = 0.01$ ), which, thus, could be a potential MA indicator. These findings suggest changes in serum TAG levels are an early indicator for the development of retained placenta and mastitis in multiparous dairy cows.

**Key Words:** dairy cow, early disease indicator, triacylglycerols

### T51 Changes in serum nonesterified fatty acids precede retained placenta and mastitis in multiparous dairy cows.

Fereshteh Zadnkari, Massimo Bionaz, Jan S. Stevens, Claudia S. Maier, and Gerd Bobe\*, Oregon State University, Corvallis, OR.

We previously documented that elevated serum nonesterified fatty acids (NEFA) before calving are a general disease risk indicator in dairy cows. Examining changes in individual or subclasses of NEFA may improve disease risk detection. Untargeted lipid profiling using ultra performance liquid chromatography-high resolution mass spectrometry (UPLC-MS) is a comprehensive technique that allowed simultaneous analysis of individual NEFA in complex biological samples. The objective of this study was to examine the association between serum levels of individual or subclasses of NEFA and subsequent development of diseases. Serum samples were collected from 161 multiparous cows 3, 2, and 1 week before calving and at calving. For this nested case-control study, serum samples of 3 groups of cows that either developed after calving retained placenta (RP; n = 8), mastitis (MA; n = 8), or remained healthy (Healthy; n = 9) were selected and serum levels of individual NEFA were measured. Using PROC MIXED, serum NEFA levels were overall and individually compared between groups. Levels of 22 individual NEFA between C14:0 and C26:0 were above the limit of detection. Overall levels of NEFA identified by UPLC-MS analysis were in good agreement with chemical analysis of total NEFA concentrations. Cows that developed RP and MA in early lactation had 1 week before calving ( $P = 0.05$ ) and at parturition ( $P = 0.005$ ) elevated overall NEFA levels compared with Healthy cows. Similar directional changes were observed for C14:0, C16:0, C16:1, C18:1, as well as the C18:1 to C18:0 ratio, which could serve as general disease risk indicators. In contrast, cows that subsequently developed diseases had lower elongation and/or very long chain fatty acid ( $\geq C20$ ) desaturation indices compared with Healthy cows. MA cows had greater  $\geq C20$  n6 to  $\geq C20$  n3 and C20:1 to C20:0 ratios and lower  $\geq C20:0$  levels compared with Healthy and RP cows, which could be used as potential MA indicators. These findings suggest changes in very long chain fatty acids levels and their ratios are an early indicator for the development of retained placenta and mastitis in multiparous cows.

**Key Words:** dairy cow, early disease indicator, nonesterified fatty acids

### T52 Distribution of most common coagulase-negative species over parity and lactation in Canadian dairy herds.

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Coagulase-negative staphylococci (CNS) are the most frequently isolated group of microorganisms from the bovine udder. Research has shown that certain CNS species can be protective, while other species are prone to decrease milk quality and production. Our objective was to describe the distribution of CNS species for the lactation period and parities in Canadian herds, as an initial effort to understand the disease. Cows were sampled randomly across Canada in 2007 and 2008. In total, 1965 cows had udder infection with CNS, being compared with a total of 6067 cows sampled in the study. Chi-squared was used to test different prevalences ( $P < 0.05$ ). The 5 most common CNS species identified in were *S. chromogenes* (49%), *S. simulans* (17%), *S. xylosum* (11%), *S. haemolyticus* (7%) and *S. epidermidis* (4%). Parity and lactation prevalences are presented in Table 1. *S. chromogenes* was predominant in heifers, and its prevalence increased during the lactation. *S. epidermidis* was mostly isolated from older cows and the prevalence did not increase

as lactation progressed. *S. haemolyticus* isolated equally often in all parities; its prevalence increased during lactation. *S. simulans* seems to be more isolated in heifers, and its prevalence did not increase over the lactation. *S. xylosum* fluctuates over parities and its prevalence increased during the lactation. The distribution of these CNS species differed over parities and lactation period. The prevalence of species such as *S. chromogenes*, which maybe less pathogenic, was very high in heifers compared with older cows. Throughout lactation, it seems that most species can persist or reinfect udder. Understanding the distribution of CNS species will possibly lead to selective management practices and may lead to identification of species or genotypes that act as protective udder pathogens.

**Table 1 (Abstr. T52).** Relative frequency (%) of CNS species distribution over parity and over lactation thirds

CNS species	Parity				Lactation thirds			
	1	2	3	$\geq 4$	1	2	3	$>305$ d
<i>S. chromogenes</i>	60	23	24	26	8	8	11	11
<i>S. epidermidis</i>	1	3	3	5	1	1	0.5	1
<i>S. haemolyticus</i>	5	5	6	5	1	1.2	2	2
<i>S. simulans</i>	15	9	10	13	4	2	2	3
<i>S. xylosum</i>	6	9	7	10	1	1	2	3

**Key Words:** coagulase-negative staphylococci, intramammary infection

### T53 Phospholipids are potential early risk indicator for retained placenta and mastitis in multiparous dairy cows.

Fereshteh Zadnkari, Massimo Bionaz, Jan S. Stevens, Claudia S. Maier, and Gerd Bobe\*, Oregon State University, Corvallis, OR.

Phospholipids such as phosphatidylcholines (PC), phosphatidylethanolamines (PE), phosphatidylinositols (PI), and phosphatidylserines (PS), have been proposed to be involved with compromised liver function in dairy cows. Examining changes in individual phospholipids may identify potential therapeutic targets of diseases in early lactation. We employed an untargeted mass spectrometry-based lipid profiling strategy to analyze simultaneously individual phospholipids in serum. The objective of this study was to examine whether individual phospholipids in serum may serve as disease risk indicators of mastitis, retained placenta, or both. Serum samples were collected from 161 multiparous cows 3, 2, and 1 week before calving and at calving. For this nested case-control study, serum samples of 3 groups of cows that either developed after calving retained placenta (RP; n = 8), mastitis (MA; n = 8), or remained healthy (Healthy; n = 9) were selected and serum levels of individual phospholipids were measured. Using PROC MIXED, serum levels of phospholipid classes and subclasses were compared between groups. Total PC, lysoPC, and lysoPE levels decreased until calving (all  $P < 0.0001$ ). At calving, cows that subsequently developed mastitis had lower lysoPC, lysoPE, and inositol phosphate levels than cows that subsequently developed RP or remained healthy (lyso PC: both  $P = 0.03$ ; lyso PE: both  $P = 0.02$ ; inositol phosphate:  $P = 0.04$  and  $P = 0.03$ ) and thus, could be used as MA indicators. In contrast, RP cows had lower levels of highly unsaturated PS (PS with 4 double bonds) than MA or Healthy cows (both  $P = 0.02$ ), which could serve as RP indicator. These findings suggest changes in phospholipids are an early indicator for the development of retained placenta and mastitis in multiparous cows and may be potential therapeutic targets for diseases in early lactation.

**Key Words:** dairy cow, early disease indicator, phospholipids

**T54 Hoof measurements before and after hoof trimmer intervention on dairy lame cows on California dairies.** Marc Pineda\*<sup>1</sup>, Ibrahim Akin<sup>2</sup>, and Noelia Silva-del-Rio<sup>1</sup>, <sup>1</sup>*Veterinary Medicine Teaching and Research Center, UC Davis, Tulare, CA*, <sup>2</sup>*Adnan Menzies University Veterinary Faculty Department of Surgery, Aydin, Turkey*.

The objective of this study was to describe lame cow hoof measurements before and after hoof trimmer intervention on California dairies. A total of 17 dairies ranging in size from 1,000 to 10,000 were enrolled in the study. Hoof trimmers were dairy workers (n = 11) or outside service providers (n = 6). Researchers collected information from rear hooves from 10 (n = 15) or 9 (n = 2) lame cows per dairy (dorsal wall length, hoof angle, heel height, and abaxial groove length) before and after the hoof trimmer intervention. Descriptive statistics were analyzed with PROC MEANS, PROC UNIVARIATE and PROC CORR of SAS 9.4. Before the hoof trimmer intervention 10.1% of the hooves had a desirable dorsal wall length (>7 to <8.5 cm). After the hoof trimmer intervention, at least 70% (n = 7) or less than 30% (n = 4) of the hooves were within the desirable range. Dorsal wall length of hooves of the same claw differed by >0.5 cm at least 5% of the time (n = 8). Dorsal wall angle was within a desirable range (<sup>3</sup>45° to <50°) on 52.2% and 50.9% of the hooves before and after hoof trimmer intervention respectively. After the hoof trimmer intervention, at least 60% (n = 4) or less than 30% (n = 3) of the hooves were within the desirable range. Dorsal wall angle of hooves of the same claw differed by 3° at least 25% of the time (n = 11). Heel height was within > 3.4 to < 4.4 cm on 20.0 and 36.8% of the hooves before and after hoof trimmer intervention respectively. After the hoof trimmer intervention, 5 herds had at least 50% (up to 65%) of hooves with heel height within > 3.4 to < 4.4 cm. Heel height of hooves of the same claw differed by >0.5 cm at least 25% of the time (n = 9). The abaxial groove length was >4.5 cm on 93.5 and 90.8% of the hooves before and after hoof trimmer intervention respectively. The correlation between heel height and abaxial groove length was significant ( $P < 0.01$ ) but with a low correlation coefficient before ( $r = 0.47$ ) and after ( $r = 0.20$ ). Our data indicates that there is an opportunity to improve hoof angle of lame cows after hoof trimmer intervention on California dairies.

**Key Words:** lameness, hoof trimmer, dairy cow

**T55 Correlation of ACTH test results with hormonal, metabolic and cardiac stress responses during stress challenge in dairy cows.** Lea Fieguth<sup>1</sup>, Lena Locher<sup>1</sup>, Anja Schacht<sup>1</sup>, Akos Kenez<sup>2</sup>, Asako Kinoshita<sup>1</sup>, Ulrich Meyer<sup>3</sup>, Sven Dänicke<sup>3</sup>, and Juergen Rehage\*<sup>1</sup>, <sup>1</sup>*Clinic for Cattle, University of Veterinary Medicine Hannover, Hannover, Germany*, <sup>2</sup>*Department of Physiology, University of Veterinary Medicine Hannover, Hannover, Germany*, <sup>3</sup>*Department of Animal Nutrition, Friedrich-Loeffler-Institute, Braunschweig, Hannover*.

Cows react to stressors with release of cortisol and catecholamines after activation of the hypothalamic-pituitary-adrenal axis (HPA) and

of the hypothalamic-adrenal medullary axis, respectively. Increases in heart rate, hyperglycemia, and hyperlipidemia are typical metabolic responses. The extent of the individual stress response depends on individual experience and stress responsiveness. ACTH-test results are assumed to reflect the individual HPA reactivity. The aim of the study was to compare results of the ACTH test with hormonal, metabolic and cardiac stress responses of cows on claw trimming as a typical acute non-painful stressor. In 18 pluriparous German HF cows claw trimming (CT) was performed in lateral recumbency 40 d postpartum (pp). Blood level of cortisol, glucose, and nonesterified fatty acids (NEFA) and heart rate (HR) and heart rate variability (HRV; standard deviation of R-R intervals: SDNN, root mean square of successive differences: RMSSD) were measured at T1: 30 min before CT in the herd, T2: standing at the surgical table, T3: end of claw trimming, T4: 30 min after CT in the herd. ACTH tests were performed on d 110 pp (80 µg/cow ACTH; repeated blood samples for cortisol measurement over 120 min) from which the area under the cortisol time curve (AUC) were assessed. AUC was correlated (SPEARMAN, SAS package) with cortisol, glucose and NEFA blood concentrations as well as HR and HRV at T1-T4 and with differences between time points during the CT procedure. AUC correlated positively with T3 cortisol ( $r: 0.76, P = 0.002$ ) and  $\text{Diff}_{T4-T1}$  cortisol ( $r: 0.75, P = 0.002$ ), and negatively with T4 NEFA ( $r: -0.64, P = 0.014$ ), T4 HR ( $r: -0.64, P = 0.014$ ), T4 SDNN ( $r: -0.59, P = 0.027$ ),  $\text{Diff}_{T4-T3}$  glucose ( $r: -0.56, P = 0.037$ ),  $\text{Diff}_{T4-T1}$  NEFA ( $r: -0.73, P = 0.003$ ),  $\text{Diff}_{T4-T1}$  HR ( $r: -0.71, P = 0.005$ ),  $\text{Diff}_{T4-T1}$  SDNN ( $r: -0.65, P = 0.012$ ). High ACTH test reactivity was associated with high cortisol level during but a quick decrease to baseline values in blood NEFA and glucose concentrations and in HR, and HRV after CT, which may indicate a close association between individual stress responsiveness and metabolic and cardiac adaptability to stress challenges in cows.

**Key Words:** ACTH test, stress reactivity, dairy cow



## ASAS Undergraduate Student Poster Competition

**T56 Weather-related cold stress on conception rates in Sim-Angus cattle.** Jessica A. Stone\* and Julie D. Weathers, *Southeast Missouri State University, Cape Girardeau, MO.*

The purpose of this study was to determine how much correlation there was, if any, between temperature, wind chill factor, and pregnancy results. It has been well documented that weather patterns and temperature will affect reproductive efficiency in cattle, as well as other species. The study was conducted in 2013 and again in 2014, using the similar methods. Because of a change in management, 2012 was not eligible for the study because of inconsistency from the following years. The same population of similarly bred Sim-Angus cattle was inseminated using a fixed timed artificial insemination program of a 14-d CO-Synch + CIDR protocol at the David M. Barton Agriculture Research Center in Gordonville, Missouri. In 2013, 41 Sim-Angus cows participated in the study and in 2014, 17 Sim-Angus cows were used. Only 17 were used in 2014 because the entire herd was split equally to create a fall and spring calving herds. By studying the weather patterns 14 d before and after the day of breeding and comparing the pregnancy results of 2013 and 2014, a correlation was found between the temperature, wind chill factor, and pregnancy results ( $r = -0.6$ ). In 2013, the animals were bred on December 6, where the average temperature was  $-5^{\circ}\text{C}$  with a wind chill of  $-12.94^{\circ}\text{C}$  at 8:53 a.m. In 2014, the day of breeding was December 12, with an average temperature of  $3.88^{\circ}\text{C}$  and a wind chill of  $0.05^{\circ}\text{C}$  at 8:53 a.m. After comparison of blood samples each year for pregnancy (BioPryn; Moscow, ID), 16 out of 41 animals were pregnant in 2013, giving a conception rate of 39%. In 2014 however, 14 of the 17 animals were bred, giving a conception rate of 82%. More research is forthcoming to determine if this correlation holds true for a larger sample of cattle and more repetitions of the experiment.

**Key Words:** conception, cold stress, temperature

**T57 Number of pigs born alive in parity-1 sows associated with lifetime performance and removal hazard in high- or low-performing herds in Japan.** Satomi Tani\*, Ryosuke Iida, and Yuzo Koketsu, *Meiji University, Kawasaki, Kanagawa, Japan.*

We compared reproductive performance across parity, lifetime performance and removal hazard of sows in commercial Japanese herds categorized into 3 sow reproductive performance groups and 2 herd productivity groups. The sow group categories were based on the lower and upper 25th percentiles of the number of pigs born alive (PBA) in parity 1, as follows: 8 pigs or fewer, 9–12 pigs and 13 pigs or more. The herds were classified into high- and low-performing herds on the basis of the 50th percentile of pigs weaned per mated female per year. We analyzed 213,514 parity records and 47,024 lifetime records of females entered into 96 herds. Multivariate and single response models were applied to reproductive performance across parity and lifetime performance, respectively, and a proportional hazard model was applied to removal hazard. Sows having 13 or more PBA in parity 1 had 1.0–1.4 more PBA in subsequent parities than sows having 8 or fewer PBA. However, there were no such differences between sow groups for weaning-to-first-mating interval across parity ( $P > 0.05$ ). Also, sows having 13 or more PBA in parity 1 had 3.4–3.7 higher lifetime average PBA than sows having 8 or fewer PBA in both the herd groups ( $P < 0.05$ ). Furthermore, the sows in the low-performing herds with 13 or more PBA in parity 1 also had 2.3 fewer lifetime average nonproductive days than sows having 8 or fewer PBA ( $P < 0.05$ ), although again no similar difference

was found for high-performing herds ( $P > 0.05$ ). The removal hazards for a sow having 13 or more PBA in parity 1 were lower than those for a sow having 8 or fewer PBA ( $P < 0.05$ ), with no difference in hazards between the 2 herd groups ( $P = 0.62$ ). In conclusion, PBA in parity 1 can be used to predict a prolific or a low PBA sow. Also, producers in low-performing herds should pay particular attention to sows having low PBA in parity 1 to reduce nonproductive days.

**Key Words:** cohort study, fertile sow, hazard model

**T58 Beneficial effects of a short-term provision of forage to intensively reared broiler chickens.** Oluwaseun S. Iyasere, Toluwatope O. Sodipo, and Anuoluwapo V. Subulokun\*, *Federal University of Agriculture, Abeokuta, Ogun State, Nigeria.*

Poultry production plays a major role in bridging the protein gap in developing countries. However, the productivity of poultry in the tropics has been limited by scarcity and high prices of the conventional protein and energy sources. In modern day free-range poultry farming, it is generally assumed that the contribution of foraging to nutritional intake is negligible despite the fact that birds still obtain some of their feed requirements from forage. It became imperative to study the effect of the provision of forage on a short-term to broilers reared intensively on deep litter. Thirty-two 5-wk-old broilers (average weight of 1.2kg) of mixed sex were divided into 2 treatments namely concentrate and concentrate-pasture fed birds, each treatment had 4 replicates. Each replicate of birds were housed in separate pens ( $1.2 \times 2.0$  cm). Birds were offered either concentrate (200g) or concentrate-pasture (100 g concentrate and 100 g *Tridax procumbens*) every other day for 30 min. The behavior of the birds was scanned at 5 min interval for 30 min, 4 times a week for 2 weeks and the percentage of birds feeding or drinking was recorded. Feed intake and water intake was monitored during the 30 min period. Weight gain was determined at the end of the 2 weeks experimental period. After checking for normality, data were analyzed using independent *t*-test of SPSS statistical package (version 16). Result showed that a greater percentage of birds offered concentrate displayed higher ( $P < 0.001$ ) drinking behavior and drank more water ( $P < 0.001$ ) compared with birds offered concentrate-pasture. The percentage of birds feeding was greater ( $P < 0.001$ ) in birds offered concentrate-pasture feed than those offered only concentrate feed. In conclusion, a short-term provision of broilers with forage (*Tridax procumbens*) alongside their concentrate feed enhanced their feeding behavior and reduced their concentrate intake by 34% without having a negative effect on their body weight gain. Hence, farmers could consider this as a strategy for conserving their expensive feed resources.

**Key Words:** behavior, body weight gain, broiler

**T59 Inosine 5'-monophosphate increases glutamic acid induced cholecystokinin release from bovine proximal small intestine.** Erin L. Doherty\*, Derek W. Brake, and George A. Perry, *South Dakota State University, Brookings, SD.*

Cholecystokinin (CCK) is a hormone secreted by the proximal small intestine and mediates pancreatic exocrine secretions in response to luminal nutrient flows. Glutamic acid and other amino acids can increase secretions of CCK among mice, dogs, and STC-1 cells. Further, inosine 5'-monophosphate (IMP) can increase amino acid induced CCK release from murine proximal intestine. Our objective was to quantify effects of

IMP on glutamic acid induced CCK release from bovine small intestine. Small intestines were collected at slaughter from 2 steers ( $638 \pm 3.2$  kg BW) fed a common dry rolled corn-based diet. The small intestine was measured after removal of digesta, and 1-m sections of duodenum, jejunum and ileum were collected. Each small intestinal section was cut longitudinally, rinsed and serosa was removed before collection of mucosa. Mucosal explants (1-cm in diameter) were incubated ( $37^{\circ}\text{C}$ ) in HBSS (containing  $1.26 \text{ mM Ca}^{2+}$ ) with  $20 \text{ mM}$  HEPES (pH 7.4) and supplemented with 0, 10, 20, or  $30 \text{ mM}$  glutamic acid and 0 or  $2.5 \text{ mM}$  IMP. After 1 h, incubation buffer was collected, centrifuged to remove cellular debris, and frozen ( $-80^{\circ}\text{C}$ ) before analyses. Cholecystokinin secretions were quantified by radioimmunoassay. Cholecystokinin release was greater ( $P < 0.01$ ) from duodenum ( $68.5 \pm 5.0 \text{ pM}$ ) than from jejunum ( $8.5 \pm 4.9 \text{ pM}$ ) and ileum ( $2.3 \pm 4.9 \text{ pM}$ ). Release of CCK was 529% greater (glutamic acid  $\times$  IMP  $< 0.01$ ) when proximal small intestine was supplemented with glutamic acid and IMP ( $2.79 \text{ pM}$  CCK released /  $\text{mM}$  glutamic acid) than when proximal small intestine was supplemented with glutamic acid alone ( $0.53 \text{ pM}$  CCK released /  $\text{mM}$  glutamic acid). cursory comparisons to previous data suggest that glutamic acid induced CCK release by proximal intestine is less in cattle than in mice; however, IMP enhanced glutamic acid induced CCK release in cattle to a greater extent than previously reported in mice.

**Key Words:** cattle, cholecystokinin, glutamic acid

#### **T60 Overexpression and inhibition of specificity protein 1 (SP1) affect milk fat formation in goat mammary epithelial cells.**

Jiangjiang Zhu\* and Jun Luo, *Shaanxi Key Laboratory of Molecular Biology for Agriculture, College of Animal Science and Technology, Northwest A&F University, Yangling, Shaanxi, China.*

The objectives of the present study are to explore the function of SP1 in regulating triglyceride accumulation, droplet formation and the expression of genes associated with fat metabolism in goat mammary epithelial cells (GMECs). Adenovirus was used for SP1 overexpression, siRNA and mithramycin A were used for SP1 silencing and inhibition respectively. GC-MS was used for fatty acid composition analysis. Three repeats was used for qPCR and fatty acid determination. Six repeats were used for triglyceride determination and oil red O staining. The results were expressed as mean  $\pm$  SD. Data of qPCR were analyzed using the  $2^{-\text{DDCt}}$  method. Group data for multiple comparisons were analyzed by ANOVA followed by Tukey's test. Significance was established at a  $P < 0.05$ . The results showed that overexpression and interference of SP1, using adenovirus and siRNA respectively, significantly reduced the content of cellular triglyceride and suppressed the accumulation of lipid droplets, and altered the expression of genes related to lipid metabolism. These data indicated that PPAR $\gamma$  and LXR $\alpha$  might have affected the role of SP1 in regulating other genes related to fatty acid uptake (CD36 and LPL), de novo fatty acid synthesis (FASN and ACACA), fatty acid elongation and desaturation (EVOLV6, SCD and FADS2), fatty acid transport (ACSL1, ACSS2 and FABP3), triglyceride synthesis (AGPAT6, LPIN1, DGAT1 and DGAT2), lipid droplet formation and secretion (TIP47, ADFP and XDH), and triglyceride hydrolysis (ATGL and HSL). But expression of SREBP1 was not affected by manipulation of SP1 despite of the crucial role in regulating de novo fatty acid synthesis. To support the results on triglyceride accumulation, adenovirus and mithramycin A, an inhibitor of SP1, was used to treat GMECs. The result showed that, similar to the decrease of triglyceride accumulation, both inhibition and overexpression of SP1 increased the relative content of saturated fatty acids, including C16:0, C14:0 and decreased the content of C18:1 which is important for triglyceride synthesis in GMECs. In conclusion, the findings provide a novel insight of SP1 function in

regulating lipid metabolism in GMECs, and also provide novel method for dairy goat breeding.

**Key Words:** dairy goat, specificity protein 1 (SP1), milk fat metabolism

#### **T61 Effects of plant-derived compounds on *Staphylococcus aureus* infection of primary bovine mammary epithelial cells.**

Ellen V. Valley\*<sup>1</sup>, Devi Jaganathan<sup>1</sup>, Kumar Venkitanarayanan<sup>1</sup>, Gary W. Kazmer<sup>1</sup>, Lynn Kuo<sup>2</sup>, Yu Bo Wang<sup>2</sup>, and Kristen E. Govoni<sup>1</sup>, <sup>1</sup>*Department of Animal Science, University of Connecticut, Storrs, CT,* <sup>2</sup>*Department of Statistics, University of Connecticut, Storrs, CT.*

Mastitis is an inflammation of the mammary gland which results in losses to the dairy industry due to discarded milk, poor milk quality and culling. Plant-derived antimicrobials (PDA) have inhibitory effects on *Staphylococcus aureus*, an economically important cause of mastitis, and have less potential to induce bacterial antimicrobial resistance. We hypothesized that (1) eugenol (EG) and *trans*-cinnamaldehyde (TC) would reduce *S. aureus* adhesion to and invasion of primary bovine mammary epithelial cells (MEC), and (2) EG and TC would alter the expression of virulence factors in *S. aureus* and immune response genes in MEC. Mammary epithelial cells were collected from a lactating dairy cow at slaughter and isolated by selective trypsinization. Four strains of *S. aureus*, Thorn 17, S35, M9175, and Thorn 15, were obtained from the Connecticut Veterinary Medical Diagnostic Laboratory. Before inoculation, MEC and *S. aureus* were incubated at  $37^{\circ}\text{C}$  for 12 and 5 h, respectively, with sub-inhibitory concentrations (SIC, concentrations not inhibiting bacterial growth) of EG (0.03%) and TC (0.006%). Mammary epithelial cells were challenged with *S. aureus* in mid log phase and incubated for 2 h at  $37^{\circ}\text{C}$ . To quantify adhesion, cells were washed with PBS to remove unadhered bacteria and lysed with Triton-X. Lysed cells were serially diluted and plated on mannitol salt agar, and colonies were counted after 48 h. To quantify invasion, cells were incubated for 1 h with gentamycin before washing, lysing, and plating as performed for the adhesion assay. Experiments included MEC and *S. aureus* as controls, MEC or *S. aureus* pre-treated with PDA, and MEC and *S. aureus* both pre-treated with PDA (9 treatments). Experiments were performed in duplicate and repeated 3 times. Data were analyzed using PROC MIXED in SAS. For all strains, adhesion was reduced 0.2 to  $0.8 \log_{10}$  cfu/mL ( $P \leq 0.05$ ) and invasion was reduced 1.7 to  $3.8 \log_{10}$  cfu/mL ( $P \leq 0.05$ ). Gene expression analysis is currently underway. In conclusion, SIC of EG and TC effectively reduced *S. aureus* infection of MEC.

**Key Words:** bovine, mastitis, plant-derived antimicrobial

#### **T62 Extended-spectrum cephalosporin, carbapenem, and fluoroquinolone-resistant gram-negative coliform bacteria present on equine environmental surfaces.**

Rachael Adams\*<sup>2</sup>, Dixie Mollenkopf<sup>1</sup>, Dimitria Mathys<sup>1</sup>, Joshua Daniels<sup>1</sup>, and Thomas Wittum<sup>1</sup>, <sup>1</sup>*The Ohio State University College of Veterinary Medicine, Columbus, OH,* <sup>2</sup>*The Ohio State University College of Food, Agricultural, and Environmental Sciences, Columbus, OH.*

Antimicrobial resistant bacteria are a rapidly growing concern in human and veterinary medicine. The rising prevalence of extended spectrum  $\beta$ -lactamase (ESBL), AmpC  $\beta$ -lactamase, carbapenemase (CRE), and fluoroquinolone-resistant *Enterobacteriaceae* continually decreases the efficiency of vital antibiotics. Moreover, antibiotic resistant enteric bacteria are zoonotic and can be transmitted between horses and people. Our objective was to evaluate the prevalence of antibiotic resistant bacteria on human contact surfaces in equine environments. Environ-

mental surfaces in 20 Ohio equine barns were sampled using 2 electrostatic cloths (Swiffer), yielding a total of 242 samples. Samples were phenotypically screened for AmpC, ESBL, CRE, and fluoroquinolone resistance using selective media. To select for cephalosporinase phenotypes, samples were incubated at 37°C in nutrient broth with 2 µg/mL cefotaxime. This broth was aseptically inoculated to MacConkey agar with 8 µg/mL cefoxitin, 4 µg/mL cefepime, and 1 µg/mL meropenem to detect AmpC, ESBL, and CRE phenotypes, respectively. Additionally, samples were incubated in nutrient broth with 16 µg/mL naladixic acid and then inoculated to MacConkey agar with 16 µg/mL naladixic acid and 2 µg/mL ciprofloxacin to detect fluoroquinolone resistance phenotypes. Genotypes were confirmed using standard PCR techniques. Of the coliform bacteria isolated from each surface, 49 (24.5%) were cefoxitin resistant, 25 (12.5%) were naladixic acid resistant, 13 (6.5%) were cefepime resistant, and 9 (4.5%) were ciprofloxacin resistant. Drains and wash stalls were most commonly contaminated at 17 resistant isolates, followed by handles of mucky equipment at 15 resistant isolates. These results indicate that equine environmental surfaces are contaminated with resistant bacteria that can potentially be transmitted between human and horse populations. Furthermore, detecting these bacteria on common human contact surfaces suggests that the environment can serve as a reservoir for antibiotic resistance genes. Identifying interventions to lower the prevalence of antibiotic resistant bacteria in equine environments will protect both animal and public health.

**Key Words:** antibiotic resistance, equine, environmental

**T63 Interleukin-6, tumor necrosis factor- $\alpha$ , insulin-like growth factor-1 and fibroblast growth factor-2 alter proliferation and differentiation of equine satellite cells.** Emma K. LaVigne<sup>\*1</sup>, Alfredo Sanchez Londoño<sup>2</sup>, and Sarah A. Reed<sup>1</sup>, <sup>1</sup>*Department of Animal Science, University of Connecticut, Storrs, CT*, <sup>2</sup>*Department of Environmental and Population Health, Cummings School of Veterinary Medicine at Tufts University, North Grafton, MA*.

Muscle growth in young horses occurs by hypertrophy of individual muscle fibers, which can be accomplished through the activation and differentiation of satellite cells. Satellite cells are muscle stem cells that reside between the sarcolemma and the basal lamina of the muscle fiber and can be stimulated or inhibited in response to different cytokines and growth factors. We hypothesized that interleukin (IL)-6, tumor necrosis factor (TNF)- $\alpha$ , insulin-like growth factor (IGF)-1 and fibroblast growth factor (FGF)-2 would alter proliferation and differentiation in satellite cells isolated from young horses. Satellite cells obtained from 10 d old foals ( $n = 4$ ) were cultured individually in the presence of 10 ng/mL IL-6, 20 ng/mL TNF- $\alpha$ , 25 ng/mL IGF-1 or 10 ng/mL FGF-2 to determine the effects on proliferation and differentiation (3 wells per treatment group per horse). Proliferation was measured by incorporation of 5-ethynyl-2'-deoxyuridine (EdU) into cells in S phase. Differentiation was measured by quantifying the fusion index (the number of nuclei present in multinucleated myofibers divided by the total number of nuclei). Data were analyzed using the MIXED procedure in SAS;  $P < 0.05$  was considered significant. Interleukin-6 and TNF- $\alpha$  decreased satellite cell proliferation compared with control cells (14.9% and 11.5%, respectively;  $P \leq 0.01$ ). Fusion into myotubes was increased 6.2% in the presence of IL-6 ( $P = 0.001$ ) but decreased 8.7% by TNF- $\alpha$  ( $P < 0.0001$ ). Satellite cell proliferation was increased 28.8% in the presence of IGF-1 ( $P < 0.0001$ ) and 73.0% in the presence of FGF-2 ( $P < 0.0001$ ) compared with controls. Insulin-like growth factor-1 increased fusion 3.5% ( $P = 0.0087$ ) and FGF-2 decreased fusion 13.1% ( $P < 0.0001$ ). By differentially stimulating or inhibiting proliferation and fusion of satellite cells, IL-6, TNF- $\alpha$ , IGF-1 and FGF-2 significantly affect muscle hypertrophy.

Dysregulation of these cytokines or growth factors, therefore, can lead to detrimental muscle degradation, inflammation, and impaired growth.

**Key Words:** satellite cells, horse, inflammation

**T64 Effects of varying anthelmintic formulations on hindgut microflora in horses.** John Rowe<sup>\*</sup>, Katelyn Barnhart, Elizabeth Share, John Mark Reddish, and Kimberly Cole, *The Ohio State University College of Food, Agricultural, and Environmental Sciences, Columbus, OH*.

Horses house a dynamic population of microbes within their hindgut that can be disrupted by diet, stress, and medication. Horses are routinely given anthelmintic drugs to reduce internal parasite loads and although their modes of action are well known, there is a lack of knowledge of their effect on the gastrointestinal microflora in horses. The objective of this study was to monitor changes in hindgut microflora after treatment with 2 pyrantel anthelmintic formulations. Ten non-pregnant Quarter Horse mares ( $8.0 \pm 6.0$  yr) were randomly assigned to 1 of 2 treatment groups: Paste or Pellet. Throughout the study, all mares continued to receive their basal diet of 0.5% BW of a 12% CP pelleted concentrate with mixed grass hay and water ad libitum. Mares in the Paste group received one dose (0.9 g per 136 kg BW) of pyrantel pamoate paste. Fecal samples were collected immediately before treatment (d 0) and on d 1, 2, 3, 7, 10, and 14 post-treatment. Mares in the Pellet group received pyrantel tartrate pellets (28.3 g per 113 kg bw) once daily for 14 d. Fecal samples were collected immediately before treatment (d 0) and on d 1, 2, 3, 7, 10, and 14 of treatment as well as d 1, 2, 3, 7, 10, and 14 post-treatment. Pooled fecal samples from d 0 served as the untreated controls. DNA was extracted from fecal samples and subjected to PCR-DGGE with universal primers specific to the V2-V3 region of the 16S rRNA gene. PCR-DGGE images were analyzed with BioNumerics software to generate dendrogram comparisons based on the position and number of bands with further evaluation using principal coordinate analysis (PCA). Dendrograms and PCA revealed clustering by time in both groups indicating that pyrantel anthelmintic treatment influenced hindgut microbial diversity. Additional research identifying specific changes in the microbial profiles is needed to better understand the influence of anthelmintic products on the hindgut microflora of horses.

**Key Words:** equine, microflora, anthelmintic

**T65 Effects of poor maternal nutrition during gestation on protein expression in the liver of lambs.** Katelyn K. McFadden<sup>\*</sup>, Maria L. Hoffman, Kristen N. Peck, Sarah A. Reed, Steven A. Zinn, and Kristen E. Govoni, *Department of Animal Science, University of Connecticut, Storrs, CT*.

Poor maternal nutrition during gestation can reduce growth and circulating growth factors secreted by the liver, as well as alter lipid metabolism; however, the mechanisms involved are not well understood. Previously, we reported that poor maternal nutrition altered liver expression of genes involved in lipid metabolism and the somatotrophic axis in the offspring. Therefore, we hypothesized that poor maternal nutrition during gestation would alter expression of key proteins involved in lipid metabolism and the somatotrophic axis in the liver of offspring. Multiparous ewes ( $n = 36$ ) were individually housed and fed 100, 60, or 140% of NRC requirements beginning at d  $31 \pm 1.3$  of gestation. Lambs were euthanized within 24 h of birth (1 d;  $n = 18$ ) or at 3 mo of age ( $n = 15$ ). Lambs from ewes fed 100, 60, or 140% will be referred to as CON, RES, and OVER, respectively. At euthanasia, whole livers were harvested, weighed and tissue samples collected. Protein was determined by Western immunob-



lot. Proteins were imaged using an Odyssey CLx Imaging System and quantified using Image Studio Lite program. Data were analyzed using PROC GLM. As previously reported, BW were 13% greater in OVER vs. CON at 1 d and 3 mo. Liver weight was 43% greater in OVER vs. CON at 1 d when adjusted for BW, but not at 3 mo. Based on previously determined changes in gene expression in the liver, sterol-regulatory element binding protein-1 (SREBP-1), a regulatory gene of hepatic lipogenesis, IGF binding protein-4 (IGFBP-4) and IGFBP-3 were evaluated. At 3 mo, IGFBP-3 expression increased 16% in OVER vs. CON ( $P = 0.03$ ) and IGFBP-4 tended to increase 88% in RES vs. CON ( $P = 0.08$ ). Maternal diet did not affect protein expression of IGFBP-3 or -4 at 1 d or SREBP-1 at either time point ( $P \geq 0.11$ ). The observed increase in IGFBP-3 protein expression in OVER parallels previously determined increases in gene expression and circulating concentrations of IGFBP-3. In conclusion, poor maternal nutrition alters gene, protein, and circulating concentrations of key growth factors, such as IGFBP-3, which may contribute to altered growth of offspring.

**Key Words:** IGF binding protein, liver, maternal nutrition

**T66 Effects of rubber covers for concrete slats on lameness in confined feedlots.** Bryant R. Chapman\*, Derrick S. Smith, Colleen N. Curtiss, Monica J. Atkin, Steven R. Rust, and Daniel L. Grooms, *Michigan State University, East Lansing, MI.*

The second most significant health issue for fed cattle in the US is lameness and is viewed as a major welfare issue in animal agriculture. Lameness tends to be more problematic for cattle housed in facilities with slatted floors. The objective of this study was to evaluate the effectiveness of rubber covered slatted floors for reduction of lameness, tail injury incidence, and improvement of overall performance. Angus-based steers (250 to 300 kg) were assigned randomly to pens with non-covered concrete slats (NC;  $n = 4$ ) or concrete slats covered with rubber (RC;  $n = 4$ ). Each pen contained 7 steers at a stocking density of 6.9 m<sup>2</sup>/steer. Cattle were on feed for 110 to 131 d. Locomotion scores, tail lesions, left carpal joint circumference, and hoof dimensions were recorded. Average daily gain, feed conversion efficiency, cleanliness, and carcass traits were collected and reported in a companion abstract. Hide cleanliness was graded on a scale of 0 to 9, with 0 being less than 5% soiled and 9 being completely soiled. Locomotion scores were recorded on a 0 to 3 scale, where 0 was a normal gait and 3 was severely lame. Tail tip injury was recorded on a scale of 0 to 3, where 0 had no visible lesions and 3 had open wounds. Locomotion scores, left carpal joint circumference, and tail lesions did not differ between treatments. The toe length of cattle housed on the RC was longer than NC (77.3 vs. 84.0;  $P < 0.01$ ). A trend for sharper angle of the hoof was observed for cattle on NC compared with RC (55.5 vs. 52.5;  $P < 0.16$ ). A positive correlation was detected between the angle of the front hoof and the carpal joint circumference ( $r = 0.77$ ;  $P < 0.03$ ). Positive correlations were detected between hide soiling vs. the front toe length ( $r = 0.71$ ;  $P < 0.05$ ) and angles between the front and rear hooves ( $r = 0.62$ ;  $P < 0.10$ ). In summary, provision of rubber covers for concrete slats had minor effects on overall cattle lameness.

**Key Words:** lameness, welfare, tail lesion

**T67 Associations between animal performance measures and rumen pH of growing feedlot steers in drought simulated conditions.** Sara E. Place<sup>1</sup>, Michelle S. Calvo-Lorenzo<sup>1</sup>, Clint R. Krehbiel<sup>1</sup>, Christopher J. Richards<sup>1</sup>, Douglas L. Step<sup>2</sup>, Kristi Allwardt<sup>1</sup>, Catherine L. Haviland<sup>1</sup>, Emily A. Andreini<sup>1</sup>, Jacob Reed<sup>1</sup>, Andrew Grimes<sup>1</sup>, Ashley Brooks<sup>1</sup>, Justin L. Lyles<sup>1</sup>, Kyre E. Larrabee\*<sup>1</sup>,

Kimberly Branham<sup>1</sup>, Megan M. Rolf<sup>1</sup>, <sup>1</sup>*Department of Animal Science, Oklahoma State University, Stillwater, OK,* <sup>2</sup>*Veterinary Clinical Sciences, Oklahoma State University, Stillwater, OK.*

Located in the drought stricken Southern Great Plains, Oklahoma is an ideal site to investigate the associations between decreased water availability and cattle performance. The objectives of this research were to investigate the effects of restricted water intake (WI) on animal performance, and associations between animal performance and rumen pH. These data are part of a larger 5 yr project that will develop selection and management tools to adapt cattle to drought conditions. The data were collected from 117 cross-bred steers (*Bos taurus*) that were blocked by BW upon arrival to the Willard Sparks Beef Research Center at Oklahoma State University in Stillwater. Steers were housed in 4 identical pens each equipped with an Insentec feed and water intake system consisting of 6 feed bunks and 1 waterer per pen, which allowed for individual DMI and WI collection. Following a 21 d adaptation, a 70 d feed and WI collection trial was conducted to establish baseline WI for each individual steer. Following the 70 d trial, steers had their WI reduced 10% of baseline weekly for 4 wks to achieve a 50% of baseline WI restriction. After 1 wk of adaptation, cattle were maintained at 50% WI restriction for 35 d to simulate reductions of WI due to decreased water quality, quantity, and increased temperature that cattle would experience during drought events. Steers were weighed every 14 d. Prior to and after water restriction, rumen fluid was collected for each steer via rumenocentesis and the pH was immediately measured. Associations between DMI, WI, ADG, and rumen fluid pH before and after water restriction and the effects of water restriction on ADG were investigated using Proc Corr and Proc GLM procedures in SAS (SAS Institute, Cary, NC), respectively. Rumen fluid pH was not associated with any steer performance measures ( $P > 0.05$ ). Water restriction significantly reduced steer ADG and DMI by 23% and 19% ( $P < 0.001$ ), respectively, and rumen pH was increased during restriction ( $P = 0.0141$ ). These data suggest drought conditions negatively affect steer performance, emphasizing the importance of selection and management tools to adapt beef cattle production to drought conditions.

**Key Words:** climate adaptation, drought, cattle

**T68 Macrominerals requirements for growth of Canindé goats.** Luana P. S. Ribeiro\*<sup>1</sup>, Ariosvaldo N. Medeiros<sup>2</sup>, Francisco F. R. Carvalho<sup>3</sup>, Elzânia S. Pereira<sup>4</sup>, Anaiane P. Souza<sup>5</sup>, José Maurício S. Neto<sup>5</sup>, Cláudio G. S. Junior<sup>2</sup>, Gabriel C. L. Branco<sup>2</sup>, and Andreia B. Bezerra<sup>2</sup>, <sup>1</sup>*Federal University of Bahia, Salvador, Bahia, Brazil,* <sup>2</sup>*Federal University of Paraíba, Areia, Paraíba, Brazil,* <sup>3</sup>*Federal University Rural of Pernambuco, Recife, Pernambuco, Brazil,* <sup>4</sup>*Federal University Ceará, Fortaleza, Ceara, Brazil,* <sup>5</sup>*State University Sao Paulo, Jaboticabal, Sao Paulo, Brazil.*

Minerals are important constituents of structural tissues, metabolism and body fluids; however, the mineral nutritional requirements remain poorly quantified in goats. In fact, the values are estimated from other species. Therefore, the objective of this study was to predict the net requirement of Ca, P, Mg, Na, and K for growth of Canindé kids. Thirty 3 castrated kids (15.65 ± 0.41 kg of initial BW) were used, 5 of which were slaughtered at the beginning of the experiment to determine the initial body composition. The 28 remaining goats were distributed in a completely randomized design, kept in individual pens, and fed 4 levels (treatments) of intake: 1) ad libitum (100%), 2) 80%, 3) 60%, and 4) 40%. The diet consisted of 55% forage (Tifton) and 45% concentrate. The goats were slaughtered when the animals fed ad libitum reached 26.55 ± 0.65 kg of BW. The carcasses and organs were weighed, homogenized and sampled

for chemical analysis. A comparative slaughter method for assessing body composition and estimation of nutritional requirements was used (Garret et al., 1959). The data were analyzed by the REG procedure of SAS. Body composition were estimated from the predicted equation obtained by regressing the logarithm of the weight of each mineral in the empty body, for animals with 15 to 25 kg. The values (g kg<sup>-1</sup> EBW) for Ca, P, Mg, K and Na ranged from 10.49 to 12.59, 8.08 to 7.16, 0.18 to 0.44, 1.63 to 1.37, and 1.59 to 4.09, respectively. The net requirements for growth were estimated by the first derivative of the allometric equations. The values (g kg<sup>-1</sup> EBW) for Ca, P, Mg, K and Na ranged from 10.91 to 13.83, 5.00 to 4.67, 0.36 to 0.93, 1.40 to 1.75 and 3.34 to 9.06, respectively. This study indicates that indigenous goats have lower requirements of Ca, Mg and Na, and greater requirements of P and K when compared with current feeding system recommendations.

**Key Words:** body composition, goat, nutrition

**T69 Back to basics: Are beef cattle good at showing estrus? An assessment of estrus response, length of sexual receptivity, follicular growth, and pregnancy rate in beef cattle following a 5-day CO-Synch + CIDR estrus synchronization program.** Victoria Morrow\*, Shannon Edwards, Stephanie Webb, Jeremy Hemmer, Jennifer Bouland, Steve Parish, and Martin Maquivar, *Washington State University, Pullman, WA.*

The development of estrus synchronization protocols that facilitate fixed time artificial insemination (FTAI) in beef cattle has resulted in variable pregnancy rates. This variability has been attributed to different factors such as expression of sexual behavior, and follicular dynamics. The objective of the present study was to assess the onset of estrus response, estrus length, follicular growth, and size at AI using the 5 d-CO-Synch + CIDR program. One hundred and 3 females (81 cows and 22 heifers) were synchronized with an administration of 100 µg of GnRH IM and an intravaginal controlled internal drug release (CIDR) at d -5. At d 0, the CIDR was withdrawn and 2 doses of PGF2-α (25 mg) were administered via IM 6 h apart. Animals were observed continuously for behavioral signs of heat after CIDR removal until either 56 h (FTAI for heifers) or 72 h (FTAI for cows). At AI all animals received 100 µg of GnRH IM. Pregnancy diagnosis was performed approximately 60 d after FTAI. Results were analyzed by PROC MIXED procedure using a 2 × 2 factorial arrangement (pregnancy status and expression of heat). No differences were observed between heifers and cows. Overall, the proportion of animals showing sexual behavior was 45% (46/103), where 22% (10/46) did not get pregnant. No differences ( $P > 0.05$ ) were detected between animals that showed estrus and became pregnant ( $n = 36$ ) or showed estrus and were non pregnant ( $n = 10$ ) at onset of sexual receptivity (52:09 h vs 46:30 h) and length of estrus (6:05 h vs 6:36 h) respectively. Non pregnant animals without estrus expression had the smallest follicle size at AI ( $P = 0.04$ ), however no differences were observed in follicular growth. Pregnancy rate at FTAI was similar among animals regardless expression of estrus. In conclusion, the lack of reproductive response in animals submitted to a FTAI synchronization program can be related to a reduction in follicular size at FTAI and absence of estrus behavior.

**Key Words:** estrus expression, beef cattle, reproductive management

**T70 Single nose ringed swine behavior in free-range production system.** Patrícia M. Gomes\*<sup>1</sup>, Amanda R. R. Cabral<sup>1</sup>, Jacqueline N. Paiva<sup>1</sup>, Karoline M. Silva<sup>1</sup>, Frederico L. Silva<sup>1</sup>, Felipe H. Soares<sup>1</sup>, Carlos A. Silva Júnior<sup>1</sup>, Julia E. G. N. Perini<sup>2</sup>, Jessica M. Araujo<sup>1</sup>, Angela P. Santana<sup>1</sup>, and Luci S. Murata<sup>1</sup>, <sup>1</sup>*University of Brasilia,*

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Single nose ring is commonly used in free-range swine production to control pasture and soil degradation. To evaluate the behavior frequency in multiparous non-pregnant sows, a trial was carried out with 2 treatments, ringed and control, with 4 sows each. The trial was divided based on cicatrization process: inflammation, proliferation and remodeling, during 23 d. The chosen method to evaluate the behavior after nose ring insertion was scan, consisting in observation every 10 min, 6 h a day (morning and evening). Twelve categories of behavior were used in scan sampling: rooting, tree rubbing, tree bark chewing, digging, wallowing, grazing, alert, resting, positive social interaction, negative social interaction, drinking and sniffing. Each group of sows were allocated in a 1000-m<sup>2</sup> paddock, limited with solar electric fences, in exotic *B. decumbens* pasture and with push-lever bowl drinker. Sows were fed with commercial regular mash diet twice a day. Results showed that ringed sows spend 60.65% of the time resting, while control group spend 47.46% ( $P < 0.01$ ). Rooting behavior was not observed in the ringed group, whereas control group demonstrated it for 0.63% of the time. Digging was seen in ringed group, in a frequency of 0.24%, and was not observed in control sows. Sniffing in ringed group was seen in 3.52% of the time and 13.17% in the control group ( $P < 0.01$ ). During inflammation phase, rooting, tree rubbing and tree bark chewing were seen in control group (0.57%, 0.43%, 0.14% respectively) and were not observed in ringed group. In proliferation phase the ringed group already showed tree bark chewing and tree rubbing (both with 0.33%) whereas for the control group the frequencies were 0.55% and 0.22% (ns). In remodeling phase control group showed 0.88% for rooting, 0.88% for tree rubbing and 1.1% for tree bark chewing, while ringed sows were not observed with the first 2 behaviors and with 0.22% (ns) of the last. The trial showed that the ring insertion changed the behavior of the sows, resulting in more time resting. Rooting behavior was absent in ringed group during observations and signals of adaptations were seen, because digging was a behavior not seen in control group and present in ring inserted sows.

**Key Words:** sustainability, rooting, welfare

**T71 Conception rates of beef heifers and cows based on facilities.** Miriam A. Snider\* and Julie D. Weathers, *Southeast Missouri State University, Cape Girardeau, MO.*

Two breeding experiments conducted over a 2-year period were conducted to determine if conception rates in beef heifers and cows were affected in response to breeding facilities. The study compared the traditional squeeze chute and the insemination barn with timed artificial insemination programs. The heifer groups were given the 14 d CIDR-PG and cow groups the 7 d CO-Synch + CIDR protocol to determine whether conception rates are affected by insemination within a squeeze chute or insemination within an insemination barn. In Experiment 1, 17 beef heifers at the Barton Agricultural Research Center in Gordonville, Missouri were artificially inseminated following standard protocols. Cattle numbers were selected at random to determine which animals would be bred in the insemination barn or within the squeeze chute. Technicians were alternated so that each technician inseminated half of the animals in each facility setting. Ultrasound technology was used to determine which heifers were pregnant. Results indicated that 87.5% of heifers bred in the squeeze chute became pregnant as compared with 37.5% ( $P \leq 0.05$ ) of heifers bred in the insemination barn. Experiment 2 largely consisted of the same process. This time, however, 17 beef cows were inseminated. BioPryn (Moscow, ID) blood tests were used to confirm pregnancy within the cow population 30 d following insemination.

Results indicated that 88.89% of the cows bred in the squeeze chute became pregnant whereas only 75% ( $P \leq 0.10$ ) in the insemination barn became pregnant. This indicates on average a 31.94% ( $P \leq 0.05$ ) higher rate of pregnancy from breeding in a traditional squeeze chute compared with the insemination barn. Future studies will include larger sample sizes of cattle and take into account technician preferences.

**Key Words:** conception, insemination barn, squeeze chute

**T72 A comparison of Dorper and Blackface lambs in growth and carcass performance.** Kayley R. Wall\* and Chris R. Kerth, *Texas A&M University, College Station, TX.*

To determine the suitability of a purebred Dorper as a meat animal in a production setting, the growth and carcass traits of Blackface April-born lambs ( $n = 8$ ) grown on a concentrate diet were compared with Dorper September-born lambs ( $n = 8$ ). All lambs were fed a concentrate diet (16% CP, 3% crude fat, 9% crude fiber) ad libitum in separate pens for 18 d before the start and for the duration of the study (62 d). The initial (0 d) and final (62 d) BW, percent change in BW, ADG, and G:F were measured. One Blackface lamb died during the feeding trial. All of the lambs were harvested on d 62 and placed into the blast chiller (4°C) for 15 h after hot carcass weights were recorded. Carcass data were collected 16 d postmortem. The carcasses were ribbed and allowed to bloom for 1 h before marbling, fat thickness, LM area, leg confirmation score, KPH %, flank streaking, and lean color ( $L^*$ ,  $a^*$ , and  $b^*$ ) were collected. Data were analyzed using a one-way ANOVA of a completely randomized design with animal as the experimental unit and  $\alpha$  set at 5%. Average daily gain and G:F were not affected ( $P > 0.47$ ) by lamb breed, but percentage change in BW tended ( $P = 0.099$ ) to be higher in Dorper sheep compared with Blackface sheep. Fat thickness, marbling, flank streaking and KPH % were higher ( $P < 0.014$ ) in the Blackface than Dorper lambs. The Blackface lambs also had heavier ( $P < 0.001$ ) live and hot carcass weights than the Dorper lambs; however, there were no significant differences ( $P > 0.13$ ) between the breeds in LM area and dressing percentage, despite the age difference of the lambs. Dorper lambs had lighter (higher  $L^*$  values;  $P = 0.001$ ) and less red (lower  $a^*$  values;  $P = 0.041$ ) lean, but  $b^*$  values were not affected ( $P = 0.62$ ) by breed. Although the Blackface lambs were fatter and had higher BW and HCW compared with the Dorper lambs, they did not outperform them in growth, dressing percent, or LM area. We believe the Dorper could be a suitable competitor to the Blackface in a production system as a meat-producing animal.

**Key Words:** Dorper, lamb

**T73 Gait score of broilers supplemented with vitamin D (25-OHD<sub>3</sub>).** Grace Alessandra Araujo Baldo\*<sup>1</sup>, Ibiara Correia de Lima Almeida Paz<sup>1</sup>, Edivaldo Antônio Garcia<sup>1</sup>, Andréa Britto Molino<sup>1</sup>, and Marlon Sávio Amadori<sup>2</sup>, <sup>1</sup>*School of Veterinary Medicine of Animal Science, UNESP, Botucatu, SP, Brazil,* <sup>2</sup>*School of Agricultural Sciences, UFGD, Dourados, MS, Brazil.*

The aim of this study was evaluated the gait score of broilers supplemented with vitamin D. For this, we used 2400 broilers distributed in a randomized blocks allocated in factorial design  $2 \times 2 \times 2$ , being males and females, strains Cobb 500 and Ross 308, supplemented or not with vitamin D (25-OHD<sub>3</sub>). The diets were formulated following the requirements of each phase and added to 69 mg of 25-OHD<sub>3</sub>/t of feed in the supplemented treatments. After 41 d evaluated the gait score and, for this, 100% of the birds were put to walk in the path of a bounded linear meter inside the boxes. The way the birds walked was ranked scores

ranging from 0 to 2, being gait score 0 bird that walked normally, gait score 1 bird which was difficult to walk and gait score 2 bird that walked with difficulty. We used the Chi-Square at a significance level of 5%. Females had higher ( $P < 0.05$ ) frequency of gait score 0 (88.41%) and males had a higher frequency of gait score 1 (16.26%). The Cobb 500 strain of birds had higher ( $P < 0.05$ ) incidence of gait score 0 (87.79%) when compared with the Ross 308 (83.10%). Supplementation with vitamin D (25-OHD<sub>3</sub>) did not affect ( $P > 0.05$ ) the gait score of birds. Therefore, it was concluded that vitamin D (25-OHD<sub>3</sub>) did not improve the way of walking (gait score) of broilers.

**Key Words:** animal welfare, poultry production, way of walking

**T74 Effects of corn particle size and diet form on nursery pig growth performance.** Ashton D. Yoder\*, Grace E. Bokelman, and Cassandra K. Jones, <sup>1</sup>*Kansas State University, Manhattan, KS.*

A total of 180 nursery pigs (PIC 327  $\times$  1050; 36 d of age; initially 16.1 kg) were utilized in a 35-d experiment to determine the effects of corn particle size and pelleting on nursery pig growth performance. All diets were nutritionally similar, but processing parameters created 4 experimental treatments in a  $2 \times 2$  factorial with 2 corn particle sizes (500  $\mu\text{m}$  vs. 750  $\mu\text{m}$ ) and 2 diet forms (mash vs. pelleted). Pigs were weaned on d 26 of age, blocked by initial BW, and fed a pelleted acclimation diet for 10 d. On d 0 of the experiment, pigs were randomly assigned to pens in a randomized complete block design with 5 pigs per pen and 9 replications per treatment. Experimental diets were fed in 2 phases: d 0 to 14 and d 14 to 35. Data were analyzed using the GLIMMIX procedure of SAS. Reducing particle size from 750 to 500  $\mu\text{m}$  did not affect growth performance ( $P > 0.44$ ). Pelleting reduced ( $P < 0.05$ ) feed intake compared with feeding mash diets during each phase and overall, but did not affect ADG or G:F ( $P > 0.11$ ). The interaction between particle size and diet form affected G:F from d 0 to 14, where finely ground mash diets or coarsely ground pelleted diets had greater feed efficiency than finely ground pelleted diets or coarsely ground mash diets ( $P = 0.04$ ; 0.72, 0.68, 0.68, and 0.74 for 500  $\mu\text{m}$  mash, 500  $\mu\text{m}$  pelleted, 750  $\mu\text{m}$  mash, and 750  $\mu\text{m}$  pelleted diets, respectively). Additionally, the interaction tended to affect ADG from d 0 to 14 and overall where pigs fed finely ground mash diets had greater ADG than those fed finely ground pelleted diets ( $P = 0.06$ ; 0.46 vs. 0.40 kg/d for d 0 to 14 and  $P = 0.10$ ; 0.53 vs. 0.48 kg/d for d 0 to 35, respectively). However, this was offset by a tendency for the interaction to affect ADFI from d 14 to 35 as pigs fed finely ground mash diets had greater feed disappearance than those fed finely ground pelleted diets ( $P = 0.08$ ; 1.18 vs. 1.08 kg/d, respectively). These findings are similar to recently published research in both nursery and finishing pigs that suggest there is little advantage of pelleting diets with a fine particle size.

**Key Words:** mash, pellet, nursery pig

**T75 Effect of herbal liniment on equine back pain over time: a preliminary study.** Shannon M. Heibeck\*<sup>1</sup>, Kelly W. Walter<sup>1</sup>, Jay A. Altman<sup>2</sup>, Brady J. Karren<sup>2</sup>, Miriam B. Altman<sup>3</sup>, and Kevin K. Haussler<sup>4</sup>, <sup>1</sup>*Agricultural Science Department, Truman State University, Kirksville, MO,* <sup>2</sup>*Arenus, Fort Collins, CO,* <sup>3</sup>*Organic Exchange Ltd., Fort Collins, CO,* <sup>4</sup>*Clinical Sciences Department, Colorado State University, Fort Collins, CO.*

Previous research established pressure algometry as a quantitative and repeatable method of assessing induced musculoskeletal back pain in horses; however there is no data evaluating large areas over time in response to pain-mitigating products. Stock-type horses ( $n = 18$ ) were



used in a completely randomized 44 d trial to investigate the efficacy of an herbal liniment solution to mitigate musculoskeletal back pain. Horses began the study after a 30 d rest and all were maintained in similar daily light work through use in university horsemanship classes and equestrian team practices for the entire 44 d. Horses were randomly assigned to treatment groups which consisted of a commercially available herbal liniment gel solution (SoreNoMore Ultra, Arenus) or a control gel solution (identical gel solution minus active ingredients) applied to the back daily following exercise at 0.02 cc per square cm. Evaluation of back pain was standardized by dividing the back into 4 equal quadrants lengthwise beginning at the highest point of the withers and ending at the sacroiliac joint. Quadrants extended 17.78 cm ventrally from the spine. The third quadrant (on left and right side) was selected as the area of interest and was clipped to maintain consistency over time. Nine data points in this quadrant were selected (top-left, mid-left, bottom-left, top-mid, center, bottom-mid, top-right, mid-right, bottom-right) for weekly evaluation using a force gage pressure meter (by a single nonblinded examiner). A negative pain response was classified as no reaction to the application of 5.9 kg pressure. Data was inverted to create a 0 to 13 pain score based on the amount of pressure applied. Data were analyzed using the PROC MIXED procedure of SAS with main effects tested being treatment, time, and treatment  $\times$  time interaction. Treated horses decreased mean pain score from beginning to end of trial (2.87 decrease;  $P < 0.001$ ) compared with control horses (0.72 decrease;  $P \leq 0.18$ ). In summary, standardized evaluation procedure using pressure algometry suggests the herbal liniment gel solution was able to mitigate pain response when applied daily over a 44-d period.

**Key Words:** horse, back pain, liniment

**T76 Evaluation of inter-day variation of horses on total fecal collection.** Elizabeth F. Miller<sup>\*1</sup>, Francesca R. Melgar<sup>2</sup>, Trevor D. Morgan<sup>2</sup>, Shanna L. Ivey<sup>2</sup>, Clint L. Loest<sup>2</sup>, Laura M. White<sup>2</sup>, and Kelly W. Walter<sup>1</sup>, <sup>1</sup>*Agricultural Science Department, Truman State University, Kirksville, MO*, <sup>2</sup>*Department of Animal and Range Sciences, New Mexico State University, Las Cruces, NM*.

Environmental changes and stress may influence passage rate and therefore digestibility in horses participating in a total fecal collection where

they are individually housed and fitted with fecal collection harnesses. Existing equine research methods sections frequently do not include descriptions of adaptation periods to allow horses to acclimate to the potential stressors of total fecal collections. Utilization of collection harnesses and confined housing could impact data collection of horse nutrition trials. Therefore, the objective of the current study is to evaluate inter-day variation of dry matter intake (DMI), output (DMO), and resulting calculation of digestibility (DMD) of horses on a 5-d total fecal collection. Twelve mature Quarter horse geldings ( $13.8 \pm 5.6$  y;  $514.4 \pm 38.9$  kg) were utilized in a completely randomized design. Horses were fitted with fecal collection harnesses and housed individually in  $3.7 \text{ m} \times 3.7 \text{ m}$  stalls with ad libitum access to water and mineral block. All horses were fed 1.85% BW coastal bermudagrass hay (AF; 8.92% CP and 33.47% ADF) and 1 kg (AF) of a pelleted concentrate supplement (divided into 2 equal meals) beginning 21 d before the start of total fecal collection and continuing through the 5 d collection. Individual daily intake was recorded, and fecal weights were obtained every 6-h during each day of collection. Subsamples of feed and feces were obtained to determine dry matter using standard AOAC methods. Individual days of collection were separated by all possible pairwise comparisons using PROC MIXED of SAS. Day of collection affected DMO, with increased DMO on d 1 compared with d 3 and 4 ( $P < 0.05$ ), and a tendency ( $P \leq 0.07$ ) for increased DMO on d 1 compared with d 2 and 5. This suggests the start of fecal collection may have altered passage rate to increase DMO. The differences of d 1 DMO affected calculated DMD between days (d 1 and 3,  $P \leq 0.04$ ; d 1 and 5,  $P \leq 0.09$ ). This highlights the importance of adapting horses to collection techniques before data collection regardless of diet or treatments to be applied, and warrants adequate description within the methods section of research reports.

**Key Words:** fecal collection, horse

# Breeding and Genetics: Application and methodology in animal breeding—Beef

**T77 Genetic parameters for breeding phase in Hereford x Aberdeen Angus crosses in grazing system of Uruguay.** Ana C. Espasandin\*<sup>1</sup>, Diego Gimeno<sup>2,1</sup>, Jorge Urioste<sup>1</sup>, and Fernando Pereyra<sup>1</sup>, <sup>1</sup>Udelar, School of Agronomy, Bernardo Rosengurt Experimental Station, Cerro Largo, Uruguay, <sup>2</sup>Uruguayan Wool Secretariat, Montevideo, Uruguay.

A diallelic experiment between Hereford (HH) and Aberdeen Angus (AA) beef breeds, developed between 1994 and 2002 at the Bernardo Rosengurt Experimental Station, College of Agriculture-University of the Republic, Uruguay (32°35' S, 54°15' W, and 94 of altitude). The data set totalized a set of 57 sires and 1365 dams. The individual ( $g_A^i - g_H^i$ ) and maternal ( $g_A^m - g_H^m$ ) breed differences, as well as individual ( $h_{AH}^i$ ) and maternal ( $h_{AH}^m$ ) heterosis were estimated using additive linear models for cow body condition score at calving (BCS), calf birth weight (BW) and weaning weight (WW), and generalized linear models for weaning percentage (%W). Effects of genetic group, year and month(year) at birth, dam age, and calf sex were included as fixed effects, and individual or sire as random effects. The effect of the difference between individual breed genetic ( $g_A^i - g_H^i$ ), as well the heterosis were not significant for BW ( $P > 0.05$ ). For WW, the difference between maternal effects ( $g_A^m - g_H^m$ ) was positive, with  $4.87 \pm 1.01$  kg ( $P < 0.0001$ ), and individual heterosis was  $5.12 \pm 1.24$  kg ( $P < 0.0001$ ). The  $h_{AH}^i$  for %W was  $0.22 \pm 0.03$ . The  $h_{AH}^m$  was significant for WW ( $11.58 \pm 1.48$  kg,  $P < 0.0001$ ). Crossbred dams were higher than pure breed by 0.04 points in BCS (3.79 vs. 3.75), being different HH and HA vs. AA and HA ( $P < 0.05$ ). At birth, calves HH, AH and HA were heavier than AA. For %W, crossbred dams averaged 24.7 points higher than pure breeds (85.0% vs. 60.3%). Crossbred breed dams weaned calves significantly heavier (means  $159.1 \pm 4.1$  kg) to purebred cows. Hereford breed dams weaned calves with significantly less weight (means  $143.3 \pm 2.7$  kg). The estimations of breed genetic effects show reductions in birth weight related to a Angus breed ( $g^i$ ), as well an increment in weaning weight due to a maternal Angus effect ( $g^m$ ). Superiority in crossbred dams were evidenced in individual and maternal heterosis.

**Key Words:** breed effect, crossbreeding, heterosis

**T78 Estimates of genetic parameters for reproductive traits in Brahman cattle breed.** Ligia Cavani\*<sup>1</sup>, Diogo Anastácio Garcia<sup>2</sup>, Luis Orlando Duitama Carreño<sup>2</sup>, Rafael Keith Ono<sup>2</sup>, Henrique Torres Ventura<sup>3</sup>, and Ricardo Fonseca<sup>1</sup>, <sup>1</sup>São Paulo State University (Unesp), Dracena, São Paulo, Brazil, <sup>2</sup>São Paulo State University (Unesp), Jaboticabal, São Paulo, Brazil, <sup>3</sup>Brazilian Association of Zebu Breeders (ABCZ), Uberaba, Minas Gerais, Brazil.

This study was designed to estimate genetic parameters for the following traits of Brahman cattle in Brazil: age at first calving (AFC), calving interval (CI), rebreeding (REB), and stayability (STAY). The data were provided by Brazilian Association of Zebu Breeders (ABCZ), and included Brahman cattle that were born between 1994 and 2012. For REB, the value 1 was assigned to heifers that rebreed and calved after first calving, and the value 0 was assigned to heifers that failed to rebreed after first calving. Likewise, for STAY, the value 1 was assigned to heifers that calved at least 3 times when they were 6-yr-old; otherwise, the value 0 was assigned. We utilized bivariate analysis using linear-threshold animal model to estimate co-variances components by using the software GIBBSF90 and THRGIBBS1F90 from BGF90. The

model included fixed effects, random effects that represented additive genetic direct effects of each animal, permanent environmental effects, included as random, and residuals. For AFC and STAY, the fixed effects were composed only by contemporary group. Besides that, for REB and CI, the fixed effects were composed by contemporary group, calf birth year and calf birth season. Also, the covariate dam age at calving was included for CI. The permanent environmental effects only were included in the model for CI. The contemporary groups were composed from farm, type of gestation (AI or natural service) and birth year of dam; and in addition, only for STAY, birth season of dam (rainy or dry). The mean  $h^2$  were 0.10, 0.02, 0.22, and 0.10 for AFC, CI, REB, and STAY; respectively. The genetic correlations were  $-0.13$  between AFC and CI,  $-0.35$  between AFC and REB,  $-0.57$  between AFC and STAY, and  $0.32$  between REB and STAY, which reveal that cows that remain productive for longer periods in the herd, also start breeding younger and present greater chances to REB; however, heifers that calved too young may present problems, such as calving difficulty, that may increase CI. The selection of Brahman cattle for reproductive traits, such as AFC, CI, REB, and STAY, will render low magnitude and long-term responses.

**Key Words:** heritability, rebreeding, stayability

**T79 Assessment of genetic variability of Brazilian Brahman cattle raised either on pasture or stabled by pedigree analysis.** Ligia Cavani\*<sup>1</sup>, Rafael Medeiros de Oliveira Silva<sup>2</sup>, Michel Marques Farah<sup>3</sup>, Rafael Keith Ono<sup>2</sup>, Luis Orlando Duitama Carreño<sup>2</sup>, Henrique Torres Ventura<sup>4</sup>, and Ricardo Fonseca<sup>1</sup>, <sup>1</sup>São Paulo State University (Unesp), Dracena, São Paulo, Brazil, <sup>2</sup>São Paulo State University (Unesp), Jaboticabal, São Paulo, Brazil, <sup>3</sup>Brazilian Agricultural Research Corporation (Embrapa), Juiz de Fora, Minas Gerais, Brazil, <sup>4</sup>Brazilian Association of Zebu Breeders (ABCZ), Uberaba, Minas Gerais, Brazil.

The aim of this study was to assess the genetic variability of Brahman cattle in Brazil by analyzing information obtained by genealogical records. The pedigree data were obtained from the Brazilian Association of Zebu Breeders and was formed by 207,747 Brahman animals from different regions of Brazil. In this data set, cattle were born between 1994 and 2012 and they were analyzed in 3 ways: considering all the pedigree information (Pt), and dividing the pedigree data according to animals raised either on pasture (Ppt) or stabled (Pst). The software R was used for data consistency and to generate the file with the following information: animal, sire, dam, birth date, and sex. The software Pedig was used to determine the inbreeding, generation intervals and the results of the parameters based on the probability of gene origin: number of founders (Nf), effective number of founders (fe), effective number of ancestors (fa) and founder genome equivalents (fg). Average inbreeding coefficients (F) were 0.31%, 0.30% and 0.22% for Pt, Ppt and Pst, respectively. However, mean F values for inbred animals were 5.94%, 5.13% and 6.52% for Pt, Ppt and Pst, respectively. In addition, average generation intervals were 4.73, 4.65 and 4.81 years for Pt, Ppt and Pst, respectively. For Pt, the results based on probability of gene origin were 13,764, 101.4, 97.07, 75.02, 1.04, 0.74 for Nf, fe, fa, fg, fe/fa and fg/fe; respectively. For Ppt, the results based on probability of gene origin were 6,906, 94.4, 91.57, 72.93, 1.03, 0.80 for Nf, fe, fa, fg, fe/fa and fg/fe; respectively. For Pst, the results based on probability of gene origin were 2,407, 68.8, 66.36, 58.42, 1.04, 0.88 for Nf, fe, fa,

fg, fe/fa and fg/fe; respectively. The number of ancestors to explain 50% of the genetic variability in Pt was 46, in Ppt was 41 and in Pst was 24. The results showed a reduction in genetic variability, because  $fe > fa > fg$ . Values close to 1 observed for fe/fa and fg/fe showed no genetic bottleneck and a small process of genetic drift. Thus, genetic variability has been reduced in Brahman cattle population, especially on those from Pst.

**Key Words:** ancestor, inbreeding

**T80 Use of a robust Bayesian regression model in genome-wide association study of growth traits in Brangus heifers.** Sunday O. Peters\*<sup>1</sup>, Kadir Kizilkaya<sup>2</sup>, Dorian J. Garrick<sup>3</sup>, Rohan L. Fernando<sup>3</sup>, Ikhide G. Imumorin<sup>4</sup>, and Milton G. Thomas<sup>5</sup>, <sup>1</sup>Berry College, Mount Berry, GA, <sup>2</sup>Adnan Menderes University, Aydin, Turkey, <sup>3</sup>Iowa State University, Ames, IA, <sup>4</sup>Cornell University, Ithaca, NY, <sup>5</sup>Colorado State University, Fort Collins, CO.

When alternative models are used for genome-wide association studies (GWAS), not a single one of them has been recognized as universally best across all traits. One explanation is that different traits may have different genomic architecture characterized by different distributions for marker effects. Further, it is known that results from BayesA and BayesB models can be heavily influenced by the value of hyperparameters assumed known, namely  $\pi$ , the scale factor and degrees of freedom. Our objective was to use a robust Bayesian regression model for GWAS that treated these hyperparameters as unknown and apply this model to 3 growth traits in Brangus (3/8 Brahman  $\times$  5/8 Angus) heifers. Genotypes for each heifer were obtained from BovineSNP50 Infinium beadchips. Phenotypes included data on birth weight (BW), weaning weight (WW) and yearling weight (YW) from 830 individuals from 67 sires. Simultaneous association of all SNP for each of BW, WW and YW were tested in a robust model that treated SNP effects as random. Fixed effects included cohort groups as class effects (defined for animals with the same calving season, location, and trait contemporary group) and dam age (in years) as classes. Several genomic locations were associated with variation in these growth traits in heifers. The top 10 regions by SNP effects were found on chromosomes 1, 2, 5, 6, 14 and 26 for BW, chromosomes 1, 2, 3, 5, 6, 16 and 28 for WW and chromosomes 5, 6, 7, 11, 12, 14, 16, 18, 20 and 29 for YW. Results confirm many previously reported regions associated with variation in these growth traits in both taurine and indicine cattle breeds but also included new associations. Results demonstrated the utility of regression models with unknown hyperparameters of  $\pi$ , scale and degrees of freedom in GWAS of growth traits in Brangus heifers.

**Key Words:** robust Bayesian regression, genome-wide association, growth

**T81 Genetic parameter estimates for fatty acid profile of *longissimus thoracis* beef from Nellore cattle finished in feedlot.** Carolyn A. Santos<sup>1</sup>, Angélica S. C. Pereira<sup>2</sup>, Fabieli L. B. Feitosa<sup>1</sup>, Marcos V. A. Lemos<sup>1</sup>, Hermenegildo L. J. Chiaia<sup>1</sup>, Mariana P. Berton<sup>1</sup>, Adrielle M. Ferrinho<sup>2</sup>, Lenise F. Mueller<sup>3</sup>, Joyce M. Furlan<sup>2</sup>, Monica R. Mazalli<sup>3</sup>, and Fernando S. Baldi\*<sup>1</sup>, <sup>1</sup>Universidade Estadual Paulista, Faculdade de Ciências Agrárias e Veterinárias, Departamento de Zootecnia, Jaboticabal, São Paulo, Brazil, <sup>2</sup>Universidade de São Paulo, Faculdade de Medicina Veterinária e Zootecnia, Departamento de Nutrição e Produção Animal, Pirassununga, São Paulo, Brazil, <sup>3</sup>Universidade de São Paulo, Faculdade de Zootecnia e Engenharia de Alimentos, Pirassununga, São Paulo, Brazil.

The present study aimed to estimate (co)variance components and genetic parameters for beef fatty acid (FA) composition in 937 Nellore bulls, finished in feedlot (period of 90 d) and slaughtered with an average age of 24 mo, weighing 500–550 kg. The FA profile was analyzed in *Longissimus thoracis* samples using a gas chromatography, with a 100-m capillary column. The contemporary groups (CG) were organized based on farm and year of birth, and management group at yearling. The following 14 FAs were quantified: C16:0, C18:0, C18:1 *cis*-9, C18:2 *cis*-6, C18:2 *cis*-9,*trans*-11 and C18:2 *trans*-10,*cis*-12, C18:3n-3, C14:0, C14:1, C18:1n-9 *trans*, 18:1 *trans*-11, C20:3n-6 *cis*-8,11,14, C20:3n-3 *cis*-11,14,17, and C20:4. The (co)variance components were estimated using an animal model which included random additive genetic effects, fixed effects (CG), and the slaughter age as a covariable (linear and quadratic effect). The restricted maximum likelihood method was used (REMLF90 computer program) and 2 multi-trait analysis were performed, one including 14 individual FA and the other the total SFA, MUFA, PUFA, n-3, n-6 and n-9. The individual FAs with the highest heritability estimates were C18:2 *trans*-10,*cis*-12 (0.38) and C18:3n-3 (0.41). The C18:0, C16:0, C18:1 *cis*-9, 18:1 t11, C18:2 *cis*-9,*trans*-11 and C20:4 FAs resulted in low heritability estimates, indicating that these FAs should respond slowly to selection. With the exception of the C18:2 *trans*-10,*cis*-12 FA, the genetic associations between individual PUFAs were high and positive. The genetic correlation estimates between the total of SFA with PUFA and n-6 were high and negative (–0.84 and –0.88), and with n-3 were moderate (–0.62). It is expected that the response to direct selection for polyunsaturated, n-3 and n-6 fatty acids would be fast. In general, the correlations between the most important fatty acids are favorable and therefore indirect selection may be used to improve the fatty acid profile. This study revealed the existence of genetic variation and hence the possibility of genetic improvement of meat fatty acid composition in zebu cattle.

**Key Words:** *Bos indicus*, genetic variation, meat

**T82 Genetic evaluation of growth traits in Brahman cattle.** A. M. Matorano<sup>2</sup>, M. E. Z. Mercadante<sup>3</sup>, J. N. S. G. Cyrillo<sup>3</sup>, L. A. L. Chardulo<sup>1</sup>, and J. A. V. Silva\*<sup>1</sup>, <sup>1</sup>Universidade Estadual Paulista, Botucatu, SP, Brasil, <sup>2</sup>Universidade Estadual Paulista, Jaboticabal, SP, Brasil, <sup>3</sup>Instituto de Zootecnia, Sertãozinho, SP, Brasil.

The aim of this study was to estimate genetic parameters and determine ratio efficiency of selection for weaning weight (W205), weight at 450 d (W450) and weight gain from weaning to 450 d (PWG450) in Brahman cattle. Traits were studied through multivariate analyses. Variance components were estimated by restricted maximum likelihood performed by the software MTDFREML. The Statistical model included the fixed effects of contemporary group (composed by farm, crop and birth season, sex, and weaning and yearling management groups) and the linear covariate age, the random effect of animal and residual. For weaning weight, the management group at yearling was excluded from the contemporary group and the fixed effect of dam at calving age (3 to 9 years and more than 9 years) and the effect of permanent environmental were included. Purpose of ratio efficiency was verifying the efficiency of genetic gain as a function of indirect selection. Observed means of W205, W450 and PWG450 were  $197 \pm 34$ ,  $320 \pm 74$ ,  $118 \pm 59$ . Estimated heritability of W205, W450, and PWG450 were 0.18, 0.16 and 0.12. W205 and W450 had very high genetic correlation (0.89). GPW450 had high genetic correlation with W450 (0.79), but relatively lower correlation with W205 (0.43). For all traits, the direct selection was more efficient. Information obtained suggests slow genetic gain for W205, W450 and GPW450. GPW450 was appropriate to use as



selection criteria because it showed less influence of weaning weight, and high correlation with W450.

**Key Words:** beef cattle, genetic correlation, weaning weight

**T83 Effect of prediction method and cross-validation approach on accuracy of DGV for feed efficiency traits.** Rafael M. O. Silva\*<sup>1</sup>, Daniela A. L. Lourenco<sup>2</sup>, Breno O. Fragomeni<sup>2</sup>, Luciana Takada<sup>1</sup>, Rafael Espigolan<sup>1</sup>, Maria E. Z. Mercadante<sup>3</sup>, Fernando Baldi<sup>1</sup>, Guilherme C. Venturini<sup>1</sup>, Joslaine N. S. G. Cyrillo<sup>3</sup>, Ignacy Misztal<sup>2</sup>, Roberto Carneiro<sup>1</sup>, and Lucia G. Albuquerque<sup>1</sup>, <sup>1</sup>Univ Est Paulista Julio de Mesquita Filho, FCAV-UNESP, Jaboticabal, SP, Brazil, <sup>2</sup>The University of Georgia, Athens, GA, <sup>3</sup>APTA Center for Beef Cattle, Animal Science Institute, Sertãozinho, SP, Brazil.

Accuracies of direct genomic values (DGV) for feed efficiency traits obtained with different methods and cross-validation approaches were compared. After quality control 437,197 SNP genotypes were available for 761 Nellore cattle provided by the Institute of Animal Science, SP, Brazil. Methods of analysis were traditional BLUP, ssGBLUP, GBLUP, and BayesC $\pi$ . The traits were residual feed intake (RFI), feed conversion ratio (FCR), average daily gain (ADG) and dry matter intake (DMI). Model included fixed effects of contemporary groups (sex, year of birth, and pen), month of birth, and the covariable age of dam (linear and quadratic effects); and, as random, the additive animal effects. Three cross-validation approaches were considered: WPRO – validation was done on animals that did not have progeny; UNREL – the data set was split into 3 less related subsets; RAN – the data set was randomly divided into 4 subsets and the validation was done in each subset at a time. The accuracy of DGV was calculated as the Pearson correlation between corrected phenotype and the DGV divided by square root of heritability. Accuracies ranged from 0.01 (with UNREL) to 0.78 (with RAN) for studied traits. The inclusion of genomic information increased more than 10% of the average accuracy of predictions over traditional BLUP; on average, GBLUP showed more accurate predictions of DGV than BayesC $\pi$  (0.33 and 0.29, respectively). For the RAN cross-validation approach, accuracies were 50% higher with ssGBLUP than GBLUP, especially for traits with high heritabilities (ADG-  $h^2 = 0.55$ , and DMI -  $h^2 = 0.58$ ). The most accurate predictions were obtained using RAN, ranging from 0.28 to 0.78. On the other hand, the UNREL cross-validation approach provided the less accurate predictions, ranging from 0.001 to 0.29. With WPRO accuracies of DGVs were from 0.12 to 0.69. These results show that accurate genomic prediction can be obtained for all analyzed traits, especially for those with high heritability. Accuracies of DGV are higher when animals in validation are more related to those in training. São Paulo Research Foundation (FAPESP) grant 2013/01228-5 associated to 2009/16118-5.

**Key Words:** beef cattle, genomic selection

**T84 Genetic correlation estimates between the profile of individual beef fatty acids of meat and carcass traits and meat in Nellore cattle finished in feedlot.** Fabieli Loise Braga Feitosa, Carolyn Aboujaoude, Marcos Vinícius Antunes de Lemos, Mariana Piatto Berton, Guilherme Costa Venturini, Rafael Lara Tonussi, Rafael Espigolan, Bianca Ferreira Olivieri, Lucia Galvão de Albuquerque, and Fernando Baldi\*, *Universidade Estadual Paulista, FCAV-UNESP, Jaboticabal, São Paulo, Brasil.*

The aim of this study was to evaluate the genetic-quantitative relationships between the beef fatty acid profile with carcass and meat traits and meat in Nellore cattle. Data from 1616 Nellore bulls finished in

feedlot (90 d), aged around 2 years old were used. Samples were taken from the *Longissimus thoracis* muscle in the region of the 12th to 13th ribs of the left half-carcasses 48 h after slaughter. The backfat thickness (BF) was measured using a caliper. The percentage of lipids (LIP) was determined by the Bligh and Dyer method. The fatty acid profile was quantified using SP-2560 capillary column (100 m  $\times$  0.25 mm diameter with 0.02 mm thickness, Supelco, Bellefonte, PA). After quality control for genotypes, a total of 470,007 SNPs from 1,556 samples remained. The (co)variances and genetic parameters were estimated using the restricted maximum likelihood method, considering an animal model (ssGBLUP). The heritability estimates for fatty acids were from low to high, ranging from 0.06 to 0.65 for saturated fatty acids, from 0.07 to 0.14 for the monounsaturated fatty acids and 0.05 to 0.62 for polyunsaturated fatty acids. Carcass and meat traits showed low to moderate heritability estimates, ranging from 0.06 (SF) to 0.17 (EGS). Estimates of genetic correlation between BF with (C14:0), (C18:0), (C18:1n-9 cis), (C18:2n-6 cis), (C18:3n-3), CLA cis-9,trans-11 and CLA cis-10,trans-12 were low to moderate, being 0.38, -0.15, -0.44, 0.18, -0.22, 0.19 and -0.10, respectively. The estimates of genetic correlations between SF with previous fatty acids were low to moderate (0.16, 0.04, 0.04, 0.02, -0.40, 0.35 and 0.60). Estimated genetic correlations between the LIP with C14:0, C18:0, C18:1n-9 cis, C18:2n-6 cis, C18:3n-3, CLA cis-9,trans-11 and CLA cis-10,trans-12 were low to high, being 0.75, 0.70, -0.02, -0.93, -0.62, -0.04 and -0.60, respectively. It is expected that selection for increasing the concentration of mono and polyunsaturated fatty acids by direct selection for carcass and meat traits is less likely to occur.

**Key Words:** lipid composition, meat quality, *Bos indicus*

**T85 Genetic parameter estimates for carcass and meat quality traits in Nellore cattle finished in feedlot.** Bianca Ferreira Olivieri<sup>1</sup>, Fabiele Loise Braga Feitosa<sup>1</sup>, Hermenegildo Lucas Justino Chiaia<sup>1</sup>, Guilherme Costa Venturini<sup>1</sup>, Carolyn Aboujaoude<sup>1</sup>, Marcos Vinícius Antunes de Lemos<sup>1</sup>, Mariana Piatto Berton<sup>1</sup>, Rafael Lara Tonussi<sup>1</sup>, Rafael Espigolan<sup>1</sup>, Joyce de Jesus Mangini Furlan<sup>2</sup>, Angélica Simone Cravo Pereira\*<sup>2</sup>, Lucia Galvão de Albuquerque<sup>1</sup>, and Fernando Baldi<sup>1</sup>, <sup>1</sup>Universidade Estadual Paulista, FCAV-UNESP, Jaboticabal, São Paulo, Brazil, <sup>2</sup>Universidade de São Paulo, FMVZ-USP, Pirassununga, São Paulo, Brazil.

The aim of this study was to estimate genetic parameters for beef fatty acids profile, carcass and meat traits in Nellore cattle. A total of 1616 Nellore bulls finished in feedlot (90 d), and slaughtered with an average of 24 mo of age were used. Samples from the *Longissimus thoracis* muscle were collected 48 h after slaughter. The proportion of saturated (SFA), monounsaturated (MUFA) and polyunsaturated fatty acids (PUFA), n-3 and n-6 were calculated. The total lipid concentration (LIP) was quantified using the Bligh and Dyer (1959) method. Fatty acids were quantified by gas chromatography (GC-2010 Plus - Shimadzu AOC 20i autosampler) using SP-2560 capillary column. The backfat thickness (BF) was measured using a caliper. Shear force (SF) was measured by the method of Wheeler et al. (1995). The (co)variances and genetic parameters were estimated using the restricted maximum likelihood method, considering an animal model (ssGBLUP), applying the remlf90 software. The animals were genotyped with the BovineSNP BeadChip (High-Density Bovine BeadChip) that contained 777,962 SNP markers. After quality control for genotypes, a total of 470,007 SNPs from 1,556 samples remained. The heritability estimates were low for SFA (0.09), MUFA 0.005, and moderate for PUFA (0.20), n3 (0.08) and n6 (0.20). For SFA, the genetic correlation estimates with SF (0.04) with BF (0.30), and LIP (0.27) were low. The genetic correlation estimates

between MUFA with carcass and meat traits were low to high, being  $-0.05$  with SF,  $-0.64$  with BF, and  $0.88$  with LIP. The genetic correlation estimates between PUFA were low with SF ( $0.22$ ), moderate with BF ( $-0.41$ ) and LIP ( $-0.64$ ). For n3, the genetic correlation estimates with were low with BF ( $-0.17$ ) moderate with SF ( $-0.37$ ) and with LIP ( $-0.40$ ). The genetic correlation estimates between omega 6 with BF and

SF were low ( $0.05$  and  $0.10$ , respectively), and high with LIP ( $-0.86$ ). It is expected that the selection for carcass and meat traits would change the beef fatty acid composition.

**Key Words:** *Bos indicus*, heritability, genetic correlation

## Breeding and Genetics: Applications and methodology in animal breeding—Dairy

### T86 Genetic and phenotypic trends for age at first calving and milk yield in daughters from imported and Thai Holstein sires.

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Age at first calving (AFC) and milk yield (MY; accumulated 305-d milk yield) are important traits for the dairy cattle industry. To genetically improve these traits under tropical conditions in Thailand, both imported Holstein (IH) and Thai Holstein (TH; purebred and crossbred Holstein-Other Breeds) sires have been used for mating with Holstein purebred and Holstein-Other Breeds crossbred cows. The objective of this study was to compare genetic changes in AFC and MY from 1991 to 2014 in first-lactation daughters of IH and TH sires. The data set included AFC and first-lactation MY from 5,390 daughters of 507 IH and 232 TH sires that calved from 1991 to 2013 in 481 dairy farms. The bivariate animal model considered herd-year-season and Holstein fraction as fixed effects, and animal and residual as random effects. Variance components were estimated using an average information restricted maximum likelihood procedure. Variance component estimates were used to compute heritabilities and correlations. Mean estimated breeding value (EBV) for daughters of IH and TH sires were computed for each calving year. Regressions of mean EBV for AFC and MY on calving years were computed to assess genetic trends. Heritability estimates were  $0.19 \pm 0.04$  for AFC and  $0.13 \pm 0.04$  for MY. Near zero genetic ( $0.09 \pm 0.18$ ) and phenotypic ( $0.07 \pm 0.02$ ) correlations were estimated between AFC and MY. Overall genetic trends were negative for AFC ( $-0.013 \pm 0.003$  mo/yr;  $P < 0.001$ ) and positive for MY ( $1.16 \pm 0.30$  kg/yr;  $P < 0.001$ ). However, non-significant negative AFC genetic trends existed for daughters of IH sires ( $-0.006 \pm 0.003$  mo/yr;  $P = 0.06$ ) and TH sires ( $-0.020 \pm 0.004$  mo/yr;  $P < 0.001$ ). Conversely, significant positive MY genetic trends occurred for progeny of IH sires ( $1.14 \pm 0.43$  kg/yr;  $P = 0.008$ ) and TH sires ( $4.40 \pm 0.46$  kg/yr;  $P < 0.001$ ). These results indicated that there was genetic improvement in the appropriate direction (negative for AFC and positive for MY) in this Thai dairy population, and that TH sires brought larger changes than IH sires, primarily for MY.

**Key Words:** dairy, selection, tropics

### T87 Genomic-polygenic evaluation for milk yield and fat yield in a multibreed dairy cattle population in central Thailand.

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Milk yield (MY) and fat yield (FY) are economically important traits for Thai dairy businesses. Genetic prediction for MY and FY in Thailand uses only pedigree and phenotypic information. Combining SNP genotypes of individual animals with pedigree and phenotypes would be expected to increase the accuracy of genetic predictions and speed up selection progress. The objectives of this study were to estimate the fraction of the genetic variance accounted for by 8,257 SNP from GeneSeek GGP-LD BeadChip and to compare the rankings of animals evaluated with a genomic-polygenic (GP), genomic (G), and polygenic (P) models for MY and FY. The data set consisted of first-lactation MY and FY records from 600 cows from 56 farms in Central Thailand collected from 2000 to 2013. The mixed model contained herd-year-season, Holstein fraction and age at first calving as fixed effects (all models).

Random effects were SNP genomic (GP and G), animal polygenic (GP and P) and residual. Variance components were estimated using GS3 software (option VCE; GP and P). Additive genetic predictions were computed with GS3 (option BLUP) for all models. The fraction of additive genetic variances explained by the 8,257 SNP from GGP-LD and computed with the GP model were 46% for MY and 45% for FY. Heritability estimates with the GP model were higher (0.37 for MY and 0.40 for FY) than those with the P model (0.28 for MY and 0.30 for FY). Rank correlations between GP and G models were the highest (0.99 for both MY and FY;  $P < 0.0001$ ), followed by correlations between GP and P models (0.91 for MY and 0.75 for FY), and the lowest correlations were between G and P models (0.89 for MY and 0.73 for FY;  $P < 0.0001$ ). Thus, SNP from GeneSeek GGP-LD not only accounted for a sizeable fraction of the additive genetic variance for MY and FY, but they also yielded animal genomic EBV whose ranking was highly correlated with rankings of both genomic-polygenic and polygenic EBV. These results indicated that utilization of GGP-LD, and perhaps higher density genotyping chips, would be advantageous for genomic-polygenic evaluation and selection in Central Thailand.

**Key Words:** dairy, genomic, tropics

### T88 Genomic evaluation, breed identification, and population structure of North American, English and Island Guernsey dairy cattle.

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Genomic evaluations of dairy cattle in the United States have been available for Brown Swiss, Holstein, and Jersey since 2009 and for Ayrshire since 2013. As of February 2015, 2,281 Guernsey bulls and cows had genotypes from collaboration between the United States (USA), Canada (CAN), England (E), and the Island of Guernsey (I). The E/I contributions were made possible by the European Commission project, Gene2Farm. Of those genotyped, 431 were males with traditional evaluations. Evaluation accuracy was assessed using 4-fold cross-validation using males only. The cross-validation was repeated 10-fold and gains in reliability over parent average were averaged. Twenty-two traits were analyzed including 5 yield traits, 3 functional traits and 14 conformation traits. Mean gain in reliability over parent average for all traits was 13.8 percentage points. The highest gains were for strength (26.7), rump width (23.6), fore udder attachment (23.2) and fat percent (21.1). Traits with the lowest gains were daughter pregnancy rate (1.0) and somatic cell score (-0.9). The average 13.8-point gain for Guernsey can be compared with averages of 9, 24, 40, and 31 for Ayrshire, Brown Swiss, Holstein, and Jersey respectively. Twenty-one single nucleotide polymorphisms were useful for Guernsey breed determination and are used in routine genotype quality control to confirm breed and identify crossbreds. Genetic differences between E/I animals and USA/CAN



animals from an admixture study using principal component analysis will be presented. No haplotypes that affect fertility were identified from the current data set however; monthly assessments are conducted. These results are expected to lead to routine genomic evaluation of Guernseys in the USA and E/I.

**Key Words:** Guernsey, genomic evaluation, single nucleotide polymorphism

**T89 Genetic relationship between final score and production traits in Brazilian Holstein cattle.** Victor B. Pedrosa\*<sup>1</sup>, Pedro G. Ribas Neto<sup>2</sup>, Silvano F. Valoto<sup>2</sup>, Luis F. B. Pinto<sup>3</sup>, and Gabrieli S. Romano<sup>3</sup>, <sup>1</sup>*Universidade Estadual de Ponta Grossa, Ponta Grossa, Parana, Brazil*, <sup>2</sup>*Associação Paranaense dos Criadores de Bovinos da Raca Holandesa, Curitiba, Parana, Brazil*, <sup>3</sup>*Universidade Federal da Bahia, Salvador, Bahia, Brazil*.

Dairy cattle farmers seek to have animals that stay longer in the herd and still produce more milk and solids with each generation. In this regard, the final score has been used as a selection criterion to be related to many linear type traits and possibly present positive correlation with dairy yield traits. Thus, this study aimed to evaluate the genetic correlations between final score, milk, fat and protein yield and thus prove the positive relationship between them to better conduct dairy cattle programs in Brazil. The database contained information on 83,176 first lactation cows born between 1991 and 2011, with 148,534 records on pedigree file. The considered final score was 50 to 97, wherein the lower score represents a weak classification and the highest score excellent. Genetic parameters estimation were obtained based on the animal model using the VCE software. Analyses were performed considering the contemporary group (same herd-year) as fixed effect and the age of cow as a covariate. The average of final score and production of milk, fat, and protein were respectively 81.50 points, 8,531.50 kg, 287.90 kg, 270.74 kg. The genetic correlation between final score and production traits were 0.12, 0.02 and 0.14, respectively for milk, fat and protein. Hence, the correlation among milk and fat, milk and protein and lastly fat and protein were 0.49, 0.84 and 0.55, respectively. The results showed that there is a low, but positive correlation between final score and production traits, demonstrating that the selection for this type feature can improve dairy production. The results of relationship between production traits are consistent with findings in the literature in Holstein breed, in which the selection to increase milk or solids will benefit other production traits, specialty among milk and protein yields. Therefore, the final score should be included in the Brazilian Holstein breeding programs because it is widely known to be positive associated with legs, feet and udder, besides genetically contributing to increase dairy production.

**Key Words:** dairy cattle, linear type trait, milk yield

**T90 Genetic trends in dairy production of Brazilian Holstein cow.** Victor B. Pedrosa\*<sup>1</sup>, Altair A. Valloto<sup>2</sup>, Jose A. Horst<sup>2</sup>, Avelino M. Figueiredo<sup>2</sup>, and Adriana S. Martins<sup>1</sup>, <sup>1</sup>*Universidade Estadual de Ponta Grossa, Ponta Grossa, Parana, Brazil*, <sup>2</sup>*Associação Paranaense dos Criadores de Bovinos da Raca Holandesa, Curitiba, Parana, Brazil*.

Production and pedigree monitoring in Holstein cattle is carried out for many years but a only a few farmers effectively use breeding programs in Brazil. In the year 2014, the Holstein Association of Paraná State in partnership with the State University of Ponta Grossa created the State breeding program of cows, to accelerate genetic progress and consequently increase dairy production. Therefore, this study aimed to

evaluate the genetic trend obtained in the last 10 years for milk, fat, and protein yield and thus increase the genetic evaluation of female population in the country. The database contained information on 54,224 first lactation cows born between 2001 and 2011, with 81,336 records on pedigree file. Genetic trends were estimated from the linear regression of annual averages of breeding values. Estimates of breeding values were obtained based on the methodology of mixed models through BLUP solutions using the PEST software. Analyses were performed considering the contemporary group (same herd-year) as fixed effect and the age of cow as a covariate. The average production of milk, fat, and protein were respectively 8,693.17 kg, 286.51 kg and 264.28 kg. The estimated genetic trend was 6.62 kg/yr for milk, 0.44 kg/yr for fat and 0.34 kg/yr for protein. The results show that even with low adherence to animal breeding programs by dairy farmers there was a significant genetic progress for the 3 studied traits. This result might be explained by the use of imported genetic material of quality bulls from markets such as the United States, Canada and the Netherlands. However, it is essential to mention the importance of the farmers to increase selection based on genetic values estimated in a national selection program, reducing possible negative effects caused by the use of imported genetic material, selected in a different environment. Thus, it is expected that the new breeding program of cows provide greater genetic and productive advances for Brazilian herds.

**Key Words:** breeding program, milk yield, selection

**T91 Association between genomic SNPs and dairy production traits in Thai multibreed dairy cattle.** Pimchanok Yodklaew<sup>1</sup>, Skorn Koonawootrittriron\*<sup>1</sup>, Mauricio A. Elzo<sup>2</sup>, and Thanathip Suwanasopee<sup>1</sup>, <sup>1</sup>*Kasetsart University, Bangkok, Thailand*, <sup>2</sup>*University of Florida, Gainesville, FL*.

Genomic chips containing large number of single nucleotide polymorphisms (SNPs) have been widely used to study marker-trait associations in dairy cattle. Identification of genes associated with economically relevant traits close to significant SNPs are subsequently identified in genomic databases. The objective of this study was to find significant SNPs and genes that were associated with milk yield (MY), initial yield (IY), peak yield (PY), day to peak (DP), persistency (PS) and age at first calving (AFC) in a Thai Holstein-Other multibreed dairy population. The data set contained 2,409 first-lactation records from 295 dairy farms located in Central, Northern, Western, and Southern Thailand that were collected between 1997 and 2014. Animals were genotyped with 1 of 4 GeneSeek Genomic Profiler BeadChips (9K, 20K, 26K, or 80K). Thus, a set of 7,357 SNPs from autosomes in common among the 4 chips were used in this study. Association between SNPs and traits were analyzed using QXPAK.5 software. The mixed model included herd-year-season, Holstein fraction, age at first calving and SNPs as fixed effects, and animal and residual as random effects. The number of significant SNPs associated with MY, IY, PY, DP, PS and AFC were 649, 491, 627, 477, 527, and 381 at  $P < 0.05$ , and 342, 225, 301, 215, 259 and 156 at  $P < 0.01$ , respectively. All significant SNPs were checked for their association to 5,320 genes in the NCBI database obtained using Map2NCBI (R package). No gene was related to all traits. However, 8 genes were associated with 5 traits (*C3H1orf87*, *LOC100848063*, *NR5A2*, *PCDH15*, *WWO30*, *NCAMI*, *GLI2* and *LOC784126*) at  $P < 0.05$ , and 7 genes were associated with 4 traits at  $P < 0.01$  (*NR5A2*, *KCNIP1*, *INSC*, *LOC784126*, *EEF1E1*, *ATRNL1* and *LOC100294923*) at  $P < 0.01$ . Results from this research emphasized the need to validate SNP-dairy trait associations under Thai tropical environmental conditions to optimize the benefits of genomic selection.

**Key Words:** SNP, multibreed, tropics

**T92 Accuracy of genomic prediction using principal component analysis on an imputed high-density SNP panel in Italian Holstein cattle.** Antonio Puledda<sup>1</sup>, Giustino Gaspa<sup>1</sup>, Ezequiel L. Nicolazzi<sup>2</sup>, Corrado Dimauro<sup>1</sup>, Paolo Ajmone Marsan<sup>3</sup>, Alessio Valentini<sup>4</sup>, and Nicolo PP Macciotta\*<sup>1</sup>, <sup>1</sup>Dipartimento di Agraria, Università di Sassari, Sassari, Italy, <sup>2</sup>Fondazione Parco Tecnologico Padano, Lodi, Italy, <sup>3</sup>Istituto di Zootecnica, Università Cattolica del Sacro Cuore, Piacenza, Italy, <sup>4</sup>Dipartimento per l'Innovazione dei sistemi biologici, agroalimentari e forestali, Università della Tuscia, Viterbo, Italy.

In this study, the effect of predictor dimensionality reduction using Principal Component analysis (PCA) on the accuracy of Direct Genomic Values (DGV) for of 2,822 Italian Holstein bulls was tested. A subset of animals (916) were genotyped with high density (HD, 800K) bead-chip; the remaining were imputed from medium density (MD, 50K) to HD. Several 617,166 markers were retained after data editing. A MD panel was simulated for all animals by merging the HD panel with the BovineSNP50v2. A total of 40,669 markers were retained for the analysis. PCA were carried out both genome and chromosome wide for the MD (MD\_GW and MD\_CHR respectively), only by chromosome for the HD (HD\_CHR) panel. Several PC explaining 90% of the total variance was retained: 1,436 (MD\_GW) 4,829 (MD\_CHR) and 5,321 (HD\_CHR) respectively. PC score effects for 32 productive, functional and conformation traits were estimated on 2,301 training bulls born before 2004. DGVs were computed for the remaining 521 validation bulls born after 2003. Accuracy of prediction (rDGV) was computed as correlation between DGVs and phenotypes. Average rDGV across 32 traits were  $0.29 \pm 0.13$ ,  $0.31 \pm 0.13$ ,  $0.36 \pm 0.14$  for MD\_GW, HD\_CHR and MD\_CHR, respectively. Such an increase may be explained by the reduction of asymmetry between the number of predictors and observations. In particular, the shrinkage of the total variance of different SNP panel size in a quite similar number of PCs occurring during PCs extraction, could be seen as possible explanation of a better repartition of variance that resulted in a gain of rDGV passing from genome to chromosome wide and from high to medium density. This research was supported by Italian Ministry of Agriculture, grant INNOVAGEN.

**Key Words:** genomic selection, principal component analysis, high-density SNP panel

**T93 Estimation of genetic parameters for calving ease of Holstein cattle in Korea.** Mahboob Alam, Kwang Hyeon Cho\*, Tae Jeong Choi, Chung Il Cho, and Yun Ho Choy, *National Institute of Animal Science, Rural Development Administration, Cheonan-si, Chungcheongnam-do, Republic of Korea.*

Calving ease (CE) is a complex reproductive trait of economic importance for dairy cattle. This study was aimed to estimate the genetic parameters of CE for Holsteins in Korea. A total of 978,674 records of CE, collected from 1992 to 2014, from first 5 parities of Holstein cows were analyzed. Age of cows at first 5 parities were ranged within 18–42 mo, 30–54 mo, 42–66 mo, 54–78 mo, and 66–90 mo, respectively. Calving ease was recorded as ordered categories ranged from 1 to 5, indicating 1 = normal (not assisted), 2 = moderate assistance (extraction by 1 person), 3 = moderate assistance (extraction by 2/3 persons), 4 = difficult (extraction by > 3 persons), and 5 = surgical assistance calving. All CE categories were converted to a continuous scale through Snell score transformation procedure. The derived Snell scores for all parities were 0.01, 3.92, 6.288, 7.485, and 8.825 for category 1 through 5, respectively. A single trait sire-maternal grandsire model (S-MGS) was considered for ANOVA and covariance components from each parity

cows using ASREML 3.0 software package. Age at calving was fitted as fixed covariate, and herd-year-season, sire and maternal grandsires of the recorded cows were fitted as random effects in the S-MGS model. Estimated direct heritabilities for parity 1 through 5 were  $0.06 \pm 0.01$ ,  $0.08 \pm 0.01$ ,  $0.10 \pm 0.01$ ,  $0.07 \pm 0.01$ , and  $0.06 \pm 0.02$ , respectively. Maternal heritabilities from the models were  $0.02 \pm 0.01$ ,  $0.04 \pm 0.02$ ,  $0.03 \pm 0.01$ ,  $0.01 \pm 0.01$ , and  $0.04 \pm 0.01$ , respectively. Genetic correlations between direct and maternal genetic components were moderately to strongly negative for first 5 parities, such as  $-0.51 \pm 0.15$ ,  $-0.50 \pm 0.16$ ,  $-0.89 \pm 0.10$ ,  $-0.97 \pm 0.30$ , and  $-0.81 \pm 0.17$ , respectively. This study being the first report on CE of Korean Holsteins, we expect that the estimated genetic parameters derived in the study would be valuable for their calving ease improvement.

**Key Words:** calving ease, genetic parameter, Holstein cattle

**T94 Modeling for estimation of genetic parameters of milk production traits using random regression models in Korean Holstein cattle.** Chung Il Cho, Tae Jeong Choi\*, Kwang Hyeon Cho, Mahboob Alam, Yun Ho Choi, and Jae Gu Lee, *National Institute of Animal Science, Rural Development Administration, Cheonan-si, Chungcheongnam-do, Republic of Korea.*

The study was aimed to estimate genetic parameters for milk production traits of Holstein using random regression models (RRM), and compare the goodness of various RRM with homogeneous and heterogeneous residual variances. A total of 126,980 test-day milk production records of the first parity Holstein cows between 2007 and 2014, from the Dairy Cattle Improvement Center of National Agricultural Cooperative Federation in South Korea, were used in this purpose. These records were milk yield (MILK), fat yield (FAT), protein yield (PROT) and solids-not-fat yield (SNF). The statistical models included random effects of genetic and permanent environments using Legendre polynomials of third to fifth order (L3-L5), fixed effects of herd-test day, and year-season at calving and, a fixed regression for the test-day record (third to fifth order). The residual variances in the models were either homogeneous (HOM) or heterogeneous (15 classes, HET15; 60 classes, HET60). Total 9 models (3 orders of polynomials  $\times$  3 types of residual variance) such as L3-HOM, L3-HET15, L3-HET60, L4-HOM, L4-HET15, L4-HET60, L5-HOM, L5-HET15, and L5-HET60 were compared using AIC and/or BIC statistic to find the best fitted model(s) for particular traits. The AIC and BIC estimates were different among models for traits. The lowest BIC value was observed for the model L5-HET15 (MILK; PROT; SNF), and L4-HET15 (FAT), and those fitted the best. The BIC value of HET15 model for a particular polynomial order was lower than that of HET60 model in most cases. The estimated heritabilities from the best-fitted models in the study ranged from 0.08 to 0.15 for MILK, 0.06 to 0.14 for FAT, 0.08 to 0.12 for PROT, and 0.07 to 0.13 for SNF according to days in milk of first lactation. Genetic variances for studied traits tend to decrease at earlier stage of lactation, which then followed by an increase in the middle, and a further decrease at the end of lactation. Estimated RRM parameters can be used in Korean national genetic evaluation system instead of lactation models.

**Key Words:** random regression model, milk production trait, heritability

**T95 Estimation of genetic trends and profitability of dairy herds using different reproductive and genetic culling strategies with a stochastic simulation model which includes multitrait genetics.** K. Kaniyamattam\* and A. De Vries, *University of Florida, Gainesville, FL.*

The objective of the study was to estimate the genetic and phenotypic performance of the herd when voluntary culling of heifers was based on estimated breeding value (EBV) of milk (1st criteria) or Net Merit (NM\$) (2nd criteria). A daily dynamic stochastic model that simulates 5000 milking dairy cows, heifers and embryos over time and which included multitrait genetics was used. Heifers were inseminated with conventional (CS) or sexed semen (SS). Surplus dairy cows were culled based on the same criteria used to rank heifers. Genetic and economic changes were measured by the changes in true breeding values (TBV) of milk and NM\$, as well as actual phenotypic milk production and profit in a decade. Each of 5 experiment was run 5 times. In the first set of experiments, only CS was used with criteria 1 and 2. The increases in mean  $\pm$  SE of TBV of milk in first parity cows were 1,243  $\pm$  9 and 1,223  $\pm$  12 kg for criteria 1 and 2 respectively. The corresponding increases in annual milk yield/milking cow were 1,628  $\pm$  31 and 1,607  $\pm$  41 kg. The increases in mean  $\pm$  SE of TBV of NM\$ in first parity cows were \$925  $\pm$  10 and \$951  $\pm$  6 for criteria 1 and 2, respectively. The corresponding increases in profit/milking cow/yr were \$752 and \$763. In the second set of 3 experiments SS was used in the first 2 inseminations in heifers with culling of the lowest 20%, 30% and 40% heifers ranked on EBV for DPR. The increases in mean  $\pm$  SE of TBV of milk in first parity cows were 1090  $\pm$  11, 1084  $\pm$  15 and 1060  $\pm$  4 kg for the 3 heifer cull rates respectively. The corresponding increases in annual milk yield were 1245  $\pm$  44, 1197  $\pm$  42 and 1087  $\pm$  30 kg. The increases in mean  $\pm$  SE of TBV of NM\$ in first parity cows were \$975  $\pm$  6, \$980  $\pm$  3 and \$974  $\pm$  6 for the 3 heifer cull rates respectively. The corresponding increases in profit/milking cow were \$680, \$685 and \$618. In conclusion, the results showed that genetic trends and profitability depended on the genetic culling and reproductive strategies. Future studies will determine optimal combinations of genetic selection and reproductive strategies.

**Key Words:** multitrait genetics, phenotype, profit

**T97 Genetic parameters and trends for dairy traits in Holstein Friesian under North African conditions.** Salem Abdalla-Bozrayda<sup>1</sup>, Emhimad A. E. Abdalla<sup>1</sup>, and Mauricio A. Elzo<sup>\*2</sup>, <sup>1</sup>*University of Benghazi, Benghazi, Libya*, <sup>2</sup>*University of Florida, Gainesville, FL.*

Few studies have investigated the performance of Holstein Friesian cattle under North African conditions. Thus, the objectives of this research were to estimate genetic parameters for milk yield (MY, kg), true herd life (THL, d), and age at first calving (AFC, mo), and to compute genetic and environmental trends for MY using information from the Ghot Al-Sultan dairy farm near Benghazi, Libya. The data set contained MY, THL, and AFC records from 1,968 Holstein Friesian first-lactation cows collected from 1986 to 2002. There were 96 sires and 1,511 dams represented in the data set. Genetic parameters were estimated using restricted maximum likelihood procedures using a 3-trait mixed model. Fixed effects were year-season (all traits) and the covariates of age at first calving (MY, THL) and lactation period (MY). Random effects were cow and residual. The relationship matrix included 2,935 animals. Computations were carried out using the AIREMLF90 program. Milk yield genetic trends for cows, sires, and dams were estimated using means of breeding values for cows, sires and dams per year as regressions of average cow, sire, and dam mean breeding values on years. The estimates of heritability for MY (0.17  $\pm$  0.04) and AFC (0.14  $\pm$  0.03)

were low, whereas the value for THL (0.05  $\pm$  0.04) was close to zero. The estimates of genetic correlations between MY and THL (0.22  $\pm$  0.01), and between MY and AFC (0.19  $\pm$  0.04) were low, and the correlation between THL and AFC was near zero (0.07  $\pm$  0.04). The MY genetic trends were 4.11  $\pm$  0.92 kg/yr for cows, 5.70  $\pm$  1.12 kg/yr for sires, and 2.32  $\pm$  0.81 kg/yr for dams. Although low, the positive MY genetic trends for cows, sires and dams suggested the existence of an effective selection program and utilization of sires with superior breeding values under Libyan conditions. Conversely, environmental trends were negative for cows, sires, and dams suggesting deterioration of nutrition and management practices during the period covered in this study.

**Key Words:** dairy cattle, genetic trends, North Africa

**T98 Genetic parameters for production traits and age at first calving in Gyr dairy cattle.** Marco Prata<sup>\*1,2</sup>, Francesca Malchiodi<sup>2</sup>, Filippo Miglior<sup>3,2</sup>, Lenira El Faro<sup>4</sup>, Aníbal Vercesi Filho<sup>5</sup>, and Vera Cardoso<sup>1</sup>, <sup>1</sup>*Departament of Genetics, Ribeirão Preto Medical School, University of São Paulo, Ribeirão Preto, SP, Brazil.*, <sup>2</sup>*Centre for Genetic Improvement of Livestock, University of Guelph, Guelph, ON, Canada*, <sup>3</sup>*Canadian Dairy Network, Guelph, ON, Canada*, <sup>4</sup>*Centro Avançado de Pesquisa Tecnológica do Agronegócio de Bovinos de Corte, Instituto de Zootecnia, Sertãozinho, SP, Brazil*, <sup>5</sup>*Centro Avançado de Pesquisa Tecnológica do Agronegócio de Bovinos de Leite, Instituto de Zootecnia, Nova Odessa, SP, Brazil.*

The selection of Gyr cattle in Brazil has been focused mostly on improving milk yield. However, after the implementation of the Brazilian Agriculture Ministry regulation policies in 2002, which established new parameters for milk quality, producers have shown an interest in increasing fat and protein yield, as well as fat and protein content of milk. Other traits of interest for producers included age at first calving, known to be elevated in Gyr cattle, and which has an important economic impact on dairy farms. The objective of this study was to estimate genetic parameters for 305d milk, fat and protein yield, fat and protein content, and age at first calving for Gyr dairy cattle. The data set consisted of 10,977 records from 8,571 Gyr dairy cows in 245 herds in Brazil. Production traits were analyzed through a linear animal model including the fixed effects of contemporary groups (herd - calving year - calving season - parity) and age at calving as covariable (linear and quadratic effects). Direct additive genetic and permanent environmental effects were included as random effects. The model for age at first calving included the effect of contemporary group (herd - year and season of birth). Estimated heritabilities (SE) for milk, fat and protein yield were 0.23 (0.02), 0.18 (0.04), and 0.13 (0.07), respectively. Fat and protein content showed heritabilities of 0.12 (0.05) and 0.08 (0.07) respectively. Furthermore, the estimated heritability of age at first calving was 0.14 (0.02). The results indicated that the selection for these traits is possible in Brazilian Gyr. However, the low heritability values suggest that environmental factors (i.e., management strategies) could have the highest potential impact on the improvement of these traits. Further analysis should be performed to build a multi-trait selection index for Gyr dairy cattle breeding program in Brazil, to obtain desirable genetic gain for these traits.

**Key Words:** Gyr cattle milk, production, age at first calving

**T99 Evaluation of genetic diversity of three indigenous Russian cattle breeds using whole-genome scanning.** Elena A. Gladyr<sup>1</sup>, Alexander A. Sermyagin<sup>\*1</sup>, Tatiana E. Deniskova<sup>1</sup>, Alexey A. Traspov<sup>1</sup>, Veronika R. Kharsinova<sup>1</sup>, Gottfried Brem<sup>2</sup>, Natalia A. Zinovieva<sup>1</sup>, and Alexey V. Shakhin<sup>1</sup>, <sup>1</sup>*L.K.Ernst Institute of Animal*



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Whole-genome analysis opens the new possibilities to characterize the genetic structure of population and to evaluate the biodiversity in agriculture animal species. The aim of our study was to investigate genetic diversity for local cattle breeds from Central and Northeast European parts of Russia and Siberia in comparison with dairy cattle breeds using the whole-genome SNP data. The total of 57 animals from 5 different breeds were genotyped by 50K Illumina BeadChip including Yaroslavlskaya (n = 5), Kholmogorskaya (n = 8) and Yakutian cattle (n = 7) as well as widespread Russian Black-and-White (n = 6) and Holstein (n = 30). Quality control and analysis was performed using PLINK (1.07), while 1119 loci with > 10% of missing data were excluded, 7069 SNPs failed test frequency minor alleles (MAF < 0.01) and one bull was removed with missingness rate > 10%. 46596 SNPs were left for further analysis. A total of 2660 heterozygous haploid genotypes with the genotyping rate 0.992 were revealed. All local breeds characterized by deficient of SNP heterozygotes. Values of index fixation (F) were positive and ranged from 0.57% in the Russian Black-and-White breed to 17.56% in the Yakutian cattle. The genomic inbreeding coefficient was 3.99%, 4.17% and 10.39% for Holstein, Kholmogorskaya and Yaroslavlskaya breeds, respectively. Runs of homozygosity (ROH) in all investigated animals were 7.64Mb in average wherein minimal ROH was observed for Kholmogorskaya, Yakutian and Yaroslavlskaya breeds (6,65Mb, 7.25Mb and 7.39Mb, respectively) while for Holsteins and Russian Black-and-Whites were characterized by the maximum value of ROH in 7.97–8.00Mb. Linkage disequilibrium in the data set was  $r^2 = 0.40$ . The principal component analysis based on pairwise identity-by-state distances confirmed that there was the clear clusterisation among animals of Russian local cattle breeds and the group of Black-and-White and Holstein breeds. The greatest genetic difference was shown between Yakutian, Yaroslavlskaya, and Kholmogorskaya breeds as well as between Yakutian and the populations, which were assigned to branch of Holstein cattle. Supported by the Federal Agency for Scientific Organizations, Theme number 19.3 and Russian Scientific Foundation, project number 14–36–00039.

**Key Words:** cattle, biodiversity, whole-genome analysis

**T100 Exploring methodology for application of genomic information in South African dairy breeds.** Bernice E. Mostert<sup>1</sup>, Robert R. Van der Westhuizen<sup>2</sup>, Este Van Marle-Köster\*<sup>1</sup>, and Brian Van Doormaal<sup>3</sup>, <sup>1</sup>Department of Animal and Wildlife Sciences, University of Pretoria, Pretoria, South Africa, <sup>2</sup>SA Stud Book and Animal Improvement Association, Bloemfontein, South Africa, <sup>3</sup>Canadian Dairy Network, Guelph, ON, Canada.

The establishment of reference populations for genomic selection for Holstein and Jersey breeds in South Africa (SA) is underway. SA breeders have been using direct genomic values (DGVs) based on foreign reference populations, depending on base year definitions and units of measurement of foreign countries to make within herd selection decisions. Due to extensive linkage with international dairy populations, potential exists for utilizing DGVs originating from foreign reference populations to provide SA breeders with genomic information, comparable to SA breeding values. International collaboration was sought for foreign genomic profiles. Genomic profiles of SA Holstein and Jersey animals were generated based on either the GGPHD78K or GGPLD23K chips and DGV on the Canadian scale were estimated, using the North American Consortium's reference population. These were converted to the SA scale using Interbull conversion equations and blended with the SA Parent Averages (PAs), based on performance of

SA cows. This blending was done according to weightings determined by the reliabilities of the DGV and SA PAs. To account for genotype × environmental interactions between Canada and SA, the reliability of the DGV was multiplied by the correlation coefficient between SA and Canada for the specific trait, thereby ensuring that DGV from traits with lower correlations received less weight in the blending with local estimated breeding values (EBV). The weighting for the blending was determined as: EBV weight = EBV reliability/(EBV reliability + DGV reliability). These blended EBVs have been released as genomically enhanced breeding values (GEBVs) and serves as the official breeding values for these animals in the genetic evaluation, which are directly comparable to EBVs of the same breed in SA. Significant increases in the reliability with implementation of this methodology have been observed, with an average increase of 27% for milk yield and 23% for functional herd life. This approach has resulted in the release of GEBVs for production, udder health, fertility, longevity and conformation traits for SA Holstein and Jersey breeders.

**Key Words:** dairy, genomic information, reliability

### **T101 Genetic parameters of fertility indicators in Holstein.**

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Reproductive performance of lactating dairy cows influences the annual calving frequency, replacement heifers available, and milk yield. The goal of this study was to estimate the genetic parameters of fertility indicators in dairy cattle. Measurements from 953 Holstein cows from one farm in Texas across 2 calving years were analyzed. Three fertility indicators were evaluated: probability of cycling at d45 post-calving (Pr\_Cyc), probability of disease diagnosis at 45d post-calving (Pr\_Sck), and probability of pregnancy after 2 AIs (Pr\_Prg). These indicators were estimated using a logistic model including the covariables: dystocia, retained placenta, body condition score at 7d and 35d post-calving (BCS7 and BCS35, respectively), and blood  $\beta$ -hydroxybutyrate (BHBA) indication of subclinical ketosis. Univariate sire models including the effects of contemporary group and lactation number were used to estimate the genetic parameters of days open (DO), BCS7, BCS35, Pr\_Cyc, Pr\_Sck, and Pr\_Prg. The percentage of cows cycling, diagnosed with a disease, and pregnant after 2 AIs were 69.7%, 14.9%, and 64.6%, respectively. The marginal probabilities indicated that cows diagnosed with dystocia, retained placenta, metritis, and mastitis were 1.7%, 6.0%, 11.4%, and 7.3% less likely to be cycling than healthy cows, respectively. Cows were 2.7% and 4% less likely to be cycling per unit increase in mucus score and BHBA, respectively. The heritability estimates (and standard errors) for Pr\_Cyc, Pr\_Sck, Pr\_Prg, DO, BCS7, and BCS35 were 0.23 (0.15), 0.29(0.17), 0.58(0.19), 0.19(0.13), 0.25(0.16), and 0.25(0.18), respectively. Our results indicate that the fertility traits considered in this study have an important genetic component and that they could be used as effective indicators of fertility in breeding and management decisions. These findings could be the foundation to develop novel fertility indices that combine the fertility indicators hereby studied to improve the accuracy of pregnancy prediction and selection for improved fertility in dairy production. These findings contribute to

a long-term multistate project database (USDA-NIFA-AFRI-003542) for direct measures of fertility.

**Key Words:** heritability, fertility, dairy cattle

**T102 Selection signature analysis in Holstein cattle identified genes known to affect reproduction.** Li Ma<sup>1</sup>, Tad, S. Sonstegard<sup>2</sup>, Curtis Van Tassel<sup>2</sup>, John B. Cole<sup>2</sup>, George R. Wiggans<sup>2</sup>, Brian A. Crooker<sup>3</sup>, F. Abel Ponce de Leon<sup>3</sup>, and Yang Da<sup>\*3</sup>, <sup>1</sup>*Department of Animal and Avian Sciences, University of Maryland, College Park, MD*, <sup>2</sup>*Animal Genomics and Improvement Laboratory, ARS-USDA, Beltsville, MD*, <sup>3</sup>*Department of Animal Science, University of Minnesota, Saint Paul, MN*.

Using direct comparison of 45,878 SNPs between a group of Holstein cattle unselected since 1964 and contemporary Holsteins that on average take 30 d longer for successful conception than the 1964 Holsteins, we conducted selection signature analyses to identify genome regions associated with dairy fertility. Several genes known to affect reproduction were located in or near genome regions with strong selection signals. These genes include the fibroblast growth factor 1 gene (*FGF1*) on Chr07; the follicle stimulating hormone receptor gene (*FSHR*) and the luteinizing hormone choriogonadotropin receptor gene (*LHCGR*) on Chr11; the KIT ligand gene (*KITLG* or *KITL*), the fibroblast growth factor 6 and 23 genes (*FGF6* and *FGF23*) and the cyclin D2 gene (*CCND2*) on Chr05; the placental growth factor gene (*PGF* or *PLGF*) and the estrogen-related receptor  $\beta$  gene (*ESRRB*) 2Mb downstream of *PGF* on Chr10; and the prolactin receptor gene (*PRLR*) on Chr20. The selection signal for the region containing *FGF1* was among the strongest selection signals we observed. According to the literature on these genes, *FGF1* is involved in broad mitogenic and cell survival activities including embryonic development, *PGF* plays a key role in embryogenesis, *ESRRB* plays an essential role in placenta development, *FSHR* is necessary for follicular development and is expressed on the granulosa cells that are closely associated with the developing female gamete in the ovary of mammals, and *LHCGR* is necessary for follicular maturation and ovulation. Mouse knockout models showed that *FSHR*, *KITLG*, *CCNG2*, and *PRLR* were involved in female fertility proteins. These known gene functions related to reproduction and the fact that these genes were in or near chromosome regions with strong selection signals indicate that these genes could be involved in the vast difference in fertility between contemporary Holsteins and the 1964 Holsteins.

**Key Words:** fertility, selection signature, Holstein

**T103 Genetic interactions for heat stress and herd yield level: predicting foreign genetic merit from domestic data.** Janice R. Wright\* and Paul M. VanRaden, *Animal Genomics and Improvement Laboratory, Agricultural Research Service, US Department of Agriculture, Beltsville, MD*.

Genetic-by-environmental interactions were estimated from national data by separately adding random regressions for heat stress (HS) and herd yield level (HL) to the US all-breed animal model to improve predictions of future records and genetic rankings in other climates and production situations. Yield data included 79 million lactation records of 40 million cows; somatic cell score, productive life, and daughter pregnancy rate were also tested but had fewer records. Coefficients for HS were the state's July average temperature-humidity index; coefficients for HL were management-level weighted means for energy-corrected milk (ECM) divided by breed-year mean ECM. Coefficients were standardized to a mean of 0 and variance of 1. Predictions of current

(August 2014) from historical (August 2011) records were tested with a model that included herd management group (absorbed), sire estimated breeding value (EBV), dam EBV, and an interaction term (HS or HL) from the truncated data; records were weighted by lactation length for records in progress and by herd heritability using the same weights as in national evaluations. Estimated regression coefficients for sire EBV and dam EBV were always near their expected values of 0.5 and did not change when HS or HL interactions were added to the model. Estimated regressions for interaction terms, expected to be near 1, were 0.80 to 0.93 for HS and 0.61 to 0.72 for HL in yield traits. Squared correlations increased by  $< 0.0003$  for both HS and HL; increases for nonyield traits were even smaller. An additional test used multitrait across-country EBV to predict rankings of the same bulls in the United States and 14 other countries with somewhat different environments. The HS coefficient was significant ( $P < 0.05$ ) in 9 of 14 countries for milk and protein and in 10 for fat; the HL coefficient was significant in 8 countries for milk, 5 for protein, and 1 for fat. Squared correlations after adding an interaction term increased by  $< 0.004$  for HL and  $< 0.01$  for HS. The small changes in rank and correlation gains when HS and HL interactions were included in national evaluations indicate that current genetic predictions perform very well in a variety of environments.

**Key Words:** heat stress, environmental interaction, random regression

**T104 Application of milk mid-infrared (MIR) spectrometry in the dairy cattle industry in Canada.** Saranya Gunasegaram<sup>\*1</sup>, Allison Fleming<sup>1</sup>, Astrid Koeck<sup>1</sup>, Francesca Malchiodi<sup>1</sup>, Mehdi Sargolzaei<sup>1,2</sup>, Milena Corredig<sup>3,4</sup>, Flavio Schenkel<sup>1</sup>, Bonnie Mallard<sup>5</sup>, Ayesha Ali<sup>7</sup>, and Filippo Miglior<sup>1,7</sup>, <sup>1</sup>*CGIL, University of Guelph, Guelph, ON, Canada*, <sup>2</sup>*Semex Alliance, Guelph, ON, Canada*, <sup>3</sup>*Gay Lea, Guelph, ON, Canada*, <sup>4</sup>*Department of Food Science, University of Guelph, Guelph, ON, Canada*, <sup>5</sup>*Department of Pathobiology, OVC, University of Guelph, Guelph, ON, Canada*, <sup>6</sup>*Department of Mathematics and Statistics, University of Guelph, Guelph, ON, Canada*, <sup>7</sup>*Canadian Dairy Network, Guelph, ON, Canada*.

In Canada, many projects are aiming to integrate milk MIR spectra to develop predictions of milk components and cow status. A proportion of milk spectral data from FOSS MIR machines at Canadian DHI partners, CanWest DHI (Guelph, ON) and Valacta (Sainte-Anne-de-Bellevue, QC), have been saved and transferred to the Canadian Dairy Network, since early 2013. With addition of roughly 60,000 milk spectra every month; from approximately 700,000 cows enrolled in milk recording programs in Canada; the database is nearing 1.8 million. These spectra will be merged with recorded cow reproduction and health events (including hoof health). Currently, University of Guelph is analyzing approximately 2,000 milk samples for their fatty acid profile, milk fat globule and casein micelle size, lactoferrin, calcium, casein, and phospholipid content to use as a reference for developing MIR prediction equations. Genetic and genomic evaluation of Canadian dairy cows and bulls will be done using these MIR predicted milk component traits, with the objective of improving the nutritional value of milk and milk products for human consumption. Use of milk MIR spectra will be incorporated in a pending Canadian project, measuring feed efficiency and methane emissions in dairy cattle as a possible means for prediction. Information available to improve both cow management and genetic evaluation in Canadian dairy industry will be increased with the implementation of MIR prediction of a variety of new milk component and cow traits.

**Key Words:** milk infrared spectra, genomic, dairy cattle

**T105 Development of a daily stochastic dynamic dairy simulation model including the 12 traits in the Net Merit Index.** K.

Kaniyamattam\*, M. A. Elzo, and A. De Vries, *University of Florida, Gainesville, FL.*

We are interested in predicting changes in genetics and profitability of dairy herds when different reproductive strategies are combined with genetic selection. Our objective was to incorporate 12 correlated genetic traits included in Net Merit Index in a dynamic, stochastic model. An existing dynamic, stochastic model that mimics the biology and management of a herd of individual young stock and cows over time was adapted as follows. A true breeding value (TBV) for each trait was calculated as the average of the sire's and dam's TBV, plus a fraction of the inbreeding and Mendelian sampling. TBV were calculated from the Cholesky decomposed genetic covariance matrix of an average unselected Holstein population given as input, multiplied with by a  $12 \times 1$  matrix of standard normal deviates. Similarly, an environmental component for each trait was calculated based on the Cholesky decomposition of the environmental covariance matrix of the same population. The environmental component was partitioned into a permanent and a daily temporary effect. The combined effect of TBV and the environmental component was converted into an effect on the phenotypic performance of each animal for 6 of the 12 traits, for example effects on milk production, fertility, and risk of culling. Hence, genetics and phenotypic performance were associated. Estimated breeding values (EBV) were calculated using a normal inverse function based on correlated random numbers, the animal's TBV and a standard deviation depending on the reliability of the estimate. The EBV were updated 3 times a year, with reliabilities depending on the age of animals. Complete technical and economic measures were calculated by the model over a period of time. Preliminary validation resulted in similar genetic changes per decade as predicted by USDA-AGIL when using the Net Merit index. The model is suitable to estimate the economic and genetic effects from using different reproductive strategies in dairy herds.

**Key Words:** multitrait genetics, stochastic modelling

**T106 Genetic correlations between days open and milk, fat, and protein yields for the Uruguayan Holstein.** Nicolas Frioni\*<sup>1</sup>,

Jorge I. Urioste<sup>1</sup>, Ignacio Aguilar<sup>2</sup>, and Gabriel Rovere<sup>1</sup>, <sup>1</sup>*Facultad de Agronomía Universidad de la República, Montevideo, Montevideo, Uruguay,* <sup>2</sup>*Instituto Nacional de Investigación Agropecuaria Las Brujas, Rincon de Colorado, Canelones, Uruguay.*

This study aims to estimate the additive genetic correlations (RA) among days open (DO), milk (MY), fat (FY) and protein yield (PY) in lactations 1 to 3 of Holstein herds on pastures-based production systems.

The database used had 546,659; 367,774 and 247,158 observations for 1st, 2nd, and 3rd lactation. Data were edited to set records to DO greater than 42 d. Traits for each lactation were considered different and they were analyzed using 6-variate models, with DO and a production trait. The fixed effects were the age at calving and the combined effect of the herd-year-season and the random additive genetic effect of the animal. Estimations were obtained via Gibbs sampling with Gibbs2f90 program. Heritabilities obtained for each lactation of DO ranged from 0.04 to 0.06, for MY the range was 0.23 to 0.22, for PY from 0.09 to 0.28 and for FY from 0.06 to 0.28. The RA between DO ranged from 0.68 to 0.96, although they were different from 1, most of the values were very high to consider them as different traits. The RA between DO and production traits is presented in Table 1. Results suggest that DO can be considered as one trait, adjusting the model with a fixed effect accounting for different lactations. Unfavorable relationship between DO and production traits may be confirmed, thus is necessary the inclusion of fertility in selection programs, and further estimation of RA analyzing all traits together with a repeatability model.

**Table 1 (Abstr. T106).** Additive genetic correlations among days open, milk yield (MY), fat yield (FY), and protein yield (PY) in lactations 1 to 3 of Holstein herds

		Days open		
		1	2	3
MY	1	0.39 (0.0375)	0.56 (0.0338)	0.51 (0.0638)
	2	0.49 (0.0357)	0.58 (0.0314)	0.59 (0.0640)
	3	0.40 (0.0335)	0.54 (0.0393)	0.63 (0.0581)
PY	1	0.43 (0.0456)	0.53 (0.0471)	0.78 (0.0282)
	2	0.44 (0.0425)	0.53 (0.0375)	0.69 (0.0372)
	3	0.46 (0.0601)	0.55 (0.0535)	0.67 (0.0543)
FY	1	0.41 (0.0751)	0.51 (0.0907)	0.69 (0.0565)
	2	0.54 (0.0604)	0.66 (0.0434)	0.80 (0.0345)
	3	0.34 (0.0620)	0.46 (0.0520)	0.63 (0.0493)

**Key Words:** fertility, heritability, dairy cattle



## Dairy Foods: Dairy products

**T107 Angiotensin-converting enzyme inhibitory activity of whey proteins obtained from the manufacture of traditional Mexican cheeses.** Yuridia S. Tarango-Hernandez, Alma D. Alarcon-Rojo, Nestor Gutierrez-Mendez, and Jose C. Rodriguez-Figueroa\*, *Universidad Autonoma de Chihuahua, Chihuahua, Mexico.*

Traditional Mexican cheese production has been increasing in recent years. Thus, the increase in cheese whey volume must be carefully dealt with. Because of its high BOD ( $27\text{--}60\text{ g L}^{-1}$ ) and DOQ ( $50\text{--}102\text{ g L}^{-1}$ ), cheese whey represents a potential pollutant to the environment. However, studies have demonstrated the bioactivity of whey proteins. Therefore, this research aimed to evaluate the angiotensin-I-converting enzyme (ACE)-inhibitory activity of Fresco, Asadero and Chihuahua cheese whey. Samples corresponded to 3 different traditional cheese-making times. The  $<1$  and  $3\text{--}1$  kDa whey fractions were obtained by ultrafiltration. The ACE-inhibitory activity was evaluated by standardized RP-HPLC methodology. Results showed that all cheese whey fractions were able to inhibit ACE activity. The highest ACE-inhibitory activity ( $67.3 \pm 2.0\%$ ) was found in the  $<1$  kDa Fresco cheese whey fraction, however it was not significant different ( $P > 0.01$ ) from the Asadero cheese whey fraction ( $65.3 \pm 3.6\%$ ). In contrast, the  $<1$  kDa Chihuahua cheese whey fraction presented the lowest ( $P < 0.01$ ) activity ( $29.3 \pm 7.5\%$ ). The  $3\text{--}1$  kDa Asadero cheese whey fraction showed high ( $P < 0.01$ ) ACE-inhibitory activity value ( $66.1 \pm 3.6\%$ ). In conclusion, Asadero and Fresco cheese whey presented relevant ACE-inhibitory activity. Therefore, whey obtained from the manufacture of these traditional Mexican cheeses may have a potential to be used as functional ingredients.

**Key Words:** ACE-inhibitory activity, traditional Mexican cheese, whey

**T108 The effect of partially hydrolyzed different milk species on xanthine oxidase activity of fetal enterocyte culture.** Hristina Kocic\*, *Medical Faculty University Maribor, Maribor, Slovenia.*

Introduction of enteral nutrition in early life may lead to autoimmune, inflammatory and allergic reactions. Oxidative damage is a contributing factor in inflammatory bowel disease, necrotizing enterocolitis and ulcerative colitis. Xanthine oxidase (XO) plays an important role in oxidative injuries, because of the generation of free radical hydrogen peroxide ( $\text{H}_2\text{O}_2$ ). It can induce oxidative modification and may activate redox-sensitive transcription factors (NF- $\kappa$ B) and proinflammatory cytokines. This study was conducted with the aim to compare the effect of hydrolyzed milk samples,  $\beta$ -casein (CS) and  $\beta$ -casomorphin-7 (CSM-7) on XO activity in culture of fetal human enterocytes (commercial H4 cells). During exposure of H4 cells to hydrolysed milk species, XO activity (U/g prot) decreased significantly treated with hydrolyzates of human colostrum ( $85.28 \pm 7.31^*$ ), mature human milk ( $80.34 \pm 5.40^*$ ), donkey colostrum ( $91.82 \pm 4.47^*$ ), mature donkey milk ( $71.73 \pm 3.89^*$ ) vs  $165.33 \pm 3.40$  of control H4 cells. The activity did not change significantly by using cow colostrum ( $182.20 \pm 5.88$ ), but decreased with mature cow milk ( $101.84 \pm 11.56^*$ ). Significant decrease was observed for Bebelac formula 0 ( $96.35 \pm 8.89^*$ ), but not of Bebelac formula for 12 mo ( $139.36 \pm 5.88$ ). Both, CS and CSM-7 decreased XO activity ( $66.80 \pm 6.48^*$  and  $67.98 \pm 5.32^*$ ). Observed activities correlated positively with NF- $\kappa$ B activity, but negatively with proliferative (mitotic activity) of H4 cells. Exclusive breast-feeding has been associated with a reduction in the incidence of chronic inflammatory conditions, including autoimmune

diseases, allergies, asthma, atopic dermatitis and inflammatory bowel diseases, what may correlate with decreased oxidative stress. The use of donkey milk seems to be a promising strategy against allergic and other immune-related reactions, regarding oxidative injury and XO activity. Well-documented antioxidative properties of CS may be due to decreased XO activity. Well-documented toxic effects of CSM-7 may not depend on XO activity.

**Key Words:** donkey milk, breast milk, cow milk

**T109 Case study: Comparison of biologically active compounds in milk from organic and conventional dairy herds.** Diane L. Van Hekken\*<sup>1</sup>, Michael H. Tunick<sup>1</sup>, Hubert J. Karreman<sup>2</sup>, Elaine R. Ingham<sup>2,3</sup>, and Peggy M. Tomasula<sup>1</sup>, <sup>1</sup>USDA, ARS, Wyndmoor, PA, <sup>2</sup>Rodale Institute, Kutztown, PA, <sup>3</sup>Soil Foodweb Inc., Corvallis, OR.

Conflicting reports of the quantities of biologically active compounds present in milk from organic grass-fed and conventional herds show that more research is required, especially as these compounds are linked to human health benefits and can improve the health value consumers place on dairy products. In collaboration with the Rodale Institute, Kutztown, Pennsylvania, a 3-year study evaluated the composition of milk obtained from 2 farms adjacent to the Rodale experimental farm: one herd transitioned to certified organic in the first year; cows averaged over 50% dry matter intake from pasture during the grazing season. The other herd was on a confined conventional farm with no access to pasture. This study provided a unique opportunity to compare milk from farms of similar soil types, climate, and weather. Over a 150-week period (3 grazing seasons), weekly milk samples were collected and assayed for composition and physical traits, and profiled for fatty acids (FAs) and proteins-peptides. The overall protein, lactose, ash, minerals, total solids contents, pH, water activity, titratable acidity, and protein profiles of the milk samples from both farms were similar ( $P < 0.05$ ). Fat content was more variable in the milk from the organic herd, especially after the seasonal grazing was implemented, with higher fat levels ( $P < 0.05$ ) obtained during the winter months. The FA profiles were the major difference between the milk from the 2 herds. Compared with the confined herd, the grazing herd milk had considerable variation in the FA profiles, which generally stabilized after 4 mo, and had higher levels ( $P < 0.05$ ) of  $\alpha$ -linolenic acid (omega-3 FA) after the second month. Once certified, the organic herd produced milk that contained 36% more omega-3 FA and had lower linoleic acid (omega-6):omega-3 FA values throughout the year, and 25–30% more conjugated linoleic (CLA) during the grazing seasons than the confined herd. Because higher CLA and omega-3 FA levels and a low omega-6:omega-3 value have been linked to being beneficial to human health, the importance of pasture grazing must be considered in supplying milk and dairy products that address consumers' demand for foods that support human health and wellness.

**Key Words:** milk, organic, omega-3 fatty acids

**T110 Evaluation of the effect of diets containing grape seed and linseed on milk fatty acid composition in Sarda sheep by principal component analysis.** Fabio Correddu, Anna Nudda\*, Giustino Gaspa, Gianni Battacone, and Giuseppe Pulina, *Dipartimento di Agraria, University of Sassari, Sassari, Italy.*

This work aimed was to study the effect of the dietary inclusion of grape seed and linseed, on milk fatty acid (FA) composition in Sarda

dairy sheep. Milk samples were collected weekly from 24 Sarda ewes assigned to 4 dietary treatments: control diet (CON), containing 300 g/d of grape seed (GS), a diet containing 220 g/d of linseed (LIN) and a diet containing both lin- and grape seed (MIX). Samples were analyzed for FA profile with GC. Data of major classes of FA were analyzed by principal component analysis (PCA) and the first 2 PC were retained for further analysis based on the proportion of variance explained by PC. The first and second PC accounted for about 90% of the total variability (78.3% and 12%, PC1 and PC2, respectively). Elements of eigenvector of PC1 were positively associated with unsaturated (0.246), monounsaturated, long, *trans* FA and CLA (0.244, 0.243, 0.233, 0.231), whereas groups characterized by short, medium chain FA, saturated FA showed negative association with PC1 (−0.231, −0.238, −0.246). PC2 loadings were positively correlated with n-6 and n-6/n-3 ratio (0.455, 0.559), and negatively with n-3 (0.470). Furthermore, PC1 presented negative loadings for atherogenic index and thrombogenic index (−0.238, −0.236). It was supposed that PC1 was positively associated with the PUFA intake, which progressively increased in CON, GS, LIN and MIX. PC2 could be related to the different source of PUFA (grape seed or linseed) and consequently, with the ratio PUFA n-6/n-3 in the diet. The observations plotted on the new system of variables defined by PC1 and PC2, revealed definite clusters for each of the 4 dietary treatments. Incremental scores in PC1 were observed for CON, GS, LIN and MIX, (−5.8, −0.3, 1.5, 4.5, respectively); PC2 discriminated only GS (positive scores, 1.9) from LIN (negative scores, −2.0). Overall, the results suggest that the inclusion of grape seed and linseed in the diet of lactating ewes could be useful to improve the quality of milk FA, especially when used in combination. The PCA has shown to be a useful tool for understanding the effect of different dietary treatments on milk FA profile.

**Key Words:** grapeseed, milk fatty acid, multivariate analysis

**T111 Interaction between whey protein and inulin in model system.** Cuina Wang<sup>1</sup>, Hao Wang<sup>1</sup>, and Mingruo Guo<sup>1,2</sup>, <sup>1</sup>*Jilin University, Changchun, Jilin, China*, <sup>2</sup>*University of Vermont, Burlington, VT*.

Effects of protein concentration (4–8%, w/v), inulin concentration (1–5%, w/v) and ionic strength (10–50 mM) on Ca<sup>2+</sup>-induced interactions between whey protein (WP) and inulin mixture (pH 7.0) heated at 85°C for 30 min were investigated. The interactions were analyzed for turbidity, particle size, zeta potential, apparent viscosity, and texture profile. Interaction properties were also compared with mixture of polymerized whey protein (PWP) and inulin in which whey protein was heated first and then mixed with inulin. Results indicated that the dispersion became more opaque with increasing protein but no significant difference was detected regarding the way inulin added ( $P < 0.05$ ). Three peaks at about 40, 900 and 5000 nm were observed in size distribution by volume. Compared with the control, there was a small shift toward larger size with increasing inulin level in the width of peak corresponding to denatured whey protein. Zeta potential values for all samples fell in the ranges of −10 to −30 mV independent of the manner in which inulin added. Increasing inulin resulted in less negative zeta potential and smaller repulsive force between particles for both types of mixtures. As protein and inulin increased, apparent viscosity increased and combination of PWP and inulin showed significant higher values ( $P < 0.05$ ) than WP and inulin mixture. It was not sufficient to form cold gels for all samples at the level of 10 mM Ca<sup>2+</sup>. Compared with the control, the mixtures of PWP with inulin showed higher water holding capacity may due to the ability of occluding water of inulin microcrystals ( $P < 0.05$ ). WP/inulin mixture showed significant higher hardness ( $P < 0.05$ ) than PWP/inulin due to possible microcrystal seeds maintained in solu-

tions when heated at 80°C for 30 min. Results indicated that interactions maybe occurred between whey protein and inulin in the model system.

**Key Words:** whey protein isolate, polymerized whey protein, inulin

**T112 Characterizing the dissolution behavior of whey protein concentrate with an ultrasonic flaw detector.** Mary Hauser\* and Jayendra Amamcharla, *Kansas State University, Manhattan, KS*.

Current methods used in the dairy industry to characterize the dissolution process are difficult to reproduce and subjective. Low-intensity ultrasound spectroscopy has the advantage of being rapid and precise, but the equipment is expensive. An ultrasonic flaw detector (UFD) is an economical alternative and allows for rapid data collection at a single frequency. This study focused on characterizing the dissolution behavior of whey protein concentrate (WPC) with an UFD. The experimental setup included an UFD (Epoch LTC) in pulse-echo mode and a 1MHz immersion transducer. Two batches of WPC80 from a commercial manufacturer were stored at 25°C and 40°C for 3 weeks. 19.74g of WPC was gradually added to 375g water. The tests were performed in duplicate. During dissolution, ultrasonic (US) data from UFD, chord length distribution from focused beam reflectance measurement (FBRM), and solubility index were collected at regular intervals for 1800s. US relative velocity and attenuation were calculated from A-scan data obtained from the UFD. The relative velocity was defined as the ratio of the US velocities of WPC solution and water at 40°C. It was observed that fresh WPC had a relative velocity of 1 after 1800s. The relative velocity at 1800s increased for powders stored at 25°C and decreased for powders stored at 40°C. After 3 weeks of storage at 25°C and 40°C, the relative velocities were 1.015 and 0.98, respectively. Attenuation had a general trend of increasing and then gradually decreasing. The storage temperature of WPC influenced the time to reach the peak and the area under the attenuation curve. Fresh powders reached the peak after 1440s. Three weeks of storage at 25°C and 40°C caused the peak time to increase to 1658s and decrease to 986s, respectively. The area had a similar trend. Fresh powders had an area of 60Np\*s/m. Powders stored at 25°C and 40°C for 3 weeks had areas of 47Np\*s/m and 91 Np\*s/m, respectively. Storage temperature significantly ( $P < 0.05$ ) effected relative velocity at 1800s, peak time, and area. FBRM data showed that powders stored at 40°C for 3 weeks had larger particles. Overall, a UFD can be a useful tool when characterizing the dissolution behavior of WPC.

**Key Words:** ultrasonic flaw detector, whey protein concentrate

**T113 Effect of pH and Ca-ion activity on the heat stability of reconstituted reduced Ca milk protein concentrate dispersions.** Gopinathan H. Meletharayil<sup>1</sup>, Anil Kommineni\*<sup>1</sup>, Chenchaiiah Marella<sup>2</sup>, and Lloyd E. Metzger<sup>1</sup>, <sup>1</sup>*Midwest Dairy Foods Research Center, Dairy Science Department, South Dakota State University, Brookings, SD*, <sup>2</sup>*Idaho Milk Products, Jerome, ID*.

The purpose of this study was to investigate the heat stability and calcium ion activity of the reconstituted MPC dispersions. Reduced calcium MPC (RCMPC) powders were produced from skim milk subjected to CO<sub>2</sub> treatment before and during the process of ultrafiltration. The CO<sub>2</sub> injection was controlled to obtain 0, 20, 30, and 40% reduction in Ca levels of MPC powders. RCMPC powders were reconstituted to 10% protein in deionized water. These dispersions were tested for heat stability in a rocking oil bath at 140°C at unadjusted, 6.5, 6.7, 6.9, and 7.1 pH. Calcium ion activity (CIA) and ionic strength measurements on Ca-standards and all dispersions were carried out using a Ca ion-selective electrode and conductivity meter. The unadjusted pH of the dispersions

varied from 6.8 in control to 5.95 in 40% RCMPC dispersions. This pH difference had an effect on the CIA of the dispersions and ranged from 1.31mM in control to 18.39mM in 40% RCMPC dispersions. Differences in the heat stability were observed in the unadjusted pH dispersions with the control MPC having the highest heat stability that decreased as the percentage of Ca reduction increased. The heat stability of control MPC dispersions showed a minimum at pH 6.7 and maximum at pH 6.9 with a decrease observed at pH 7.1. The CIA of RCMPC dispersions decreased with an increase in pH. A steady increase in the heat stability of 30 and 40% RCMPC dispersions was observed with increase in pH which correlated with decrease in the calcium ion activity. Dispersions from 20% RCMPC showed a heat stability profile similar to the control MPC dispersion but had higher heat stability values at all adjusted pH values. The CIA activity had a direct correlation on the heat stability of the unadjusted MPC dispersions, but at a higher pH of 7.1, no correlation was evident, even though, all the dispersions had a low CIA. Heat stability of 30 and 40% RCMPC dispersions increased with an increase in the pH showing highest heat stability at a pH of 7.1. From this study, it can be concluded that pH and Ca-ion activity influences the heat stability of RCMPC dispersions.

**Key Words:** milk protein concentrate, Ca ion activity, heat stability

**T114 Rennet coagulation and cold gelation properties of recombined highly concentrated micellar casein concentrate and cream.** Ying Lu<sup>\*1</sup>, Donald McMahon<sup>1</sup>, Lloyd Metzger<sup>2</sup>, Anil Kommineni<sup>2</sup>, and Almut Vollmer<sup>1</sup>, <sup>1</sup>Western Dairy Center, Utah State University, Logan, UT, <sup>2</sup>Midwest Dairy Foods Research Center, South Dakota State University, Brookings, SD.

Highly concentrated micellar casein concentrate (HC-MCC), a potential ingredient for cheese making, containing ~20% casein with ~70% of whey proteins removed by microfiltration, and diafiltration of skim milk, and then further concentrated by vacuum evaporation. Our objective was to investigate rennet coagulation and cold gelation properties of recombined thawed frozen HC-MCC and cream under different conditions. The HC-MCC was recombined with cream using low shearing at 50°C for 30 min, followed by rheological measurements. Cold gelling temperature [the temperature at which storage modulus ( $G'$ ) = loss modulus ( $G''$ )], was positively correlated with casein levels from 8.6% to 11.5% ( $R^2 = 0.71$ ) and pH from 6.6 to 7.0 ( $R^2 = 0.96$ ). Gelation occurred at ~-35, ~-26 and ~-13°C with 11%, 10% and 9% casein concentration, respectively. This process was reversible with a hysteresis effect observed depending on whether the mixture was being heated or cooled. Rennet coagulation of HC-MCC and cream mixture at rennet level of 20 mg/100 g was studied by testing coagulation time (CT, the time when  $G' = G''$ ) and curd firming rate (the ratio of  $G'$  at 1.5 times of coagulation time and at coagulation time). Increasing casein level from 3.3 to 10.9% significantly increased ( $P < 0.001$ )  $G'$  from 0.1 to 2 Pa, and slightly reduced CT ( $P < 0.05$ ) by 1.3 min from casein level from 3.3% to 5.7%, without change of CT at higher casein level. Decreasing pH of mixture from 6.6 to 6.2 significantly reduced CT ( $P < 0.001$ ) by 5.5 min, and increased firming rate ( $P < 0.05$ ) from 35 to 107. Decreasing coagulation temperature from 31 to 25°C significantly decreased firming rate ( $P = 0.001$ ) from 160 to 76, increased CT ( $P < 0.001$ ) by 2 min, and  $G'$  ( $P < 0.05$ ) from 2 Pa to 4 Pa. Addition of citrate to the mixture significantly decreased ( $P < 0.05$ ) firming rate from 35.2 to 4.6, increased  $G'$  ( $P < 0.05$ ) from 3 Pa to 6 Pa, with no significant effect ( $P > 0.05$ ) on CT. Therefore, decreasing coagulation temperature, or adding citrate may make rennet coagulation properties of recombined HC-MCC and cream at high casein level more suitable for cheese making by increasing coagulation time and reducing curd firming rate. Understanding rennet coagulation and

cold gelation properties can help potentially use recombined HC-MCC and cream for cheese making.

**Key Words:** rennet coagulation, micellar casein, microfiltration

**T115 Formation of sodium caseinate-maltodextrin conjugates and the rheological properties of acid gels.** Shuwen Zhang<sup>\*1,2</sup>, Yuansheng Gong<sup>1</sup>, Som Khanal<sup>1</sup>, Yanjie Lu<sup>1</sup>, and John A. Lucey<sup>1</sup>, <sup>1</sup>Department of Food Science, University of Wisconsin-Madison, Madison, WI, <sup>2</sup>Institute of Agro-Product Processing Science and Technology, Chinese Academy of Agricultural Sciences, Beijing, China.

Conjugation of polysaccharides to proteins utilizing the Maillard reaction leads to significant improvement in the functional properties of proteins, such as solubility, emulsification, and thermal stability. However, no gelation properties of conjugates have previously been reported. In this study, we studied the conjugation reaction between sodium caseinate (NaCN) and maltodextrin100 (Md100) in aqueous solutions via the initial stage of the Maillard reaction. The rheological properties of acid-induced gels made from NaCN-Md conjugates prepared by Maillard reaction were investigated. Acidification was performed by the addition of glucono-delta-lactone at 30°C. Covalently linked conjugates of NaCN and Md100 were produced by incubating aqueous solutions containing 5% NaCN and 5% Md100 at 90°C for 10 h. The covalent attachment of Md100 to NaCN was confirmed by SDS-PAGE with both protein and carbohydrate staining. The Schiff base formation of NaCN-Md conjugates was monitored by maximum absorbance peak at ~300 nm using UV spectroscopy. Conjugation resulted in faster gelation (higher pH) but lower storage modulus and shear stress values of acid gels compared with gel made with NaCN only. An increase of loss tangent was also observed in acid gel made with conjugates. These results indicate that conjugates modify protein gelation and a potential use for these NaCN-Md could be as specialty functional food ingredients.

**Key Words:** sodium caseinate, maltodextrin, conjugate

**T116 Edible packaging films from calcium-caseinate and citric pectin.** Laetitia M. Bonnaille\* and Peggy M. Tomasula, USDA/ARS/NEA/ERRC, Wyndmoor, PA.

Casein-based films have excellent gas-barrier properties and strength appropriate to form a variety of edible food packaging, but do not constitute strong moisture barriers. In prior work, we used humidity-controlled DMA (DMA-RH) to show that solvent-cast films from calcium caseinate (CaCas, 75% dry basis) and glycerol plasticizer (Gly, 25% dry basis) of 0.03–0.05 mm thickness are extremely sensitive to ambient relative humidity (RH) and temperature (T) and melt around  $T_m \sim 45^\circ\text{C}$  at 50% RH, or  $RH_m \sim 67\%$  at  $20^\circ\text{C}$ ;  $T_m$  also decreases at higher RH, which limits the usefulness of CaCas packaging films in warm and/or humid conditions. Citric pectin (CP, 0.1 to 2% dry basis) was explored as an edible additive to crosslink CaCas/Gly films with the objective to improve their environmental resistance. CP was added to water/Gly solutions, stirred for 1h, then CaCas was added and stirred for 1 h. Dynamic T-ramps (10–70°C) with an AR2000 rheometer showed that at 25°C, CP% strongly increased the viscosity and storage modulus,  $G'$ , of the solutions near-exponentially, signifying strong binding of CP with CaCas; in addition, new crosslinks were activated between ~42–60°C for 1–2% CP. DMA-RH (Q800) T-ramps of dried films at 50% RH showed that 0.1–0.5% CP softened the films and reduced  $G'$  due to steric hindrance of isolated CP particles throughout the CaCas network, while



1 to 2% CP increased  $G'$ , demonstrating a higher crosslink-density in the films.  $T_m$  increased to  $\sim 50^\circ\text{C}$  with 1% CP and  $\sim 60^\circ\text{C}$  with 2% CP. Optical microscopy imaging confirmed strong changes in the CaCas/Gly/CP network as a function of CP content. Moisture sorption RH-ramps at  $20^\circ\text{C}$  (Q5000SA) indicated that CaCas films absorb  $\sim 10\%$  of their weight in water at 50% RH and up to  $\sim 28\%$  at 80%RH; adding up to 2% CP reduced water-sorption by up to 15% at high RH. High CP% (0.75–2%) slowed dissolution kinetics of the films in water at  $20^\circ\text{C}$ , while low CP% (0.1–0.5%) accelerated solubilization slightly. CaCas films strengthened with  $> 1$ –2% CP are more environmentally-resistant and have the potential to replace or improve some synthetic food packaging to reduce waste and/or add functionality and nutrition to food products.

**Key Words:** casein, hydrophilic film, dynamic rheology

#### **T117 Changes in volatile compounds in whey protein concentrate stored at elevated temperature and humidity.**

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Whey protein concentrate (WPC) has been recommended for use in emergency aid programs, but it is often stored overseas without temperature and relative humidity (RH) control, which may cause it to be rejected because of yellowing, off-flavors, or clumping. Therefore, the volatile compounds present in 14.0-kg bags of WPC34 (36% protein) and 18.7-kg bags of WPC80 (79% protein) were analyzed during storage over 18 mo in environmental chambers at 25, 30, and  $35^\circ\text{C}$  at RH levels of 70, 80, and 90%. An ambient chamber at around  $21^\circ\text{C}$  and 45–65% RH served as the control. The concentrations of dimethyl disulfide increased with time as the sulfur-containing amino acids cysteine and methionine were degraded. Five aldehydes (hexanal, heptanal, octanal, nonanal, and benzaldehyde) and 2 ketones (2-butanone and 2-heptanone) appeared as a result of lipid oxidation. Lactose breakdown led to the presence of butanol, hexanol, and furaneol. The levels of volatiles were higher in the WPC80 and tended to increase with temperature and RH. Although the color and flow properties were affected by the conditions, the generation of volatile compounds did not have a noticeable effect on product odor.

**Key Words:** whey protein concentrate, volatile compounds

#### **T118 Characterization of milk, soymilk, and almond milk through consumer affective and emotional responses.**

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Plant-based milk beverages, notably soymilk and almond milk, are competing with dairy milk in the refrigerated retail case. It is unknown if product sensory differences affect consumer behaviors and purchasing decisions. Dairy and plant-based milk beverages were evaluated for acceptability and characterization of product attributes and emotional response for dairy milk, almond milk, soymilk, and soymilk with added titanium dioxide for whitening. We hypothesized that appearance would affect acceptability; soymilk treatments and almond milk, with a darker visual appearance, would have different acceptability and emotional characterization than dairy milk. Untrained panelists ( $n = 49$ ) received each product for evaluation of overall acceptability, just-about-right (JAR) scale, and check-all-that-apply (CATA) emotional terminology

(EsSense) ballot. Mean acceptability of all treatments was between “neither like or dislike” and “like slightly” on a 9-point hedonic scale (mean = 5.33); however, all products appeared to have bimodal distributions. CATA emotional terms calm, content, good, and mild were chosen by  $>20\%$  of panelists (frequently selected terms) for all treatments. Of the frequently selected terms across products, calm was selected less for soymilk, good was chosen more for almond milk, and the terms happy and pleasant were not chosen for dairy milk. Mild (almond, soymilk with titanium dioxide, and dairy milk) and quiet (soymilk, soymilk with titanium dioxide) terms were associated with decreased hedonic score based on penalty life analysis. Pleased (soymilk) and satisfied (dairy milk) were most closely associated with high hedonic scores. Dairy milk had a more optimal appearance than all plant-based milk products based on JAR characteristics. Almond milk and soymilk have different product characteristics than dairy milk, based on JAR, that influence acceptability. Plant-based beverages, while competing for shelf-space in the dairy retail case, do not match consumer just-about-right expectations for sensory quality as well as dairy milk. Understanding consumer emotional perception of dairy milk compared with plant based milk beverages can add value to marketing and promotion of fluid milk in a competitive beverage market.

**Key Words:** milk, emotions, just-about-right scale

#### **T119 Effect of storage temperature on physico-chemical and sensory attributes of ready-to-drink breakfast smoothie.**

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Shelf-life is an important consideration for ready-to-drink (RTD), long-shelf-life beverages such as smoothie. For any new product, product developer would be interested to know the shelf-life of the products when stored at different temperature. The objective of the present study was to determine the physico-chemical and sensory stability of formulated smoothie at 3 different temperatures ( $4^\circ\text{C}$ ,  $30^\circ\text{C}$  and  $40^\circ\text{C}$ ). Sensory analysis using trained panel as well as physico-chemical attributes such as expressible serum (ES), acidity, pH, viscosity were studied to determine the shelf-life. An expert panel consisting 10 member evaluated the products for its color and appearance (C&A), flavor, consistency, sweetness and overall acceptability (OA) based on 9-point hedonic scale at the interval of every 15 d. Effect of storage period on acceptability of smoothie at 3 different temperatures was analyzed using IBM SPSS 20 software and study was done in triplicate. ES and acidity values were increased significantly ( $P < 0.05$ ) at all temperatures during storage and highest increment for both parameters were found at  $40^\circ\text{C}$  while lowest increment were at  $4^\circ\text{C}$ . Viscosity was increased during storage at  $4^\circ\text{C}$  but reduction was found at rest of 2 temperatures ( $P < 0.05$ ). pH of product decreased at all temperatures significantly ( $P < 0.05$ ). Sensory attributes; C & A, flavor, sweetness and OA scores were significantly ( $P < 0.05$ ) reduced during storage at all temperatures. Highest reduction in scores was found at  $40^\circ\text{C}$  while lowest reduction was at  $4^\circ\text{C}$ . In case of consistency, score was increased during storage at  $4^\circ\text{C}$  while reduction in score was observed at  $30^\circ\text{C}$  and  $40^\circ\text{C}$  ( $P < 0.05$ ). Shelf life studies revealed that the product remained an acceptable up to 90, 75 and 60 d at  $4^\circ\text{C}$ ,  $30^\circ\text{C}$  and  $40^\circ\text{C}$  respectively.

**Key Words:** ready-to-drink, smoothie, storage study

#### **T120 Physico-chemical profile and mineral content of yogurts and whey beverages available in the Brazilian market.**

Thaiza Serrano, Simone Lorena, Adriano Cruz, and Renata Raices\*, Federal

The physico-chemical profile and the minerals content in yogurt and whey beverage marketed in Brazilian market were investigated. Twenty samples (Ten yogurt and 10 whey beverages) presented in the label Federal Inspection Service (SIF) were submitted to physicochemical analysis (moisture, proteins and lipids levels) minerals contents (Ca, Fe, K, Mg, Na, Cd, Cr, Cu, Mn and Pb) being the latter identified and quantified from Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES). Absence of difference was noted in moisture content, as the yogurt samples presented mean value  $82.12 \pm 0.21\%$  while and the dairy beverages presented  $83.08 \pm 0.06\%$  w/w ( $P > 0.05$ ). Both products presented protein content of yogurt above the minimum required by the legislation ( $4.87 \pm 0.42\%$  w/w and  $2.72 \pm 0.13\%$  w/w, yogurt and whey beverage, respectively) which present an advantage on the nutritional point of view. Regards the lipid levels, the yogurt presented high values compared with whey beverages ( $2.00 \pm 0.07$  and  $1.23 \pm 0.06\%$  w/w, respectively,  $P < 0.05$ ). Calcium was the mineral with the highest concentration in both product categories (mean values  $1.36 \pm 0.36$  and  $1.07 \pm 0.36$  mg/g, respectively,  $P < 0.05$ ), followed by potassium (mean values  $1.60 \pm 0.29$  and  $1.41 \pm 0.21$  mg/g, respectively,  $P > 0.05$ ), sodium (mean values  $0.74 \pm 0.15$  and  $0.68 \pm 0.09$  mg/g, respectively,  $P > 0.05$ ), magnesium (mean values  $0.16 \pm 0.04$  and  $0.11 \pm 0.01$  mg/g, respectively,  $P > 0.05$ ) and iron ( $0.01 \pm 0.00$  mg/g for both products categories). It was not detected minority constituents (Cd, Cr, Cu, Mn and Pb), in both products.

**Key Words:** yogurt, whey beverage, characterization

**T121 Probiotic fermented milk with banana flour: Understanding the prebiotic effect.** Aline Silva, Renata Raices, Marcia Cristina, Luciana Nogueira, and Adriano Cruz\*, *Federal Institute of Science and Technology of Rio de Janeiro (IFRJ), Rio de Janeiro, Brazil.*

This study aimed to investigate the effect of adding increased amounts of banana flour (0, 0.5, 1.5 e 3 w/w %, I, II, III, IV) in probiotic fermented milk processing. Microbiological (*Streptococcus thermophilus*, *Lactobacillus acidophilus* and *Bifidobacterium* counts), Physico-chemical analysis (gross composition, resistant starch, pH, proteolysis and firmness) were performed weekly along 21 d. While *S. thermophilus* presented always values about 8 log cfu/g for all samples, probiotic microorganisms presented different behavior. While *L. acidophilus* presented a decrease in the viable counts in the samples added with 3 w/w % banana flour (from 8.03, d 1, to 7.14 log cfu/g, d 21,  $P < 0.05$ ), *Bifidobacterium* present increase along the storage time (6.78, d 1 to 7.14 log cfu/g, d 21,  $P < 0.05$ ). Toward the physico-chemical parameter, pH did not present difference among the samples and related to storage time (values ranged 4.60 to 4.67,  $P > 0.05$ ), while increased proteolysis level was observed at the fermented milks added with intermediate levels of banana flour (values ranged from 0.430 to 0.667, sample II and 0.455 to 0.662, sample III) in relation to sample III (values ranged from 0.345 to 0.378,  $P < 0.05$ ). For the firmness, it was observed direct relation with the banana flour addition (values ranged to 1.23, sample I to 1.88 N, sample IV) without relation to the storage time. The gross composition reported absence of difference among the fat and proteins levels while for resistant starch content, sample III presented increased decrease (30.11%) compared with sample IV (20.07%), respectively. Our findings suggest the need to adjust the banana flour dosage during

probiotic fermented milk processing as well as the probiotic culture to be added at the formulation to obtain the prebiotic effect.

**Key Words:** banana flour, probiotic fermented milk

**T122 Probiotic yogurt with glucose oxidase: Performance with commercial products and potential advantages.** Aline Batista, Marcia Silva\*, Renata Raices, Luciana Nogueira, and Adriano Cruz, *Federal Institute of Science and Technology of Rio de Janeiro (IFRJ), Rio de Janeiro, Brazil.*

The quality parameters of probiotic yogurt with addition of glucose oxidase (250 and 500 ppm, GOXI and GOXII) compared with commercial products available in the Brazilian market were investigated. Microbiological (probiotic bacteria count), physical-chemical (pH, proteolysis) and metabolic activities (production of organic acids, flavor and aroma compounds and fatty acid profile) were performed. High counts were observed for all microorganisms, with values above 9 log cfu/mL for *S. thermophilus*, and 8 log cfu/mL for *L. bulgaricus*, *L. acidophilus*, and *Bifidobacterium* ( $P > 0.05$ ). In addition, they presented lower post-acidification values and the highest proteolysis values, which ranged from 4.17 to 4.21 and 0.796 to 0.788, respectively,  $P < 0.05$ ), as well as exhibited similar results for lactic acid and acetic acid (from 1.34 to 1.37, and 0.72 to 0.79 mg/mL,  $P > 0.05$ ), and higher values for diacetyl and acetaldehyde. Regards the fatty acid profile, it were observed intermediate values for short- and medium-chain fatty acids (SCFA and MCFA, in the range of 1.82 to 2.20, and 16.3 to 17.07 g/100 g lipids,  $P < 0.05$ ) and higher values for long-chain fatty acids especially for the yogurt GOX1 (LCFA, in the range of 80.73 to 81.92 g/100 g lipids,  $P > 0.05$ ). In addition the yogurt GOX1, presented higher values for monounsaturated and polyunsaturated fatty acids (MUFA and PUFA, in the range of 30.14 to 34.26, and 4.04 to 4.12 g/100 g lipids,  $P < 0.05$ ), as well as, higher values for trans-vaccenic acid and for linolenic and conjugated linoleic acids (C18:1t, C18:2 and CLA, in the range of 3.71 to 3.72, 2.84 to 2.89, and 1.28 to 1.27 g/100 g lipids, respectively,  $P > 0.05$ ). In a functional food perspective, the addition of glucose oxidase to probiotic yogurts may be an interesting technological option for small and medium-size dairy enterprises to enter to the market of functional dairy foods.

**Key Words:** probiotic yogurt, glucose oxidase

**T123 Physical characteristics of set-type probiotic yogurt produced from mixtures of cow and sheep milk.** Felipe S. Vianna, Celso Fasura Balthazar\*, and Adriana C. O. Silva, *Department of Food Technology, Veterinary College, Federal Fluminense University, Niterói, Rio de Janeiro, Brazil.*

Bovine milk is the most important from commercial and industrial point of view, because the milk supplied from cows is cheaper and more plentiful than sheep milk. Thus, large-scale industrialization of the dairy sheep sectors in many countries is limited by low volume and seasonal cyclicality of individual milk production. As well, it is known that milk is more rich in milk total solid than bovine milk and has interesting features concerning texture technological aspects, thus this milk is used to produce dairy products. To combine the benefits of sheep milk plus the more affordable bovine milk, the purpose of this study was to develop a functional set-type probiotic yogurt from the mixture of cow and sheep milk, to establish a comparison among the mixtures (25/75; 50/50; 75/25 v/v sheep/cow milk) and control treatments through evaluation of yogurt physical composition. Sheep milk yogurt (SMY) obtained the highest ( $P < 0.05$ ) firmness value. Thus, an increase in the

amount of sheep milk for yogurt increased the firmness of mixtures. Different sheep milk ratios in cow milk did not influence ( $P < 0.05$ ) the apparent viscosity of the yogurt mixtures. Concerning the water-holding capacity, SMY obtained the highest values, whereas CMY exhibited the lowest values ( $P < 0.05$ ). The yogurts produced with mixed milk (50/50 v/v) presented an intermediate value when compared with the other treatments. All treatments presented significant differences in syneresis index and the greater the amount of sheep milk in the yogurt resulted in lower index of syneresis. Thus intermediate mixed milk yogurts showed satisfactory results for physical analyses.

**Key Words:** firmness, sheep milk, water-holding capacity

**T124 Physical characteristics of set-type yogurts produced using milk from different ruminants species.** Felipe S. Vianna, Celso Fasura Balthazar\*, and Adriana C. O. Silva, *Department of Food Technology, Veterinary College, Federal Fluminense University, Niterói, Rio de Janeiro, Brazil.*

Most published studies involving dairy products used cow milk because of large volume and economic importance, however, many countries with specific climatic (desert) and terrain features (mountainous terrain) favor dairy goat and sheep farming, making this activity essential for national economy. The present study purpose was to compare physical properties of yogurts obtained from sheep (SMY), cow (CMY) and goat (GMY) milk for 28 d of storage. This research found that SMY were firmer ( $P < 0.05$ ) than those of CMY and GMY. The firmness of CMY was greater ( $P < 0.05$ ) than GMY in the 14th and 28th day of storage. In most of cases, the firmness was not altered by the storage time. Apparent viscosity varied depending on the type of milk used to produce yogurts. The apparent viscosity (AV) of SMY was higher ( $P < 0.05$ ) than CMY and GMY at 14th and 28th day. Moreover, CMY had greater ( $P < 0.05$ ) levels of viscosity than GMY. There was not, however, a significant difference during storage ( $P < 0.05$ ) for each yogurt type. The water-holding capacity (WHC) of GMY were lower ( $P < 0.05$ ) than that obtained from CMY and SMY, which had the highest levels. Furthermore, there was no influence in this ratio during the storage time ( $P < 0.05$ ). The syneresis index (SI) was significantly affected by the milk type, however it was not altered by the storage time ( $P < 0.05$ ). In contrast with WHC, the lowest syneresis was observed in SMY than in CMY and GMY. Therefore, SMY achieved the greater firmness, AV and WHC, with lower SI than CMY and GMY. On the other hand, despite the proximate composition of CMY being similar to GMY, GMY generally had the lowest firmness, AV and WHC, but highest SI.

**Key Words:** cow milk, goat milk, sheep milk

**T125 Fatty acid profile in prebiotic sheep milk yogurt.** Celso Fasura Balthazar\*, Jeremias Moraes<sup>2</sup>, Hugo Leandro A. Silva<sup>1</sup>, Renata S. L. Raices<sup>2</sup>, and Adriano G. Cruz<sup>2</sup>, *Department of Food Technology, Veterinary College, Federal Fluminense University, Niterói, Brazil, <sup>2</sup>Instituto Federal de Educação, Ciência e Tecnologia do Rio de Janeiro, Rio de Janeiro, Brazil.*

The fatty acids (FA) present in sheep milk have beneficial compounds, which might prevent cardiovascular diseases. Inulin fiber was added to sheep milk to enhance the prebiotic biological effects to this milk. This study aimed to verify the FA profile, atherogenic (AI) and thrombogenic indices (TI) in prebiotic sheep milk yogurt (PSMY) with added inulin (0, 2, 4 and 6%) during 28 d of storage. Thus, CLA *trans*-10,*cis*-12 were detected, but below qualification limit, meaning they were in low amounts in PSMY. The predominant FA were palmitic, oleic, myristic

and capric. There was an increase ( $P < 0.05$ ) of saturated fatty acids (SFA) during storage in PSMY. Therefore, palmitic appeared with approximately 33% (C16:0 and SFA:  $r = 0.582$ ), myristic with approximately 20% (C14:0 and SFA:  $r = 0.894$ ) and capric with approximately 16% (C10:0 and SFA:  $r = 0.929$ ). Oleic acid indicated an antiatherogenic (C18:1n9 and AI:  $r = -0.845$ ) and antithrombotic (C18:1n9 and TI:  $r = -0.897$ ) properties and represented around 92% of MUFA. CLA *cis*-9,*trans*-11 represented approximately 78% of PUFA and 2% of FA, which indicated that PSMY are a great source of it. There could be a direct link between CLA and oleic acid ( $r = 0.978$ ), as well CLA increased ( $P < 0.05$ ) along storage. Still, ALA was present in low levels, corresponding to about 22% of PUFA and 1% FA. There was a drop ( $P < 0.05$ ) in ALA level at the first storage measure, which was maintained until the last storage measurement. ALA has nutritional importance due to its anti-thrombogenic effect. PSMY might be considered a food with low atherogenic and thrombogenic risk, because of the low levels of AI and TI. Plus, sheep milk yogurt may have beneficial prebiotic effects with inulin addition.

**Key Words:** CLA, fermented milk, inulin

**T126 Acidity in sheep milk yogurt: The inulin effect.** Celso Fasura Balthazar\*, Rodrigo B. A. Oliveira<sup>1</sup>, and Adriano G. Cruz<sup>2</sup>, *Department of Food Technology, Veterinary College, Federal Fluminense University, Niterói, Brazil, <sup>2</sup>Instituto Federal de Educação, Ciência e Tecnologia do Rio de Janeiro, Rio de Janeiro, Brazil.*

The widespread inulin use in food industry is based on their technological and nutritional properties. This study analyzed pH, titratable acidity (TA), lactose and lactic acid in prebiotic sheep milk yogurt (PSMY), with different inulin content (0, 2, 4 and 6%) immediately after incubation and during 28 storage days at  $4 \pm 2^\circ\text{C}$ . The bacteriological count was performed at the 1st and 28th days of storage. On the 1st day, *Streptococcus thermophilus* counts were: 9 log, 10 log, 11 log, 11 log cfu.  $\text{mL}^{-1}$ , whereas *Lactobacillus delbrueckii* subs. *bulgaricus* were 8 log, 8 log, 8 log, 9 log cfu.  $\text{mL}^{-1}$ . On the 28th day, *S. thermophilus* were 8 log, 8 log, 8 log, 9 log cfu.  $\text{mL}^{-1}$ , and *L. bulgaricus* were 6 log, 7 log, 7 log, 7 log cfu.  $\text{mL}^{-1}$  for PSMY treatments, respectively. There was a significant decrease ( $P < 0.05$ ) in both lactic acid bacteria during storage, due to inulin protective effect. As expected, the pH decreased and TA increased significantly (pH and TA:  $r = -0.835$ ;  $P < 0.05$ ) of PSMY during storage. There was a significant difference ( $P < 0.05$ ) between PSMY with different inulin content, being 0% the most acidic and 6% the less acidic. As well, pH values were not significantly influenced ( $P < 0.05$ ) by the inulin addition. Lactose had a significant decrease ( $P < 0.05$ ) expected through storage ( $r = -0.759$ ) in PSMY. Inulin content had a significant impact ( $P < 0.05$ ) in lactose concentration, because lactose amount was higher in PSMY 6%, dropping in each treatment until PSMY 0%, which had a significantly lower concentration ( $P < 0.05$ ) of lactose during measurement days. This statement suggested that the acid lactic bacteria consumed inulin instead of lactose proportionally with inulin amounts, meaning that lactose was proportionally degraded by lactic acid bacteria into lactic acid (lactose and lactic acid:  $r = -0.826$ ). Thus, lactic acid concentration of PSMY 0% was higher ( $P < 0.05$ ) compared with others. Lactic acid had a significant increase during storage ( $r = +0.936$ ;  $P < 0.05$ ) in PSMY. Moreover, inulin amount delayed the acidity increase in PSMY (lactic acid and TA:  $r = +0.919$ ), suggesting that inulin fiber retarded the acidity growth in yogurts, which is pleasant for consumer and dairy industry.

**Key Words:** ferment milk, inulin, lactic acid.



**T127 A clean label approach for manufacture of yogurts using tailored milk protein interactions.** Gopinathan H. Meletharayil\* and Hasmukh A. Patel, *South Dakota State University, Brookings, SD.*

Tailoring protein interactions can be used as a strategy to improve textural properties that can potentially replace stabilizers in yogurts. The objective of the present study was to investigate the influence of functionalized milk proteins in yogurt formulations of varying protein content on the rheological and textural properties of yogurt. Functionalized retentate (FUR) was obtained by injecting CO<sub>2</sub> prior to and during the ultrafiltration of skim milk (SM) at ~10°C. Control and test yogurt formulations containing 3.0, 4.0 and 5.0% w/w of total protein were prepared with control containing proteins from SM and test formulations containing proteins from SM and FUR at 1:1 protein ratio. Formulations were heated at 90°C for 10 min, followed by cooling to 42°C. Following inoculation with yogurt culture, formulations were incubated at 42°C for 4 h in cups. After overnight storage at 4°C, the samples were analyzed for hardness using a texture analyzer and flow behavior using a rheometer. Experimental data were tested for ANOVA and statistical significance ( $P < 0.05$ ) was determined, using statistical software SAS. The hardness values significantly increased ( $P < 0.05$ ) from 189, 305 and 426 g in control yogurts to 236, 351 and 511 g in yogurts containing functional proteins at 3.0, 4.0 and 5.0% protein respectively. Similarly, the consistency coefficient, K, increased significantly ( $P < 0.05$ ) from 6.1, 10.3 and 18.7 Pa.S<sup>n</sup> in control yogurts to 10.68, 21.2 and 25.80 in yogurts containing functionalized milk proteins. Apparent viscosity ( $\eta_{70}$ ) significantly increased ( $P < 0.05$ ) from 0.45, 0.67 and 1.1 Pa.S in control yogurts to 0.51, 1.08 and 1.25 Pa.S in yogurts containing functional milk proteins at 3, 4, and 5% protein respectively. These results showed that there was an increase in the hardness, consistency coefficient and apparent viscosity of yogurts containing functionalized milk protein at all the three protein levels, which could be attributed to greater protein interactions in the serum phase. The results of the present study clearly indicate that the use of functionalized milk in yogurt formulation can be used as a potential approach to replacing stabilizers such as gelatin in yogurts and thus achieving a step towards clean label products

**Key Words:** clean label, yogurt, carbon dioxide

**T128 Application of computing technology in simulation of consumer acceptance of typical hard ice creams during storage** Maryam Bahram-Parvar\*<sup>1,2</sup>, Fakhreddin Salehi<sup>2</sup>, and Seyed Razavi<sup>2</sup>, <sup>1</sup>*University of Guelph, Guelph, ON, Canada,* <sup>2</sup>*Ferdowsi University of Mashhad, Mashhad, Khorasan Razavi, Iran.*

Sensory evaluation is widely used in the food industry, especially for quality inspection, product design and marketing. Ice cream acceptability by consumers is mainly perceived by means of texture and flavor. Various ingredients contribute to ice cream's complex colloidal structure and make its perception difficult. In addition, it requires training the judges and proper environmental conditions during assessment. Furthermore, one distinctive characteristic of sensory responses is that they are ambiguous and imprecise; that is, they are fuzzy. Then, by normal statistical analysis of sensory data obtained through subjective evaluation, a complex idea of a product quality is often generated, which makes it nearly impossible to determine the strength and weakness of the product concerning its sensory attributes. Therefore, application of fuzzy set concept could be useful in this area. Combination of fuzzy concept and artificial neural network is of great importance among various combinations of methodologies in soft computing. There are some reports dealing with the sensory evaluation of ice cream based on texture perception. However, there are no or few studies available concerning

the use of computing technology for prediction of consumer acceptance of ice cream. Hence, the objectives of this work were to investigate the efficiency of genetic algorithm-artificial neural network (GA-ANN) and adaptive neuro-fuzzy inference system (ANFIS) simulations for predicting acceptability of hard ice creams during storage. For this purpose, GA-ANN and ANFIS were fed with 4 inputs of flavor, iciness, wateriness and creaminess for prediction of overall acceptability of ice cream. Both models were trained with experimental data. The developed GA-ANN, which included 16 hidden neurons, could predict total acceptance with correlation coefficient of 0.93. The overall agreement between ANFIS predictions and experimental data was also very good ( $r = 0.92$ ). Results of present research showed that both GA-ANN and ANFIS models' predictions agreed well with testing data sets and could be useful for understanding and controlling factors affecting palatability of ice cream.

**Key Words:** ice cream, quality, storage

**T129 Evaluation of textural property changes in three types of low-fat goat milk ice creams during 8 weeks of frozen storage.** Christopher E. McGhee, Brittany I. Davis, Krishna P. Bastola\*, Jolethia O. Jones, and Young W. Park, *Fort Valley State University, Fort Valley, GA.*

Although ice cream is a popular frozen food around the world, low-fat ice creams have problems of low flavor and low textural quality compared with traditional ice creams. Three types of low-fat soft-serve goat milk ice creams were manufactured using whole milk (3.64% fat), 2% fat and skim (0.71% fat) goat milk, and stored at -18°C for 0, 2, 4, 8 weeks to evaluate the effect of extended frozen-storage on textural properties of the caprine products. A commercial cow milk based powdered vanilla flavor pre-mix containing 2.5% fat (Alpha Freeze, Tampa, FL) was formulated into the 3 types of fluid goat milk base for ice cream manufacture, where the final fat contents of the whole, 2% and skim milk mixes were 5.31, 3.84, 1.87%, respectively. Textural characteristics of the ice cream were evaluated using a texture analyzer (TA-XT2 Texture Technologies Corp., Scarsdale, NY). A cylindrical probe, which was made of acrylic material and 2.5 cm in diameter and 3.5 cm in height, was used for detecting shear force (g). The results showed that mean values (g force) of firmness and consistency for freshly made soft serve whole milk, 2% and skim milk goat ice creams were: 208.7, 297.3; 182.8, 261.8; 183.5, 255.0, respectively. There were significant increases ( $P < 0.001$ ) in firmness and consistency of all 3 types of soft-serve low-fat goat ice creams after 1 d frozen-storage. The similar trend of increase was observed in cohesiveness and index of viscosity for all tested products, probably due to the hardening of the texture of the frozen products after 1 d storage. Regardless of fat level of the low-fat caprine milk ice creams, all tested textural properties after 56 d frozen-storage revealed substantial increases ( $P < 0.0001$ ), especially in firmness and consistency traits compared with those of the original soft-serve goat ice creams with extreme high variations. It was concluded that all textural properties of the experimental low-fat goat milk ice creams were very significantly increased after 8 weeks extended frozen-storage.

**Key Words:** goat ice cream, frozen storage, textural property

**T130 Dulce de leche: Characterization by physicochemical and instrumental methods.** Leonardo Gaze<sup>2</sup>, Carlos Conte-Junior<sup>2</sup>, Adriano Cruz<sup>1</sup>, Renata Raices\*<sup>1</sup>, and Monica Freitas<sup>2</sup>, <sup>1</sup>*Federal Institute of Science and Technology of Rio de Janeiro (IFRJ), Rio de Janeiro, Brazil,* <sup>2</sup>*Federal University Fluminense, Rio de Janeiro, Brazil.*

The physicochemical profile of dulce de leche (DL) was determined by both routine analysis and others techniques (HPLC, GC-MS and ICP-OES). Seven Brazilian commercial brands were characterized for moisture content, protein, fat, ash, pH and titratable acidity, mineral content (sodium, potassium, calcium, and phosphorus) and instrumental analysis (carbohydrates content and volatile compounds). Protein, lipid, ash, moisture, lactic acid content and pH ranged respectively from 3.51 to 7.12; 3.56 to 6.99; 1.31 to 2.05; 17.49 to 29.67; 0.23 to 0.50; and 6.14 to 6.37 w/w ( $P < 0.05$ ), with a significant variability in all parameters. Regards the mineral content, it is also reported difference between these parameters was observed for all samples, ranging

from 0.12 to 0.16 g/100 g; 0.28 to 0.40 g/100 g; 0.19 to 0.36 g/100 g; 0.14 to 0.24 g/100 g for sodium, calcium, potassium and phosphorus, respectively ( $P < 0.05$ ). In concordance, a great variation was observed for glucose content, which ranged from 0.09 to 0.75 g/100 g for all the samples, suggesting heterogeneity of the technological processing of DLs. 32 volatile compounds were identified belonging to 10 different chemical families of which only 2 were not present in all samples. In this sense, an increase of intrinsic quality of DL is related closely the standardization of operational parameters using during the manufacture.

**Key Words:** dulce de leche, quality, control quality

## Dairy Foods: Microbiology

**T131** *Lactobacillus plantarum* L67 suppresses allergic inflammation. Sooyeon Song<sup>\*1</sup>, Anna Jeong<sup>1</sup>, Geun-Bae Kim<sup>2</sup>, Dong-June Park<sup>3</sup>, and Sejong Oh<sup>1</sup>, <sup>1</sup>Div. of Animal Science, Chonnam National University, Gwangju, South Korea, <sup>2</sup>Dept. Animal Science, Chung-Ang University, Anseong, South Korea, <sup>3</sup>Korea Food Research Institute, Seongnam, South Korea.

Bisphenol A (BPA) is a widely used monomer of polycarbonate plastics and epoxide resin that has been implicated in allergy related disease. Allergy is an abnormal immune response to an allergen. Type I hypersensitivity is an immunoglobulin (Ig) E-mediated allergic disorder. This study investigated the inhibitory effect of the *Lactobacillus plantarum* L67 extract on BPA induced allergic inflammatory response in RBL-2H3 cells. RBL-2H3 cells were treated with bisphenol A (50  $\mu$ M/mL) and co-treated with the *L. plantarum* L67 extract (5–100  $\mu$ g/mL) for 30 min. histamine releasing,  $\beta$ -hexosaminidase and intracellular  $\text{Ca}^{2+}$  level were estimated in the medium. IgE production and activities of PKC, iNOS were measured ELISA and Western blotting separately. Also IL-4 and TNF- $\alpha$  were measured by RTq-PCR. The histamine releasing of treatment with the *L. plantarum* L67 extract considerably decreased in a concentration dependent manner. When the cells were treated with the 100  $\mu$ g/mL *L. plantarum* L67 extract, the values of histamine releasing decreased 0.48-fold compared with the BPA treatment alone. The addition of BPA caused a 1.43-fold increase in IgE levels compared with control, while the values of IgE amounts decreased by 1.31, 1.28, 1.14, 1.08 and 1.04 at addition *L. plantarum* L67 extract (5–100  $\mu$ g/mL) in the presence of BPA, compared with that for the BPA treatment alone. Our results showed that *L. plantarum* L67 extract inhibited the histamine releasing,  $\beta$ -hexosaminidase, productions of IgE, the intracellular  $\text{Ca}^{2+}$  level, and activity of iNOS in BPA treated RBL-2H3 cells but also the results indicated that the *L. plantarum* L67 extract inhibits expression of cytokines related to allergy such as TNF- $\alpha$  and IL-4 on the degranulation stage of mast cell. The *L. plantarum* L67 extract can inhibit to occur allergy caused by environmental hormone such as BPA.

**Key Words:** bisphenol A,  $\beta$ -hexosaminidase, *Lactobacillus plantarum*

**T132** Metal-chelating and ACE-inhibitory activity of a milk fermented with bacteria isolated from double cream cheese of Chiapas, Mexico. Claudia Y. Figueroa<sup>\*</sup>, Gustavo F. Gutiérrez, and Humberto Hernández, *Escuela Nacional de Ciencias Biológicas-IPN, México City, México.*

During the fermentation of milk with lactic acid bacteria, biologically active sequences, known as bioactive peptides, can be produced. Among the most important biological activities attributed to these peptides are metal-chelating (mainly iron and calcium) and ACE-inhibitory activities. Three probiotic strains (*Lactobacillus plantarum*, *Lb. pentosus* and *Lb. acidipiscis*) were isolated from double cream cheese produced in Chiapas (Mexico). This study aims to assess the metal-chelating (iron and calcium) and ACE-inhibitory activity during the milk fermentation by each microorganism and by a mixed culture. Fermentations were performed using reconstituted skim milk powder. Milks were inoculated with 2% of each of the microorganisms or mixed culture. Fermentations were performed at 37°C for 48 h. Samples were taken every 8 h for measurement of microbial growth, protein concentration, degree of proteolysis (TNBS method), metal-chelating (calcium and iron) and ACE-inhibitory activity. Variations on free amino group concentration with respect to the

initial concentration that could indicate proteolysis during fermentation were observed. The initial iron-chelating activity was high (68  $\pm$  2%), and was attributed to the fact that some milk proteins have the ability to bind iron. However, during the fermentation the iron-chelating activity increased (99  $\pm$  0.6% of iron-chelating activity in *Lb. plantarum* fermentation). The other fermentation batches showed a lower percentage of iron-chelating activity. In the case of the calcium-chelating activity, an initial activity of 5  $\mu$ M  $\text{Ca}^{2+}$ /mg protein which increased up to 18  $\mu$ M  $\text{Ca}^{2+}$ /mg protein in *Lb. acidipiscis*, *Lb. plantarum* and the mixed fermentations could be measured. *Lactobacillus plantarum* fermentation showed the lowest ACE inhibitory activity (its maximum value is 78  $\pm$  2.3% of ACE-inhibition) while other fermentations reached 97  $\pm$  3% of ACE-inhibition activity. The initial ACE-inhibition activity was 35  $\pm$  3%. All the fermentation batches performed generated metal-chelating (calcium and iron) and ACE-inhibitory activities in these in vitro tests which could be important for the production of functional dairy products.

**Key Words:** bioactive peptides, metal-chelating activities, ACE-inhibitory activity

**T133** Virulence and regulator gene expression in *Bacillus* spp. from ultrapasteurized organic milk. Alyssa Grutsch<sup>\*</sup> and John McKillip, *Ball State University, Muncie, IN.*

We isolated a *Bacillus amyloliquefaciens* strain present in ultra-high temperature (UHT) pasteurized organic whole milk to ascertain virulence determinants present in this species, as well as the pattern of virulence gene expression over time in a model food (UHT milk) system compared with the type strain *Bacillus cereus* ATCC14579. The overall goal of this project was to genotypically and phenotypically characterize thermotolerant *B. amyloliquefaciens* virulence potential and the presence of the global regulator effector PlcR. Recovery of bacteria from milk required an enrichment in brain-heart infusion broth, incubated aerobically for >24 h, after which time samples were spread-plated onto tryptic soy agar (TSA) plates to recover *Bacillus* spp. Resulting colonies were streak plated onto TSA to ensure purity of culture before catalase testing, Gram and spore-staining to presumptively identify to the genus level. Pure cultures were biochemically identified to the species level using the Microgen *Bacillus* ID system (Hardy Diagnostics), yielding an identification of *Bacillus firmus*, and validated further using fatty acid profiling and 16S rDNA sequencing, which revealed the isolate to be a strain of *B. amyloliquefaciens*. To confirm presence of the target genes in each isolate, DNA was extracted from pure cultures during late log phase. Quantified DNA template was used in real-time (SYBR Green-based) PCR with primers specific for each of the target genes: *plcR*, *codY*, *nheA* and *hblC*, and the 16S rRNA gene as a control. PCR results indicated no significant difference existed between average melting temperatures ( $T_m$ ) of 16S rRNA ( $P = 0.435$ ) and *plcR* ( $P = 0.341$ ) of *B. amyloliquefaciens* and *B. cereus*. The applications of this project will be to determine if parameters regarding shipment, storage, and shelf life of UHT organic milk should be revisited, to ensure quality before consumption of product that may harbor thermotolerant toxigenic *Bacillus* spp.

**T134** Sequencing and annotation of novel plasmids from *Lactobacillus curvatus*. Jordan Hendricks<sup>1</sup>, Craig Oberg<sup>\*1</sup>, Michele Culumber<sup>1</sup>, Taylor Oberg<sup>2</sup>, Donald McMahon<sup>2</sup>, and Jeff Broadbent<sup>2</sup>, <sup>1</sup>Weber State University, Ogden, UT, <sup>2</sup>Utah State University, Logan, UT.



*Lactobacillus curvatus*, a nonstarter lactic acid bacteria (NSLAB), is rapidly becoming the most prevalent NSLAB strain isolated from aged Cheddar cheese. Plasmids often carry genes that confer advantageous traits, which provide a survival advantage to the organism. Understanding the genotype of *Lb. curvatus* plasmids could provide important information concerning genes that provide *Lb. curvatus* an advantage in the cheese environment. Plasmids were isolated from 2 different strains of *Lb. curvatus*, WSU-1 and LFC-1. These strains were isolated from distinct geographical areas. WSU-1 contained 7 plasmids while 4 plasmids were detected in LFC-1. Similarities, at least in size, exist between the largest (40kb) and smallest (1240 bp) plasmids in both *Lb. curvatus* strains. The genomes of both strains have been sequenced and several putative plasmid contigs identified. These sequences were paired with similarly sized plasmids isolated from the organisms and the identified open reading frames compared. Each strain contained a plasmid, of different sizes, with nearly identical genes that coded for an anti-toxin part of a toxin-antitoxin system. Plasmid contigs also coded for cation transport proteins that could promote survival in aging cheese. Variation in plasmid profiles between the 2 *Lb. curvatus* strains also suggests multiple strains may be circulating in cheese plants. WSU-1 and LFC-1 strains carry a greater complement of plasmids than are typically found in dairy lactobacilli. Maintaining a large set of plasmids has a high metabolic cost to the cell, indicating these plasmids contain genes of value to the organism.

**Key Words:** NSLAB, plasmids, *Lactobacillus curvatus*

**T135 Comparative analysis of prebiotics on growth kinetics, fermentation, and antioxidant activity of probiotics.** Evelyn Puspitasari\*<sup>1</sup>, Chi Kong Yeung<sup>2</sup>, and Marie Yeung<sup>1</sup>, <sup>1</sup>Biological Sciences Department, California Polytechnic State University, San Luis Obispo, CA, <sup>2</sup>Dairy Science Department, California Polytechnic State University, San Luis Obispo, CA.

Prebiotics are nondigestible oligosaccharides that selectively stimulate the growth of beneficial bacteria in the human intestine. Fructooligosaccharide (FOS) and inulin are among the most common prebiotics used in food products and dietary supplements. Previous studies examining the bioactivity of lactulose suggest that it also has a prebiotic potential. Lactulose is a derivative of lactose in which the glucose moiety is isomerized to fructose in the presence of heat; and hence can be found in heated milk. The goal of this study was to establish growth kinetics of common probiotics cultured in FOS, inulin or lactulose as the sole carbohydrate source. Fermentation of the prebiotics and the antioxidant activity of spent medium were also characterized. Dextrose and non-probiotic species were included for comparison. Eight commercial and ATCC strains were cultured in a semi-defined modified MRS, modified MRS plus L-cysteine-HCl, or peptone medium containing 1% carbohydrate. When incubated at 37°C in aerobic condition, except for *Lactobacillus casei*, probiotic strains cultured in prebiotics did not reach maximum growth rate or yield compared with dextrose. Multiple pairwise comparisons showed that lactulose tended to produce better growth than FOS and inulin in *L. rhamnosus* and *L. acidophilus*. In anaerobic condition, non-probiotic species were able to catabolize all carbohydrate sources, but displayed weaker lactic acid production (0.13 ± 0.06%) relative to *Bifidobacterium* spp. (0.30 ± 0.09%) or *Lactobacillus* spp. (0.56 ± 0.27%), as measured by titratable acidity. Antioxidant activity, with Trolox as the reference standard, was inversely correlated with the pH of the spent medium after fermentation ( $P = 0.022$ ). Among the 3 prebiotic substrates, lactulose yielded the lowest pH in *L. acidophilus* and *B. infantis*, while FOS appeared to be preferred by *L. casei* and *B. bifidum*. Antioxidant activity was overall the highest from lactulose

fermentation. In conclusion, lactulose is a promising prebiotic ingredient that can be incorporated in functional food products.

**Key Words:** prebiotic, probiotic, lactulose

**T136 Heat tolerance of *Lactococcus lactis* with prior subjection to mild heat stress.** Ingrid Osorio\* and Kayanush J. Aryana, School of Animal Sciences, Louisiana State University Agricultural Center, Baton Rouge, LA.

*Lactococcus lactis* has been associated with cheese manufacturing. It is important that the cultures used are able to survive to adverse heat conditions in manufacturing of probiotic process cheese. The hypothesis was whether prior exposure to mild heat would enhance heat tolerance of *L. lactis*. The objective was to evaluate the effect of prior exposure to mild heat on the growth of *Lactococcus lactis*. *L. lactis* R-604 was subjected to heat shock at 40 or 50°C for 1 h. Control was not subjected to heat shock. Cultures were subsequently incubated for 24 h at 30°C followed by subjecting them to batch pasteurization (for ice cream mix) at 71.11°C for 30 min. M17 Agar with 10% w/v lactose was used for plating. Plates were incubated aerobically at 30°C for 48 h. Each experiment was conducted 3 times. Counts observed after subjecting mildly heat treated *L. lactis* to batch pasteurization were 4.4 Log for 40°C, 4.3 Log at 50°C, and 1 Log for the control. Exposure of *L. lactis* R-604 to mild heat enhanced its tolerance to heat (batch pasteurization).

**Key Words:** heat tolerance, dairy culture

**T137 Effect of ultraviolet light exposure and mild heat shock on the salt tolerance of *Lactococcus lactis*.** Ernesto E. Gonzalez-Duran\* and Kayanush J. Aryana, School of Animal Sciences, Louisiana State University Agricultural Center, Baton Rouge, LA.

*Lactococcus lactis* is a dairy culture bacterium widely used in dairy products which contain salt (NaCl) such as cheese and salted butter. Osmotic conditions generally hinder the growth of both pathogen and desirable bacteria. It has been observed that exposure to an environmental stress can develop resistance to several stresses. Some studies have shown that short UV (UV) light exposure build up resistance to acid, ethanol, hydrogen peroxide and heat induced stress. There are many proteins and low molecular weight compounds that are produced under more than one stress condition which protects the cell. If salt tolerance is enhanced in desirable bacteria they would survive better compared with pathogens in salty environments. The hypothesis was whether salt tolerance of *Lactococcus lactis* can be enhanced. The objective was to study the influence of UV light and heat shock on salt tolerance of *L. lactis*. *L. lactis* R-604 cells were exposed to mild stresses of UV light (245 nm) for 5 min or a heat shock at 50°C for 25 min. A control sample was run without any stress. Samples were transferred to M17 broth with 5 concentrations of NaCl (0, 1, 3, 5 and 7% w/v) and incubated aerobically at 30°C for 5 d. Plating was conducted immediately after inoculation and every 24 h for 5 d in M17 agar with 0.5% of lactose and incubated aerobically at 30°C for 48 h. Three replications were conducted. On d 1 and 2 no differences were observed for either UV light or heat shock treated cells when exposed to 0, 1 and 3% w/v NaCl, however growth inhibition was observed using UV light or heat shock at 7% w/v NaCl. On d 1, 2 and 3, resistance to salt was improved using UV light or heat shock treated cells when exposed to 5% w/v NaCl compared with control. On d 5, mild heat shock and UV light exposure had no effect on salt tolerance. Until d 4, different amount of NaCl and days of salt exposure had a differential effect on growth of *L. lactis* R-604.

**Key Words:** salt tolerance, dairy culture

**T138 Influence of osmotic adaptation and lactose deprivation on the salt tolerance of *Lactococcus lactis*.** Ernesto E. Gonzalez-Duran\* and Kayanush J. Aryana, *School of Animal Sciences, Louisiana State University Agricultural Center, Baton Rouge, LA.*

*Lactococcus lactis* is often used in cheese and salted butter manufacture. Mediterranean cheeses can have up to 7% NaCl. Osmotic conditions generally slow down the growth of both pathogen and desirable bacteria. Studies have shown that vegetative cells exposed to a mild stress become more resistant to lethal doses of the same stress since the first exposure starts the defense mechanisms of the cells creating an effect of cross-protection. The hypothesis was whether salt tolerance of *Lactococcus lactis* can be enhanced. The objective was to study the influence of lactose deprivation and osmotic adaptation on salt tolerance of *L. lactis*. *L. lactis* R-604 was subjected to mild stress induced by lactose deprivation (grown with no lactose in M17 broth) or prior osmotic adaption (grown with 3% w/v NaCl in M17 broth) for 24 h aerobically at 30°C. A control was run without stress (grown in M17 broth with lactose and no NaCl and incubated aerobically for 24 h at 30°C). Lactose deprived or osmotic adapted cells were transferred to M17 broth with 5 concentrations of NaCl (0, 1, 3, 5 and 7% w/v) and incubated aerobically at 30°C for 5 d. Plating was conducted immediately after inoculation and every 24 h for 5 d in M17 agar with 0.5% of lactose and incubated aerobically at 30°C for 48 h. Three replications were conducted. After the 24 h incubation in M17 broth with no lactose or 3% NaCl an increase in 4 log cfu/mL was observed. Lactose deprived cells exposed to 0, 1, 3 and 5% w/v salt had a stationary phase at 11 logs for d 1 and 2. While use of 7% w/v salt reduced cells by 2 logs by d 2. On d 5 there were no differences in counts of lactose deprived cells exposed to all salt concentrations. Osmotic adapted cells exposed to 5% salt had a stationary phase at 11 logs for d 1 and 2, while cells exposed to 0, 1, 3 and 7% NaCl slowly declined until d 5. On d 5 there were no differences in counts of osmotic adapted cells exposed to all salt concentrations. Different amount of NaCl and days of salt exposure had a differential effect on growth of *L. lactis*. From d 3 until d 5, prior exposure to either mild stress did not have an effect on salt tolerance of *L. lactis* R-604.

**Key Words:** salt tolerance, dairy culture

**T139 *Lactobacillus wasatchii* WDC04 associated with late gas production in aged Cheddar cheese.** Lauren Montierth<sup>1</sup>, Craig Oberg<sup>1</sup>, Michele Culumber\*<sup>1</sup>, Donald McMahon<sup>2</sup>, Fatih Ortakci<sup>2</sup>, and Jeff Broadbent<sup>2</sup>, <sup>1</sup>Weber State University, Ogden, UT, <sup>2</sup>Utah State University, Logan, UT.

A new species of nonstarter lactic acid bacteria (NSLAB), called *Lactobacillus wasatchii* WDC04, was identified in aged Cheddar cheese manufactured in northern Utah. This bacterium has been linked to gas formation in the latter stages of Cheddar cheese ripening. It is an obligate heterofermentative NSLAB shown to produce gas in broth cultures under the conditions of cheese aging. WDC04 prefers growth on ribose at low pH (5.0–5.5). It grows slowly at cold temperatures, which could play a role in its ability to create gas defects during cheese ripening. In aging cheese, gas formation causes swelling of the packaging and splitting of the cheese, making it unfit for consumer use. Twenty-seven aged Cheddar cheeses from around the world were tested for WDC04 using MRS medium (pH 5.2) amended with 1.5% ribose and incubated for 1 to 4 weeks. Isolates were identified using 16S rRNA gene sequencing then compared with the GenBank database and to the 16S rRNA gene from *Lb. wasatchii* WDC04. No *Lb. wasatchii* were detected in cheeses without gas defects. WDC04 was found, however, in several distinct aged commercial Cheddar cheeses produced in facilities geographically distant from the original isolation location. These results indicate *Lb.*

*wasatchii* is more widespread than previously thought, and appears to be a causative agent of late gas defect in aged Cheddar cheeses.

**Key Words:** NSLAB, gas production, cheese

**T140 The effect of xenon pulsed-light technology on biofilm adhered to stainless steel surfaces.** Stephanie Jacquez\* and Rafael Jimenez-Flores, *California Polytechnic State University, San Luis Obispo, CA.*

In food processing, inadequate sanitation procedures lead to the formation of biofilms, in which bacteria attach to surfaces and aggregate in a hydrated polymeric matrix of their own synthesis. Formation of these sessile communities and their inherent resistance to existing sanitation agents are at the root of the risk of bacterial infections for consumers. Based on this evidence, an effective method for reducing biofilm formation in dairy processing equipment is necessary. UV pulsed light technology has proven effective in eliminating microorganism populations on food products. The objective of this work is to evaluate the effect of pulsed light technology on a biofilm consisting of different dairy media (e.g., whey protein concentrate (WPC) and pure lactose) as well as 3 strains of spore forming *Bacillus* species (*B. subtilis*, *B. coagulans*, and *B. licheniformis*) adhered to square, 2.5 × 2.5, ASI 304, stainless steel coupons. Four treatment levels (no treatment, 5 bursts, 10 s, and 20 s) were applied to the coupon surfaces using the Xenon model RC847 machine. Each coupon was placed at a distance of 10.5 cm away from the UV lamp. The pulsed light effect was evaluated using the pour plate technique with 0.2% (w/v) starch TSA and incubated for 48h at 55 C. The adhesion of *Bacillus* to stainless steel in water as matrix was 1000 to 3000/cm<sup>2</sup> as measured in our laboratory. When compared with the No treatment group, there was a maximum of a 3.96 delta log kill rate in the biofilm created with whey protein concentrate when exposed to 20 s of pulsed UV light. Results indicate a difference between kill rates at 20 s with biofilms created with 5% whey protein concentrate and 5% lactose.

**T141 Slime production by *Bacillus* strains affects biofilm formation on dairy separation membranes.** Nuria Garcia-Fernandez\*<sup>1,2</sup>, Ashraf Hassan<sup>1</sup>, and Sanjeev Anand<sup>1,2</sup>, <sup>1</sup>Dairy Science Department, South Dakota State University, Brookings, SD, <sup>2</sup>Midwest Dairy Foods Research Center, South Dakota State University, Brookings, SD.

Our previous research showed that the hydrophobicity of the extracellular polysaccharides produced by lactic acid bacteria plays an important role in biofilm formation on dairy separation membranes. Thermotolerant bacilli possess a major challenge for the dairy industry, due to their resistance to heat and cleaning agents. The objective of this study was to evaluate the effect of slime production by *Bacillus* spp. on biofilm formation on separation membranes. Two slime-producing strains; *Bacillus mojavensis* (Bc) and *Bacillus licheniformis* (K1), isolated from dairy powder and one non-slime producing variant from each of the 2 strains (K1-1 from K1 and Bc-1 from Bc) produced by spontaneous mutation were used to study attachment (in the absence of growth) and biofilm formation on polyamide RO membrane pieces. Parameters related to bacterial adhesion (cell charge, capsule production, and hydrophobicity) were evaluated to determine their contribution to differences in biofilm formation among strains. The number of viable cells on biofilm formed by the hydrophobic Bc was more than 1 log cfu/cm<sup>2</sup> greater than its less hydrophobic slime-negative mutant (Bc-1) ( $P < 0.05$ ). However, counts lower by about 0.7 log cfu/cm<sup>2</sup> were found in biofilm formed by the hydrophilic K1 than its slime-negative mutant ( $P > 0.05$ ). Bc and K1 slime precipitated by ethanol contained only 3.6

and 6.5% of total carbohydrates. Bacterial cell surface hydrophobicity was the only parameter that strongly related to both attachment and biofilm formation on RO membranes. To confirm its role, cell surface hydrophobicity was modified by surfactants with different hydrophilic lipophilic balance (HLB) values and attachment of the altered cells was then studied. Tween 20 (high HLB) resulted in lower attachment of Bc while Span 80 (low HLB) improved biofilm formation by K1 ( $P < 0.05$ ). In conclusion, hydrophobic slime produced by *Bacillus* enhanced attachment and biofilm formation on RO membranes. Cleaning strategies that decrease cell hydrophobicity or increase membrane hydrophilicity would reduce risk of biofilm formation by the potential spoilage and disease-causing *Bacillus* strains.

**Key Words:** biofilm, *Bacillus*, slime

**T142 Identification of gram-negative bacteria in cooling tanks of dairy farms.** Magali Soares Santos Pozza\*<sup>1</sup>, Gilberto Henrique Simões<sup>2</sup>, Maximiliane Alarvase Zambom<sup>2</sup>, Maichel Lange<sup>2</sup>, and Grasielle Scaramal Madrona<sup>1</sup>, <sup>1</sup>Universidade Estadual de Maringá, Maringá, Brazil, <sup>2</sup>Universidade Estadual do Oeste do Paraná, Marechal Cândido Rondon, Paraná, Brazil.

The main parameter to verify the quality of milk is the microbiological profile and microbial contamination index. The aim of this study was to isolate and identify gram-negative proteolytic bacteria in different dairy production systems in western Paraná, Brazil. Thirty-five milk samples collected directly from milk tanks evaluated for flow cytometry by somatic cells count (SCC) and total bacteria count (TBC). Samples were diluted in peptone water and seeded in caseinate agar. The colonies were inoculated into *Escherichia coli* broth for 48 h at 35°C. Subsequently were inoculated Eosin Methylene Blue Agar (EMB) and inoculated in Bactray I and II Kit, for the purpose of identification of gram-negative oxidase negative bacteria. Twenty 7 samples showed isolation and identification of some kind of agent, with 77.1% of bacteria isolated. The mean values obtained for TBC and SCC were 1.867.000 cfu/mL and 944.000 cels/mL respectively, values considered above the current legislation. The frequencies of isolated species were *Escherichia coli* (8.6%), *Escherichia fergusonii* (8.6%), *Klebsiella oxytoca* (8.6%), *Yersinia enterocolitica* (8.6%), *Hafnia alvei* (5.7%), *Serratia liquefaciens* (5.7%) *Serratia odorifera* (5.7%), *Citrobacter freundii* (2.85%), *Klebsiella ornithinolytica* (2.85%), *Klebsiella pneumoniae* (2.85%), *Proteus mirabilis* (2.85%), *Providencia rustigiani* (2.85%), *Providencia Stuart II* (2.85%), *Salmonella* (2.85%), *Serratia rubidaea* (2.85%) and *Shigella flexneri* serogroup B (2.85%). Of isolates identified 42.85% of them belonged to the family *Enterobacteriaceae*, poor hygiene indicators in manufacturing processes. Proteinases produced by this microbiota are thermostable, remains intact and active after heat treatment, and the considerable economic losses, and one should trace their sources of contamination as hazards to public health.

**Key Words:** isolating, microorganisms, milk

**T143 Can probiotic bacteria survive in a beverage made from “acid whey” from Greek yogurt?** Alexis Duferene\*, Dasom Park, Douglas Olson, and Kayanush J. Aryana, *School of Animal Sciences, Louisiana State University Agricultural Center, Baton Rouge, LA.*

Over the last 2 years, Greek yogurt sales and consumption have grown very rapidly leading to a \$2 billion per year industry. The by-product of Greek yogurt manufacture is “acid whey,” in which the industry needs to decide its utilization. For every 3 parts of milk, 1 part of Greek yogurt and 2 parts of acid whey are formed. The probiotics industry is rapidly

growing since the health benefits of probiotics are widely known. Typically, probiotic bacteria do not thrive in acidic conditions. In a product such as acid whey, which naturally contains proteins, amino acids and sugars (lactose) that are needed for lactic acid bacterial growth, it will be beneficial to determine if the lactic acid probiotic *Lactobacillus acidophilus* can survive. The objectives were 1) to manufacture a probiotic acid whey beverage, 2) to determine the growth of the probiotic *Lactobacillus acidophilus* in the acid whey beverage and 3) to study any changes in pH, viscosity and titratable acidity over 5 wk of refrigerated storage. Plain yogurt was manufactured, and whey was separated from the plain yogurt to yield Greek yogurt and acid whey. Acid whey was batch pasteurized, cooled, sweetened, flavored with pineapple flavoring, inoculated with *Lactobacillus acidophilus* to 10<sup>7</sup> cfu/mL and stored at 4°C for 5 wk. The *L. acidophilus* counts declined from 3.2 × 10<sup>7</sup> immediately after manufacture to 1.4 × 10<sup>3</sup> at wk 3 and 90 cfu/mL at wk 5. There were no changes in pH, TA and viscosity of the flavored probiotic acid whey over storage for 5 wk which is desirable as it indicates product stability over shelf life. *L. acidophilus* survived in the flavored acid whey, although counts rapidly declined over 5 weeks. This suggests future research on methods to enhance acid tolerance of probiotic *L. acidophilus*.

**Key Words:** acid whey, probiotic

**T144 Evaluation of microbial quality of raw goat and ewe’s milk produced in Sabrata, Libya.** Yahiah Abojnah<sup>1</sup>, Nahed Khatabi<sup>2</sup>, Said Gnan<sup>2</sup>, and Marvin Moncada\*<sup>3</sup>, <sup>1</sup>University of Tripoli, Tripoli Libya, <sup>2</sup>School of Science, Academy of Graduate Studies, Tripoli, Libya, <sup>3</sup>School of Animal Sciences, Louisiana State University, Baton Rouge, LA.

The objective of this project was to evaluate the microbial quality of the raw goat and ewe’s milk in the region of Sabrata, Libya. One hundred random samples of bulk tank raw ewe’s and goat’s milk (50 samples each) were collected from different farms in Sabrata. All samples were subjected to microbiological tests which included total plate counts (TPC), total coliform counts, yeast and mold counts, *Staphylococcus aureus*, psychrotrophic, thermophilic, proteolytic and lipolytic bacteria counts as well as pH measuring. There was a significant ( $P < 0.05$ ) differences between all samples analyzed on total plate counts, thermophilic, coliform and lipolytic bacteria counts. The obtained results showed that the mean TPC log cfu/mL were 6.36 and 4.62 respectively. Psychrotrophic and thermophilic counts were detected in 90 and 88%; 88 and 76% of the samples with mean values in log cfu/mL of 3.67 and 3.18; 2.79 and 3.56 respectively. Results also indicated that *Staphylococcus aureus* and coliforms were present in 100 and 100%; 98 and 98% of the samples with mean values in log cfu/mL 2.82 and 3.28; 3.63 and 3.18 respectively. On the other hand proteolytic and lipolytic bacteria were present in 100 and 100%; 98 and 94% of the samples with mean values in log cfu/mL 2.88 and 3.17; 2.61 and 2.61 respectively. Yeast and mold were detected in 88 and 88% of the samples with mean values in log cfu/mL < 1.17 and < 1.17. The mean values of pH in all samples analyzed were 6.46 and 6.5. In conclusion, the relatively high microbial count reflecting the poor sanitation and hygienic practices in the region evaluated.

**Key Words:** microbial, goat-ewe, milk

**T145 Quantification of spoilage and contaminants bacteria in samples of raw milk.** Magali Pozza\*<sup>1</sup>, Gilberto Simões<sup>2</sup>, Maximiliane Zambom<sup>2</sup>, Marcelo Neumann<sup>2</sup>, and Paulo Pozza<sup>1</sup>, <sup>1</sup>Universidade



The majority of the mesophilic bacteria found in raw milk, has a big involvement in the refrigerated milk degradation and its dairy products producing enzymes with proteolytic and lipolytic action. This research was conducted in the state of west region of Parana- Brazil. The milk samples from 35 properties were collected directly from the cooling tanks and analyzed by flow cytometry for TBC and SCC. The proteolytic and lipolytic microorganisms were plated on caseinate agar and tributyrin agar both incubated at 35°C for 48 h. For all milk samples was also evaluated in the presence of *Staphylococcus* spp. at 3 critical points of production: from milking, hands-of-milkers, milk cooling tank and plated on Baird Parker agar being held coagulase test. It was possible to stratify the milk samples into 4 distinct treatments according to the values of somatic cells count and total bacteria count: Treatment 1 (>600.000 cells/mL), Treatment 2 (>600.000 cfu/mL), Treatment 3 (>600.000 cells/mL and cfu/mL) and Treatment 4 (<600.000 cells/mL and cfu/mL). The treatments showed mean values of proteolytic microorganisms:  $2.7 \times 10^3$  cfu/mL,  $1.8 \times 10^4$  cfu/mL,  $1.9 \times 10^4$  cfu/mL,  $1.4 \times 10^4$  cfu/mL and lipolytic microorganisms  $1.0 \times 10^5$  cfu/mL,  $2.1 \times 10^5$  cfu/mL,  $2.0 \times 10^5$  cfu/mL,  $6.2 \times 10^4$  cfu/mL, with no significant difference by Tukey test ( $P > 0.05$ ). The dairy production systems had unsatisfactory hygienic conditions. Moderate contamination were obtained from *Staphylococcus*  $9.6 \times 10^3$  cfu/mL,  $2.2 \times 10^3$  cfu/mL,  $1.4 \times 10^4$  cfu/mL and  $3.8 \times 10^3$  cfu/mL cfu/mL; for milking, hands-of-milker, cooling tank and milk respectively. The results show the presence of *Staphylococcus* agent in dairy production systems.

**Key Words:** microbiology, milking, cooling tank

**T146 The identification of lactic acid bacteria in the traditional Carpathian ewe's cheese.** Orysya Tsisaryk<sup>\*1</sup>, Iryna Slyvka<sup>1</sup>, and Tomasz Boczer<sup>2</sup>, <sup>1</sup>Lviv National University of Veterinary Medicine and Biotechnology, Ukraine, <sup>2</sup>Rzeszow University, Poland.

Our purpose was to isolate and identify lactic acid bacteria (LAB) from the traditional ewe's cheese from the Carpathian region (Ukraine), which were unknown. The objective of the study was to determine LAB from ewe's cheese by molecular genetic methods. 95 pure cultures of LAB were isolation from 3 samples of cheese. Culture media MRS and M17 were used for following isolation of pure cultures of LAB. Genomic DNA was prepared by using a set of Genomic Mini according to the instructions. Primer 1254 was used for RAPD-PCR. Primers EGE1 and 1492R were used for RFLP-PCR (Sigma-Aldrich). Two endonucleases were used for digestion amplification product: *RsaI* and *HinfI* (Roche). Taxonomic position of LAB strains was established under the complex cultural, morphological and biochemical properties. Seven strains of LAB of genera *Enterococcus*, 9 strains of genera *Lactobacillus*, 6 strains of genera *Lactococcus* and 6 strains of genera *Leuconostoc* were isolated from cheese N1. Twenty-eight strains of genera *Lactococcus* and 8 strains of genera *Enterococcus* were isolated from sample N2. Twenty-six strains of genera *Lactococcus* and 5 strains of genera *Enterococcus* were isolated from sample N3. Sixteen groups of LAB with heterogeneous properties were obtained by the method RAPD-PCR.

Thirteen groups of LAB were obtained using endonuclease *RsaI* and 11 groups of LAB using endonuclease *HinfI*. Thirty-five cultures of LAB for heterogeneous differences were selected to determine their nucleotide sequences of the 16S rRNA gene. Taxonomic position of 20 isolates of LAB of 98-99% homology, 4 isolates of LAB with 96-94% homology, 1 isolates with 93% and 1 isolate with 90% were established by the results of the 16S rRNA gene sequencing. Taxon belonged to 7 species of *Lactobacillus plantarum* ssp., *Leuconostoc mesenteroides* ssp., *Lactococcus lactis* ssp. *lactis*, *Lactococcus garvieae* ssp., *Enterococcus faecium* ssp., *Enterococcus faecalis* ssp., *Enterococcus durans* ssp. The results of the LAB identification demonstrated a complex LAB population of Carpathian ewe's cheeses. Our examinations of LAB populations in raw ewe's milk products from Carpathian cheeses revealed 7 distinct species, some of which could be used in the dairy industry.

**Key Words:** Carpathian cheese, LAB, PCR-RFLP

**T147 Studies of microbiological parameters of cultured butter during storage.** Orysya Tsisaryk\* and Lubov Musiy, Lviv National University of Veterinary Medicine and Biotechnology, Ukraine.

The aim of this work was to study the stability and growth ability of probiotic strains *Lactobacillus acidophilus* (La-5) with *Lac. lactis* ssp. *cremoris*, *Lac. lactis* ssp. *lactis*, *Lac. lactis* ssp. *diacetylactis* and *Leu. mesenteroides* ssp. *cremoris*, as Flora Danica (FD) Chr. Hansen commercial starters. Four groups of cultured butter (CB) were produced: 1 – the cream fermented at 30°C by FD (CB1), 1:1 FD and La-5 (CB2) and La-5 (CB3); 2 – the cream fermented at 37°C by FD (CB4), 1:1 FD and La-5 (CB5) and La-5 (CB6); 3 (Alnarp's Winter method) – 8 ° → 20 ° → 12 ° and added starter FD (CB7), 1:1 FD and La-5 (CB8) and La-5 (CB9); 4 – added starter FD (CB10), 1:1 FD and La-5 (CB11) and La-5 (CB12) to the butter grain. The initial concentration of starter cultures in cream was 6 log cfu/mL. The experiment was replicated 3 times. Cultured butter was packed in polystyrene cups and stored in a refrigerator at the temperature of 4–6°C for 42 d. The cell viability of Flora Danica and *Lactobacillus acidophilus* La-5 was analyzed during storage on the d 1, 7, 14, 21, 28, 35, and 42. Viable cell counts were performed by preparing serial decimal dilution in 0.1% peptone water. The *Flora Danica* strains were subsequently counted by plating (in duplicate) into M17 agar (Merck). The plates were incubated aerobically for 72 h, at 30°C. The La-5 strains were subsequently counted by plating (in duplicate) into MRS agar (Merck). The plates were incubated aerobically for 72 h, at 37°C. The results were recorded as colony forming units (cfu) per g of product. During 14 d of storage at 4–6°C the number of viable cells *Flora Danica* and La-5 has increased. After 14 d all samples showed a drastic decrease of the number of viable cells. The samples CB1 (7.1 log cfu/g) and CB2 (7.3 log cfu/g) possessed the best viable cells *Flora Danica* properties after 35 d of storage. The sample CB2 (7.7 log cfu/g) possessed the best viable cells La-5 properties after 35 d of storage. The best combination with a significant growth of probiotic strains *Lactobacillus acidophilus* La-5 was observed in CB2. We recommend the storage period of cultured butter less than 35 d at 4–6°C for ensure the survival the probiotic strains.

**Key Words:** cultured butter, Flora Danica, La-5

## Extension Education

**T148 Utilization of fixed-time artificial insemination (TAI) to reduce breeding season length and its effects on subsequent calf value: A case study.** Vitor R. G. Mercadante\*, Darren D. Henry, Francine M. Ciriaco, Guilherme H. L. Marquezini, Tera E. Black, Kalyn M. Waters, Pedro L. P. Fontes, Nicolas DiLorenzo, and G. Cliff Lamb, *North Florida Research and Education Center, University of Florida, Marianna, FL.*

The development of TAI protocols has resulted in the opportunity for increased application of AI in commercial cattle operations. However, the long-term production and economic impact of implementing a TAI protocol in beef cattle operations has not been evaluated. Therefore, during an 8-yr period we evaluated the impacts of TAI to reduce the length of the breeding season (BS) and its effects on subsequent calving distribution, calf value, and BS pregnancy rates. The North Florida Research and Education Center consists of a beef herd containing 300 cows of Angus, Brangus, and Braford breed origin. During the 2006 and 2007 BS, the cows were exposed to a 120 d BS by natural service. In 2008, and every subsequent BS to 2013, all females were exposed to TAI using either the 5-d or 7-d CO-Synch+CIDR protocols. Initially, calving season length resulted in cows being inseminated in 3 TAI groups (in the 2008 and 2009 BS), subsequently reduced to 2 TAI groups (in the 2010 and 2011 BS), and eventually to a single TAI group (in the 2012 and 2013 BS). Following the initial TAI for each group, females were detected for estrus and inseminated artificially after an observed estrus until d 23 after TAI. On d 23 after TAI, bulls were introduced and cows were naturally mated for the remainder of the BS. All bulls passed a breeding soundness examination before being introduced to cows. The BS length was reduced from 120 to 70 d between the 2008 and 2013 BS. Calf distribution and subsequent weaning performance were determined. Overall pregnancy rates increased from 81% and 86% in the 2006 and 2007 BS, respectively, to 94% and 93% in 2012 and 2013, respectively. Mean calving date from the first calf born during each calving season was reduced from 80.9 d from the 2007 BS to 38.9 d from the 2013 BS. Utilizing a similar calf value across years of \$0.91/kg, the mean value per calf increased by \$87 per calf resulting from the 2008 BS to \$169 per calf resulting from the 2013 BS. We conclude that exposing beef females to TAI and reducing the BS length for 6 years altered calving distribution, increased breeding season pregnancy rates, and increased calf value.

**Key Words:** fixed-time artificial insemination, economics, calving distribution

**T149 Silage safety issues for large-scale bunker silos and drive-over piles: Avalanches.** Ruth E. Bolsen and Keith K. Bolsen\*, *Kansas State University, Manhattan, KS.*

Few farming operations invite as many different opportunities for injury or fatality as a silage program. One of these is an avalanche or collapsing silage. It only takes a fraction of a second for part of a silage face to silently break off and fall, and the result can be deadly for anyone located beneath it. Two silage avalanche tragedies are documented and 5 common sense ways they can be avoided are presented. In October 2013, Matthew Winkelbauer was buried in a silage avalanche on a feedlot near Norfolk, Nebraska. Winkelbauer, who owned the feedlot, was pronounced dead at the scene. He was standing in front of the feedout face, which was about 4 m tall, and the avalanche pushed the falling silage more than twice that distance from the face. In January

2014, Jason Leadingham, a silage haul-back driver, was working alone in a bunker silo near Roswell, New Mexico, when 10 to 15 tons of corn silage collapsed on him. Leadingham's body was not recovered from the silage until 2 h later, and the cause of his death was mechanical asphyxia. Many bunkers and piles are too large to be safe for the crew filling them and the one feeding the silage out. Common sense tells us that a 6-m-tall silage face is far more dangerous than one that is only 3 m tall. Here are guidelines that can decrease the chance of having a fatality or serious accident caused by a silage avalanche: (1) Never allow people to stand near the feedout face, (2) a rule-of-thumb is never stand closer to the feeding face than 3 times its height, (3) suffocation is a primary concern and a likely cause of death in any silage avalanche, so follow the "buddy rule" and never work alone in a bunker or pile, (4) post warning signs, "Danger! Silage Face Might Collapse", around the perimeter of bunkers and piles, and (5) avoid being complacent and never think that an avalanche cannot happen to you. We cannot stop avalanches from happening, and they are impossible to predict, but we can prevent people from being under them. Every feedlot and dairy should have safety policies and procedures for their silage program, and they should schedule regular meetings with all their employees to discuss safety.

**Key Words:** silage, avalanche, fatality

**T150 Production system characteristics related to technology transfer facility to improve milk quality of small dairy farms.**

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The milk production by small-scale farmers has great importance in Brazilian agribusiness, however this sector is deficient in terms of technology to ensure minimum quality standards. Many aspects are needed to achieve these quality criteria, but among these, an important factor is the transfer of technology through technical assistance programs. The study was conducted in State of São Paulo, Brazil, and had the objective of identify the characteristic of small-scale dairy farms that are related to technology transfer facility to improve milk quality. During 12 mo, 60 farms were monitored for milk composition, somatic cell count and total bacterial count, and received technical assistance. At the beginning of the study, the producers were interviewed with the use of a semi-structured questionnaire. After 12 mo, the farms were divided into 2 groups: greater improvement and lesser improvement of milk quality, and a multiple correspondence analysis was performed to identify the characteristics that differentiate the groups. The results show that properties had average herd of 38 cattle, 180 L/d milk, total area of 30 ha and use of direct family manpower. Multivariate analysis enabled to identify some characteristics that helped in improvement of milk quality. The group with greater improvement used artificial insemination without technical advice with the choice of bulls, were in quality-based payment programs, used formulated diets with no silage. These characteristics were evaluated at the beginning of the study, and altered according to the needs of each property, resulting in improvement of milk quality. The changes suggested by the extensionists in nutrition, mastitis control, and milking hygiene had great importance, however the participation of quality-based payment programs had the greatest impact on improvement of milk quality. These information are useful and could help to develop and guide technical assistance

programs and the transfer of technology to small-scale milk producers. Financial support: FAPESP.

**Key Words:** extension program, milk production, small-scale dairy farming

**T151 Participation in the 2015 Margin Protection Program by Idaho dairy producers.** Richard J. Norell\*<sup>1</sup> and Benjamin M. Eborn<sup>2</sup>, <sup>1</sup>University of Idaho, Idaho Falls, ID, <sup>2</sup>University of Idaho, Driggs, ID.

A mail-in survey was conducted to evaluate participation in the Margin Protection Program (MPP) and to gather producer input for future MPP education. The survey was mailed to every registered dairy in Idaho (n = 515) and 107 surveys were returned (20.7% response rate). Survey data were compared with Proc GLM and Proc Freq in SAS (SAS Inst. Inc., Cary, NC). Dairies were categorized by annual milk sales: small (<1,814 tonnes, 40.2%), medium (1,814 to 5,443 tonnes, 23.3%), large (5,444 to 11,399 tonnes, 19.6%) or very large (>11,399 tonnes, 16.8%). Eighty-two percent of respondents described their MPP knowledge as moderate to very high and their decision to participate as not difficult to somewhat difficult. On-line MPP decision tools were used by 30% of respondents and 28% calculated an average historical margin basis (herd margin - MPP margin) for their dairy. Sixty-eight percent of respondents elected to participate in MPP and 31 out of 73 elected to buy up coverage over basic (\$4.00/45 kg). Participation rate was greater ( $P < 0.01$ ) for dairies producing >11,399 tonnes but the percentage of producers buying up coverage was higher ( $P < 0.01$ ) for those producing <5,443 tonnes. The overall mean margin and coverage rate were  $\$5.03 \pm 0.16$  and  $84 \pm 2\%$ , respectively, and averaged  $\$6.42 \pm 0.16$  and  $76 \pm 4\%$  for those purchasing higher coverage. Mean buy up coverage was similar ( $P > 0.20$ ) between herd size categories. Producers indicated an interest in learning how to calculate and interpret margin basis plus training on how to use and interpret the on-line MPP decision tools. Producers preferred future MPP training with the following methods: producer meetings ( $76 \pm 4\%$ ), newsletters ( $70 \pm 4\%$ ), magazine articles ( $60 \pm 4\%$ ), web based materials ( $48 \pm 4\%$ ), and webinars ( $24 \pm 4\%$ ). Preference for educational method did not differ between herd size categories ( $P > 0.25$ ). We conclude that the MPP is an important management tool for Idaho dairy producers and further MPP training is desired by the industry.

**Key Words:** Margin Protection Program, risk management

**T152 Comparing lying behaviors on commercial Croatian dairy farms.** Nicole L. Eberhart\*<sup>1</sup>, Pero Mijic<sup>2</sup>, Goran Vuckovic<sup>2</sup>, and Peter D. Krawczel<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Tennessee, Knoxville, TN, <sup>2</sup>Faculty of Agriculture, Josip Juraj Strossmayer University of Osijek, Osijek, Croatia.

Lying behavior can be a useful tool for evaluating the suitability of dairy cows' housing environment. The objective of this study was to evaluate the lying behavior of high-producing Holstein dairy cows on commercial Croatian farms with different management systems. Data were collected from 303 dairy cows across 4 farms in eastern Croatia for at least 3 d. There were some general differences in farm management. One farm used bedded pack housing and a parallel parlor (BP), one used older freestalls (>30 yrs. old) and a herringbone parlor (FH), one was a forced-traffic robotic farm with freestalls (FRb), and one used new freestalls (built in 2013) and a rotary parlor (FRt). Behaviors (lying times (min/d), lying bout duration (min/bout), lying bouts (n/d) and laterality of lying) were recorded by dataloggers recording at 1-min intervals. Acceleration data were summarized into lying behaviors for

each individual cow using SAS (v9.3) and the MIXED procedure was used to determine differences in lying behavior across farms. Lying times differed ( $P < 0.001$ ) with values ranging from (farm mean  $\pm$  SE)  $735.1 \pm 17.2$  (BP) to  $623.7 \pm 22.8$  (FRb) min/d. Lying duration values ranged from  $95.7 \pm 5.2$  (FH) to  $60.1 \pm 5.4$  (FRt) min/bout, differing across farms ( $P < 0.01$ ). Lying bouts differed ( $P < 0.001$ ) with values from  $13.4 \pm 0.4$  (FRt) to  $9.1 \pm 0.6$  (FRb) n/d. Right side lying duration differed ( $P < 0.001$ ) with values from  $87.7 \pm 3.9$  (FH) to  $58.1 \pm 4.1$  min/bout (FRt). Right side lying bouts ranged from  $6.7 \pm 0.3$  (FRt)  $4.1 \pm 0.3$  (FH) n/d and differed ( $P < 0.001$ ). Left side lying times differed ( $P = 0.01$ ) ranging from  $381.1 \pm 15.8$  (BP) to  $291.7 \pm 21.1$  (FRb) min/d. Left side lying duration ranging from  $92.3 \pm 5.7$  (FH) to  $63.3 \pm 6.0$  (FRt) min/bout differed ( $P = 0.006$ ). Left side lying bouts differed ( $P < 0.001$ ) ranging from  $6.7 \pm 0.3$  (FRt) to  $4.5 \pm 0.5$  (FRb) n/d. BP had longer daily lying times than any of the farms with freestalls suggesting greater cow comfort. FRt had more lying bouts per day and the shortest lying durations suggesting reduced comfort. These data suggest there may be differences among farm systems to be addressed when developing standards for cow comfort.

**Key Words:** lying behavior, dairy cow, dairy management

**T153 A demonstration on the use of small ruminants for unwanted vegetation management.** Enrique N. Escobar\*<sup>1</sup>, Jorge J. Rodriguez<sup>2</sup>, and Harry Taylor<sup>2</sup>, <sup>1</sup>University of Maryland Extension-1890 Program, University of Maryland Eastern Shore (UMES), Princess Anne, MD, <sup>2</sup>Department of Agriculture, Food and Resource Sciences, University of Maryland Eastern Shore (UMES), Princess Anne, MD.

The practice of using small ruminants to manage unwanted vegetation (noxious weeds, invasive species, weeds, etc.) is considered an environment favorable practice. Sheep and goats have a small environmental footprint and combined with their size and maneuverability allure their use as tools to manage unwanted vegetation. The main objective of this extension demonstration project was to develop a module with multiple components to propose solutions for management of unwanted vegetation using sheep and goats on Delmarva. During the spring and summer of 2014, 2 groups of sheep and goats were selected to be part of a vegetation management demonstration at the University of Maryland Eastern Shore. The animals were confined using cattle panels and steel posts to 2 main targeted areas. Area 1 consisted of 2 drain canal sites with a total area of 2160.24 m<sup>2</sup> and contained mixed understory, predominantly multiflora rose (*Rosa multiflora*). Area 2 consisted of steep banks in 2 lagoons with a combined target area of 1,670.97 m<sup>2</sup>. The vegetation was predominantly horseweed (*Coniza canadensis*). To estimate dry matter available for consumption, vegetation samples were collected from random sites (1 m<sup>2</sup>). The collected samples were weighed, air-dried and sent to a laboratory for analysis. The vegetation's total digestible nutrients for the ditch and lagoon locations were 54.25% and 51.40%, respectively. Percent protein was higher in Area 1 than the lagoon location, 9.25% and 4.98%, respectively. Acid detergent fiber percentage was similar, 45.90% for the canals and 45.98% for the lagoon banks. In Area 1 there were 419.3 kg Dry Matter (DM) available for consumption and 1019.3 kg DM on Area 2. The results obtained showed that in Area 1 it took 29 d to obtain an average disappearance of unwanted vegetation of 83% using 10 does and 10 ewes (average BW 35.6 kg). While in Area 2, it took 40 sheep and goat wethers (average BW 41.3 kg) 28 d remove 95% of unwanted vegetation. The measurement of vegetation disappearance is a key component for



future studies to estimate the number of sheep and goats necessary to manage vegetation in targeted areas.

**Key Words:** unwanted vegetation, small ruminant, environmental impact

**T154 Bedding characteristics are associated with milk quality in Illinois dairy farms: A Dairy Focus Team approach.** Maria I. Rivelli\*<sup>1</sup>, Katie J. Haerr<sup>1</sup>, Sarah Y. Morrison<sup>1</sup>, Saige A. Sulzberger<sup>1</sup>, Cassandra S. Skenandore<sup>1</sup>, Leo L. Timms<sup>2</sup>, and Felipe C. Cardoso<sup>1</sup>, <sup>1</sup>University of Illinois, Urbana, IL, <sup>2</sup>Iowa State University, Ames, IA.

The ultimate goal of this project is to improve Illinois dairy farms through education, accomplished through the science based innovation of the University of Illinois research and extension. The specific aim of this study was to investigate the association between bedding material and milk quality of dairy cows. Twenty dairy farms in Illinois located in the central (C), north (N), and south (S) regions were visited. During the visits, a questionnaire, DHI records along with the individual farm data set (PCDART or DAIRYCOMP305) were collected and a final data set with the combined information was built in excel for analysis. Cow's bedding sample was a composite sample made from 3 different spots in the bed (front, center, and back). Bedding quality was evaluated by quantifying dry matter (DM) percentage, organic matter (OM) percentage, and particle size (PS). Dry matter analysis was performed in all types of bedding; OM and PS analyses were performed only in sand bedded farms (n = 12). Three replications for each sample were performed for PS to obtain particle size ( $\mu\text{m}$ ), standard deviation, and particles per gram (p/g). Statistical analysis was performed using the UNIVARIATE and GLM procedures of SAS (v9.4). Sixteen farms (80%) used sand bedding; 32% of those farms combined sand with a different type of bedding such as straw or compost. Among the other 4 farms (20%), there were 2 farms (10%) with straw bedding, 1 farm (5%) was using sawdust, and only one farm (5%) had their cows on pasture. Differences for DM and OM were found between and within regions. On average the 12 farms that used sand as bedding had a  $92 \pm 8\%$  of DM,  $3 \pm 2.7\%$  of OM, milk yield per cow/d was  $35.17 \pm 6\text{kg}$ , and somatic cell count (SCC) tank was  $204 \pm 91$  cells/mL. However there was a linear correlation ( $P = 0.05$ ,  $R^2 = 0.32$ ,  $\text{SCC} = 16.28\text{OM} + 134.1$ ) between OM and SCC, and no correlation ( $P = 0.3827$ ) between DM and SCC, Southern IL had lower DM ( $88.4 \pm 9$ ) and OM ( $1.9 \pm 0.8$ ) percentage and higher somatic cell count ( $261.2 \pm 179$ ) than N or C. In conclusion, our results confirm the premise that farms with high OM bedding are associated with higher milk tank SCC.

**Key Words:** sand, bed, organic matter

**T155 On-farm field days as a tool to demonstrate and educate about dairy waste management practices.** Mario E. de Haro-Marti\*<sup>1</sup>, Mireille Chahine<sup>2</sup>, Lide Chen<sup>2</sup>, and Howard W. Neibling<sup>3</sup>, <sup>1</sup>University of Idaho, Gooding, ID, <sup>2</sup>University of Idaho, Twin Falls, ID, <sup>3</sup>University of Idaho, Kimberly, ID.

Teaching best management practices (BMP) or introducing new agricultural waste management practices to dairy producers and crop farmers is a challenge. A series of on-farm field days were designed to deliver information and demonstrate several waste management techniques. During these field days, extension personnel presented each technique and offered written information on how to apply them. Presentations by the livestock producers and farmers who are already applying the techniques and hosted each field day at their farms were a powerful tool to spark interest and conversations with attendees. Five field days

were delivered in 2012 and 2013. The objectives were to demonstrate techniques aimed to reduce ammonia, greenhouse gases, and odor emissions, increase nitrogen retention from manure, reduce run-off risks, manage mortalities, and generate usable by-products from livestock and other agricultural wastes. Topics addressed on each field day were (a) dairy manure collection and composting; (b) dairy slurry manure land application using a drag hose and injection system; (c) two field days on grape vine prunings and dairy manure composting; (d) mortality and offal on-farm composting at a sheep and goat dairy farm. A total of 142 individuals attended the field days. In all cases, farm owners and their managers presented and were available to answer attendees' questions, sharing their experience, and opinions regarding the demonstrated practices. Many attendees expressed their interest and willingness to adopt some of the demonstrated practices. On-farm field days are an excellent tool to increase understanding and adoption of BMP and new technologies. Hearing experiences first hand from producers applying the techniques and being able to see them in action are excellent outreach tools. On-farm field days also fit the fast pace, busy schedule of modern producers who can later visit with extension, industry, and other institutions personnel if they need more details, information, and help on how to adopt the techniques they are interested in.

**Key Words:** extension, field day

**T156 Reproductive performance in dairy farms throughout Illinois: A Dairy Focus Team approach.** Maria I. Rivelli\*<sup>1</sup>, Tonja Egan<sup>1</sup>, Diego A. Velasco Acosta<sup>1,2</sup>, Katie J. Haerr<sup>1</sup>, Sarah Y. Morrison<sup>1</sup>, Saige A. Sulzberger<sup>1</sup>, Cassandra S. Skenandore<sup>1</sup>, and Felipe C. Cardoso<sup>1</sup>, <sup>1</sup>University of Illinois, Urbana, IL, <sup>2</sup>Universidade Federal de Pelotas, Pelotas, Brazil.

The ultimate goal of this project is to improve Illinois dairy farms through education. This is accomplished through the science-based innovation of the University of Illinois research and extension, and also gives students hands-on experience evaluating commercial dairy farms. Fertility in dairy cows has been declining in recent years. Therefore, the specific aim of this study was to survey the association between milk yield and reproductive status of dairy cows in selected dairy farms in Illinois. Twenty dairy farms in Illinois located in the central (C), north (N), and south (S) regions were visited. During the visits, a questionnaire, DHI records along with the individual farm data set (PCDART or DAIRYCOMP305) were collected and a final data set with the combined information was built in Excel for analysis. Yearly pregnancy rates (PR), first service conception rate (FSC), and services per conception (SC) were assessed on cows and heifers from 12 farms. Statistical analysis was performed using the UNIVARIATE procedure in SAS (v9.4). Heifer's PR average overall was  $14 \pm 8.4\%$ , and cow's PR was  $16.7 \pm 4.9\%$ . As expected, FSC was higher for heifers ( $56.5 \pm 16.5\%$ ) than cows ( $40.8 \pm 9.0\%$ ). Heifer's SC was  $2.2 \pm 1.4$ , and cow's was  $2.6 \pm 0.6$ , and average milk yield per cow/d was  $35.3 \pm 6.1\text{kg}$ . In Northern Illinois, heifer's PR was higher ( $20.2 \pm 4.3\%$ ) than Central Illinois ( $13.5 \pm 8\%$ ). Furthermore, in Northern Illinois cow's PR was higher ( $19.2 \pm 4.9\%$ ) than Central Illinois ( $17.7 \pm 2.6\%$ ). Moreover, similar differences were found for FSC, and SC. Average milk yield per cow/d was  $38.0 \pm 6.6\text{kg}$  for N;  $32.9 \pm 5.6\text{kg}$  for C; and  $36.2 \pm 6.4\text{kg}$  for S. In conclusion, there were differences for PR, FSC, and SC percentages between and within regions. These results collectively, although limited in sample size, suggest that geographical aspects may play a role in reproductive success in Illinois dairy farms. The results of this study would imply customized recommendations to each farm to improve its reproductive status.

**Key Words:** reproductive success, Illinois, pregnancy rate

**T157 Evaluation of seed corn hybrids to provide nutrients that are highly digestible for South Dakota livestock operations.** David P. Casper\*, Sara Sontag, Laura McMartin, William Weich, and Jonathan Kleinjan, *South Dakota State University, Brookings, SD.*

South Dakota cropping operations are historically well known historically, as is the SD State University (SDSU) Crop Performance Testing (CPT) program, for focusing on grain production. Recent expansion of the SD dairy industry has created interest in selecting the right seed hybrid for either corn silage and/or grain production. A unique SDSU relationship developed to expand the CPT program to evaluate hybrids for forage production and quality. This information will be crucial for the Interstate-29 SD dairy industry to compete globally. In the 2013 growing season, 45 grain hybrids were evaluated for both corn silage and grain. Hybrids were planted in 4 row field test plots at the SDSU Volga research farm. Corn plants from the center rows were hand harvested at a min of 15 cm from the ground. The plants were weighed, chopped (chipper/shredder), inoculated with Silo King (0.55 kg/ton), packed in

duplicate 18.9 L buckets, sealed, weighed, and allowed to ensile for >90 d at which time, buckets were weighed, opened, and samples submitted for measurement of nutrient concentrations and digestibility (Analab Inc.). In the 2014 growing season, 31 silage specific silage hybrids were evaluated. Procedures were the same, except buckets were opened at 60 and 90 d. In the 2013, the grain hybrids were drier at harvest (range: 28.7 – 56.5; mean =  $39.1 \pm 3.21\%$  DM), compared with 2014 hybrids being wetter (range: 21.8 - 39.6; mean =  $29.9 \pm 2.62\%$  DM). For the 2013, the hybrids varied in digestible DM (range: 60.6 – 74.3; mean =  $69.8 \pm 3.21\%$  DM), while 2014 hybrids were (range: 65.4 – 72.5; mean =  $69.6 \pm 2.54\%$ ) were less variable. The 2014 DM silage yield (range: 18.4 - 23.8; mean =  $20.2 \pm 2.51$  t/ha) varied dramatically, while digestible DM yield (range: 12.4 - 16.8; mean =  $14.1 \pm 2.02$  t/ha) varied as well. Selecting the appropriate seed corn hybrid can improve corn silage yield and digestibility, thereby the right hybrid being able to meet a great percentage of the nutrient requirements of the lactating dairy cow.

**Key Words:** hybrid, corn silage, forage

# Food Safety

**T158 Consumer perception regarding beef quality and food safety.** Maribel Ruiz-Leon, Karina Leon-Lucio, Gilberto Aranda-Osorio\*, and Agustin Ruiz-Flores, *Universidad Autonoma Chapingo, Chapingo, Texcoco, Mexico.*

The aim of this study was to evaluate consumers' perception and attitude regarding beef quality and food safety and the characteristics that are taken into account for purchasing, considering socio-economic factors such as income and education level. A survey integrated by 70 questions, grouped into 6 sections, was applied to 347 administrative and academic staff belonging to the University of Chapingo. The sample population was divided into 4 monthly income levels (strata): 1) Low (less than 7,000 pesos), 2) Medium (from 7,000 to 14,000 pesos), 3) High (from 14,000 to 21,000 pesos) and 4) Very high (more than 21,000 pesos). USD exchange ratio: 1USD = 13.50 pesos. Data were processed by discriminatory canonical analysis to identify the main factors that affect beef consumption; after that, association tests between the income level and beef consumption variables were performed using the ANOVA and the statistic Chi-squared of SPSS. The results showed dependence ( $P < 0.05$ ) between income and education level to: preference for purchasing place (butcheries > supermarkets > street markets) and meat type (poultry > beef > pork) according to the conservation method (chilled > not chilled > frozen); perception and preference for TIF (Federal Inspection Type) seal (42%); preference for the amount of fat (60%, low) and beef color (80%, bright red); knowledge about the use of clenbuterol on finishing cattle (90%) and its negative effect on consumer health (62%); willingness for purchasing beef-free of clenbuterol (100%); knowledge about the concept of food safety (35%); perception of the importance of safe beef and its relation to human health (32%). In conclusion, the awareness about beef safety positively grows with the income and education levels of the consumers, encouraging them to search for safe markets, which ensure beef safety and quality.

**Key Words:** beef quality, food safety, consumer characteristic

**T159 Levels of aflatoxin M<sub>1</sub> in dairy products from Londrina supermarkets and its estimated daily intake.** Joice Sifuentes dos Santos\*, Ana Beatriz C. Ribeiro, Vanessa R. França, Shiguely Katto, and Elsa Helena W. Santana, *University North of Paraná, Londrina, Paraná, Brazil.*

Aflatoxins are fungi secondary metabolites that contaminate cereals and other products of vegetable origin. Aflatoxin B<sub>1</sub> (AFB<sub>1</sub>) is the most common and the most toxic aflatoxin. After the ingestion of AFB<sub>1</sub> contaminated feeds, a part is degraded in the rumen, resulting in the formation of aflatoxicol. The remaining fraction is absorbed in the digestive tract by passive diffusion and is hydroxylated in the liver to aflatoxin M<sub>1</sub> (AFM<sub>1</sub>). Circulating AFM<sub>1</sub> can be excreted in the urine or appear in milk. AFM<sub>1</sub> excretion is also observed in human milk. AFM<sub>1</sub> was originally classified as a Group 2B – possibly carcinogenic to humans, in 1993, but subsequent evidence of its cytotoxic, genotoxic and carcinogenic effects led to a new categorization of AFM<sub>1</sub> as Group 1. The occurrence of aflatoxin M<sub>1</sub> (AFM<sub>1</sub>) was evaluated in 42 milk (pasteurized, ultra-high treated (UHT) milk and milk powder) randomly collected in July 2014 from supermarkets in Londrina, Paraná State, Brazil and this rate of occurrence was used to estimate AFM<sub>1</sub> exposure. AFM<sub>1</sub> determination was carried out by ELISA. AFM<sub>1</sub> was detected in 100% samples, with levels ranging from 0.01 to 0.81 µg/kg, and a mean of 0.13 µg/kg. Differences were observed in AFM<sub>1</sub> levels in milk

powder samples (0.61 µg/kg) compared with pasteurized (0.02 µg/kg) and UHT milk (0.04 µg/kg;  $P < 0.05$ ). None of the samples presented AFM<sub>1</sub> above the maximum permitted level by Brazilian Legislation (0.5 µg/kg for fluid milk and 5 µg/kg for milk powder). Determination of the exposure degree is one of the most important parameters to assess risk from chemical compounds. The Estimated Daily Intake (EDI) of AFM<sub>1</sub> from fluid milk and milk powder was obtained using the amount of food consumed and the corresponding mean concentrations of AFM<sub>1</sub> detected in each food group, taking into account the mean body weight of the age groups. The estimated daily intake (EDI) of AFM<sub>1</sub> was evaluated, and the average intake was 0.468 ng/kg body weight (BW) for adolescents, 0.384 ng/kg BW for adults and 0.559 ng/kg BW for the elderly, values that pose a toxicological risk to the population investigated.

**Key Words:** mycotoxin, dairy product, ELISA

**T160 Occurrence of aflatoxin M<sub>1</sub> and somatic cell count in milk from farms in São Paulo, Brazil.** AF Rosa<sup>1</sup>, MS Miranda<sup>1</sup>, JRP Arcaro<sup>1</sup>, R. Braghini<sup>2</sup>, E. Pinatti<sup>3</sup>, and CR Pozzi\*<sup>1</sup>, <sup>1</sup>*Instituto de Zootecnia, Nova Odessa, São Paulo, Brazil,* <sup>2</sup>*Instituto de Ciências Biomédicas, São Paulo, São Paulo, Brazil,* <sup>3</sup>*Instituto de Economia Agrícola, São Paulo, São Paulo, Brazil.*

Milk needs to be of good quality and a low somatic cell count (SCC) and absence of contaminants such as aflatoxin M<sub>1</sub> (AFM<sub>1</sub>) are important factors in the assessment of the health status of herds. The objective of this study was to evaluate the presence of aflatoxin M<sub>1</sub> and SCC in milk and to determine the correlation between aflatoxin M<sub>1</sub> and SCC. Fifteen lactating cows were monitored at intervals of 15 d over a period of 45 d ( $n = 4$ ) on 9 dairy farms in the state of São Paulo ( $n = 540$ ). Milk samples were collected from the glass collecting jars ( $n = 15$ ) and refrigeration tanks ( $n = 38$ ). Aflatoxin M<sub>1</sub> was determined on immunoaffinity columns using separation and quantification by high-performance liquid chromatography (HPLC). Somatic cell counting was performed by flow cytometry in a Somacount 300. Data were analyzed using a one-way ANOVA with Tukey's test ( $P \leq 0.05$ ). Pearson correlation test was used between AFM<sub>1</sub> and CCS. Aflatoxin M<sub>1</sub> was detected in 12% ( $n = 64$ ) of the milk samples collected from the glass bottles. Mean contamination of the samples ranged from  $0.03 \pm 0.09$  to  $0.746 \pm 1.85$  µg/kg, with a maximum level of 9.83 µg/kg. Of the 38 milk samples obtained from the refrigeration tanks, 16% ( $n = 6$ ) were contaminated with aflatoxin M<sub>1</sub>, with levels ranging from 0.44 to 2.65 µg/kg; 83% ( $n = 4.98$ ) of these samples contained levels that exceeded the limit permitted by the Brazilian legislation (0.5 µg/kg). Mean SCC in the glass collecting bottles ranged from 283.78 ( $\pm 1,302.12$ ) to 1,124.58 ( $\pm 1,844.9$ ) x 1,000 cells/mL. Mean SCC in the refrigeration tanks ranged from 452.75 ( $\pm 220.7$ ) to 2,057.75 ( $\pm 1,233$ ) x 1,000 cells/mL. All farms had tanks with SCC above the limit permitted by the current Brazilian legislation ( $500 \times 1,000$  cells/mL). There was no correlation between the detection of aflatoxin M<sub>1</sub> and SCC in milk samples from the glass collecting jars or refrigeration tanks. The presence of aflatoxin M<sub>1</sub> and high SCC indicate deficiency in the sanitation management of the farms and highlight the need for public policies to improve these patterns.

**Key Words:** aflatoxin M<sub>1</sub>, somatic cell count, milk

**T161 Risk assessment of seven toxic elements residues in raw milk in China.** XueYin Qu<sup>1,2</sup>, Nan Zheng<sup>1,2</sup>, JiaQi Wang\*<sup>1,2</sup>, XueWei Zhou<sup>1,2</sup>, and SongLi Li<sup>1,2</sup>, <sup>1</sup>*Ministry of Agriculture-Laboratory of*



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The pollution of toxic elements has been serious since the rapid development of industrialization in China. There were no data about toxic element levels in raw milk in China. The object of this study was to survey the levels of 7 toxic elements residues in raw milk in China and assess the potential health risk of those residues. The 178 raw milk samples were collected from 8 main milk-producing provinces and from 3 types of milk stations in China, and were analyzed for arsenic (As), lead (Pb), cadmium (Cd), chromium (Cr), mercury (Hg), aluminum (Al) and nickel (Ni) using inductively coupled plasma-mass spectrometry (ICP-MS). Al, Pb, Hg, Ni, Cr, As were detected in 47.8%, 29.2%, 28.1%, 23.6%, 12.4% and 9.0% of total milk samples, respectively, and Cd was not detected in any samples. The levels of detected toxic elements were all below China's regulated limits of As  $\leq 0.1$  mg/kg, Pb  $\leq 0.05$  mg/kg, Cr  $\leq 0.3$  mg/kg, Hg  $\leq 0.01$  mg/kg. The residue levels of the samples from the processing plants were lower than that from the large-scale farms and small farm cooperatives by the results of Nemerow pollution index analysis method and this could be related with the different manufacturing practices. The regional difference analysis results indicate that the raw milk samples from heavy industry provinces have relative high residue levels. The need for further attention to the raw milk of heavy industry regions is crucial. In all detected samples, the risks of the concentrations were far below the reference values. The HQ analysis showed that the residues of As, Pb, Hg, Cr, Al and Ni in the raw milk samples were not presenting potential risk to Chinese adult, if the daily intake was 300 mL.

**Key Words:** raw milk, toxic elements, China

#### **T162 Concentration of 22 elements in milk, feed and water of dairy cow, goat, and buffalo from different regions of China.**

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Milk is the most diversified natural food contained more than 20 trace elements that are necessary for human health. However, metals such as Pb, Cr, Hg, As and so on, may contaminate the environment and thus the feed and water, passing into milk and causing health problems. In our study, the content of 22 elements, including Fe, Mn, Cu, Zn, Co, Al, V, Cr, Ni, Ga, As, Se, Rb, Sr, Ag, Cd, Cs, Ba, Hg, Tl, Pb and U, in cow milk (40), goat milk (40) and buffalo milk (20) in China were analyzed. These 22 elements in their corresponding feed and water were also examined using inductively coupled plasma-mass spectrometry (ICP-MS) after microwave-assisted acid digestion. Significant differences ( $P < 0.05$ ) with mean values were analyzed by Tukey's HSD test in SPSS 17.0. The mean spiked recovery of analytical method was 70.96–113.41% in milk, 71.2–114.8% in feed, and 86.50–123.30% in water. In milk, the mean content of Rb and Zn showed high levels ( $>1000\mu\text{g/L}$ ), followed by Al, Fe, Sr and Ba ( $100\text{--}1000\mu\text{g/L}$ ), and then Mn, Cu, Se and Cs ( $10\text{--}100\mu\text{g/L}$ ). The mean values of V, Cr, Co, Ni, Ga, As, Ag, Cd, Hg, Tl, Pb and U were less than  $10\mu\text{g/L}$ . Co, Rb, Sr, Cs and Tl levels showed significant difference ( $P < 0.05$ ) among cow, goat and buffalo milk. The content of Rb, Sr, Pb, Cs, As, Ni, Tl in drinking water were

significant ( $P < 0.05$ ) correlated with those in milk. However, elements in feed were not ( $P > 0.05$ ) related to milk. It suggested that elements in water might contribute the content of elements in milk. The levels of As, Cd, Hg and Pb were under the MRL of China and did not pose threat to consumers.

**Key Words:** element, milk, ICP-MS

**T163 Effect of HTST and UHT processing on the stability of cephalosporin residues in milk.** Meixia Chen<sup>1,2</sup>, Nan Zheng<sup>1,2</sup>, Fang Wen<sup>1,2</sup>, Hui Wang<sup>1,2</sup>, Songli Li<sup>1</sup>, and Jiaqi Wang<sup>\*1,2</sup>, <sup>1</sup>*Ministry of Agriculture Laboratory of Quality & Safety Risk Assessment for Dairy Products (Beijing), Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China,* <sup>2</sup>*Ministry of Agriculture-Milk and Dairy Product Inspection Center, Beijing, China,* <sup>3</sup>*State Key Laboratory of Animal Nutrition, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China.*

Presence of antibiotics in raw milk threatens human health and dairy production. Heat treatments are expected to inactivate various pathogens or reduce the amount of other undesired substances such as antibiotics before milk reach consumers. As a common part of cephalosporins' molecular structure,  $\beta$ -lactam ring makes cephalosporins susceptible to chemical reaction. Hence, cephalosporins are very likely to experience decrease once heated. To date, few information about cephalosporins' stability during conventional dairy process such as high temperature short time pasteurization (HTST) and ultrahigh temperature process (UHT) has been reported. This study was designed to study the effects of HTST and UHT on cephalosporins. So heat treatment were performed on raw milk samples spiked with 5 cephalosporins—cefoperazone (PER), cefquinome (QUI), cephapirin (PIR), ceftiofur (TIO), cefazolin (ZOL)—on the level of maximum residues limits (MRLs). Of these treatments,  $65^\circ\text{C}/15\text{s}$  was used for preheating raw milk, 20M Pa for homogenization,  $75^\circ\text{C}/15\text{s}$  for HTST and  $140^\circ\text{C}/4\text{s}$  for UHT. Concentration of 5 cephalosporins in milk was determined by UPLC-MS/MS simultaneously before and after heat treatment. Recovery test was conducted to validate the analytical methods used in this study for determination of 5 cephalosporins in our lab. SAS 9.2 statistical software package were applied to carry out the statistical analysis. Data of recovery ranges from 85.8% to 99.8% with RSD lower than 10%, which demonstrates good recovery was obtained. Degradation percentages of cephalosporins obtained after HTST is as following: 2.6% for ZOL, 8.6% for PIR, 12.2% for QUI and 12.3% for TIO with the highest 20.5% for PER. Whereas UHT process showed higher reduction level of cephalosporins in all cases. As is shown, cephalosporins' loss is 15.7% for PIR, 36.3% for PER, 43.9% for QUI, 50.0% for TIO with the highest 77.4% for ZOL after UHT. It is noteworthy that degradation results obtained in our work is higher than the previous data predicted by M. Roca (2011) using the prediction model based on the kinetic equation. In addition, the effect of HTST and UHT displays significant difference ( $P < 0.01$ ). In conclusion, cephalosporins studied in this work is resistant to HTST but is unstable in UHT.

**Key Words:** cephalosporins, HTST, UHT

**T164 Toxins in milk of cows fed with transgenic maize.** Geraldo Neto Balieiro<sup>\*1,3</sup>, Keila Maria Roncato Duarte<sup>2</sup>, Roberto Botelho Ferraz Branco<sup>1</sup>, and Acyr Vanderley de Paula Freitas<sup>1</sup>, <sup>1</sup>*São Paulo State Agency Agribusiness Technology, Ribeirão Preto, São Paulo, Brazil,* <sup>2</sup>*São Paulo State Agency Agribusiness Technology, Nova Odessa, São Paulo, Brazil,* <sup>3</sup>*Research Supported by FAPESP, São Paulo, Brazil.*

The goal of this study was to evaluate the presence Cry toxin in milk of cows feeding transgenic maize hybrids. Twenty-four Jersey cows were allotted into 2 groups: control (n = 12), feeding conventional diet, or test, consuming grain and silage from maize with *cry* gene from *Bacillus thuringiensis* (n = 12). A paired *t*-test was used to determine whether there were significant differences between the 2 treatments. Were used 0.5 g of maize leaves transgenic lyophilized, macerated in liquid nitrogen, resuspended in 1 mL of methanol (80%) and diluted with 4 mL of PBS buffer as immunoassay antigen. Two female New Zealand rabbits were used to produce polyclonal antibodies against Cry1A105, Cry2Ab2, Cry1F, Cry1Ab and VP3Aa20 toxins from DKB 390 VT Pro II, AG 8088 Pro II, Biomatrix 2B655 Hx, Syngenta Impacto TL TG and Syngenta 7205 Viptera, respectively, to be used as a screening for maize-derived food products, by plate-trapped antigen (PTA)-ELISA. Immunogen was prepared bioconjugating Cry toxin to carbodiimides. Immunizations were performed each 15 d. At the end of 5 immunizations the rabbits were bled and sera were kept at -80°C. Antibodies were purified in immunoaffinity columns between 50 and 100 kDa. Milk samples, collected 3 time each 14 d during 56 d, were frozen and lyophilized. Antigen (100 mL) were placed into micro wells in triplicates and incubated 1 h at 37°C. Blocking were done using 200 mL of PBS buffer added to 1% BSA for 1 h 37°C. Plates were dried and 100 mL of anti-sera dilution (1:500) were placed, for 1 h at 37°C. Plates were washed (PBS) and anti-rabbit conjugated HRP sera dilution (1:1000) was added. After 1 h at 37°C, plates were washed (PBS-T-G) and PNPP (*p*-nitrophenyl phosphate) revealed, at 450 nm. The presence Cry toxin analysis did not differ between milk of cows fed by transgenic maize hybrids and its nonbiotech counterpart. Results showed that the milk system production using silage and ration with transgenic maize hybrids that contains cry gene is safe and the antibodies produced through leaves as antigen are highly sensitive and specific to Cry toxin and can be used on an enzyme immunosorbent with low cost, high sensibility and specificity.

**Key Words:** GMO, food security, plate-trapped antigen (PTA)-ELISA

**T165 Aflatoxin B<sub>1</sub> and aflatoxin M<sub>1</sub> induced cytotoxicity and DNA damage in differentiated and undifferentiated Caco-2 cells.** Jie Zhang<sup>1,2</sup>, Nan Zheng<sup>1,3</sup>, Fadi Li<sup>2</sup>, Songli Li<sup>1,3</sup>, and Jiaqi Wang\*<sup>1,3</sup>, <sup>1</sup>Ministry of Agriculture Laboratory of Quality & Safety Risk Assessment for Dairy Products (Beijing), Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>2</sup>College of Animal Science and Technology, Gansu Agricultural University, Lanzhou, China, <sup>3</sup>State Key Laboratory of Animal Nutrition, Institute of Animal Sciences, Chinese Academy of Agricultural Sciences, Beijing, China.

Aflatoxin B<sub>1</sub> (AFB<sub>1</sub>) and aflatoxin M<sub>1</sub> (AFM<sub>1</sub>) are natural mycotoxins that frequently found in food and feed. Numerous reports have indicated that these toxins pose serious risks to human and animal health. However, there are few data in the literature regarding the impairment of AFB<sub>1</sub> and AFM<sub>1</sub> on intestine. The present study therefore was undertaken to investigate the cytotoxic effect and DNA damage of these toxins on human colon carcinoma cell line Caco-2, especially the differentiated cells that resemble mature small intestinal enterocytes. Both undifferentiated (UC) and differentiated (DC) cell were treated with AFB<sub>1</sub> and AFM<sub>1</sub> at various concentrations for 24, 48 and 72 h. Cell viability, lactate dehydrogenase (LDH) release and reactive oxygen species (ROS) production were determined, and DNA damage was accessed by comet assay. Statistical analysis of data was carried out using SAS9.2, statistical software package. Data showed that AFB<sub>1</sub> and AFM<sub>1</sub> inhibited

UC and DC cell growth and increased the LDH release in a time- and dose-dependent manner. Besides, AFB<sub>1</sub> treatment resulted in an evident increase in cytotoxicity over AFM<sub>1</sub> at the high dosage ( $P < 0.05$ ). Moreover, DC were more sensitive to toxins compared with UC ( $P < 0.05$ ), particularly after exposure of 72 h at the dose of 1 µg/mL, as indicated from the lower cell viability (48% vs. 67%, AFB<sub>1</sub> treatment) and the higher LDH release (118% vs. 186%, AFM<sub>1</sub> treatment; 142% vs. 194%, AFB<sub>1</sub> treatment). Marked impacts in the genetic damage were observed after treatment with 2 toxins on UC and DC, even higher than those of H<sub>2</sub>O<sub>2</sub> treatment (positive control). Compared with UC, DC also had more DNA damage ( $P < 0.05$ ), which might due to the alteration of cells during differentiation. All these cytotoxic effects might associate with the strong intracellular ROS generation in the presence of toxins. The present study provided the first experimental evidence of the in vitro DNA damage of DC induced by AFB<sub>1</sub> and AFM<sub>1</sub>.

**Key Words:** aflatoxin B<sub>1</sub>, aflatoxin M<sub>1</sub>, DNA damage

**T166 Interaction of aflatoxin M<sub>1</sub>, ochratoxin A, zearalenone, and α-zearalenol combinations on Caco-2 cells.** Yanan Gao<sup>1,2</sup>, Nan Zheng<sup>1,2</sup>, Songli Li<sup>1,2</sup>, Yangdong Zhang<sup>1,2</sup>, and Jiaqi Wang\*<sup>1,2</sup>, <sup>1</sup>Ministry of Agriculture Laboratory of Quality & Safety Risk Assessment for Dairy Products (Beijing), Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>2</sup>State Key Laboratory of Animal Nutrition, Institute of Animal Sciences, Chinese Academy of Agricultural Sciences, Beijing, China.

It is common to find the co-occurrence of aflatoxin B<sub>1</sub> (AFB<sub>1</sub>), ochratoxin A (OTA), zearalenone (ZEA) in feed. These mycotoxins can be metabolized or transferred to raw milk in the forms of Aflatoxin M<sub>1</sub> (AFM<sub>1</sub>), ochratoxin A (OTA), ZEA, and α-zearalenol (α-ZOL), which are stable in processing of dairy products. The aim of the present study is to investigate the cytotoxicity of combined mycotoxins of AFM<sub>1</sub>, OTA, ZEA, and α-ZOL on the human colon adenocarcinoma cell lines (Caco-2) by using the tetrazolium salt (MTT) assay and the isobologram analysis. Statistical analysis of data was carried out using SAS9.2 statistical software package. Our results demonstrated the significant cytotoxic effects ( $P < 0.05$ ) of the 2-toxin, 3-toxin and 4-toxin combination on Caco-2 cells in a time- and concentration-dependent manner. The results from MTT assay demonstrated that AFM<sub>1</sub> had the similar cytotoxicity as OTA (IC<sub>50</sub> = 4.10 µM) upon exposure for 72 h, which was much higher than that of ZEA and α-ZOL. The cytotoxic effect of the all the mycotoxin combinations increased in the following order: OTA+α-ZOL < ZEA+α-ZOL < OTA+ZEA < AFM<sub>1</sub>+ZEA < AFM<sub>1</sub>+α-ZOL < AFM<sub>1</sub>+OTA < OTA+ZEA+α-ZOL < AFM<sub>1</sub>+ZEA+OTA < AFM<sub>1</sub>+ZEA+α-ZOL < AFM<sub>1</sub>+OTA+α-ZOL < AFM<sub>1</sub>+ZEA+OTA+α-ZOL. Isobologram analysis was further indicated that the existence of AFM<sub>1</sub> brought additive effect at IC<sub>50</sub> and even synergism at IC<sub>25</sub>. The synergistic effect of the 4-toxin group is the highest of all mycotoxin combinations investigated (Combination Index = 0.17 ± 0.05 at IC<sub>25</sub>). Other cell lines will be further studied in the future to obtain an appropriate health risk assessment of milk.

**Key Words:** aflatoxin M<sub>1</sub>, cytotoxicity, interaction

**T167 Clean-in-place cleaning validation at lower temperatures with alkaline chlorinated detergent Cool CIP.** Gary Smith\*, John Partridge, and Zey Ustunol, Michigan State University, East Lansing, MI.

Traditionally removal of organic soils in a clean-in-place (CIP) cleaning cycle by alkaline, chlorinated detergents is carried out at temperatures

ranging from 50 to 65°C. The objective of this research was to determine the cleaning efficiency of a newly formulated CIP solution called Cool CIP, a chlorinated alkaline cleaner developed by DeLaval Cleaning Solutions designed to function at low temperatures. Cool CIP is formulated to function at temperatures ranging from 32 to 40°C. The study was conducted over a 1-year period at the Michigan State University Dairy Plant. CIP cleaning cycles of a raw milk tanker truck and receiving lines, raw milk lines, and raw milk storage tanks were chosen to be included in the study. Effectiveness of Cool CIP was evaluated on each cleaning cycle by analysis of solution temperature, chlorine level, pre-wash and post-wash turbidity, Adenosine Tri-Phosphate (ATP) bioluminescence counts, as well as visual confirmation of overall cleanliness. Samples were collected at the beginning and end of the chemical recirculation step of each CIP cycle. ATP swabs were taken inside the vessel of each cycle, before and after each complete cleaning cycle. The results showed that Cool CIP was able to clean effectively at temperatures as low as 30°C as careful visual inspection of all systems passed after each cleaning cycle. The results also showed that all ATP tests were passable post-cycle on tests at 30°C. The raw line tanker truck receiving lines (n = 14) had 1011.6 ± 943.4 ATP units pre-cleaning, and 0 ± 0 ATP units post-CIP cleaning cycle. The raw milk lines (n = 7) had 10611.3 ± 25620.2 ATP units pre-cleaning, and 1.5 ± 4.8 ATP units post-CIP cleaning cycle. The raw milk tanks (n = 12) had 1381.1 ± 1116.2 ATP units pre-cleaning, and 0 ± 0 ATP units post-CIP cleaning cycle. This paper will present Cool CIP as an effective substitute for traditionally accepted high-temperature alkaline CIP cleaning regimens.

**Key Words:** clean-in-place (CIP)

**T168 Metagenomic evidence of the prevalence and distribution patterns of antimicrobial resistant genes in dairy agroecosystems.** Dipti Pitta\*, Sanjay Kumar, Nagaraju Indugu, Zhengxia Dou, John Toth, Bonnie Vecchiarelli, and Bhima Bhukya, *Department of Clinical Studies, School of Veterinary Medicine, New Bolton Center, University of Pennsylvania, Kennett Square, PA.*

Antimicrobial resistance (AR) is a global problem with serious implications for public health. AR genes are prevalent on animal farms but little is known about their origin and distribution patterns in animal farm agroecosystems. In this study, a total of 20 resistomes (collections of AR genes), 5 each from animal feces, manure, near and far soil, on dairy farms were analyzed using shotgun approach. Antimicrobial resistance genes were detected on all 5 farms and only 2% was annotated to AR genes, but varied with sample type ( $P < 0.05$ ). Despite variations in abundance, majority of AR genes (*acrB*, *bcrA*, *macB*, *mdtF*, *pbp1a*, *tetL*, *tetM*, *tetO*, *tetQ*, *tetS*, *tetW*, *vanrA*, *vanrB*) that conferred resistance to multiple antibiotics, including macrolides,  $\beta$ -lactams, amphenicols, and tetracyclines, were common to all samples. Additionally, fecal and manure resistomes contained AR genes specific for quinolones, sulfonamides and aminoglycosides. Across all farms, manure was identified as a “Hot Spot” with the greatest abundance and diversity of AR genes. We compared AR genes from present data set with publically available data sets using hierarchal clustering. Multidrug resistant AR genes were prevalent across different animal species and humans, implicating an exchange of AR genes between unrelated microbial niches that may be potentiated by human activities. This study reports the prevalence of AR genes across different farm sectors and identifies potential pathways for horizontal transmission of AR genes (animal-manure-soil) in dairy agroecosystems.

**Key Words:** antimicrobial resistance, agroecosystems, antimicrobial resistance (AR) genes

**T169 Antimicrobial resistance of *Escherichia coli* isolates from cheese made from unpasteurized milk in Brazil.** Laryssa Freitas Ribeiro\*<sup>1,2</sup>, Mayhara Martins Cordeiro Barbosa<sup>3</sup>, Fernanda de Rezende Pinto<sup>4</sup>, Renato Pariz Maluta<sup>5</sup>, Mônica Costa Oliveira<sup>2</sup>, Viviane de Souza<sup>6</sup>, Maria Izabel Merino de Medeiros<sup>7</sup>, Lucimara Antonio Borges<sup>2</sup>, Priscila Arrigucci Bernardes<sup>1</sup>, Luiz Augusto do Amaral<sup>2</sup>, and John Morris Fairbrother<sup>1</sup>, <sup>1</sup>*Faculté de Médecine Vétérinaire, Université de Montréal, Saint-Hyacinthe, QC, Canada*, <sup>2</sup>*Universidade Estadual Paulista, Jaboticabal, São Paulo, Brazil*, <sup>3</sup>*Instituto Federal de Educação, Ciência e Tecnologia do Ceará, Quixadá, Ceará, Brazil*, <sup>4</sup>*Universidade Federal de Pelotas, Pelotas, Rio Grande do Sul, Brazil*, <sup>5</sup>*Universidade de Campinas, Campinas, São Paulo, Brazil*, <sup>6</sup>*Embrapa Caprinos e Ovinos, Sobral, Ceará, Brazil*, <sup>7</sup>*Instituto Tecnológico de Alimentos, Campinas, São Paulo, Brazil*.

The production of cheeses from unpasteurized milk is a public health problem, due to the use of raw milk and the associated poor hygienic conditions. Contamination may occur from several sources, involving several different pathogenic microorganisms, including *Escherichia coli*. The use of antimicrobials in animals has led to emergence of resistant microorganisms, contributing to the ineffectiveness of these products. The purpose of the current study was to investigate the presence of antimicrobial resistance in *E. coli* isolates in raw milk cheese in Brazil and to identify the potential risk to public health. A total of 83 cheeses from 3 different cities, Uberaba, Minas Gerais (30), Ribeirão Preto, São Paulo (22) and Aracaju, Sergipe (31) were cultured. From each cheese, 5 colonies were examined and a total of 169 *E. coli* isolates, 51, 25, and 93 from Uberaba, Ribeirão Preto, and Aracaju respectively were obtained. Ninety-five randomly selected isolates were tested for susceptibility to the 15 antimicrobials: amoxicillin + clavulanic acid (20 + 10  $\mu$ g), ceftiofur (30  $\mu$ g), ceftriaxone (30  $\mu$ g), ciprofloxacin (5  $\mu$ g), amikacin (30 $\mu$ g), ampicillin (10  $\mu$ g), ceftiofur (30  $\mu$ g), gentamicin (10  $\mu$ g), kanamycin (30  $\mu$ g), nalidixic acid (30  $\mu$ g), streptomycin (10  $\mu$ g), tetracycline (30  $\mu$ g), chloramphenicol (30  $\mu$ g), sulfisoxazole (0.25 mg) and trimethoprim + sulfamethoxazole (1.25 + 23.75  $\mu$ g) used for testing generic *E. coli* in the Canadian Integrated Program for Antimicrobial Resistance Surveillance (CIPARS), by the disk-diffusion (Kirby-Bauer) method. The greatest number of isolates with a resistance pattern (one antimicrobial class or more) was observed in Uberaba, most being resistant to 1–2 antimicrobial classes. Multidrug resistance (resistance to 3 or more classes of antimicrobial agents) was much more frequently observed in isolates from Uberaba and Ribeirão Preto (12 to 14%) than in those from Aracaju (2%). The highest prevalence of resistance in all regions was to tetracycline. Strikingly, prevalence of resistance to ampicillin and amoxicillin/clavulanic acid was much higher in Uberaba than in Ribeirão Preto and Aracaju. For example, 58.85% of Uberaba samples were amoxicillin/clavulanic acid resistant while Ribeirão Preto and Aracaju were 18.18% and 3.57% respectively. For ampicillin, the prevalence in samples were 61.54%, 27.27% and 14.29% to Uberaba, Ribeirão Preto and Aracaju respectively. All 95 isolates were susceptible to ceftiofur, amikacin, gentamicin, and chloramphenicol. Resistance to ceftiofur was found in Ribeirão Preto, suggesting the therapeutic use this antimicrobial. For statistics analysis, the sample was considered resistant to the antimicrobial when at least one of the isolates demonstrated resistance and it was used exact chi-squared. So, with the test,  $P$ -value was 0.001571 and it showed that the city is not resistant independent, thus they do not have the same resistance. And it showed that Uberaba was different from Ribeirão Preto and Aracaju and Aracaju and Ribeirão did not differ ( $P < 0.05$ ). The variation of resistance to antimicrobials between regions observed in the present study indicates



that the cheese made with unpasteurized milk in Brazil may contain *E. coli* that can be a risk for public health.

**Key Words:** public health, microbiology, antimicrobial resistance

**T170 Ecoepidemiology of *Staphylococcus* spp. in small-scale goat milk dairy plants in northeastern Brazil.** Candice de Leon<sup>1</sup>, Celso Oliveira\*<sup>1</sup>, Iara Siqueira<sup>2</sup>, Maria G. Carvalho<sup>2</sup>, and Denis Spricigo<sup>3</sup>, <sup>1</sup>Federal University of Paraíba (UFPB), Brazil, Areia, PB, Brazil, <sup>2</sup>Federal University of Campina Grande, Patos, PB, Brazil, <sup>3</sup>LANAGRO, Porto Alegre, RS, Brazil.

The microbiological quality of goat milk needs to be monitored to ensure the milk and milk products are safe for consumers. Our previous studies have demonstrated high contamination of goat milk by coagulase-positive and coagulase-negative *Staphylococcus* spp. that frequently harbor enterotoxin-producing genes. *Staphylococci* have been associated with food-poisoning outbreaks in humans and because *staphylococci* are commonly isolated from goat milk and are have an important effect on public health, these organisms are used as indicators of contamination in goat dairy plants. Therefore, this study aimed to evaluate the epidemiology of *Staphylococcus* spp. at different points of small-scale processing plants of goat milk in northeastern Brazil, the leading goat milk-producing region in the country. A longitudinal study was performed in 2 goat milk processing plants; samples of raw and pasteurized milk and swabs of the inner surface of equipment, walls, and handlers' hands were collected. Samples were collected in the morning before milk processing to detect residual contamination. *Staphylococcus* spp. isolates (n = 36) were subjected to genotyping by REP-PCR using the primer RW3A. Indistinguishable genotypic profiles were observed among *Staphylococcus* spp. collected from milk samples, equipment, and handlers' hands, indicating the presence of residual contamination during milk processing and the need to improve cleaning and disinfection procedures. The *Staphylococcus* spp. isolates from handlers' hand swabs had high genotypic relatedness with isolates from equipment and pasteurized milk, indicating that workers can play a role in the cross-contamination by *Staphylococcus* spp. in milk processing plants.

**Key Words:** food safety, genotyping, pathogen

**T171 Effects of purple prairie clover (*Dalea purpurea* Vent.) on feed intake, nutrient digestibility and fecal shedding of *Escherichia coli* O157: H7 in lambs.** Qianqian Huang<sup>1,2</sup>, Long Jin<sup>1</sup>, Zhong Xu<sup>1</sup>, Ruth Barbieri<sup>1</sup>, Surya Acharya<sup>1</sup>, Tianming Hu<sup>2</sup>, Tim McAllister<sup>1</sup>, Kim Stanford<sup>3</sup>, and Yuxi Wang\*<sup>1</sup>, <sup>1</sup>Lethbridge Research Centre, Agriculture and Agri-Food Canada, Lethbridge, AB, Canada, <sup>2</sup>College of Animal Science and Technology, Northwest A&F University, China, <sup>3</sup>Alberta Agriculture and Rural Development, Lethbridge Agriculture Centre, Lethbridge, AB, Canada.

This study was conducted to assess the effects of purple prairie clover (PPC, *Dalea purpurea* Vent.) on feed intake, nutrient digestibility, fecal shedding of *Escherichia coli* O157: H7 and blood metabolites of lambs. Three groups of lambs (6 per group) were individually fed green chop alfalfa (Alf), a mixture of Alf and PPC in a ratio of 40:60 (DM basis; Mix) and Mix supplemented with polyethylene glycol (Mix-P) for 45 d. Polyethylene glycol (PEG) was included to neutralize the biological activity of condensed tannins (CT) in PPC. Total-tract digestibility was determined using AIA as an indigestible marker. Blood samples were collected to measure serum metabolites and activity of antioxidant enzymes. All lambs were orally inoculated with a 5-strain mixture of *E. coli* O157:H7 after determination of diet digestibility and fecal samples

were enumerated for *E. coli* O157:H7 after 1, 2, 3, 4, 7, 14, 21 and 28 d of inoculation. Feed intake was similar among lambs fed the 3 diets. The OM digestibility of Mix ( $P < 0.05$ ) and CP digestibility of both Mix and Mix-P ( $P < 0.01$ ) were lower than Alf. However, the 3 diets had similar NDF digestibility. After inoculation, lambs fed Mix shed less *E. coli* O157:H7 than lambs fed Alf between d 1 and d 7 ( $P < 0.05$  to 0.006), and lambs fed Mix-P between d 1 and d 3 ( $P < 0.05$  to 0.012). Lambs fed Alf shed more *E. coli* O157:H7 than lambs fed Mix ( $P < 0.001$ ) or Mix-P ( $P < 0.01$ ) over the 28-d post-challenge period. Lambs fed Mix had lower levels ( $P < 0.05$ ) of serum creatinine and urea nitrogen than Alf, but both Mix and Mix-P had higher ( $P < 0.05$ ) levels of serum P. Mix fed lambs had greater ( $P < 0.05$ ) serum total antioxidant capacity but lower ( $P < 0.05$ ) serum superoxide dismutase activity than lambs fed Alf. Incorporation of PPC in lamb diets at 60% of DM reduced fecal shedding in lambs challenged with *E. coli* O157:H7 as compared to Alf. The fact that shedding of *E. coli* O157:H7 was also reduced with Mix-P suggests that the anti- *E. coli* O157:H7 response of PPC is not solely due to CT.

**Key Words:** purple prairie clover, *E. coli* O157: H7, condensed tannins

**T172 Reduction of biological hazards in animal feed mills via a decontamination protocol.** Anne R. Huss\*, Roger A. Cochrane, Aiswariya Deliephan, Charles R. Stark, and Cassandra K. Jones, Kansas State University, Manhattan, KS.

Animal feed and ingredients have been shown to be potential vectors of pathogenic bacteria in to the human food chain. Introduction of contaminated materials can lead to facility contamination, which can be easily spread to non-contaminated materials, including finished feeds. Upon detection of contamination, facility decontamination is necessary to prevent potential cross-contamination to feedstuffs. The purpose of this experiment was to evaluate a standardized protocol to decontaminate an animal feed manufacturing facility using *Enterococcus faecium* (ATCC 31282) as an indicator. A pelleted swine diet inoculated with *E. faecium* was manufactured at the Kansas State University Feed Safety Research Center. Environmental samples including swabs, RODAC plates and air samples were collected and consisted of: 1) baseline before inoculation, 2) after inoculated feed production, 3) after production physical clean using pressurized air, 4) after chemical clean with a quaternary ammonium-glutaraldehyde blend, 5) after chemical clean with sodium hypochlorite, 6) after facility heat-up to 60°C for 24 h, 7) 48 h, and 8) 72 h. Air samples collected on the exterior of the facility confirmed pathogen containment as *E. faecium* concentrations were equal to or lower than baseline levels at each sample location. Decontamination step and its associated interactions were the only factors that affected *E. faecium* incidence ( $P < 0.0001$  vs.  $P > 0.22$ ). After production of the inoculated diet, 85.7% of environmental samples were positive for *E. faecium*. Interestingly, physical cleaning had no effect on contamination ( $P = 0.32$ ). Chemical cleaning with a quaternary ammonium-glutaraldehyde blend and sodium hypochlorite each reduced ( $P < 0.0001$ ) *E. faecium* contamination to 28.6% and 2.4% of tested surfaces. All samples were negative after 48 h of heating. In summary, both wet chemical cleaning and facility heating resulted in substantial *E. faecium* decontamination, but not physical cleaning. In addition, this experiment confirmed both successful containment and decontamination of biological pathogens in the tested pilot-scale feed mill.

**Key Words:** feed safety, pathogen, decontamination

### T173 Evaluation of select bacterial populations in poultry excreta and potential treatments for their disinfection. C.

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Because poultry litter has been used as feed supplement for cattle, there is interest in learning more about any unwanted bacteria present in poultry excreta. The objective of this study was to test for the presence of select bacterial populations in UACH poultry excreta for total aerobes, *Salmonella* spp., *E. coli* and coliforms, and *Campylobacter* spp. Additionally, sodium chlorate and nitroethane were evaluated as bactericidal agents to reduce bacterial concentrations. Approximately 45 kg of poultry excreta were mixed with 16 L of water and distributed equally to 9 buckets. Treatments were sodium chlorate (10 mM concentration), nitroethane (12 mM concentration) and a control. Excreta temperature and pH were measured. Samples were collected at 0, 6 and 24 h and processed for bacteriological enumeration. Enumeration of total aerobes, *Salmonella* spp., *E. coli* and coliforms, and *Campylobacter* spp. were performed with 3M Petrifilm Total Aerobe Count Plates, brilliant green agar supplemented with novobiocin, 3M Petrifilm E.coli/Coliform Count Plates and Campy-Cefex Agar, respectively. Data were analyzed by means of an ANOVA design, with time and treatments as main effects and their interactions. There was found a main effect of time ( $P < 0.0001$ ) but not treatment or an interaction ( $P > 0.05$ ) on litter temperature and pH. A main effect of time ( $P < 0.0001$ ) was observed but treatment and the time x treatment interaction ( $P > 0.05$ ) for litter temperature and pH was not significant. A main effect of time ( $P < 0.01$ ) but not treatment or the interaction ( $P > 0.10$ ) on total aerobes or on *Salmonella* spp. was observed. A treatment x time interaction ( $P = 0.05$ ) on *E. coli* and total coliforms was observed. *Campylobacter* spp. was present but no effects of treatment or time ( $P < 0.15$ ) on incidence of culture-positive samples was observed. *Campylobacter* concentrations were not sufficient across all samples to perform quantitative analysis. In conclusion, modest effects of chlorate and nitroethane treatment were observed on *E. coli* and total coliforms but not on total aerobes or *Salmonella*.

**Key Words:** poultry excreta, *Salmonella* and *E. coli*, chlorate nitroethane

### T174 *Trypanorhyncha* cestodes in *Brachyplatystoma rousseauxii*, *Cynoscion leiarchus*, *Cichla* spp., and *Colossoma macropomum*, captured in coast of Amazon/Brazil. Raquel L. Salgado\*<sup>1</sup> and Josemir S. Gonçalves<sup>2</sup>, <sup>1</sup>Universidade Federal Fluminense, Niterói, Rio de Janeiro, Brazil, <sup>2</sup>Universidade Federal Rural do Semi Árido, Mossoró, Rio Grande do Norte, Brazil.

Fish consumption in the Amazon region is one of the world's largest; however, many species of fish commonly show high parasitism rates in muscles, making them unfit for consumption. In this context, the aim of this study was to evaluate the diversity of the parasitic fauna of 4 different species of fish captured in the northern coast of Amazon/Brazil. During the months from January to December 2009, 60 specimens of each species (*Brachyplatystoma rousseauxii*, *Cynoscion leiarchus*, *Cichla* spp., and *Colossoma macropomum*) were weighed, measured, and identified according their anatomic traits. They were filleted and with the aid of a candling table, their muscles were evaluated for the presence of parasites. The *Trypanorhyncha* larvae were released from their blastocysts, placed in Petri dishes containing distilled water for 24 h in the refrigerator. Subsequently, they were fixed in A.F.A. (95 parts of 70% alcohol, 5 parts of formalin, 2 parts of acetic acid) for 24

h. The cestodes identification was based in their morphologic traits. The amount of parasites in each specimen and overall were used to calculate the prevalence (P), infection intensity (I), mean infection intensity (MI) and mean abundance (MA). Of the 60 specimens of *B. rousseauxii* analyzed, 2 were infested by cestodes parasites, one by *Poecilancistrum caryophyllum*, P (1.60%), I (1), MI (1.00), MA (0.02) and one by *Pterobothrium heteracanthum* P (1.60%), I (1), MI (1.00), MA (0.02). Three specimens of *Cynoscion leiarchus* exhibited multiple parasitism by *Anisakis* spp. and *Poecilancistrum caryophyllum* P (7.50%), I (1–2), MI (1.50), MA (0.10). Six specimens of *Cichla* spp. were infested by *Clinostomum complanatum*, P (10.00%), I (2–3), MI (2.50), MA (0.92) and 2 are also infested by *Contracecum* spp. P (17.00%), I (1–3), MI (1.70), MA (0.27). No specimen of *Colossoma macropomum* was infested by any parasites. Despite they do not have zoonotic potential, the high prevalence of *Trypanorhyncha* represents a serious risk to human health, through the possibility of causing allergic reactions in humans.

**Key Words:** fish, human health, parasite

### T175 Helminth with zoonotic potential in fish of Amazon/Brazil. Raquel L. Salgado\*<sup>1</sup> and Josemir S. Gonçalves<sup>2</sup>, <sup>1</sup>Universidade Federal Fluminense, Niterói, Rio de Janeiro, Brazil, <sup>2</sup>Universidade Federal Rural do Semi Árido, Mossoró, Rio Grande do Norte, Brazil.

Among the parasites with zoonotic potential, nematodes from the family Anisakidae are most involved in cases of infection due to the consumption of fish. The aim of this study was to evaluate the presence of parasites with zoonotic potential in *Brachyplatystoma rousseauxii*, *Cynoscion leiarchus*, *Cichla* spp., and *Colossoma macropomum* captured in the northern coast of Amazon/Brazil. During the months from January to December 2009, 60 specimens of each specie were weighed, measured, and identified according their anatomic traits. They were filleted and with the aid of a candling table, their muscles were evaluated for the presence of parasites. Nematode larvae were placed in Petri dishes containing distilled water and subsequently fixed in A.F.A. (95 parts of 70% alcohol, 5 parts of formalin, 2 parts of acetic acid) for 24 h. The nematodes identification was based in their morphologic traits. The amount of parasites in each specimen and overall were used to calculate the prevalence (P), infection intensity (I), mean infection intensity (MI) and mean abundance (MA). Nematodes of the genus *Anisakis* spp. were observed parasitizing 15 specimens of *Cynoscion leiarchus* P (37.50%), I (3–42), MI (18.06), MA (6.77). Three specimens exhibited multiple parasitism by *Anisakis* spp. and *Poecilancistrum caryophyllum* P (7.50%), I (1–2), MI (1.30), MA (0.10). Twelve specimens of *Cichla* spp. were infested, of which, 10 by nematodes of the genus *Contracecum* spp. P (17.00%), I (2–12), MI (2.50), MA (0.92) and 2 by *Contracecum* spp. and by *Clinostomum complanatum* P (17.00%), I (1–3), MI (1.70), MA (0.27). Nine specimens of *B. rousseauxii* were infested by *Anisakis* spp. P (15.00%), I (4–32), MI (12.00), MA (1.80). No parasite species were found in the 60 specimens of *C. macropomum* analyzed. The parasites found may be responsible for allergic and gastric reactions and may even lead to death in humans. The high prevalence of parasites with zoonotic potential combined with the small number of studies on the parasite fauna of the Amazon fish, represents a serious risk to human health, and reinforce the need for a larger number of studies in this area.

**Key Words:** Anisakidae, human health, parasite

# Forages and Pastures: Silages and forages in beef and sheep production systems

**T176 Tiller population stability index from the spring in palisadegrass previously used in deferred grazing.** Simone Pedro da Silva<sup>\*1</sup>, Manoel Eduardo Rozalino Santos<sup>2</sup>, Bruno Humberto Rezende Carvalho<sup>2</sup>, Ludiêmilem Keith Parreira da Costa<sup>2</sup>, Denis Douglas Pessoa<sup>2</sup>, Heron Alves de Oliveira<sup>2</sup>, Róger Carvalho Cardoso<sup>2</sup>, Miriã Gonçalves Simplício<sup>2</sup>, and Dilermando Miranda da Fonseca<sup>3</sup>, <sup>1</sup>Instituto Federal Goiano, Hidrolândia, Goiás, Brazil, <sup>2</sup>Universidade Federal de Uberlândia, Uberlândia, Minas Gerais, Brazil, <sup>3</sup>Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.

We evaluated the effect of pasture condition previously deferred and on late winter on regrowth of *Brachiaria brizantha* 'Marandu' (palisadegrass). From October 2013 to January 2014, the palisadegrass pastures were evaluated at the Federal University of Uberlândia, located in the State of Minas Gerais, Brazil. The experimental site was located at 776 m altitude, 18°30' S and 47°50' W. Annual precipitation was around 1.584 mm. Medium temperature was 22.3°C. The tiller dynamics was evaluated in deferred pastures with 4 conditions to late winter. The pastures conditions were low (15.1 cm), medium (23.2 cm), high (31.4 cm) and high/cutting (31.3 cm and cutting to 8.0 cm). The experiment was carried out using a randomized completely design with 3 repetitions and subdivided plots. In each experimental unit (paddocks), all tillers inside the 3 frames with 0.0625 m<sup>2</sup> were counted and marked with colored plastic. Every 30 d, all tillers were recounted and new tillers were marked with a different wire color. The collected data were used to calculate the tiller population stability index (SI). During spring and early summer, all pastures were kept with 30 cm in continuous stocking, with sheep. There were interactions ( $P < 0.05$ ) of months with pasture conditions for SI. In October, the low (2.3) and high/cutting (2.2) pastures showed ( $P < 0.05$ ) higher SI, followed by medium (1.7) and high (1.4) pasture, respectively. For other months, there was no effect of pasture conditions on SI. The SI was higher in low and high/cutting pastures, due to high tillering these pastures. The SI was less than or equal to one unit only in November (0.9) and December (1.0). The high development of shoots in tillers in October may have generated higher shading at plant base in November and December, causing decrease in SI. The lower and deferred palisadegrass pastures in late winter have a higher SI from early spring, compared with higher pastures. We conclude that cutting of high palisadegrass pasture in late winter improves the tiller population stability index from the spring.

**Key Words:** grazing, seasons of the year, sward height

**T177 Performance of crossbred cows on Tanzania grass pastures subjected to two pasture-management strategies.** Alberto Magno Fernandes<sup>\*1</sup>, Ricardo Augusto Mendonça Vieira<sup>1</sup>, Tadeu Silva de Oliveira<sup>1</sup>, and Fermino Deresz<sup>2</sup>, <sup>1</sup>Universidade Estadual do Norte Fluminense, Campos dos Goytacazes, Rio de Janeiro, Brazil, <sup>2</sup>EMBRAPA-Gado de Leite, Juiz de Fora, Minas Gerais, Brazil.

The objective of this study was to evaluate nutrient intake and yield and components of milk from Holstein × Zebu cows on Tanzania grass using a fixed resting period (FR) or according to 95% light interception (LI, by accuparLP-80) for managing Tanzania-grass pastures. As the LI of the herbage mass reached 95%, the animals were allowed to graze each paddock for 3 d; and (2) 30 d defoliation interval with a 3-d grazing period (FR). Experimental unit consisted of 11 paddocks grazed by 5

Holstein-Gir cows allowing for 2 replicates per defoliation treatment in the first year with the same areas assigned to the same treatment the following year grazed by 4 Holstein-Gir cows per experimental unit. Intake and digestibility were estimated using the external marker chromic oxide (Cr<sub>2</sub>O<sub>3</sub>). Each grazing animal received 5 g of Cr<sub>2</sub>O<sub>3</sub> administered orally in paper cartridges twice daily at 12-h intervals for 12 d. The cows were milked twice daily, at 06.00 and 15.00 h. Milk samples were analyzed every 14 d to determine protein, fat, lactose, energy, and total solids (TS) contents. The management strategies imposed on the Tanzania-grass pasture did not affect ( $P > 0.05$ ) the intake or digestibility of nutrients. Milk yield and milk components were not affected ( $P > 0.05$ ) by the management strategies, but treatment LI95 yielded 2,067 ± 288 kg milk/ha while FR was less at 1,861 ± 288 kg milk/ha ( $P < 0.05$ ). In conclusion, adopting forage management utilizing a 95% light interception defoliation interval improves milk production per land unit and efficiency of land resources.

**Key Words:** grass, intake, milk

**T178 Performance of cows and calves grazing limpograss pastures as affected by creep-feeding protein supplements.** J. M. D. Sanchez<sup>\*1</sup>, J. M. B. Vendramini<sup>1</sup>, P. A. Lancaster<sup>1</sup>, J. K. Yarborough<sup>1</sup>, F. A. Kuwahara<sup>2</sup>, and V. C. Gomes<sup>3</sup>, <sup>1</sup>Range Cattle Research and Education Center, University of Florida, Ona, FL, <sup>2</sup>Faculdade de Medicina Veterinária e Zootecnia, Universidade Estadual Paulista, Botucatu, Sao Paulo, Brazil, <sup>3</sup>Instituto de Zootecnia, Nova Odessa, Sao Paulo, Brazil.

Creep-feeding soybean meal (SBM) to suckling calves grazing limpograss (*Hemarthria altissima*) pastures has been an effective management practice to improve calf average daily gain (ADG); however, it is not clear if the positive effect is result of a greater CP and energy intake or specifically the additional rumen degradable protein (RDP) supplied by the SBM. The objective of this study was to investigate the effects of creep-feeding different amounts of RDP to cow-calf pairs grazing limpograss pastures. The experiment was conducted at the Range Cattle Research and Education Center, Ona, FL from May to August 2014. Treatments were 400 g/d of SBM (65% RDP) or SoyPlus (40% RDP) distributed in a randomized complete block design with 4 replicates. Brangus crossbred cow-calf pairs (n = 16; calf initial BW = 160 ± 5 kg) were randomly assigned to 8 limpograss pastures (1.0 ha/pasture; 2 pairs/pasture). Average daily gain of cows and calves was recorded every 28 d. Body condition scores (BCS) of cows was measured at the beginning and ending of the experiment. Herbage mass (HM) and CP concentration were measured every 14 d. Data were analyzed using the Proc Mixed of SAS with treatment and month as fixed effects and block as random effect. There were no differences in HM ( $P = 0.48$ , 6800 ± 1200 kg DM/ha) and CP ( $P = 0.20$ , 12.2 ± 3.2% CP) between treatments. The CP concentration decreased ( $P < 0.01$ ) from May to July and was similar in July and August (from 15 to 10% CP). There was no difference in ADG of cows ( $P = 0.71$ , 0.12 ± 0.14 kg/d) and calves ( $P = 0.24$ , 0.74 ± 0.03 kg/d) between treatments. Calf ADG decreased from May to August ( $P < 0.01$ , from 1.03 to 0.30 kg/d). There were no differences in the final cow BCS ( $P = 0.90$ , 5.34 ± 0.30) and change in BCS ( $P = 0.73$ , 0.51 ± 0.31) during the experimental period. In conclusion, the increase in performance of calves creep-fed with SBM observed in



previous studies may be due to increase in CP and energy intake rather than the additional RDP provided by the SBM.

**Key Words:** soybean meal, SoyPlus, limpoggrass

**T179 Decreased reproductive rates in sheep fed a high selenium diet.** Thomas Z. Davis\*<sup>1</sup>, Bryan L. Stegelmeier<sup>1</sup>, Kip E. Panter<sup>1</sup>, and Jeffery O. Hall<sup>2</sup>, <sup>1</sup>United States Department of Agriculture-Agricultural Research Service, Poisonous Plant Research Laboratory, Logan, UT, <sup>2</sup>Utah State University Veterinary Diagnostic Laboratory, Logan, UT.

High Se-containing forages grow on seleniferous soils in many parts of the United States and throughout the world. Selenium is an essential trace element that is required for many physiological processes but can also be either acutely or chronically toxic to livestock. Anecdotal reports of decreased reproductive rates in livestock grazing seleniferous forages have been reported and it has been speculated that reproductive failure is one of the initial changes of Se poisoning. The objective of this study was to determine the effect of high Se forages on reproductive rates in sheep. The study was conducted in 2 phases. Phase 1, the ewes (n = 3–5) were fed a Se-containing alfalfa pellet (made by adding western aster containing approximately 2000 ppm Se) that contained <0.3 (control), 10, 20, 30, 45, or 60 ppm Se for 12 weeks. Feeding of the pellets began 6 weeks before exposing the ewes to rams. Each ewe was exposed to 2 rams twice each day for 2 complete reproductive cycles. At 35 to 45 d post-exposure the pregnancy status of the ewes was determined by ultrasound imaging. Each group fed Se-containing pellets had fewer pregnant ewes than the control group (pregnant ewes/ewes exposed): control (3/3), 10 ppm Se (3/5), 30 ppm Se (0/5), 45 ppm Se (1/5), 60 ppm Se (1/5). Throughout the study hair samples, blood samples, liver biopsies, and muscle biopsies were collected to monitor the Se status of the ewes. In phase 2, ewes were divided into 3 groups (n = 10) and were fed Se-containing pellets containing <0.3 (control), 10, and 30 ppm Se in the same manner and for the same period. However liver and muscle biopsies were not performed. After the first cycle, significantly ( $P < 0.05$ ) more ewes were pregnant in the control group (10/10) than in the 10 ppm Se (6/10) and 30 ppm Se (6/10) groups. After a second cycle 9/10 and 6/10 were pregnant in the 10 and 30 ppm Se groups, respectively. None of the ewes in either phase of the study demonstrated any clinical signs of chronic Se poisoning during the study. In summary, high selenium forages containing 10 ppm Se or greater decreased reproduction rates in ewes when fed for 6 weeks before exposure to rams.

**Key Words:** selenium, sheep, reproduction

**T180 Total nitrogen in Marandu-grass pastures under different grazing intensities in southeast Brazil.** Mariana Vieira Azenha\*<sup>1</sup>, Liziane Figueiredo Brito<sup>2</sup>, Andre Alves Oliveira<sup>2</sup>, Elisamara Raposo<sup>2</sup>, Estella Rosseto Januskiewicz<sup>2</sup>, Ricardo Andrade Reis<sup>2</sup>, and Ana Claudia Ruggieri<sup>2</sup>, <sup>1</sup>EMBRAPA Pecuaria Sudeste, São Carlos, Sao Paulo, Brazil, <sup>2</sup>UNESP / FCAV, Jaboticabal, Sao Paulo, Brazil.

Pasture productivity relies heavily on exchange of several different compounds between roots and shoots. The study aimed to evaluate nitrogen content on Marandu-grass pasture managed under 3 grazing management heights. The heights of 15, 25 and 35 cm were managed by beef steers grazing under continuous stocking and occurred from January to November, 2011. Data were analyzed by repeated measurements with grazing intensity, period and interactions. Roots and shoots samples were collected monthly. The collected samples were washed, dried and

ground for further analysis. Total nitrogen concentration in the shoots was higher during the summer (14.77 g kg<sup>-1</sup> DM) and lower during the drier seasons (7.47 g kg<sup>-1</sup> DM) ( $P < 0.05$ ). This decrease in concentration was more marked for the 35 cm pasture height ( $P < 0.05$ ). Total nitrogen concentration in the roots followed the opposite pattern, increasing in drier seasons (7.29 g kg<sup>-1</sup> DM), and had no consistent correlation with pasture height. Root nitrogen content was also negatively correlated with precipitation and temperature, while shoot nitrogen was positively correlated ( $P < 0.05$ ). During the summer, fertilizer application along with temperature, precipitation and insolation favor plant growth. With a higher quantity of new leaves in this season there was an increase in shoot nitrogen content. During the winter, limited growth favors nitrogen storage in roots. Our results indicate that roots provide a storage site for nitrogen during drier, less favorable seasons. These reserves were directed to shoot regrowth during the summer. We found a negative linear association between pasture heights and shoot nitrogen content in autumn and winter ( $P < 0.05$ ). In more intensely managed pastures, greater tissue renewal occurs as tillers develop in an environment with reduced competition for light. Indeed, intensely managed palisadegrass pastures have higher rates of tiller appearance and death, and higher defoliation frequency of individual tillers. In addition, taller pastures carry a higher proportion of stem and dead material in relation to shorter pastures, contributing to lower nitrogen concentrations. Root nitrogen did not show a consistent response pattern in relation to pasture height.

**Key Words:** pasture management, roots, shoots

**T181 Effects of conservation method on condensed tannin content, ruminal and intestinal digestion characteristics of purple prairie clover.** Qianqian Huang<sup>1,2</sup>, Tianming Hu<sup>1</sup>, Long Jin<sup>2</sup>, Surya Acharya<sup>2</sup>, Tim McAllister<sup>2</sup>, Alan Iwaasa<sup>3</sup>, Mike Schellenberg<sup>3</sup>, and Yuxi Wang\*<sup>2</sup>, <sup>1</sup>College of Animal Science and Technology, Northwest A&F University, Yangling, Shaanxi, China, <sup>2</sup>Agriculture and Agri-Food Canada, Lethbridge Research Centre, Lethbridge, AB, Canada, <sup>3</sup>Agriculture and Agri-Food Canada, Semiarid Prairie Agricultural Research Centre, Swift Current, SK, Canada.

Purple prairie clover (PPC, *Dalea purpurea* Vent.) is a forage that contains high levels of phenolics including condensed tannins (CT) and exhibits anti-*Escherichia coli* O157: H7 activity. The objectives of this study were to evaluate the effects of forage conservation method on the CT content, ruminal and intestinal digestion characteristics of PPC. Whole PPC was harvested at the full flower stage and either immediately freeze-dried to represent green chop (FG), ensiled in mini-silos for 45 d (Silage), or sun cured in the field to DM >85%, baled and stored under shed for 45 d (Hay). In situ DM, NDF and CP degradabilities were determined by incubating the conserved forage in 3 rumen cannulated heifers for 0, 1, 2, 4, 8, 12, 24, 48 and 72 h, and analyzing data for Degradability =  $a + b(1 - e^{-c(t-L)})$  and effective degradability (ED) =  $a + [bc/(c+k)]e^{-(c+k)L}$ . Intestinal DM and CP digestion were estimated by incubating 12-h ruminal in situ residues in a modified 3-step in vitro procedure. Silage exhibited decreased ( $P < 0.001$ ) extractable CT, but increased ( $P < 0.001$ ) protein and fiber-bound CT, whereas Hay only increased ( $P < 0.01$ ) protein-bound CT. The ED of FG was greater ( $P < 0.01$ ) than Silage or Hay, with ED of Hay being lower ( $P < 0.01$ ) than Silage. However, the ED of NDF and CP as influenced by conservation method ranked as Silage > FG > Hay ( $P < 0.01$ ). Conservation method did not affect intestinal DM digestion of 12-h in situ residues, but the CP intestinal digestion of Silage was higher ( $P < 0.01$ ) than FG and Hay. Ensiling increased proportions of protein- and fiber-bound CT, but also increased ED of both CP and NDF as well as in vitro intestinal digestibility of CP. These findings indicate that the biological activity

of CT and the formation of CT-protein complexes were altered by the ensiling process. Silage is a better conservation method than sun cured hay for PPC in terms of ruminal and intestinal digestion of DM, NDF, and CP while the biological activity of CT is higher in Hay than in Silage.

**Key Words:** purple prairie clover, forage conservation, condensed tannins

**T182 Nutrient content of *Atriplex canescens* (Pursh Nutt) as a function of soil electrical conductivity.** Aracely Zúñiga\*<sup>1</sup>, Luz M. Tejada<sup>1</sup>, Juan C. Martínez-Alfaro<sup>2</sup>, and Miguel Mellado<sup>1</sup>, <sup>1</sup>Autonomous Agrarian University Antonio Narro, Saltillo, México, <sup>2</sup>Regional Academic Center UAAAN, Chiapas, México.

Arid and semi-arid areas of northern Mexico are characterized for having little availability of fodder. Another limitation in some desert areas is the presence of soils with high amounts of salts, which limits the development of crops. Four-wing saltbush (*Atriplex canescens*) is a shrub of arid and semi-arid areas adapted to soils with high salinity, it remains green throughout the year, and has a high nutritional content, which represents a good forage for herbivores in these areas. For this study, *A. canescens* samples (leaves) from 20 plots (3 × 3 m) were collected during November in northern Mexico (24°N) and the nutrient content was analyzed. Soil samples near stems of these shrubs were also collected, at depths of 0–30 cm. Crude protein of this fodder shrub was 9.87%. Soil testing showed slight levels of salinity, with an electrical conductivity of 2 to 4 mMhos/cm. The association between nutrient content of *Atriplex canescens* and electrical conductivity of soil were analyzed by nonlinear regression with the CurveExpert program. It was found that the crude protein of *A. canescens* was positively affected by ascending levels of electrical conductivity ( $y = 10.9 \times 0.092^{1/x}$ ;  $r^2 = 0.17$ ). Calcium levels increased with electrical conductivity ( $y = 4.85 + 0.34x - 0.32/x^2$ ;  $r^2 = 0.31$ ). The association between electrical conductivity and phosphorous was weak ( $y = 7.29 - 0.94x - 0.5/x^2$ ;  $r^2 = 22$ ). The variable with the highest association with electrical conductivity was potassium ( $r^2 = 0.44$ ). It was concluded that neither dry matter digestibility nor structural carbohydrates are associated with electrical conductivity of soil and that the increase in electrical conductivity of soil mainly affect the mineral content of this fodder shrub.

**Key Words:** *Atriplex canescens*, electrical conductivity, crude protein

**T183 Nutritional composition of a *Musa* sp. fodder bank located in the central part of Costa Rica.** Pablo Chacon Hernandez\*, Carlos Boschini Figueroa, and Ricardo Russo Andrade, *Universidad de Costa Rica, San Pedro, San José, Costa Rica.*

Bananas are widely grown in tropical areas and a large proportion of the production is related to the pseudo-stem which is not normally utilized. We hypothesized that this material could be used depending on its nutritional quality. Using a completely randomized block design with 3 replicates, the nutritional composition was evaluated on a *Musa* sp. fodder bank located at the University of Costa Rica's Alfredo Volio Mata Experimental Station during the rainy season of 2012. Five adult plants were harvested on each block at 20–25 cm above ground and divided into 5 sections for sampling (base, center and tip of the pseudo-stem and blades and petioles of the leaves). For the whole plant mean values of 9.22% of DM, 8.57% of CP, 58.02% of NDF, 37.59% of ADF, 9.25% of LIG, 28.34% of CEL, 20.42% of HEMI, 2.35% of EE, 14.17% of ASH, 6.91% of NDICP and 6.91% of NFC were obtained; also, statistical differences among plant parts were found ( $P < 0.05$ ). Despite the low dry

matter content, banana plant portions may be utilized as ingredient in ruminant diets, especially in areas where the crop is commonly grown.

**Table 1 (Abstr. T183).** Nutritional composition of the *Musa* sp. plant

Bromatological composition (%)	Pseudo-stem			Leaves	
	Base	Center	Tip	Petiole	Lamina
DM	3.93 <sup>e</sup>	4.97 <sup>d</sup>	6.13 <sup>c</sup>	10.38 <sup>b</sup>	20.68 <sup>a</sup>
CP	5.61 <sup>b</sup>	5.87 <sup>b</sup>	5.99 <sup>b</sup>	5.56 <sup>b</sup>	19.82 <sup>a</sup>
NDF	50.63 <sup>d</sup>	55.71 <sup>c</sup>	54.29 <sup>c</sup>	61.34 <sup>b</sup>	68.11 <sup>a</sup>
ADF	33.82 <sup>c</sup>	37.50 <sup>b</sup>	36.15 <sup>bc</sup>	44.24 <sup>a</sup>	36.24 <sup>bc</sup>
LIG	6.84 <sup>c</sup>	7.57 <sup>c</sup>	7.15 <sup>c</sup>	9.90 <sup>b</sup>	14.79 <sup>a</sup>
CEL	26.98 <sup>c</sup>	29.94 <sup>b</sup>	29.01 <sup>bc</sup>	34.34 <sup>a</sup>	21.46 <sup>d</sup>
HEMI	16.81 <sup>c</sup>	18.21 <sup>b</sup>	18.14 <sup>b</sup>	17.10 <sup>bc</sup>	31.86 <sup>a</sup>
EE	1.62 <sup>b</sup>	1.96 <sup>b</sup>	2.07 <sup>b</sup>	1.85 <sup>b</sup>	4.27 <sup>a</sup>
ASH	19.30 <sup>a</sup>	16.73 <sup>b</sup>	13.99 <sup>c</sup>	11.74 <sup>d</sup>	9.12 <sup>e</sup>
NDICP	3.83 <sup>b</sup>	3.72 <sup>b</sup>	3.83 <sup>b</sup>	3.89 <sup>b</sup>	19.26 <sup>a</sup>
NFC	26.68 <sup>a</sup>	23.45 <sup>a</sup>	27.49 <sup>a</sup>	23.41 <sup>a</sup>	17.96 <sup>b</sup>

<sup>a-e</sup>Statistical differences in the same row ( $P < 0.05$ ).

**Key Words:** fodder bank, *Musa*, bromatology

**T184 Litter lignin and phosphorus content in different grazing intensities of Marandu-grass pasture in Southeast Brazil.** Mariana Vieira Azenha<sup>1</sup>, Elisamara Raposo<sup>2</sup>, Andre Alves Oliveira<sup>2</sup>, Liziane Figueiredo Brito<sup>2</sup>, Estella Rosseto Januszkiewicz<sup>2</sup>, Ricardo Andrade Reis<sup>2</sup>, and Ana Claudia Ruggieri\*<sup>2</sup>, <sup>1</sup>EMBRAPA Pecuaria Sudeste, São Carlos, Sao Paulo, Brazil, <sup>2</sup>UNESP/FCAV, Jaboticabal, Sao Paulo, Brazil.

The objective of this study was to evaluate lignin and phosphorus content in a Marandu-grass pasture managed under 3 heights of pasture. The pastures were managed by beef steers grazing under continuous stocking. Evaluations of litter decomposition occurred in 2011, at Jaboticabal, SP, Brazil. The litter bag technique was used to evaluate the decomposition of senescent plant material. Incubation times were 4, 8, 16, 32, 64, 128 and 256 d. Nylon bags were used. At the end of each incubation period, the bags were analyzed. The lignin and phosphorus content was analyzed and compared using Proc Mixed from SAS. Polynomial models were used to fit the curves for each pasture stubble height according to incubation times. A significant linear increase in litter lignin with increasing pasture stubble height was observed in the litter incubated ( $P < 0.01$ ), with means of 16.32, 18.50, and 19.84%, respectively, for pasture stubble heights 15, 25, and 35 cm. A relative increase in litter lignin (67%) with decomposition time was observed. Since increasing pasture stubble height is associated with stem elongation, hence higher cell-wall and lignin content than leaves, this could explain the positive association observed between litter lignin and pasture stubble height. The magnitude of differences in lignin content according to pasture stubble height is, however, expected to decrease over the course of the incubation period as the relative content of other components becomes progressively lower while lignin remains undecomposed, since it is chemically very insoluble. Since more soluble components, such as cellulose and hemicelluloses, are primarily used by microorganisms as energy source, a relative increase in litter lignin with incubation time is expected. Phosphorus in the incubated litter was 0.02% on average, and did not differ among pasture stubble heights ( $P > 0.05$ ). The relative increase in litter phosphorus content with incubation time ( $P < 0.01$ ) was expected. As phosphorus immobilization occurs when C/P ratio is

higher than 200, this process prevailed throughout the incubation period, when C/P ratios ranged from 1600 to 825.

**Key Words:** litter decomposition, height, pasture management.

**T185 Adding medium quality hay to the diet of stocker calves grazing annual ryegrass did not improve beef production.**

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Strategic use of hay may allow increasing the stocking rate of small areas of high nutritive value pastures, benefiting producers by keeping a greater number of calves for stocker programs. The objective was to compare performance and blood metabolite concentrations of beef steers continuously stocked on annual ryegrass (*Lolium multiflorum* Lam. 'Marshall') or grazing and strategically receiving bermudagrass (*Cynodon dactylon* 'Jiggs') hay (CP = 9.6%; TDN = 50%; NDF = 67%). For 3 consecutive years, 54 steers (BW = 229 ± 2.9 kg) were used. Treatments (3 replicates; n = 6 steers) evaluated were: 1) steers (CON) continuously stocked on ryegrass (1.33 ha; 4.5 steers/ha; 1040 kg BW/ha); 2) steers grazed 50% of the area assigned to CON (0.67 ha; 9 steers/ha; 2081 kg BW/ha) on alternate days (HAY24) and fed hay in a barn; 3) steers grazed 50% of the area assigned to CON (0.67 ha; 9 steers/ha; 2079 kg BW/ha) for 96 h, then were hay-fed in the barn for 72 h (HAY72). Steers were individually fed hay using Calan gates. Blood samples were obtained on 4 consecutive days (d1 after 24 h on ryegrass). Data were analyzed using Proc mixed. Pastures grazed by HAY24 and HAY72 steers had greater ( $P < 0.05$ ) forage mass than CON from d 28 until the end of the grazing season. Concentrations of ADF (27 and 23%) and NDF (41 and 37%) were greater and IVDMD (74 and 78%) was lower ( $P < 0.05$ ) in ryegrass grazed by HAY72 vs. CON, respectively. Production (kg/ha) and ADG (kg) were greater ( $P < 0.05$ ) for CON (498 and 1.2, respectively), intermediate for HAY24 (418 and 0.8, respectively), and lowest for HAY72 (352 and 0.6). Plasma urea nitrogen (PUN) concentration was greatest ( $P < 0.001$ ) for CON (28 mg/dL), intermediate for HAY24 (24 mg/dL), and lowest for HAY72 (21 mg/dL). NEFA were similar for CON and HAY24 (0.32 mEq/L) but lower ( $P < 0.001$ ) than HAY72 (0.47 mEq/L). Concentration of PUN decreased (26 to 23mg/dL) and NEFA increased (0.31 to 0.45 mEq/L) linearly ( $P = 0.003$ ;  $P < 0.001$ , respectively) from d1 to d4 of sampling. Steers in HAY72 consumed 13% less hay ( $P < 0.05$ ) than those on HAY24. In the present experiment, hay feeding strategies negatively affected animal performance.

**Key Words:** hay, ryegrass, stocking rate

**T186 Cow-calf performance and gain per ha from bermudagrasses overseeded with 'Apache' arrowleaf clover or 'TAM-90' annual ryegrass and stocked at three intensities.**

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From 2002 through 2013, both Coastal (COS) and common (COM) Bermudagrass (*Cynodon dactylon*) (BER) pastures were overseeded in Oct each year with either 'Apache' arrowleaf clover (*Trifolium vesiculosum*) (CLV) without N-fertilization or 'TAM-90' annual ryegrass (*Lolium multiflorum*) (RYG) plus N for the 7-mo stocking period. F-1 (Hereford × Brahman) cows with fall-born Simmental-sired (SIM) calves were stocked on all pastures from late Feb. to early June (SPG), and with winter-born SIM calves from mid-June to late Sept. (SUM). All pastures were stocked continuously at 3 intensities (STK) to achieve different forage mass. Proc Mixed was used to determine treatment dif-

ferences. Calf (steers and heifers) ADG was greater ( $P < 0.01$ ) during growth of CLV and RYG in SPG compared with SUM. During SPG, calf ADG was greater ( $P < 0.01$ ) from RYG (1.24 kg d<sup>-1</sup>) compared with CLV (1.05 kg d<sup>-1</sup>); and ADG was different ( $P < 0.01$ ) at all 3 STK for low (LO) 1.40 kg d<sup>-1</sup>, medium (ME) 1.20 kg d<sup>-1</sup>, or high (HI) 0.83 kg d<sup>-1</sup>; and no difference for BER. During SUM, calf ADG ( $P < 0.01$ ) was different at all STK at LO (1.05 kg d<sup>-1</sup>), ME (0.81 kg d<sup>-1</sup>), and HI (0.58 kg d<sup>-1</sup>). Cow ADG during SPG was greater ( $P < 0.01$ ) for RYG (0.40 kg d<sup>-1</sup>) vs CLV (0.20 kg d<sup>-1</sup>), and all 3 STK were different ( $P < 0.01$ ) for LO (0.74 kg d<sup>-1</sup>), ME (0.30 kg d<sup>-1</sup>), and HI (-0.15 kg d<sup>-1</sup>). In SUM, cow ADG was affected by STK ( $P < 0.01$ ), BER ( $P < 0.04$ ), and STK × BER ( $P < 0.04$ ). Cows lost weight on COM at all 3 STK (-0.19, -0.53, and -0.87 kg d<sup>-1</sup>); whereas cow ADG on COS was different at each STK of LO, ME, and HI, respectively, at 0.15, -0.47, and -0.89 kg d<sup>-1</sup>. Calf gain ha<sup>-1</sup> for combined SPG and SUM was greater ( $P < 0.01$ ) from COS (720 kg ha<sup>-1</sup>) than COM (638 kg ha<sup>-1</sup>); and from either HI or ME STK (758, 724 kg ha<sup>-1</sup>) compared with LO STK (557 kg ha<sup>-1</sup>). The RYG + N resulted in total calf gains of 804 kg ha<sup>-1</sup> compared ( $P < 0.01$ ) to CLV + no N at 556 kg ha<sup>-1</sup>. This 11 yr study showed calf gains per ha of more than 550 kg ha<sup>-1</sup> on BER with CLV and no-N. Apache CLV proved to be a reliable, productive legume for overseeding BER in the southeastern US without use of N.

**Key Words:** cow-calf, clover, ryegrass

**T187 Performance of steers raised on pastures of elephant and mombasa grasses.**

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The performance of steers raised on elephant and mombasa grass pastures was evaluated from October to December 2006. A rotational grazing system with a regulated forage offer was adopted. The experimental area consisted of 2 block containing 18 paddocks of 0.25 ha divided into 9 paddocks with elephant grass and 9 with mombasa grass. Forage species, area block, the interaction among these effects, paddocks within the interaction, and the experimental error were the effects of the treatments. This model was used to analyze pasture attributes. For analyses of intake, digestibility and gain, 24 steers were randomly assigned to the respective interactions among forage vs. block vs. grazing system, totaling 3 animals per triple interaction. The grazing systems consisted of ad libitum (AL) and restricted (PR) grazing. Elephant-grass showed the highest ( $P < 0.05$ ) total dry matter biomass (TB) and green leaf dry matter biomass (GLDM). There were no differences ( $P > 0.05$ ) among forage plants for production of biomass of green leaf dry matter; however, leaf proportion was the greatest ( $P < 0.05$ ) in mombasa grass. Extrusas showed similar ( $P > 0.05$ ) bromatological composition. Only crude protein intake, which was the highest for mombasa pasture, differed ( $P < 0.05$ ) among forage plants. Ad libitum feeding enabled a higher ( $P < 0.05$ ) intake of all nutrients from both pastures. Digestibility of nutrients was higher for elephant-grass but it did not differ ( $P > 0.05$ ) among feeding levels:restricted grazing allowed for greater plant crude protein concentration while ad libitum grazing resulted in greater gross energy consumption. In conclusion, despite qualitative and quantitative differences among forage plants, differences of weight gain per animal and per area were not found. For animal with ad libitum feeding, average body mass gain was 850 g/day and gain per area was 246 kg/ha during the experimental period. Funded by CAPES, CNPq, FAPEMIG, and FAPERJ.

**Key Words:** digestibility, growth, intake



**T188 Diurnal time to move animals to a new paddock: Forage nonstructural carbohydrates concentration, grazing patterns, and performance of beef cattle.** F. C. Leite de Oliveira<sup>\*1</sup>, C. O. Rocha<sup>1</sup>, J. M. D. Sanchez<sup>2</sup>, R. S. Ferigato<sup>3</sup>, L. E. T. Pereira<sup>1</sup>, C. G. Lima<sup>1</sup>, P. H. C. Luz<sup>1</sup>, and V. R. Herling<sup>1</sup>, <sup>1</sup>University of São Paulo, Faculty of Animal Science and Food Engineering, Department of Animal Science, Pirassununga, Sao Paulo, Brazil, <sup>2</sup>Range Cattle Research and Education Center, University of Florida, Ona, FL, <sup>3</sup>Anhanguera Faculties, Department of Veterinary, Leme, Sao Paulo, Brazil.

Although the physiology of substances produced in photosynthesis is known, few studies with tropical forage species have been done to provide information about animal grazing patterns and performance. The objective of this study was to investigate the effects of diurnal time of allocating animals to a new paddock on ingestive behavior, forage nonstructural carbohydrates concentration (NSC: sugar and starch) and performance of beef heifers grazing *Brachiaria brizantha* 'Marandu'. The experiment was conducted in southeastern Brazil (21° 59' N; 47° 26' W). Treatments were daily time of allocating animals to new paddocks at 0600 h (AM) and 1500 h (PM) in a randomized complete block design with 3 replicates. Heifers (n = 60, initial BW = 230 kg) were managed by 78 d in rotational stocking, with 1 d grazing period and 25 d resting period during the summer of 2012/2013. All variables were measured every 26 d. Treatments had similar forage allowances ( $P > 0.05$ ,  $3.7 \pm 0.1$  kg DM/kg BW). Times spent grazing, ruminating and other activities were acoustically recorded during a 24 h period. Animals were weighed after a 14–16 h fasting. NSC was determined in forage samples hand-plucked at 0600 and 1500 h, in both treatments. Data were analyzed with Proc Mixed of SAS, with treatment and month as fixed effects and block as a random effect. Total daily grazing time was greater for AM ( $P < 0.05$ , 490 vs. 450  $\pm$  5.4 min), however, PM concentrated diurnal grazing time in the afternoon ( $P < 0.05$ , ~66%), whereas AM did not concentrate grazing activities in the morning when compared with the afternoon ( $P > 0.05$ , ~50% in each period). There was no difference of NSC between treatments for a given time of the day ( $P > 0.05$ , 36 and 79  $\pm$  2.7 g/kg DM at 0600 and 1500 h, respectively), although PM had 50 g NSC/kg DM more than AM at 1500 h ( $P < 0.05$ ). Thus, compensating between grazing time and forage NSC throughout the day resulted in similar ADG (0.75  $\pm$  0.04 kg/d). The schedule for allocating beef cattle in new paddocks may be done according to the convenience of each farm, as it does not affect performance.

**Key Words:** daytime, ingestive behavior, sugar

**T189 Sequential and mixed grazing of stockpiled toxic tall fescue by fall-calving Angus cows and Katahdin ewes.** Taylor N. Drane\*, Richard E. Daugherty, James D. Caldwell, Bruce C. Shanks, Chris L. Boeckmann, Cindy A. Deornellis, Amy L. Bax, Abbey J. Kempker, and Jason D. Walker, Lincoln University, Jefferson City, MO.

There are many potential benefits with multi-species grazing including possible performance improvements for one or both species involved. For this reason, there is renewed interest in evaluating mixed versus sequential grazing schemes; however, little research has been done in hair sheep and cattle especially grazing stockpiled toxic tall fescue [*Lolium arundinaceum* (Schreb.) Darbysh; E+]. Therefore, the objective of this study was to determine performance and reproductive measurements by fall-calving Angus cows and Katahdin ewes grazing stockpiled E+ under a mixed or sequential grazing scheme. Over 3 consecutive yr, fall-calving Angus cows (n = 60; 499.8  $\pm$  56.4 kg initial BW; 5.7  $\pm$  0.7 initial BCS) and Katahdin ewes (n = 121; 26.1  $\pm$  6.9 kg initial BW; 2.9

$\pm$  0.4 initial BCS) were stratified by BW and age within species and allocated randomly to 1 of 4 groups representing 2 treatments: 1) sequential grazing (SG; 6 replications) or 2) mixed grazing (MG; 6 replications). Each year, a total of 8.16 ha (0.68-ha per group) were grazed 40 d for yr 1 and 2 and 42 d for yr 3. In SG, cows always followed ewes and all groups were rotated based on available forage. Initial weight, initial BCS, end weight, end BCS, total gain, and ADG did not differ ( $P \geq 0.15$ ) between treatments. Cow and ewe pregnancy rates did not differ ( $P \geq 0.96$ ) between treatments. Calf birth date and calf birth weight did not differ ( $P \geq 0.33$ ) between treatments. Calf start weight was heavier ( $P = 0.03$ ) for MG vs. SG, but calf end weight, total gain, and ADG were similar ( $P \geq 0.13$ ) between treatments. Lamb birth date, birth weight, and number of lambs born did not differ ( $P \geq 0.12$ ) between treatments. Therefore, sequential grazing compared with mixed grazing may not improve performance and reproductive measurements in fall-calving Angus cows and Katahdin ewes. Thus, the extra labor associated with a multi-species, sequential grazing scheme may not be justified.

**Key Words:** fescue, mixed grazing, sequential grazing

**T190 Performance and reproductive measurements by spring-born Katahdin ewes grazing stockpiled toxic tall fescue, non-toxic tall fescue, or Persist orchardgrass.** H. L. Bartimus<sup>\*1,2</sup>, B. C. Shanks<sup>1</sup>, J. D. Caldwell<sup>1</sup>, A. L. Bax<sup>1</sup>, L. S. Wilbers<sup>1</sup>, A. J. Kempker<sup>1</sup>, J. D. Walker<sup>1</sup>, C. A. Clifford-Rathert<sup>1</sup>, and A. K. Busalacki<sup>1</sup>, <sup>1</sup>Lincoln University, Jefferson City, MO, <sup>2</sup>University of Arkansas, Fayetteville, AR.

Stockpiled forages are an alternative to expensive feedstuffs and may extend the grazing season through the winter months for small ruminant producers. Our objective was to evaluate the effects of stockpiled endophyte-infected tall fescue [*Lolium arundinaceum* (Schreb.) Darbysh; E+], novel endophyte-infected tall fescue (NE+), and Persist orchardgrass (*Dactylis glomerata* L.; OG) on performance and reproductive measurements by spring-born Katahdin ewes. Over 2 consecutive years, Katahdin ewes (n = 116; 27.7  $\pm$  0.57 kg initial BW, 2.8  $\pm$  0.05 initial BCS) were stratified by weight and BCS and allocated randomly to 1 of 3 treatments consisting of: 1) E+ (5 replications), 2) NE+ (5 replications), 3) OG (5 replications). Each year, excess forage was grazed in early August, 54 kg N/ha was applied in mid-September, and beginning in early November grazing was initiated. Data were analyzed using PROC MIXED of SAS with 2 preplanned orthogonal contrast statements to evaluate ewe performance and reproductive measurements: 1) the mean of E+ vs. the mean of NE+ and OG and 2) the mean of NE+ vs. the mean of OG. Pregnancy rates and percentage of multiple births were analyzed by the Chi-squared procedure of SAS. Ewe BW, BCS, total gain, and ADG did not differ ( $P \geq 0.12$ ) across treatments at the initiation of the study, at breeding, end of breeding, or end of the study. Pregnancy rates did not differ ( $P \geq 0.19$ ) across treatments. Percentage of multiple births tended ( $P = 0.07$ ) to be greater from NE+ and OG compared with E+ (19 vs. 0%, respectively) and tended ( $P = 0.07$ ) to be greater from NE+ compared with OG (19 vs. 0%, respectively). Therefore, grazing spring-born Katahdin ewes on stockpiled endophyte-infected tall fescue, novel endophyte-infected tall fescue, or Persist orchardgrass during the winter months may result in similar performance and pregnancy rates. However, grazing ewes on stockpiled novel endophyte-infected tall fescue may result in more multiple births.

**Key Words:** Katahdin, grazing, performance

# Graduate Student Competition: ADSA Dairy Foods Graduate Student Poster Competition

**T191 Characteristics of cheese powders and the role of color on consumer perception of cheese flavor.** Ni Cheng\*, Yeijin Jo, and M. A. Drake, *Southeast Dairy Foods Research Center, North Carolina State University, Raleigh, NC.*

Cheese powder is applied in many food applications including sauces and snacks. Cheese flavor is a consumer expectation of these applications. The objective of this study was to characterize the properties of cheese powders and to explore the role of color on consumer perception of cheese flavor on cheese powder applications. Duplicate samples of 11 commercial cheese powders were collected. Proximate analyses (moisture, fat, salt, color, pH) were conducted. Cheese powders were rehydrated and evaluated with a trained panel using an established cheese flavor lexicon. Volatile compounds were extracted by solid phase micro-extraction (SPME) followed by gas chromatography-mass spectrometry (GC-MS). Seven representative powders were selected from trained panel sensory results, applied to potato chips, and evaluated by 113 consumers. Consumers evaluated expected liking and cheese flavor before tasting (visual only) and after tasting. Moisture, fat, salt and pH of cheese powders varied (1.40–4.73%, 0.77–47.74%, 1.68–13.99% and 4.56–6.88, respectively). Color varied from white to dark orange. Cheese powders were characterized by a variety of natural cheese flavor attributes including milky, whey, diacetyl, sulfur, brothy, nutty, and free fatty acid and basic tastes. The major aroma-active volatile compounds in cheese powders were dimethyl sulfide, dimethyl disulfide, dimethyl trisulfide, methional, 2/3-methylbutanal, guaiacol, 1-octen-3-ol, diacetyl and volatile free fatty acids. Consumers associated orange color with higher cheesy color liking and higher expected cheese flavor compared with white cheese powders ( $P < 0.05$ ). Diacetyl and whey flavors with salty and sour tastes were perceived as “cheesy” by consumers. Orange color in cheese powders enhanced consumer cheese flavor expectations while diacetyl and whey flavors and salty and sour tastes increased cheese flavor on potato chips.

**Key Words:** cheese powder, cheese flavor, color

**T192 The effect of raw milk cooling on sensory perception and shelf life of pasteurized skim milk.** Andy Lee<sup>\*1</sup>, D. M. Barbano<sup>2</sup>, and M. A. Drake<sup>1</sup>, <sup>1</sup>*Southeast Dairy Foods Research Center, North Carolina State University, Raleigh, NC,* <sup>2</sup>*Cornell University, Ithaca, NY.*

The cooling rate of raw milk may influence sensory properties and pasteurized shelf life. Under the Pasteurized Milk Ordinance (PMO) for grade A milk, raw milk may be cooled instantaneously by on-farm heat exchangers but is also acceptable if “cooled to 10°C or less within four (4) hours of the commencement of the first milking.” The objective of this study was to determine the effect of raw milk cooling on consumer perception and shelf life. Raw milk (18–21°C) was obtained and transported within 1 h of milking to North Carolina State University. After comingling, the batch was split and a plate heat exchanger was used to quickly cool one treatment to <6°C for all milkings. The second treatment was stored in a jacketed bulk tank and slowly cooled over 4 h to <10°C. Three consecutive milkings were collected every 12 h with subsequent milkings added to the previous collections. The bulk milk was kept below 10°C while adding milk for the slow cool milk treatment. After 72 h, each whole milk was separated, pasteurized at 73 or 78°C for 20 s, homogenized, and held at 4°C. Difference tests ( $n = 75$ )

and consumer acceptance tests ( $n = 100$ ) were conducted to determine if consumers could detect differences among milks. Descriptive analysis (DA) and microbial testing for aerobic, psychrotrophic, and spore counts were conducted through shelf life. The entire experiment was repeated in triplicate. Raw milks averaged 4.29 Log cfu/mL by aerobic plate count, 52 cfu/mL coliforms, 300,000 SCC and  $3.15 \pm 0.7\%$  protein. Spores were < 20 cfu/mL in raw milk. After processing, consumers could not detect differences ( $P < 0.05$ ) between cooling treatments of the same pasteurization temperature nor between different temperatures of the same cooling treatment. Milks reached sensory failure 35–42 d after processing, and aerobic counts were between 5 and 7 log cfu/mL. Higher pasteurization temperature decreased shelf life. Cooling treatment had no effect on shelf life. These results suggest that pasteurized milk quality is due to a combination of many factors. Raw milk cooling rate is not the largest effect on milk quality.

**Key Words:** fluid milk, cooling, quality

**T193 Reducing protein bar hardening via whey protein-polyphenol ingredients.** Margaret Schneider\*, Mary Ann Lila, and E. Allen Foegeding, *Department of Food, Bioprocessing and Nutrition Sciences, North Carolina State University, Raleigh, NC.*

Whey proteins serve as a structural component and source of protein in foods. When used in protein bars, whey proteins can contribute to a reduction in shelf life as a consequence of bar hardening. Bar hardening has been associated with water migration between phases and formation of a protein network in bars over time. One approach to ameliorate bar hardening is to form whey protein meso-structures that are not susceptible to water migration or protein network formation. We hypothesize such structures can be formed by using berry polyphenols to initiate whey protein aggregation that leads to formation of inert particles. Whey protein-cranberry polyphenol (WP-CP) particles were formed by adding 200 g of whey protein isolate (WPI) to 1 L of cranberry juice during continuous stirring. After mixing for 4 h, particles were pelleted (7,000 x g for 40 min), re-suspended at 0.01 g/mL in water, and spray dried. Bars were made with WP-CP particles or unmodified WPI. Textural properties were assessed using transient creep-recovery testing which measured the maximum compliance ( $J_{max}$ , indicating bar firmness) and percent creep-recovery (bar viscoelasticity). The WP-CP particles contained up to 2 mg/g of total phenolics, and the particle size ranged from 10 to 100  $\mu\text{m}$ . Bars produced with WPI had a smaller  $J_{max}$  (more rigid) of 0.000053 Pa<sup>-1</sup> compared with 0.0023 Pa<sup>-1</sup>  $J_{max}$  for bars formulated with WP-CP particles. Bars formed with the WP-CP had a reduced percent recovery of 2.5% compared with 28.6% for bars made with WPI. Greater percent recovery indicates a more elastic system and possible network formation. Bars were placed in accelerated shelf life tests that imitate 3 weeks of storage at room temperature. The bars with WP-CP particles remained less rigid and more viscous ( $J_{max}$  of 0.0039 Pa<sup>-1</sup> and 10.9% creep recovery) than control whey protein bars ( $J_{max}$  0.000012 Pa<sup>-1</sup> and 57.5% creep recovery). All differences between bars were found to be statistically significant at the  $P < 0.01$  level. Rheological properties are consistent with WP-CP particles acting as inactive fillers and preventing formation of a protein network. These results suggest that WP-CP particles can be used to extend shelf life of protein bars and offer additional health benefits from plant polyphenols.

**Key Words:** whey protein, polyphenol, protein bar

**T194 Development and characterization of whey protein nanoparticles for beverage applications.** Ty B. Wagoner<sup>\*1</sup>, Loren S. Ward<sup>2</sup>, Chris W. Pernell<sup>1</sup>, and E. Allen Foegeding<sup>1</sup>, <sup>1</sup>North Carolina State University, Raleigh, NC, <sup>2</sup>Glanbia Nutritionals, Twin Falls, ID.

Whey protein consumption has been linked to several health benefits including increased satiety and metabolic regulation; therefore, there is interest in increased consumption of foods rich in whey proteins. Meal replacement beverages and sports drinks are categories of foods that contain whey proteins. However, low thermal stability—especially near the protein isoelectric point (pI)—limits the pH range and protein concentrations at which beverages can be formulated. Studies have shown that at a narrow pH range close to the pI, whey proteins and pectin self assemble into soluble complexes (SCs) that have improved colloidal stability. Our objectives were to 1) determine the conditions required to form stable whey protein-pectin SCs, 2) characterize physical properties of the SCs, and 3) evaluate the effect of SCs on bulk rheological properties. Soluble complexes were formed at various protein concentrations (1, 4, 5 and 6% wt/wt) with a constant protein to pectin ratio of 8:1 by adjusting the pH from 7 to 5. After complexation, particles were stabilized by heating at 85°C for 25 min. The properties of unheated and heat-set SCs were characterized via intrinsic viscosity, particle size distribution, and rheological analysis. Laser diffraction particle size analysis revealed that heating shifted sizes toward a monomodal distribution with mean diameter of ~100 nm for all protein concentrations. Heat-stabilization significantly reduced intrinsic viscosity of SCs from 93.6 mL/g to 79.5 mL/g ( $P < 0.05$ ), suggesting conformational changes that favor a smaller hydrodynamic size. Decreased viscosity of heat-set SCs was consistent with intrinsic viscosity and particle size results. Increasing protein concentration had very little effect on size of heat-set SCs, suggesting applications in beverages where a high protein concentration is desired. Heat-set SCs remained as 100 nm particles and did not aggregate over the pH range of 3–6. Moreover, they remained stable in this pH range after thermal processing. These results indicate that whey protein-pectin SCs can be heat-set into 100 nm particles with enhanced colloidal stability for applications in beverages.

**Key Words:** whey protein, beverage, thermal stability

**T195 Casein and exopolysaccharide degrading activities of *Bacillus* strains isolated from the dairy environment.** Dipakkumar Mehta<sup>\*1</sup>, Hasmukh Patel<sup>1</sup>, Ashraf Hassan<sup>2</sup>, and Brandon Nelson<sup>2</sup>, <sup>1</sup>South Dakota State University, Brookings, SD, <sup>2</sup>Daisy Brand, Garland, TX.

Milk proteins and exopolysaccharides (EPS) play an important role in texture formation and stabilization of yogurt. Many *Bacillus* strains have the ability to breakdown proteins and degrade polysaccharides. To our knowledge, the ability of the *Bacillus* spp. to degrade EPS from starter cultures has not been tested. The objective of this study was to quantify proteolytic and the EPS-degrading activity of *Bacillus* strains. Twenty-five *Bacillus* strains isolated from the pasteurizer balance tank of a dairy company were pre-screened for their proteolytic ability using skim-milk agar (SMA) at 42°C. The strains that showed proteolytic activities were inoculated individually at  $10^3$  cfu/mL in heated (90°C for 10 min) rehydrated nonfat dry milk (11% w/v) which was then incubated at 42°C for 48h. The evidences of proteolytic activity were quantified by SDS-PAGE and measuring the non-casein nitrogen (NCN). To study EPS degradation, 2 *Bacillus* strains that were able to degrade a wide range of polysaccharides in a previous study were selected. The reduction in viscosity of a whey protein concentrate (WPC) solution (10% w/v) fermented with yogurt cultures producing high, medium, or low EPS in

the presence of the test *Bacillus* strains ( $10^3$  cfu/mL) indicated positive results. The viscosity of the fermented media was measured using the Stress-Tech rheometer at 42°C and a shear rate of  $100 \text{ s}^{-1}$ . Out of the 25 *Bacillus* strains, 20 strains showed proteolytic activities on SMA. Of these 20 strains, 8 strains showed significant ( $P < 0.05$ ) 30–50% degradation of caseins (predominantly  $\beta$ - and  $\kappa$ -CN) as determined by SDS-PAGE and 4 fold increase in NCN contents. Highly proteolytic strains also caused sweet curdling in skim milk. Used Two *Bacillus* strains showed 20–30% reduction in viscosity was observed in the WPC medium containing the low EPS producing culture whereas only 8–10% decrease in viscosity was found in the medium fermented with the high EPS-producing culture. The results of the present study showed the potential of *Bacillus* strains to degrade EPS and reduce viscosity of the fermented dairy products. It also provided useful information on the prevalence of highly proteolytic *Bacillus* strains in a relatively large number of isolates from a dairy plant.

**Key Words:** *Bacillus*, protein, EPS

**T196 Chemosensory analysis of light-emitting diode and fluorescent light on fluid milk volatiles.** Kemia N. Amin<sup>\*</sup>, Maria A. Hadley, Susan E. Duncan, and Kumar Mallikarjunan, Virginia Polytechnic Institute State University, Blacksburg, VA.

Fluid milk deteriorates rapidly under lighted storage, affecting milk sensory and nutritional quality before purchase. Energy conservation efforts have shifted commercial retail cases from fluorescent to light emitting diode lights (LED). The effect of LED lights on milk quality has not been determined. Efficient milk freshness validation methods are needed in the dairy industry to predict sensory quality. The purpose of this study was to determine volatile differences of 2% milk under LED and fluorescent lights using a commercial conducting polymer electronic nose (ENose). This study focused on changes in volatiles within 72 h of storage under refrigerated retail case conditions. Fresh 2% milk (half gallon high density polyethylene;  $n = 60$ ) was purchased from a local grocery store immediately upon delivery. Bottles (treatments: light protected control [LP; foil overwrap]; light-exposed (LE)) were randomly placed in a commercial retail case with LED and fluorescent lighted sections. Milk was tested on receipt and at 4, 8, 24, 48, and 72 h. Samples ( $n = 3$  bottles per LP and LE treatment) were collected at each time interval and tested with the ENose and thiobarbituric acid reactive substances (TBARS). Canonical distribution ( $P < 0.05$ ) of volatile changes showed 100% separation of volatiles components detected by ENose, for each time under each light treatment for LE milk compared with LP control. As time increased, separation between LED and fluorescent clustered ENose data moved further away from the control and each light treatment resulting in greater differences. By 72 h, LED LE cluster data were farther from the ideal fresh milk than the fluorescent cluster. This suggests that milk under LED may have more differences in volatiles than milk under fluorescent light at 72 h. TBARS values were not significantly different ( $P > 0.05$ ) between the interval light treatments. ENose technology could be more sensitive to volatile changes than TBARS. The volatile differences detected through the ENose may affect product integrity and consumer response. Understanding different light effects on milk and ENose application in the dairy industry could improve sensory and quality testing of milk.

**Key Words:** electronic nose (ENose), milk, light oxidation



**T197 Inactivation of thermoduric bacterial endospores in milk by combined effect of cavitation and thermal treatment.** Dikshi Bawa\*, Sanjeev Anand, Harsh Dahiya, and Hasmukh Patel, *South Dakota State University, Brookings, SD.*

*Bacillus* endospores can survive milk pasteurization, and later germinate and grow during further processing of milk. Their resistance to various physical and biochemical treatments, dormancy, ability to adhere to the surfaces of process equipment resulting in biofilm formation, and prompt germination under favorable conditions, make them important contaminants in dairy processes. The objective of this research was to investigate the effect of hydrodynamic cavitation in a continuous mode, and its combination with thermal treatment, to reduce *Bacillus* spores in milk. We hypothesized that cavitation would induce germination of spores and any post cavitation heat treatment would lead to their inactivation. Spores of *Bacillus licheniformis* (ATCC 6634) were produced in lab by incubating on brain-heart infusion (BHI) agar plates for about 2 weeks, and were harvested from plates by washing and centrifugation. The vegetative cells in spore suspension were inactivated by heating at 85°C for 10 min. The resulting spores were inoculated in sterile skim

milk to a level of log 2 cfu/mL. Inoculated milk samples at 10°C were passed through APV Cavicator (SPX, Denmark) at 60 Hz frequency using 4-row rotor in 6-mm housing. The exposure time of 22 s per pass was provided at 200 L/h flow rate with a back pressure of 120 kPa. The milk containing endospores was recirculated for 25 min (a total exposure time of 183 s) with a temperature rise to 99°C. Samples were cooled and held for 3 h at 30°C to let the germination process occur. Samples were then heated at 85°C for 15 min, and were plated on BHI agar medium. Experiments were conducted as replicates of 2, and were repeated thrice. Statistical significance of the data at  $P < 0.05$  was determined using the SAS software. A significant difference was observed in the endospore counts after the treatments. Heat treatment alone did not result in inactivation of the spores. Whereas, cavitation with holding followed by heat treatment caused a reduction with only 0.60 log survivors. In conclusion, it was evident that a combined process with cavitation effect, holding for germination, followed by thermal treatment can be effectively used to inactivate thermoduric endospores.

**Key Words:** cavitation, endospore, germination

# Graduate Student Competition: ADSA Production Division Graduate Student Poster Competition, PhD

**T198 Effects of estrus number on milk yield and estrus expression in Holstein cows managed for extended lactation.** Charlotte Gaillard\*, Mogens Vestergaard, and Jakob Sehested, *Aarhus University, Foulum, Tjele, Denmark.*

Cows managed for a 16-mo extended lactation have, at least, 8 estrus periods before being inseminated. The aims were to determine the effect of estrus number on milk yield and estrus expression, and to compare conception rates with the rates obtained during the previous 10 mo lactation. It was hypothesized that (1) milk yield will decrease during the day of estrus, and this loss will diminish with increasing estrus number; (2) postponing the insemination (AI) will increase the expression of estrus leading to a better detection and a higher conception rate. A total of 62 Holstein cows (30% 1st parity) were managed for 16 mo extended lactation. The 8 first estrus periods were determined by milk progesterone levels (<3 ng/L) and the day of estrus by visual observations (mucus consistency, blood in genital area, cow jump/stand/lay). Milk yield at 'estrus day' was compared with 'normal day' yield (average yield of d -4, -5 and -6 before 'estrus day'). The percentage of cows expressing estrus was calculated using the behaviors mounting, being mounted or standing as criteria. Milk yield was analyzed using a linear mixed effects model while a chi-squared test was used for evaluating estrus expression and conception rate. The results showed a milk loss of  $0.56 \pm 0.19$  kg per estrus day ( $P < 0.001$ ) with no differences between the 8 estruses ( $P = 0.3$ ). This negative estrus effect on milk yield is small compared with the well-known pregnancy effect. Moreover, 70% of the cows expressed the 8th estrus compared with 37% for the 1st estrus ( $P = 0.02$ ) and 40% for the 2nd estrus ( $P = 0.06$ ). However, the conception rates of the 1st and 2nd AI did not differ from those of the previous 10 mo lactation (respectively + 2.9%,  $P = 0.8$  and + 3.4%,  $P = 0.7$ ). To conclude, milk yield decreases similarly during each of the 8 estrus days studied whereas the proportion of cows expressing estrus nearly doubles from the 1st and 2nd to the 8th estrus.

**Key Words:** estrus number, milk yield, mounting

**T199 Effects of canola meal as a source of rumen-undegraded protein on ruminal fermentation using a dual-flow continuous-culture system.** Eduardo Marostegan de Paula\*<sup>1</sup>, Lorryny Galora da Silva<sup>1</sup>, Pedro Del Bianco Benedetti<sup>1,2</sup>, Hugo Monteiro<sup>1,3</sup>, Yenling Yeh<sup>1</sup>, Teshome Shenkoru<sup>1</sup>, Glen A. Broderick<sup>4</sup>, and Antonio Faciola<sup>1</sup>, <sup>1</sup>University of Nevada, Reno, NV, <sup>2</sup>Federal University of Viçosa, Viçosa, MG, Brazil, <sup>3</sup>Maringa State University, Maringa, PR, Brazil, <sup>4</sup>Broderick Nutrition & Research, Madison, WI.

Previous research indicated that there were significant differences in rumen-undegraded protein (RUP) among canola meals (CM). These differences could influence the nutritional value of CM. The objective of this study was to evaluate the effects of feeding CM with different RUP on ruminal fermentation, nutrient digestion, and microbial growth using a dual-flow continuous-culture system. Diets were randomly assigned to 6 fermenters in a replicated  $3 \times 3$  Latin square with three 10-d experimental periods consisting of 7-d for adaptation and 3-d for sample collection. Treatments were solvent soybean-meal (SBM); low-RUP CM (LCM); and high-RUP CM (HCM). Fermenters were fed 72 g/d, divided in 4 feedings. Diets were prepared as 3 concentrate mixes that were combined with orchardgrass hay and wheat straw. Liquid and solid flow rates were adjusted to 11 and 5.5%/h, respectively. Samples

were collected for digestibility, ruminal fermentation, and microbial growth. Statistical analyses were performed using SAS. Orthogonal contrasts were used to compare effects of different protein sources (SBM vs. LCM + HCM), and (LCM vs. HCM). Partial data are presented in the table below. Ruminal  $\text{NH}_3\text{-N}$ , total VFA concentration, molar proportion of acetate, propionate, butyrate, and isobutyrate were not affected by treatments. Molar proportion of valerate was greater when SBM was fed, whereas molar proportions of isovalerate, and total BCVFA were lower for CM diets (Table 1). Isovalerate, an intermediate product of leucine degradation, is associated with protein degradation; therefore, this may indicate that CM diets had higher RUP than SBM.

**Table 1 (Abstr. T199).** Effects of feeding canola meals (with different RUP LCM and HCM) on ruminal fermentation

Item	Treatment				Contrast <i>P</i> -values	
	SBM	LCM	HCM	SEM	SBM vs. LCM + HCM	LCM vs. HCM
$\text{NH}_3\text{-N}$ , g/d	0.51	0.57	0.53	0.02	NS	NS
Total VFA, mmol	122.6	116.2	116.6	2.54	NS	NS
Acetate, %	64.6	65.4	66.5	1.25	NS	NS
Propionate, %	19.9	20.7	21.1	0.86	NS	NS
Butyrate, %	11.7	10.9	9.4	0.64	NS	NS
Isobutyrate, %	0.44	0.48	0.44	0.02	NS	NS
Valerate, %	1.5	1.6	1.6	0.03	0.02	NS
Isovalerate, %	1.3	1.1	1.1	0.08	0.02	NS
Acetate: Propionate	3.3	3.2	3.2	0.18	NS	NS
Total BCVFA, mmol	2.1	1.8	1.8	0.11	0.04	NS

**Key Words:** canola meal, continuous culture, RUP

**T200 Comparison of lying times of lame versus sound dairy cattle using a leg-based accelerometer.** Barbara A. Wadsworth\*, Lauren M. Mayo, Nicky I. Tsai, Amanda E. Stone, Denise L. Ray, Joey D. Clark, and Jeffrey M. Bewley, *University of Kentucky, Lexington, KY.*

Lameness is a painful disease that affects cattle performance. Lame cattle may lay down longer than sound cattle. The objective of this study, conducted at the University of Kentucky Coldstream Dairy from March 18 to December 10, 2014, was to determine lying times of lame cows versus sound cows using a leg-based accelerometer. All cows ( $n = 96$ ) were housed in 2 freestall barns and were balanced for parity and days in milk. Each individual freestall barn stocking density never exceeded 100%. One barn was equipped with sawdust covered Dual Chamber Cow Waterbeds (Advanced Comfort Technology, Reedsburg, WI) and another with sawdust covered rubber-filled mattresses (Promat Inc., Woodstock, Ontario, Canada). All cows were equipped with an AfiAct Pedometer Plus (Afirmilk, Kibbutz Afikim, Israel), which recorded daily lying time. The AfiMilk milking system (Afirmilk) recorded daily milk yield. The Hobo U23 Pro V2 External Temperature Relative Humidity Data Logger (Onset, Bourne, MA) determined temperature and relative humidity (THI) in each barn every 15 min. Ambient conditions were categorized as low and high if THI was below or above 68, respectively.

Cows gait was assessed weekly using a 1 (sound cow) to 5 (severely lame cow) scale for general symmetry, speed, head bobbing, spine curvature, tracking, and abduction and adduction. Final gait score was calculated as the mean of all gait aspects for each cow. Cows scored  $\geq 3$  were determined as lame. The MIXED procedure in SAS (Version 9.3, Cary, NC) was used to evaluate factors influencing lying times. Stepwise backward elimination was used to remove non-significant interactions ( $P \geq 0.05$ ). Lying times of lame cows were greater than sound cows ( $P < 0.01$ ; 10.96 h/d and 9.51 h/d, respectively). All cows lied down longer in the cold season versus the warm season ( $P < 0.01$ ; 10.43 h/d and 10.04 h/d, respectively). Milk yield was not different ( $P \geq 0.05$ ) between lame and sound cows.

**Key Words:** lying time, lameness, accelerometer

**T201 Survey of management of reproduction on Canadian dairy farms.** José Denis-Robichaud<sup>\*1</sup>, Ronaldo L. A. Cerri<sup>2</sup>, Andria Jones-Bitton<sup>1</sup>, and Stephen J. LeBlanc<sup>1</sup>, <sup>1</sup>*Department of Population Medicine, University of Guelph, Guelph, ON, Canada*, <sup>2</sup>*Faculty of Land and Food Systems, University of British Columbia, Vancouver, BC, Canada*.

The objective of this study was to survey attitudes and management practices for reproduction in a representative sample of Canadian dairy farms. A questionnaire was distributed online and by mail to Canadian dairy farmers from March to May 2014. Respondents were asked to give their percentage of artificial inseminations (AI) based on visual heat detection, timed artificial insemination (TAI) program, or automated activity monitoring (AAM), for first and subsequent AI separately. The main management practice was the one that used for  $>50\%$  of inseminations, or “combined” if no one practice was used for  $>50\%$  of AI. Out of the approximate 9,000 invitations to participate to the survey, a total of 833 surveys were completed (response rate estimated at 9%). The average number of lactating cows per herd was 77 (median = 55, interquartile range = 40–88), and the herds were located in all Canadian provinces. Lactating cows were housed in tie stall (61%) and freestall barns (39%). Visual heat detection was reported to be the main management practice in 51% and 44% of herds for first and subsequent AI, respectively. Respondents reported observing cows for heat signs 3.3 times per day for a total of 36 min, on average. Timed AI was the main management practice in 21% and 22% of herds for first and subsequent AI, respectively. Although 74% of respondents agreed that reproductive hormones were safe for consumers of dairy products, fewer (64%) agreed that routine use of synchronization programs was acceptable to them, or to consumers (44%). Ten percent of respondents, mainly housing lactating cows in free stall barns, reported using AAM as their main management practice for all AI. They reported using the system to flag cows in heat, but also using observed signs of estrus to decide to inseminate. Sixteen and 20% of all respondents used combined practices for first and subsequent inseminations, respectively. Bulls were used by 4% of respondents. These results suggest that visual heat detection is the principal management practice in many Canadian herds, but that TAI and AAM systems are also important in the management of reproduction.

**Key Words:** dairy cow, reproduction management, survey

**T202 Lactational performance of early lactation, high-producing dairy cows fed corn silage produced by different seed corn hybrids.** Ishwary Acharya<sup>\*1</sup>, Mark Kirk<sup>2</sup>, and David Casper<sup>1</sup>, <sup>1</sup>*Dairy Science Department, South Dakota State University, Brookings, SD*, <sup>2</sup>*Masters Choice, Anna, IL*.

Twenty-one (6 primiparous and 15 multiparous) high producing early lactation Holstein cows were fed one of 3 experimental corn silages harvested from the planting of 3 different seed corn hybrids from wk 4 through wk 14 postpartum. Corn hybrids (Control (C): Dekalb and Masters Choice: MC1 and MC2) were planted and harvested as corn silage during the 2012 growing season. Corn silage was harvested using a kernel processor silage harvester, inoculated, and ensiled in either a bunker or individual Ag Bags. Total mixed rations were formulated to be isonitrogenous at 17.5% CP consisting of 15.9% alfalfa hay, 35.1% concentrate mix and 48% of the respective experimental corn silage (DM basis). Cows were blocked by calving date and parity and randomly assigned to 1 of 3 treatments in a randomized complete block design. Data collected the third wk postpartum was used as a covariate in least squares analysis of covariance via the PROC MIXED procedure (SAS Institute). Dry matter intake (22.9, 23.5, and 22.4 kg/d for C, MC1, and MC2, respectively), milk yield (35.6, 34.8, and 36.1 kg/d), 3.5% fat-corrected milk (FCM) yield (38.7, 36.5, and 37.6 kg/d), energy corrected milk yield (38.2, 36.1, and 38.1 kg/d), feed efficiency (1.79, 1.61, and 1.67 kg/kg; 3.5% FCM/DMI), milk fat (4.17, 3.94, and 3.71%), milk protein (3.12, 3.09, and 3.03%), lactose (4.93, 4.92, and 4.92%), solid-not-fat (8.96, 8.92, and 8.85%), body weight change (–0.10, –0.06, and –0.08 kg/d), and body condition score change (–0.05, –0.04, and –0.05 score/d) were similar for early lactation dairy cows fed all corn silage hybrids. Milk urea nitrogen was significantly ( $P < 0.05$ ) lower for cows fed C (13.64) compared with cows fed MC1 and MC2 corn silage (15.0, and 15.0 mg/dL). This study demonstrated that different seed corn hybrids for the production of corn silage were similar in lactational performance.

**Key Words:** corn silage, corn hybrid, high-producing dairy cow

**T203 Nitrogen utilization and growth effects in Holstein dairy calves fed a moderately high protein or conventional milk replacer.** Colleen E. Chapman<sup>\*1</sup>, Thelton M. Hill<sup>2</sup>, and Peter S. Erickson<sup>1</sup>, <sup>1</sup>*University of New Hampshire, Durham, NH*, <sup>2</sup>*Provimi North America, Brookville, OH*.

Studies have shown that calves fed milk replacers (MR) with crude protein (CP) concentrations greater than 20% typically found in conventional MR have higher dry matter intakes (DMI) and greater average daily gains (ADG), but consume less starter which can lead to stress during weaning and reduced rumen development. The greater amount of CP being fed to preweaned calves may alter their nitrogen (N) balance and excess N may be excreted in the urine. The objective of this study was to determine N utilization in preweaned calves fed diets varying in the amount of CP and MR fed. This study used 24 newborn dairy heifer calves blocked by birth and randomly assigned to 1 of 3 treatments: (1) 454 g of a conventional MR (C; 20% CP, 20% fat), (2) 680 g of a moderately high protein MR (MMR; 26% CP, 17% fat), or (3) 908 g of a MMR (HMR; 26% CP, 17% fat). All calves had free choice access to starter and water. Both MR and starter were medicated with decoquinate. During weaning (d 43–49), AM MR feeding ceased. On d 50, all MR feedings ended; however, starter and water intakes were continuously recorded until d 56. At 5 wk of age, urine was collected using urinary catheters for 3 d and chromium oxide was administered by bolus at 2 g/d for 7 d to estimate N efficiency. Calves fed MMR and HMR had similar starter intakes, feed efficiencies (FE), and ADG; with the combined treatments having reduced starter intakes (258 g/d vs. 537 g/d), greater ADG (674 g/d vs. 422 g/d), and improved FE (0.57 vs. 0.45) compared with the C calves preweaning ( $P < 0.05$ ). However, DMI and water intake were similar across all treatments. Results from the N utilization phase showed that MMR and HMR treatments had



similar but lower N efficiency than C calves (45.5% vs 52.7%;  $P < 0.05$ ). This could be due to MMR and HMR fed calves having greater urine volume; and thereby, greater combined urine N output compared with C calves (17.6 g/d vs 12.1 g/d;  $P < 0.05$ ). In summary, feeding a moderately high protein MR increased ADG and improved FE during the preweaning period, but reduced starter intake and lowered N efficiency.

**Key Words:** high protein, milk replacer, nitrogen efficiency

**T204 Blood calcium changes after prophylactic subcutaneous treatment with calcium.** Cynthia L. Miltenburg\*, Elizabeth Scholtz, Dorothee Bienzle, Todd F. Duffield, and Stephen J. LeBlanc, *University of Guelph, Guelph, ON, Canada.*

Prophylactic calcium supplementation immediately after calving is a common strategy to prevent clinical and subclinical hypocalcemia in parturient dairy cows. The objective of this study was to evaluate the effect of prophylactic administration of Theracalcium on blood calcium concentration at 24 and 48 h after treatment, in cows without clinical hypocalcemia. Cows ( $n = 128$ ) from 4 farms were blocked by parity and randomly assigned to receive either calcium gluconate (35% w/v) in combination with calcium glucoheptonate (10% w/v; Theracalcium, Vétoquinol Canada Inc., Lavaltrie, Quebec) or a placebo (medication vehicle solution with no calcium) at first contact with each cow after calving and again 12–24 h later when available for lockup. Each dose was 120 mL injected subcutaneously over 2 sites. Total serum calcium concentration (tCa) was measured from coccygeal blood samples before (time 0) and 24 and 48 h after first treatment. There was no significant difference in tCa at time 0 ( $2.05 \pm 0.02$ ) between groups ( $P = 0.18$ ). Serum tCa at time 0 was highly correlated with parity ( $r = -0.7$ ) therefore pre-treatment tCa but not parity was used as a covariate in the models. A mixed model was constructed to measure tCa at 24 h after first treatment. For cows that had received 1 injection of calcium before the blood sample at 24 h ( $n = 95$ ), tCa was significantly higher in the treated cows ( $P = 0.01$ ): mean  $\pm$  SE  $2.03 \pm 0.03$  versus  $1.90 \pm 0.03$  mmol/L, accounting for tCa at time 0 and a treatment by tCa at time 0 interaction. At 48 h there was no significant difference in tCa between treatment and control (mean  $\pm$  SE  $2.12 \pm 0.02$  and  $2.10 \pm 0.03$  mmol/L, respectively) accounting for tCa at time 0 and farm as a fixed effect. With this subcutaneous prophylactic calcium treatment regimen, blood calcium levels were temporarily increased at 24 h after treatment. Further studies of disease and production in the postpartum period are required to determine if such a rise in blood calcium translates into improvements in transition cow health.

**Key Words:** calcium supplementation, hypocalcemia

**T205 Moved to Ruminant Nutrition: General II (page 475)**

**T206 Quantification of select ruminal bacterial in Holstein bull calves treated with daily oral sodium bicarbonate.** Taylor T. Yohe\*<sup>1,2</sup>, Rene R. Delgado-Peraza<sup>1</sup>, Hannah L. M. Tucker<sup>1,2</sup>, and Kristy M. Daniels<sup>1,2</sup>, <sup>1</sup>The Ohio State University, OARDC, Wooster, OH, <sup>2</sup>Virginia Tech, Blacksburg, VA.

Dairy calves commonly have a rumen pH below 5.5 and it is unknown if this is problematic or normal for rumen development. The objective was to test if oral administration of sodium bicarbonate ( $\text{NaHCO}_3$ ) to young calves can affect ruminal: pH, lactate concentration, and populations of select ruminal bacteria. We hypothesized that administration of  $\text{NaHCO}_3$  would increase rumen pH, decrease lactate concentration, and

affect populations of lactate-producing (*Lactobacillus* spp.) and lactate-utilizing (*Megasphaera elsdenii*) bacteria. Twelve Holstein bulls ( $40.2 \pm 1.6$  kg BW) were randomized into treatments, arranged factorially. The factorials were treatment and route of treatment. The treatments were: CON-drench (water), BICARB-drench (1 M  $\text{NaHCO}_3$  in water), CON-bolus (gelatin capsule), BICARB-bolus ( $\text{NaHCO}_3$  gelatin capsule).  $\text{NaHCO}_3$  was administered twice daily, adjusted weekly, and reached a maximum of 48 g/d. Calves were fed 543g DM/d of a 22% CP, 20% fat milk replacer and 20% CP starter. Rumen contents were sampled at wk 1 and 8 for pH, lactate, and bacteria measurement. Rumen pH did not depend on treatment or route, but was different by week (wk 1, pH =  $6.00 \pm 0.15$ ; wk 8 pH =  $5.19 \pm 0.14$ ;  $P = 0.007$ ). Overall D- and L- lactate concentrations did not differ ( $7.45 \pm 2.15$  mM and  $7.33 \pm 1.98$  mM, respectively); although, concentrations of each were numerically higher at wk 8 as opposed to wk 1. Populations of *Lactobacillus* spp. and *M. elsdenii* were calculated as a percentage of total 16S rDNA gene copies. *Lactobacillus* spp. abundance was unaffected by treatments or their interactions, but abundance of *M. elsdenii* was affected by the interaction of treatment and week (wk 1 CON = 0.05%, wk 1 BICARB = 0.00%, wk 8 CON = 0.19%, wk 8 BICARB = 0.26% of total bacteria, respectively;  $P = 0.05$ ). Administration of  $\text{NaHCO}_3$  by one of 2 oral routes did not affect rumen pH, lactate, or *Lactobacillus* spp. numbers, but in combination with time, did have an effect on *M. elsdenii* abundance. This suggests that *M. elsdenii* was able utilize lactate as a substrate and can withstand low rumen pH. These results support the idea that low rumen pH may be part of the normal rumen development of a dairy calf.

**Key Words:** dairy calf, rumen, bacteria

**T207 Cost of days open equations accounting for variable market and dairy herd conditions.** Karmella A. Dolecheck\* and Jeffrey M. Bewley, *University of Kentucky, Lexington, KY.*

The objective of this study was to develop equations for estimating farm-specific cost of days open. The equations were constructed using a whole farm stochastic simulation model previously described by Bewley et al. (2010) and Liang (2013). Ten thousand iterations were run for lactations 1 to 5 with the mean cost per day open as an output. Stochastic variables expected to have potential effects on cost per day open were collected from each iteration. Those variables included: rolling herd average milk production, age at first calving, mature cow live weight, heifer calf value, bull calf value, semen cost, days in milk dictating an open cow as a reproductive cull, milk production level dictating an open cow as a production cull, veterinarian costs, discount rate, milk price, feed price, replacement price, cull cow price, voluntary waiting period, estrus detection rate, and conception rate. The GLMSE-LECT procedure of SAS 9.3 (SAS Institute, Inc., Cary, NC) was used to analyze the effect of stochastic variables and 2-way interactions on the mean cost per day open for each lactation. Variables remained in the model when significant at  $P < 0.05$ . The  $R^2$  of the resulting models were 0.57, 0.54, 0.64, 0.85, and 0.63 for lactations 1 to 5, respectively. The models were used to develop deterministic, lactation-specific equations for cost of days open. These equations are available in an online spreadsheet at: <http://afsdairy.ca.uky.edu/CostOfDaysOpen>. To demonstrate use, mean US Holstein herd data from 2015 DairyMetrics (Dairy Records Management Systems, Raleigh, NC), 2014 Food and Agricultural Research Policy Institute (Columbia, MO), and published literature were entered into each equation. Mean cost per day open for lactations 1 to 5 was \$2.44, \$2.82, \$4.42, \$4.54, and \$3.32, respectively. These new, robust regression equations for cost of days open account for the complexities of varying market and herd conditions. The equations

can estimate cost of days open in partial budgets without the costs or computing time required for stochastic simulations.

**Key Words:** days open, cost of days open, stochastic model

**T208 Checks and balances: Evaluating reliability of dairy nutrient management data to better protect groundwater resources.** Christine Miller\* and Deanne Meyer, *University of California, Davis, Davis, CA.*

To protect groundwater from further nitrate contamination, California regulations prohibit dairy producers from applying more than 140% of the nitrogen (N) that their crops remove. The regulations require copious annual reporting of crop field management, farm infrastructure, and animal population. The data collected in these annual reports could be integral to evaluating and improving both farm practices and the regulations themselves. Data reliability and accuracy must be assessed to use the information responsibly. Annual Reports from 18 dairies were obtained to assess reliability. Mass balance calculations were performed to check the self-consistency of data within a facility. The results of mass balance calculations show that the data do not account for a remarkably large percentage of the nutrients being produced on the farms. Literature suggests that over 60% of N and 90% of P should be recovered; however, a median of only 25% of both N and P in cattle manure was recovered based on annual report data. This could be due to many different causes including inaccurate nitrogen sampling and analysis techniques, systematic reporting errors, or fraudulent reporting. Given that the accuracy of the majority of the recommended sampling and analysis protocols has not been assessed, it is likely that these methods are a significant source of error. Projects that should improve data collection protocols in both the short and long-term are in progress. Online decision trees are being developed to help farmers self-assess their current data collection practices, and provide personalized suggestions for improvement. Additionally, I will use a statistical modeling approach paired with in-field measurements to examine the uncertainty in these recommended protocols (and thereby the overall uncertainty in regulations). By separating the various sources of measurement error, the model will identify the best ways to improve data collection and regulation efficacy. Results of this and future studies will influence future nutrient management regulations in California and other states with active livestock industries.

**Key Words:** dairy waste management, nitrate leaching

**T209 Distribution of quarter-level SCC across the dry and early post-partum period.** Stephanie A. Metzger\*, Laura L. Hernandez, and Pamela L. Ruegg, *University of Wisconsin, Madison, WI.*

The aim of this study was to characterize changes in quarter-level SCC for cows at dry off and the first 2 weeks postpartum. Milk samples were collected at dry off (S1) and postpartum wk 1 (S2) and wk 2 (S3) from quarters (n = 660) of 185 cows at the UW dairy farm. Microbiological analysis of milk samples was performed at the UW Milk Quality Laboratory. The SCC at each sampling period was used to determine postpartum categories: (1) Healthy (SCC < 150,000 at S1,S2,S3), (2) DryCure (S1 ≥ 150,000; S2 & S3 < 150,000), (3) Chronic (S1,S2,S3 all ≥ 150,000), (4) PostChronic (S < 150,000; S2 & S3 ≥ 150,000), (5) LateCure (S1 & S2 ≥ 150,000, S3 < 150,000), (6) LateInf (S1 & S2 < 150,000, S3 ≥ 150,000), (7) ShortNew (S1 & S3 < 150,000, S2 ≥ 150,000). Univariate analyses were performed to determine risk factors associated with Chronic quarters. Average DIM was 6.1 and 13.6 for S1 and S2, respectively. Median SCC were 117,000, 42,000, and

18,000 cells/mL for S1, S2 and S3, respectively. The prevalence of SCC ≥ 150,000 cells/ml was 43.6%, 13.5%, and 9.4% for S1, S2, and S3, respectively. The distribution of quarters was: Healthy (n = 321; 48.6%), DryCure (221; 33.5%), Chronic (20; 3.0%), PostChronic (13, 2.0%), LateCure (33, 5.0%), LateInf (29, 4.4%), and ShortNew (23, 3.5%). The occurrence of a clinical case in the lactation before dry off was associated with postpartum category ( $P = 0.004$ ). Fewer quarters that had previously experienced a clinical case were categorized as healthy and a greater proportion were categorized as LateCure and PostChronic. Parity was associated with postpartum category ( $P < 0.001$ ). As compared with younger cows, quarters of cows in parity ≥ 3 were 10.9 (3.6–34.6) times more likely to be categorized as chronic (rather than healthy). The number of days dry was greater for quarters categorized as Chronic (66.8d) and LateCure (66.4) as compared with quarters categorized as DryCure (59.1), PostChronic (57.6), LateInf (54.2), Healthy (53.1), or ShortNew (50.5) ( $P < 0.001$ ). Only 8.6% of Chronic quarters were microbiologically positive at dry off. Results of this study demonstrate the dynamic nature of quarter SCC values during the dry and immediate postpartum period.

**Key Words:** dryoff, fresh cow, SCC

**T210 Protein nutrient supply and feed milk value of two newly developed genotypes of transgenic alfalfa compared with non-transgenic alfalfa in dairy cattle.** Xinxin Li\*<sup>1,2</sup>, Yonggen Zhang<sup>2</sup>, and Peiqiang Yu<sup>1</sup>, <sup>1</sup>*Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, Saskatchewan, Canada,* <sup>2</sup>*College of Animal Science and Technology, Northeast Agricultural University, Harbin, China.*

Two newly developed transgenic alfalfa with transformed *TRANSPARENT TESTA 8 (TT8)* and *HB12* genes were developed by scientists at Agriculture and Agri-Food Canada. Our study was conducted to (1) predict the extent of ruminal and intestinal digestion of protein, (2) evaluate the nutrient supply to dairy cattle and (3) determine the feed milk value in dairy cattle. *HB12* transgenic alfalfa, *TT8* transgenic alfalfa and unmodified control genotype alfalfa were cultivated and harvested from the greenhouse of Agriculture and Agri-Food Canada. There were 2 experimental samples for each treatment. Total rumen degraded protein (TRDP), total rumen undegraded protein (TRUP) and intestinal digestible protein fractions were predicted. The parameters evaluated included: truly absorbed rumen microbial protein (AMCP), truly absorbed rumen undegraded feed protein in the small intestine (ARUP), endogenous protein loss in the small intestine (ENDP), total metabolizable protein (DVE) and degraded protein balance (OEB). The PROC MIXED procedure of SAS 9.3 was used for data analysis. Significance was declared at  $P < 0.05$  and trends at  $P \leq 0.10$ . The results showed that *TT8* alfalfa had highest TRDP value compared with *HB12* and control alfalfa ( $P < 0.05$ ). *TT8* alfalfa trended to be higher in TRUP than *HB12* alfalfa, with control alfalfa intermediate ( $P < 0.10$ ). Intestinal digestible PB1 was higher in *TT8* alfalfa than the other 2 alfalfa plants ( $P < 0.05$ ). *TT8* alfalfa had a greater content of microbial protein production ( $MCP_{RDP}$ ,  $P < 0.05$ ). No significant difference of OEB value was found between *HB12* alfalfa and control alfalfa, but the OEB value was higher in *TT8* alfalfa ( $P < 0.05$ ). No significant differences in DVE and feed milk value (FMV) were found among 3 different genotypes of alfalfa plants ( $P > 0.05$ ). In conclusion, altering the expression of lignin-related *TT8* and *HB12* genes in alfalfa plant could affect its nutrient supply to dairy cattle.

**Key Words:** lignin-related gene, alfalfa, feed milk value

### T211 Probiotic *Enterococcus faecium* increased the propionate and total volatile fatty acids on in vitro rumen fermentation.

Lovelina L. Mamuad, Ashraf A. Biswas, and Sang Suk Lee\*, *Sunchon National University, Suncheon, Jeonnam, South Korea.*

*Enterococcus faecium* had been widely used as direct fed microbial due to their capability to produce antibacterial compounds. Thus, this study was conducted to determine the beneficial effects of *E. faecium* on in vitro rumen fermentation, methane concentration, microbial diversity and population. Ruminant samples were collected from ruminally cannulated Holstein Friesian cattle and 40:60 rice straw to concentrate ratio were used as substrate at 1g dry matter (DM) per 100mL buffered rumen fluid. Fresh culture of *E. faecium* ( $7.5 \times 10^8$  cfu/ml) at different inclusion rates were investigated using in vitro rumen fermentation. The following treatments were non addition, 0.1%, 0.5% and 1.0% of *E. faecium* and, hereafter referred to as control, treatments 1, 2, and 3, respectively. Increased in total gas production and ammonia nitrogen concentration over time were found in control and all treatments while the opposite was observed in pH. Addition of *E. faecium* had significant effect on total gas production after 48h of incubation having the highest ( $P < 0.05$ ) in T2 followed by T4, T3 and control with 93.33, 91.75, 89.50 and 82.20 ml, respectively. pH was not affected by addition of *E. faecium* and lactate was only detected at 0h and was not detected after 12, 24 and 48h of incubation. Propionate was found highest ( $P < 0.05$ ) in T1 after 12h of incubation with 14.15 mM/L. Higher ( $P < 0.05$ ) concentrations in treated than control were found in acetate after 12h, propionate after 48h, butyrate after 12, 24 and 48h and total volatile fatty acids (TVFA) after 12 and 48h of incubations. Pyrosequencing, methane concentration, and methanogen quantity are still on going. So far, addition of *E. faecium* did not affect the pH but increased the propionate and TVFA concentrations.

**Key Words:** *Enterococcus faecium*, in vitro, pyrosequencing

**T212 Effects of replacing soybean meal with canola meal or treated canola meal on performance of lactating dairy cows.** E. M. Paula<sup>1</sup>, M. A. C. Danes<sup>2</sup>, N. E. Lobos<sup>2</sup>, G. I. Zanton<sup>3</sup>, G. A. Broderick<sup>4</sup>, and A. Faciola<sup>1</sup>, <sup>1</sup>University of Nevada, Reno, NV, <sup>2</sup>University of Wisconsin, Madison, WI, <sup>3</sup>USDA-Agricultural Research Service, US Dairy Forage Research Center, Madison, WI, <sup>4</sup>Broderick Nutrition & Research, Madison, WI.

Canola meal (CM) has been shown to be a more effective CP source than soybean meal (SBM) for lactating dairy cows. Treating CM may increase its RUP fraction and improve the amount of absorbable AA. The objective of this study was to evaluate the effects of feeding treated CM (TCM) on performance of dairy cows. Forty-five Holstein cows were blocked by parity and DIM and used in a study of randomized complete block design. Cows were fed a control diet for a 2-week covariate period and then switched to the experimental diets for a 12-week study. Treatments differed only in CP source and were SBM, CM, and TCM. All diets contained (DM basis) 30% alfalfa silage, 30% corn silage, 4% soy hulls, 2.4% mineral-vitamin premix and 16% CP. SBM diets contained 25% high moisture corn (HMC) and 8.6% SBM; CM diets contained 22% HMC and 11.4% CM. Data were analyzed using the mixed procedure of SAS. Orthogonal contrasts were used to compare effects of different protein sources (SBM vs. CM + TCM) and (CM vs. TCM). There were no statistical differences in DMI and milk yield among diets; however, CM diets had numerically higher milk yields (Table 1). Compared with SBM both CM diets decreased MUN concentration ( $P = 0.02$ ). There were no significant differences in milk composition and yields among treatments. Results from this study suggest that CM diets may improve N utilization, as indicated by reduced MUN. Although the large numeric

differences in milk production were not statistically significant, these differences may be important from a practical standpoint.

**Table 1 (Abstr. T212).** Dry matter intake, milk production, and composition results

Item	Treatment			SEM	Contrast probability	
	SBM	CM	TCM		SBM vs. CM + TCM	CM vs. TCM
DMI, kg/d	25.9	26.5	27.1	0.90	0.44	0.70
Milk, kg/d	39.4	40.3	41.9	1.34	0.32	0.46
FCM, kg/d	41.2	45.2	43.7	1.96	0.16	0.66
FCM/DMI	1.64	1.67	1.70	0.06	0.53	0.79
Fat, %	4.16	4.18	4.14	0.15	0.99	0.93
Fat, kg/d	1.55	1.60	1.67	0.08	0.33	0.63
Protein, %	3.18	3.12	3.22	0.06	0.95	0.34
Protein, kg/d	1.18	1.21	1.30	0.05	0.26	0.33
Lactose, %	4.83	4.91	4.89	0.05	0.26	0.80
Lactose, kg/d	1.77	1.85	2.00	0.10	0.20	0.41
SNF, %	8.91	8.95	8.95	0.07	0.72	0.99
SNF, kg/d	3.32	3.43	3.68	0.17	0.25	0.41
MUN, mg/dL	14.0	12.9	12.5	0.40	0.02	0.49

**Key Words:** canola meal, MUN, soybean meal

**T213 Transport of a fluorescent analog of glucose (2-NBDG) by rumen bacteria.** Junyi Tao\*, Rebecca K. Diaz, and Timothy J. Hackmann, *University of Florida, Gainesville, FL.*

Our objective was to determine if cultured strains of rumen bacteria would transport a fluorescent analog of glucose (2-NBDG) with the same specificity and kinetics as glucose. Our rationale was that 2-NBDG could be used to identify uncultured, glucose-utilizing bacteria if it were transported similarly to glucose. Pure cultures of bacteria were harvested in the mid-to-late log phase, washed, and dosed with 2-NBDG or radio-labeled sugar (0 to 100  $\mu$ M). Transport was halted by adding  $-5^{\circ}$ C stop buffer and filtering through a membrane. The membrane was taken for fluorometry or liquid scintillation counting. Transport of 2-NBDG could be detected within 2 s of 2-NBDG addition for *Streptococcus bovis* and *Selenomonas ruminantium* (2 strains each), but it was not detected at any time for 6 other glucose-fermenting species. Genomes of *S. bovis* and *S. ruminantium* strains were found to possess genes for the mannose phosphotransferase system, whereas the other species had genes for other glucose transporters. For *S. bovis* JB1, the Michaelis constant ( $K_m$ ) for 2-NBDG transport was 10.6-fold lower than that for [ $^{14}$ C]-glucose transport ( $P = 0.006$ ). The maximum velocity ( $V_{max}$ ) was 2.9-fold lower than that for [ $^{14}$ C]-glucose, but this difference was not significant ( $P = 0.100$ ). In another set of experiments, transport of 2-NBDG at a single concentration (100  $\mu$ M) was compared with that of [ $^{14}$ C]-glucose, [ $^3$ H]-mannose, and [ $^{14}$ C]-deoxy-2-glucose. For *S. bovis* JB1, transport of 2-NBDG was 3.2-fold lower than that of [ $^{14}$ C]-glucose ( $P = 0.002$ ) but similar to that for [ $^3$ H]-mannose ( $P = 0.992$ ) and [ $^{14}$ C]-deoxyglucose ( $P = 0.955$ ). 2-NBDG could identify uncultured, glucose-utilizing bacteria, but only those with a mannose phosphotransferase system (not other glucose transporters). Its transport may more closely reflect that of mannose and deoxy-2-glucose than glucose.

**Key Words:** rumen bacteria, 2-NBDG, transport



**T214 Characteristics of dairy cows with a greater or lower risk of subacute rumen acidosis: Volatile fatty acid absorption, rumen digestion kinetics and consistency of the risk.** Xiaosheng Gao\* and Masahito Oba, *University of Alberta, Edmonton, AB, Canada.*

The objectives of this study were to examine if lactating dairy cows with a greater or lower risk of subacute ruminal acidosis (SARA) have differences in volatile fatty acid (VFA) absorption rate, rumen digesta passage rate, and in situ rumen digestion, and determine if cows identified to have a greater risk of SARA in mid-lactation consistently have lower rumen pH in late-lactation than cows with a lower risk of SARA. Fourteen ruminally-cannulated dairy cows (DIM =  $119 \pm 47.2$ ; BW =  $640 \pm 47.9$  kg) were fed a high-grain diet consisting of 30% forage to induce SARA. Eight cows with the lowest acidosis index (area below pH 5.8 normalized for DMI;  $0.10 \pm 0.16$  pH·min/kg) and 5 with the highest acidosis index ( $3.72 \pm 0.19$  pH·min/kg) were classified as animals with lower (LOW) and higher (HIGH) risk of SARA, respectively. All response variables were evaluated for the group effect using the PROC TTEST procedure of SAS (version 9.2, SAS Institute Inc., Cary, NC). Minimum ( $5.75$  vs.  $5.33$ ;  $P < 0.01$ ) and mean ruminal pH ( $6.33$  vs.  $5.98$ ;  $P < 0.01$ ) was higher for LOW compared with HIGH animals. However, there were no differences in VFA absorption rate, rumen digesta passage rate, and in situ ruminal digestibility of starch and NDF between HIGH and LOW cows. Nine of these 14 animals (4 HIGH and 5 LOW) were fed the same high-grain diet at a later stage of lactation (DIM =  $243 \pm 19.6$ ) for 21 d. Similar to the mid-lactation, minimum ( $5.73$  vs.  $5.32$ ;  $P = 0.02$ ) and mean ruminal pH ( $6.32$  vs.  $6.07$ ;  $P = 0.01$ ) was higher for LOW compared with HIGH animals. These results suggested that variable risk of SARA among lactating dairy cows fed a high grain diet cannot be attributed to the differences in VFA absorption rate, rumen digesta passage rate, and ruminal digestibility, and that stage of lactation does not affect the risk category of animals in developing SARA.

**Key Words:** acidosis, VFA absorption, rumen digestion kinetics

**T215 Systems nutrition in dairy cattle: Integrating hepatic metabolomics and transcriptomics in late pregnancy to better understand postpartal ketosis.** Khuram Shahzad\*<sup>1</sup>, Johan Osorio<sup>2</sup>, Daniel Luchini<sup>3</sup>, and Juan J. Loo<sup>1</sup>, <sup>1</sup>*University of Illinois, Urbana, IL*, <sup>2</sup>*Oregon State University, Corvallis, OR*, <sup>3</sup>*Adisseo NA, Alpharetta, GA.*

‘Omics’ and bioinformatics were used to identify unique signatures characterizing liver of cows with postpartal ketosis relative to healthy cows fed rumen-protected methionine during late-pregnancy. Transcriptomics and metabolomics data were generated from liver tissue ( $n = 8$ /group, d -10 relative to parturition) of cows overfed a higher-energy diet during the dry period and classified as follows based on postpartal health: healthy (OVE), ketosis (K), or OVE plus Smartamine M (SM) or MetaSmart (MS). Data integration was via Ingenuity Pathways Analysis. Network construction included transcription regulators (TR) within the transcriptomics database and metabolites obtained through GC/MS-LC/MS. By comparing the different groups we obtained 21, 6, 10, 3, 11 and 15 transcription regulators (TR) out of 2908, 832, 1261, 922, 1573 and 1033, respectively, differentially expressed genes from K vs. OVE, K vs. SM, K vs. MS, SM vs. OVE, MS vs. OVE and SM vs. MS. Out of 313 known biochemical compounds, we detected 25, 34, 33, 20, 21, and 48 affected metabolites in the respective comparisons. As an example, using the TR (*HIF1A*, *HIF3A*, *SIRT1*, *HDAC4*) along with affected metabolites (cholic acid, D-erythro-dihydrosphingosine, lactic acid, malic acid, xylitol) in K vs. OVE the bioinformatics analyses revealed alterations in pathways related to tissue growth, and glucose and lipid metabolism. In regards to the methionine-supplemented groups,

using the TR (*HDAC2*, *SOX10*, *STAT1*) along with affected metabolites (arginine, inosine) in SM vs. OVE the bioinformatics analyses revealed alterations in pathways related to regulation of liver regeneration and metabolism. Unique patterns also were detected between SM and MS, analysis of the TR (*CREBBP*, *GATA2*, *NFKB2*, *STAT1*) along with affected metabolites (arachidonic acid, arginine, chenodeoxycholic acid, docosahexaenoic acid, oleic acid) revealed alterations in pathways involved in cell signaling, immune response, and cholesterol synthesis. Results indicate that ‘omics’ data integration could be helpful in better understanding the link between nutrition and incidence of disorders after calving.

**Key Words:** systems biology, ketosis, network reconstruction

**T216 Laboratory validation of a prototype cow-side instrument for the measurement of blood ionized calcium concentrations in dairy cattle.** Rafael C. Neves\*, Tracy Stokol, and Jessica A. A. McArt, *Department of Population Medicine and Diagnostic Sciences, Cornell University, Ithaca, NY.*

There is currently no efficient and inexpensive method for field measurement of blood calcium concentrations. Ionized calcium (iCa) is the homeostatic form of the mineral and is thought to have greater biological relevance over that of total calcium. The objective of this study was to evaluate the linearity and precision of a prototype cow-side instrument (Horiba, Japan) for measuring blood iCa concentrations. Blood (300 mL) was collected from the right jugular vein of a multiparous dairy cow (4 d-in-milk) into lithium heparin tubes immediately before (T0) and 5 min after (T5) intravenous administration of 500 mL of 23% calcium borogluconate. The iCa concentrations were determined using a blood-gas analyzer (ABL-800 FLEX, Radiometer) as a gold-standard. The T0 sample was diluted using 0.9% saline to create a sample with low iCa (reference interval = 1.10 to 1.35  $\mu\text{mol/L}$ ). The diluted T0 sample was then mixed with the T5 sample in different ratios (100/0, 75/25, 50/50, 25/75, 0/100) to obtain 5 levels of iCa concentrations (0.69, 1.0, 1.28, 1.58, and 1.82  $\mu\text{mol/L}$ ). Each mixture was then analyzed in triplicate using 3 different prototypes under one-point (1P) and 2-point (2P) calibration with the means compared with results from the blood-gas analyzer. Cumulative sum tests for linearity from Passing and Bablok regressions showed no deviation from linearity for the combined results of all 3 prototypes under 1P vs. the gold-standard ( $P = 0.19$ ) and under 2P vs. the gold-standard ( $P = 0.19$ ). Instrument precision (coefficient of variation; CV) was determined by 10 repeat measurements of the diluted T0 sample, T0, and T5 samples under 1P and 2P calibrations. The CV ranged from 1.3 to 5% for the 3 prototypes. Laboratory results indicate good accuracy and precision for a cow-side instrument at the tested iCa concentrations. Investigation of the instrument under field conditions is warranted.

**Key Words:** ionized calcium, cow-side instrument, dairy cattle

**T217 Validation of a hand-held meter for measuring  $\beta$ -hydroxybutyrate in plasma and serum of dairy cows.** Arnulfo Pineda\* and Felipe C. Cardoso, *University of Illinois, Urbana, IL.*

The aim of this study was to compare serum (sBHBA) and plasma (pBHBA) BHBA concentrations analyzed using either a laboratory method (Randox Laboratories Ltd., UK, Cat # RB1007 as “gold standard”; LM) or a hand-held meter (PX; Precision Xtra). Results from 187 samples taken from Holstein cows from 11 d before to 5 d after parturition were used for the analysis. Statistical analysis was performed using the MIXED, REG, and LOGISTIC procedures of SAS (v9.4). A

linear mixed model with repeated measures was performed for LM and PX. A regression analysis was completed to estimate the relationship between the 2 methods. Cross-validation by randomly splitting the data in model building and validation sets was performed to estimate and validate the equation that predicted the LM results using PX. Receiver operation characteristic (ROC) curves were made to estimate the sensitivity and specificity of PX at different threshold levels. The PX yielded higher ( $P < 0.01$ ) pBHBA and sBHBA than LM, 1.45 vs. 0.95 and 1.63 vs. 1.00 mmol/dL, respectively. Adjusted  $R^2$  between both methods for pBHBA was 97.1% and 97.8% for sBHBA. The equation from the model building data set that predicted pBHBA results using PX was  $y = -0.05353 + 0.67842x$ , while the equation that predicted sBHBA results was  $y = 0.05542 + 0.56862x$ , where  $y$  = predicted LM BHBA and  $x$  = PX BHBA. Mean square error (MSE) yielded by the model building data set for pBHBA and sBHBA were 0.006 and 0.005, respectively. Mean squared predictor error (MSPE) from the validation data set was 0.007 for pBHBA and 0.006 for sBHBA. Highest sensitivity and specificity for PX was achieved when threshold was set to 1.8 for pBHBA and 2.1 mmol/dL for sBHBA. The area under ROC curve (AUC) was 0.97 for pBHBA and 0.96 for sBHBA. In conclusion, the small difference between MSE and MSPE suggest that PX can be used to predict pBHBA and sBHBA by using the aforementioned equations. However, the threshold for ketosis requests to be higher than the commonly used in the field (1.2 mmol/dL) when using PX. The AUC close to 1 suggests greater ability of PX in predicting ketosis under the indicated thresholds.

**Key Words:** BHBA, dairy cow, Precision Xtra

**T218 Effects of feeding calcium hydroxide-treated corn stover on milk production and milk composition in lactating Holstein cows.** Brittany A. Casperson<sup>\*1</sup>, Aimee E. Wert-Lutz<sup>2</sup>, and Shawn S. Donkin<sup>1</sup>, <sup>1</sup>Purdue University, West Lafayette, IN, <sup>2</sup>ADM Alliance Nutrition, Quincy, IL.

Chemical treatment may improve the nutritional value of corn stalk residues and their potential use as an alternative forage source for lactating dairy cows. The objectives of this study were to determine the effect of prestorage hydration and treatment with 6.6% Ca(OH)<sub>2</sub> on feeding value of corn stalks as an alternative forage source on milk production, milk composition, and DMI. Mid-lactation multiparous Holstein cows ( $n = 30$ ) were stratified by parity and milk production and randomly assigned to 1 of 3 diets. Corn stalks were chopped, hydrated, and treated with 6.6% Ca(OH)<sub>2</sub> (DM basis) and stored in Ag-bag silos. Treated corn stover was fed in a TMR at 0, 15, and 30% of the diet DM. Treated corn stover replaced either alfalfa haylage (15% stover diet) or replaced alfalfa haylage and an additional portion of corn silage (30% stover diet). Cows were individually fed in tie stalls for 10 weeks. Milk production was not altered by treatment ( $P = 0.80$ ). Compared with 0% stover diet, DMI was reduced when the 15% stover diet was fed ( $25.9$  vs.  $22.7 \pm 0.88$  kg/d,  $P < 0.05$ ) and tended to be reduced ( $25.9$  vs.  $23.1 \pm 0.88$  kg/d,  $P = 0.08$ ) when cows were fed the 30% stover diet. Milk production per unit DMI (kg/kg) tended to increase for cows fed 15% stover diet compared with the 0% stover diet ( $1.41$  vs.  $1.62 \pm 0.07$ ,  $P = 0.08$ ) but was not different between cows fed the 0% and 30% stover diets ( $1.41$  vs.  $1.50 \pm 0.07$ ,  $P = 0.62$ ). Milk composition, energy corrected milk production, and energy corrected milk produced per unit of DMI (kg/kg) was not different ( $P > 0.05$ ) among treatments for the 10-week feeding period. Cows fed the 15% and 30% diets had stable DMI and daily milk production over the 10-week treatment period but DMI for cows fed 0% stover increased slightly (time  $\times$  treatment effect,  $P < 0.05$ ). These data indicate that corn stover processed through prestorage hydration with Ca(OH)<sub>2</sub> results in an alternative forage source

for lactating dairy cows that when fed to mid-lactation cows tends to improve the efficiency of conversion of feed to milk without altering milk production or milk composition.

**Key Words:** corn stover, alternative forage, milk fat

**T219 Increased stocking density at the feed bunk may affect the welfare and productivity of dairy cows and growth of their heifer calves.** Jessica A. Pempek<sup>\*</sup>, Maurice L. Eastridge, Kathryn L. Proudfoot, Gregory G. Habing, Lohendy M. Muñoz Vargas, and Danielle N. Coleman, *The Ohio State University, Columbus, OH.*

Overstocking the feed bunk during late-gestation has behavioral and physiological consequences, both of which negatively affect the welfare of the cow. However, it is unknown as to whether overstocking the dam may also affect the growth of the developing calf. The aim of the present study was to investigate the effect of increased stocking density at the feed bunk during different stages of the dry period on metabolic health and productivity of dairy cows, as well as the postnatal growth of their calves. One hundred twenty nonlactating Holstein dairy cows were blocked and assigned to 1 of 4 treatment groups with different stocking densities at the feed bunk (Overstocked (OS): 0.88 headlocks/cow; Understocked (US): 1.17 headlocks/cow). The 4 treatments were: OS from 60 to 1 d (OS), OS from 60 to 26 d and US from 25 to 1 d (OS-US), US from 60 to 26 d and OS from 25 to 1 d (US-OS), and US from 60 to 1 d (US) before calving. The heifer calves' treatment reflected the treatment assignment of their dam ( $n = 13, 18, 16,$  and  $11$ , respectively). Blood samples were obtained from the cow at  $-60, -30, -14, -7,$  and  $+7$  d relative to calving to determine concentrations of nonesterified fatty acids (NEFA). Colostrum quantity and quality were recorded from the cow's first milking. Calf body weight (BW) was measured once per wk through weaning at 5 wk of age. Data were analyzed using repeated measures and the Mixed model procedure of SAS (2012). NEFA concentrations were similar among treatment groups across periods (0.36, 0.42, 0.37, and 0.42 mEq/L, respectively;  $P > 0.05$ ). There was a tendency for colostrum quantity to differ among treatments (7.52, 5.70, 7.03, and 8.76 kg, respectively;  $P = 0.11$ ), primarily due to the difference between OS-US and US. However, there was no difference in colostrum quality (weighted Brix value of 7.24%). Using initial BW as a covariate, calf BW was similar among treatment groups (53.7, 51.9, 52.2, and 54.1 kg, respectively;  $P > 0.05$ ), and there was no treatment by week interaction. In conclusion, moderate increases in stocking density at the feed bunk did not appear to compromise the metabolic status of dairy cows or the growth of their heifer calves.

**Key Words:** stocking density, feed bunk, dry period

**T220 Unraveling the mechanisms that regulate activation of  $\beta$ -defensin antimicrobial peptide responses in cattle.** Mercedes F. Kweh<sup>\*</sup>, Kathryn E. Merriman, and Corwin D. Nelson, *University of Florida, Gainesville, FL.*

Bovine  $\beta$ -defensin peptides exhibit antimicrobial properties against bacterial pathogens, and expression of several  $\beta$ -defensin genes has been reported in the udder during mastitis. In cattle, toll-like receptor (TLR) and vitamin D signaling pathways induce expression of multiple  $\beta$ -defensin genes; however,  $\beta$ -defensin expression in mammary epithelial cells is only activated via the TLR pathway and is much lower compared with neutrophils and macrophages. The  $\beta$ -defensin gene cluster on bovine chromosome 27 contains multiple potential DNA methylation sites, suggesting the hypothesis that epigenetic mechanisms contribute to induction of the  $\beta$ -defensin response in cattle. The objective of this

study was to investigate the effects of DNA methylation and histone deacetylation inhibitors, 5-aza-2'-deoxycytidine (5-Aza) and Trichostatin A (TSA), respectively, on TLR and 1,25-dihydroxyvitamin D<sub>3</sub> (1,25D)-induced expression of  $\beta$ -defensins in primary bovine mammary epithelial cells (bMEC). Primary bMEC cultures obtained via mammary biopsy were treated with 5-Aza (1  $\mu$ M, 72 h), TSA (80 nM, 16 h) or control in combination with lipopolysaccharide (LPS; 100 ng/mL; 16 h) and 1,25D (10 nM, 16 h). Expression of  $\beta$ -defensin 3 (*BNBD3*), *BNBD4*, *BNBD7*, *BNBD10*, and lingual antimicrobial peptide (*LAP*) genes was determined using qPCR. The 5-Aza treatment resulted in a > 10-fold increased expression of each gene ( $P < 0.001$ ). There was also an interaction between LPS and 5-Aza for expression of *BNBD3*, *BNBD10*, and *LAP* genes ( $P < 0.001$ ). The TSA treatment also increased expression of the *BNBD7*, *BNBD10*, and *LAP* genes approximately 5-fold compared with control ( $P < 0.01$ ), and there was TSA by LPS interaction for each of the genes ( $P < 0.05$ ). In contrast, there were no interactions between 1,25D and 5-Aza or TSA for any of the  $\beta$ -defensin genes. These data suggest that DNA methylation and histone acetylation both contribute to  $\beta$ -defensin expression, and that epigenetic mechanisms may affect TLR activation of the  $\beta$ -defensin antimicrobial response of mammary epithelial cells.

**Key Words:**  $\beta$ -defensins, mammary immunity, vitamin D

**T221 Ellipsoid equation improves accuracy and efficiency of estimating protozoal volume.** Benjamin A. Wenner\*, Brooklyn K. Wagner, and Jeffrey L. Firkins, *Department of Animal Sciences, The Ohio State University, Columbus, OH.*

Previous observations of protozoa in cultures treated with essential oils or ionophores indicated possible cell shrinkage due to deleterious effects on cell function. Cell volume reduction by formaldehyde preservation combined with visually flattened or tapered morphology of rumen protozoa limited our ability to detect volume differences using common cylindrical derivations for protozoa. The advent of affordable, high definition imaging equipment enables recording of live protozoa from cultures treated with various additives that potentially shrink cells. We hypothesized that using still frames from video of protozoa swimming would improve accuracy of volume predictions by optimizing an approach to measure one maximal longitudinal measurement and both minimum and maximum diameter measurements perpendicular to the longitudinal axis, thus yielding a 3-dimensional estimation of protozoal volume. An ellipsoid formula ( $E, \frac{4}{3}\pi abc$ ) was compared with previously published estimations using cylindrical ( $C, L\pi(\frac{W}{2})^2$ ) or species coefficient ( $SP, XLW^2$ ) calculations. Testing this method on inanimate objects shaped similarly to protozoa demonstrated that the ellipsoid is more accurate in predicting volume as measured by displacement. True displacement was 11.8 mL for 10 large particles, and estimated volumes were 12.7 to 27.1, 7.7 to 16.6, and 12.3 mL for C, SP, and E, respectively. For smaller particles with more surface area, true displacement was 4.5 mL, and estimated volumes were 5.6 to 13.1, 3.2 to 7.5, and 5.6 for C, SP, and E, respectively. Rumen fluid sampled from 2 lactating Jersey cows was flocculated and wet-mounted on a microscope fitted with an

HD (1080p) camera. Mean entodiniomorphid (ENTO) volumes were 109,665 ( $\pm 55,912$ ), 99,145 ( $\pm 51,704$ ), and 79,830 ( $\pm 39,859$ )  $\mu\text{m}^3$  for C, SP and E, respectively. Mean epidinium (EPI) volumes were  $1.74 \times 10^5$ ,  $2.47 \times 10^5$ , and  $1.45 \times 10^5$  for C, SP, and E, respectively. Regression of SP on E demonstrated that SP more likely overestimates volume for ENTO ( $Y = 1.187X + 0.8567$ ) or EPI ( $Y = 1.467X + 38528$ ) than for teardrop-shaped isotrichids ( $Y = 0.833X + 346,973$ ). This ellipsoid method offers potential to advance prediction of treatment effects on protozoal viability and volume.

**Key Words:** protozoa, imaging, rumen

**T222 Milk yield at dry-off and other factors affecting risk of intramammary infections at calving.** Paige N. Gott\*<sup>1</sup>, Päivi J. Rajala-Schultz<sup>1</sup>, Gustavo M. Schuenemann<sup>1</sup>, and Joseph S. Hogan<sup>2</sup>, <sup>1</sup>The Ohio State University, Columbus, OH, <sup>2</sup>The Ohio State University, Wooster, OH.

Increased milk yield at dry-off has been associated with increased risk of intramammary infections (IMI) at calving. Abrupt cessation of milking is widely practiced although gradual cessation has been shown to significantly decrease milk yield. The objective of this study was to evaluate the effect of milk cessation method (MilkCess) and Dairy Herd Improvement Association final test day (FTD) milk yield on IMI at calving. Cows in 8 Ohio dairy herds were enrolled 7 to 14 d before expected dry-off and randomly assigned either to ABRUPT or to GRADUAL cessation of milking. The GRADUAL group was milked once daily for the final week of lactation while the ABRUPT group kept the farm's normal milking schedule. Aseptic quarter foremilk samples were collected at enrollment, at the final milking (DRY), and within 7 d of calving (CALV). Samples were cultured following NMC guidelines. Isolation of  $\geq 10$  colonies of similar morphology was considered an IMI while isolation of  $\geq 3$  morphologies was called contamination. Coagulase-negative staphylococci and *Corynebacterium* spp. were considered minor (MIN) pathogens. Other species were considered major (MAJ) pathogens. The association between quarter IMI status at CALV and MilkCess and FTD milk yield was evaluated using PROC GLIMMIX in SAS. Separate models were run with MAJ and MIN IMI at CALV as the outcomes. Data from 392 cows were analyzed. MilkCess was not significant, but was forced into each model. For every 4.5-kg increase in FTD milk yield, the odds of IMI at CALV increased (MAJ = 39%,  $P = 0.0007$ ; MIN = 18%,  $P = 0.0498$ ). The odds of MAJ IMI at CALV for quarters with MAJ IMI at DRY were 17.2 times the odds of quarters uninfected at DRY ( $P < 0.0001$ ). The odds of MIN IMI at CALV for quarters with MIN IMI at DRY were 1.9 times the odds of quarters uninfected at DRY ( $P = 0.0316$ ). For every 7d increase in DIM at dry-off above 305 DIM, the odds of MIN IMI at CALV increased 3.1% ( $P = 0.0328$ ). On average, dry periods lasted 58 d and for every 7-d increase in days dry above 58 d, the odds of MIN IMI increased 28% ( $P = 0.0125$ ). In conclusion, decreasing milk yield before dry-off could help improve udder health at calving.

**Key Words:** milk yield, IMI, dry-off



# Growth and Development I

**T223 Exogenous estradiol decreases relative endometrial gene expression for *ERα* and *ERβ* in pre-pubertal dairy heifers.** Chelsee T.L. Holloway\*<sup>1</sup>, Adam J. Geiger<sup>1</sup>, Jennifer Malkus<sup>1</sup>, Alan D. Ealy<sup>2</sup>, Rebecca R. Cockrum<sup>1</sup>, and Robert M. Akers<sup>1</sup>, <sup>1</sup>Virginia Polytechnic Institute and State University, Department of Dairy Science, Blacksburg, VA, <sup>2</sup>Virginia Polytechnic Institute and State University, Department of Animal and Poultry Sciences, Blacksburg, VA.

The objectives of this study were to determine (1) the effect of early nutrition on reproductive development and (2) response of reproductive tissues to estradiol ( $E_2$ ) in pre-pubertal heifers. Holstein heifer calves ( $n = 23$ ; 7 d of age) were fed either a restricted milk replacer diet (20% CP, 20% fat, 454 g/d) or accelerated diet (28% CP, 25% fat 1,333 g/d) for 8 wks. After the initial 8 wks, heifers were partitioned into a  $2 \times 2$  factorial design and treated for an additional 2 wks accordingly: restricted diet + placebo implant (CON;  $n = 6$ ), accelerated diet + placebo (ACC;  $n = 5$ ), restricted diet +  $E_2$  implant (CONEST;  $n = 6$ ) and accelerated diet +  $E_2$  (ACCEST;  $n = 6$ ). Estrogen receptor  $\alpha$  (*ERα*) and  $\beta$  (*ERβ*) gene expression was determined from RNA isolated from harvested endometrial, ovarian, and hypothalamic tissues using Real-Time qPCR. Fold changes were analyzed using the GLM procedure in SAS with treatment used as a fixed effect and initial body weight used as a covariate. The main effect of diet did not differ ( $P \geq 0.127$ ) for any of the tissues. However,  $E_2$  treatment differed for the endometrium ( $P \leq 0.001$ ), but not for the ovary or hypothalamus ( $P \geq 0.528$ ). In the endometrium, *ERα* was upregulated ( $P \leq 0.024$ ) in ACC heifers ( $8.44 \pm 1.21$ ) as compared with ACCEST ( $2.69 \pm 1.43$ ), and CONEST heifers ( $2.63 \pm 1.22$ ). Gene *ERβ* was also upregulated more ( $P \leq 0.052$ ) in ACC heifers ( $4.16 \pm 0.52$ ) than ACCEST ( $2.11 \pm 0.61$ ) or CONEST heifers ( $1.46 \pm 0.52$ ) in the endometrium. Additionally, *ERβ* was upregulated ( $P < 0.001$ ) in CON heifers ( $4.56 \pm 0.52$ ) as compared with CONEST heifers, and tended ( $P = 0.071$ ) to be upregulated in ACCEST heifers. Overall, results indicated that diet alone did not affect expression of *ERα* or *ERβ* in reproductive tissues of pre-pubertal heifers. However, treatment with  $E_2$  in pre-pubertal heifers affected relative gene expression of key genes within the hypothalamic-gonadal pathway. Determining the effect of estrogen on the development of reproductive tissues is key to understanding signaling pathways that activate the onset of puberty.

**Key Words:** estradiol, gene expression, nutrition

**T224 Plane of nutrition affects muscle fiber hypertrophy and satellite cell activity in neonatal bull calves.** Jennifer S. Bradley\*, Meghan E. MacGhee, Sarah R. McCoski, Amanda M. Reeg, Alan D. Ealy, and Sally E. Johnson, Virginia Polytechnic Institute and State University, Blacksburg, VA.

Muscle growth in newborn calves is a product of protein accretion concomitant with addition of nuclei to the muscle fiber. The objective of the experiment was to examine the effect of diet on live animal performance, muscle fiber hypertrophy and satellite cell activity in young calves. Neonatal Holstein bull calves were fed either a high nutritional plane (HNP; 27% CP, 10% fat) or low nutritional plane (LNP; 20% CP, 20% fat) milk replacer. Daily intake of GE for the LNP group was 50% of HNP, but exceeded the NRC requirement for NEm and NEg. Groups of HNP ( $n = 4$ ) and LNP ( $n = 4$ ) were slaughtered at 2, 4 and 8 wk. The LM was removed for fiber morphometric evaluation and satellite cell isolations. Data were analyzed by ANOVA using PROC GLM for the main effects of diet and time and their interaction. Calves

fed HNP gained more ( $P < 0.05$ ) BW at 2, 4 and 8 wks than LNP with the largest difference at 8 wks (HNP = 749 g/d, LNP = 134 g/d, pooled SEM = 0.05). Fiber cross-sectional area was larger ( $P < 0.05$ ) in HNP than LNP at 8 wk ( $1502 \pm 242 \mu\text{m}^2$  vs.  $356 \pm 25 \mu\text{m}^2$ ). The numbers of Pax7 immunopositive satellite cells per fiber did not differ ( $P > 0.05$ ) between the groups with time or diet. Satellite cell isolates were cultured in growth media and pulsed for 2 h with EdU, a thymidine analog, to measure proliferation rate. Isolates from HNP at 2 wks contained a greater ( $P < 0.05$ ) percentage of proliferating cells during log phase growth than LNP at equal cell densities (51.7 and 35.2%, respectively, SEM = 2.1). At 4 wks, the percentage of proliferating HNP satellite cells was less ( $P < 0.05$ ) than that of LNP isolates (21.5 vs. 39.1%, SEM = 1.6) and tended ( $P = 0.10$ ) to remain lower at 8 wk. No differences were observed in the numbers of myofibers formed from HNP or LNP satellite cell isolates. These results demonstrate that calves fed a higher plane of nutrition gain more weight and experience greater muscle fiber hypertrophy than calves fed at a lower level. The larger HNP fiber may be attributed to greater satellite cell proliferation and their subsequent fusion into the adjacent fiber.

**Key Words:** muscle fiber, hypertrophy, Pax7

**T225 Effects of heat-treatment of colostrum on absorption and gut development in neonatal bull calves during the first 12 hours of life.** Coral Kent-Dennis<sup>1</sup> and Thomas McFadden\*<sup>2</sup>, <sup>1</sup>University of Alberta, Edmonton, AB, Canada, <sup>2</sup>University of Missouri, Columbia, MO.

Heat-treatment of colostrum reduces transmission of pathogens to neonatal calves but may alter the bioactivity of colostrum. The objective was to determine the effects of heat-treating colostrum on absorptive function and gut development in neonatal calves. First-milking colostrum was collected in batches and stored at  $-20^\circ\text{C}$ . Batches were thawed, pooled, mixed thoroughly, and divided into equal volumes. Half was re-frozen in 1L aliquots and the other half was heat-treated at  $60^\circ\text{C}$  for 60 min then re-frozen in 1L aliquots. Holstein bull calves were enrolled in the study as they were born and received 2L of either heat-treated ( $n = 9$ ) or unheated ( $n = 8$ ) colostrum via esophageal tube feeder within 1.5h of birth. Blood samples were taken at 0 (pre-colostrum), 3, 6, 9 and 12h after birth for analysis of IgG and  $\beta$ -lactoglobulin (BLG) concentrations. Six calves from each treatment group were euthanized at 12h after birth and ileal tissue was collected for analysis of villus height, crypt depth, and goblet cell count. Statistical analysis was conducted using PROC MIXED of SAS with batch as a random effect and repeated measures where appropriate. Colostral IgG concentration did not differ between heated and unheated pools. Plasma IgG concentration tended to be higher in calves fed unheated versus heated colostrum at 9 h and the difference was significant at 12h ( $15.8 \pm 1.4$  versus  $12.9 \pm 1.4$  mg/mL;  $P = 0.02$ ). Plasma BLG concentrations were significantly higher in calves fed unheated versus heat-treated colostrum at 6 h ( $324.3 \pm 52$  versus  $239.9 \pm 51.0$  mg/mL;  $P = 0.04$ ) and 9h ( $258.2 \pm 52.0$  versus  $126.3 \pm 51.9$  mg/mL;  $P = 0.002$ ). By 12 h, there was no difference between the 2 treatment groups. These data suggest that different colostrum proteins have different rates of absorption and biological half-lives. Villus height and crypt depth did not differ significantly between treatment groups. The average number of goblet cells on ileal villi was higher in calves fed heat-treated versus unheated colostrum ( $63.4 \pm 3.3$  versus  $48.8 \pm 3.3$ ;  $P = 0.02$ ). Results indicate that heat-treatment of colostrum altered

absorption of colostrum proteins and stimulated development of mucus-producing cells but did not affect mucosal growth.

**Key Words:** colostrum, pasteurization, calf health

**T226 Direct-fed microbial efficacy and milk plan: Effect on heifer growth in an automated feeding system.** Melissa C. Cornett\* and Amy L. Stanton, *University of Wisconsin-Madison, Madison, WI.*

Automated feeding systems (AF) offer flexible nutrition programs for pre-weaned dairy calves. The technology offers the option of changing daily milk allotment as calves grow and providing feed additives, such as direct-fed microbial (DFM). This study determined the effect of DFM efficacy and 2 milk plans (SLW, FST) on growth. Holstein heifers ( $n = 104$ ) from a commercial Wisconsin herd were enrolled at birth in a  $2 \times 2$  factorial design. Factor 1 was a DFM protocol versus a placebo (PLAC) from birth to weaning. Factor 2 compared 2 feeding plans (SLW and FST) offering 543L pasteurized whole milk over 53 feeder days (FD). The SLW initially offered (7L/d), and peaked milk at 28 FD (13L/d). The FST initially offered 11L/d, and peaked milk at 21 FD (15L/d). Both plans stopped milk at 53 FD. Rate of passive transfer was 86%. Calves were housed individually before AF. Ability of calf to drink milk unassisted triggered AF move. During AF, ad libitum Ampli-Calf (Land O' Lakes, MN) calf starter was provided in a bunk. No individual starter intakes were recorded. Calf weight (WT) was measured  $2 \times /wk$  from age  $3 \pm 2$  d (Mean  $\pm$  SD) until 1 wk post-weaning. The effect of treatments on WT was evaluated using PROC MIXED in SAS, controlling for initial WT, age, and study period, study month was a random effect and calf nested in treatment for repeated measure. Study period was divided into 4 times: Period 1 birth to AF (P1), Period 2 FD1 – FD28 when FST offered more milk/d than SLW (P2), Period 3 FD29-FD53 (P3), and Period 4, the 7 d post-weaning (P4). Average milk consumption was  $430.9 \pm 89.0L$  for SLW and  $378.5 \pm 78.7L$  for FST. Initial feeder enrollment age was  $7 \pm 2$  d. Milk plan, DFM, and study period interacted ( $P = 0.03$ ). In P2, FST DFM calves weighed  $3.6 \pm 1.5$  kg (LSM  $\pm$  SEM) more than FST PLAC ( $P < 0.02$ ). In P3, FST DFM calves weighed  $3.2 \pm 1.5$  kg more than FST PLAC ( $P < 0.04$ ). In P3, SLW PLAC weighed  $3.0 \pm 1.5$  kg more than FST PLAC ( $P < 0.05$ ). In P4, FST DFM tended to weigh  $2.9 \pm 1.6$  more than FST PLAC ( $P = 0.07$ ). These results suggest DFM effectively improves growth in dairy calves on a rapid increase milk plan. Calves provided peak milk at 28FD had higher growth than calves with peak milk at 21FD. Growth can be optimized by AF and DFM.

**Key Words:** calves, growth, probiotic

**T227 Anti-oxidative status and inflammatory response in neonatal calves fed quercetin with or without colostrum.** Harald M. Hammon\*<sup>1</sup>, Jeannine Gruse<sup>1</sup>, Manfred Mielenz<sup>1</sup>, and Siegfried Wolfram<sup>2</sup>, <sup>1</sup>Leibniz Institute for Farm Animal Biology (FBN), Dummerstorf, Germany, <sup>2</sup>Christian Albrechts University, Kiel, Germany.

Flavonoids such as quercetin are known to exert anti-inflammatory and anti-oxidative effects that could improve neonatal health. We hypothesized that quercetin feeding during the first week of life may improve anti-inflammatory and antioxidative status in neonatal calves and that quercetin feeding compensates for an inadequate colostrum supply. Twenty-eight new-born male Holstein calves were randomly assigned to 2 feeding groups receiving same amounts of either colostrum (C;  $n = 14$ ) or a milk-based formula (F;  $n = 13$ ) with same nutrient density as C, but no biologically active factors (e.g., growth factors), during the first 2 d of life. From d 3 to d 7, all calves were fed milk replacer at

12% of BW. From d 2 on, groups were subdivided each into a treatment group receiving  $150 \mu\text{mol}$  quercetin/(kg BW  $\times$  d) as quercetin aglycone with milk and a control group without quercetin. For characterization of anti-oxidative status, trolox equivalent antioxidative capacity (TEAC), ferric reducing ability of plasma, thiobarbituric acid reactive substances (TBARS) and prostaglandin-like substances were analyzed in blood plasma taken on d 1, 4, and 7. Liver biopsy was taken 2 h after morning meal on d 8 to determine mRNA abundances of anti-oxidative enzymes (catalase; glutathione peroxidase; superoxide dismutase) and inflammation markers (tumor necrosis factor, TNF $\alpha$ ; serum amyloid A2, SAA2; C-reactive protein, CRP) using quantitative real-time PCR. Data were evaluated by Proc Mixed (blood) or Proc GLM (mRNA) of SAS with milk and quercetin as fixed effects. TEAC and TBARS were greater in C- than in F-fed calves ( $P < 0.05$ ). Relative mRNA abundance of TNF $\alpha$  was greater in quercetin-fed groups ( $P < 0.05$ ); CRP was greater ( $P < 0.05$ ) and SAA2 tended to be greater ( $P < 0.1$ ) in F- than in C-fed calves. Absence of C feeding was associated with a reduced anti-oxidative status and increased hepatic mRNA abundance of acute phase proteins, which underlines the importance of C feeding for neonatal health. Results further indicate that quercetin supplementation barely affects the anti-inflammatory and antioxidative status in neonatal calves.

**Key Words:** calf, quercetin, antioxidative status

**T228 Effect of milk aroma flavor on starter consumption, growth, and feed conversion in female Holstein calves.** Yaqi Wu, Yanliang Bi, Shengli Li\*, Yajing Wang, and Zhijun Cao, *College of Animal Science and Technology, China Agriculture University, Beijing, China.*

Milk aroma flavor (Luctarom1866, Lucta, Spain) was added to calf starter to investigate its effects on starter consumption (SC), growth and feed conversion in dairy calves. Forty-four female Holstein calves (6 wk of age;  $56 \pm 6$  kg of BW) were randomly assigned to 2 groups, which were fed with 2 g or 0 g milk aroma flavor per kg starter. All calves consumed the same milk and starter from birth to the beginning of the experiment (wk 0–6). The experiment lasted 4 weeks (wk 7–10) and calves were provided same milk from the beginning of the experiment (wk 7) and weaned at 8 wk of age. SC was recorded daily and each calf was weighed weekly. The data were analyzed by ANOVA and LSD test at 5% and 1% probability utilizing SAS statistical program. The results indicated that SC of calves fed flavor had no significant differences in wk 7, 8, 9 and during the entire experimental period, while it increased by 0.38 kg per day ( $P < 0.05$ ) in wk 10. Compared with the calves fed starter with no flavor, the calves fed flavor gained more weight in wk 9 (0.2 kg per day ( $P < 0.05$ )), wk 10 (0.3 kg per day ( $P < 0.01$ )), and during the whole experimental period (0.1 kg per day ( $P < 0.05$ )). From wk 9 to wk 10, the ratio of weight gain to SC was 0.42 and 0.38 for calves fed with and without flavor ( $P > 0.05$ ). Overall, milk aroma flavor in starter could improve SC and average daily gain (ADG) in dairy calves and tend to encourage feed conversion.

**Key Words:** milk aroma flavor, average daily gain, weight gain:starter consumption

**T229 Caloric restriction reduces protein accretion in skeletal muscle by attenuating IGF-I signaling in young calves.** Yue Lu<sup>1</sup>, Jennifer S. Bradley<sup>1</sup>, Sarah R. McCoski<sup>1</sup>, John M. Gonzalez<sup>2</sup>, Adam J. Geiger<sup>1</sup>, R. Michael Akers<sup>1</sup>, Alan D. Ealy<sup>1</sup>, and Sally E. Johnson\*<sup>1</sup>, <sup>1</sup>Virginia Polytechnic Institute and State University, Blacksburg, VA, <sup>2</sup>Kansas State University, Manhattan, KS.

Caloric restriction decreases skeletal muscle mass in mammals, principally due to a reduction in fiber size. This study examined the consequences of reduced caloric intake on the metabolic properties of skeletal muscles in young Holstein heifer calves. Newborn heifers were assigned to a control (CON, n = 5; 28% CP, 25% fat) or caloric restricted (CR, n = 5; 20% CP, 20% fat) milk replacer diet from birth to 8 wks of age. Grain (25% CP, 4% fat) was pair-fed beginning at wk 5. Diets supported ADG of 771 g/d and 220 g/d for CON and CR calves, respectively. Heifers were euthanized at 8 wks and the LM was collected for muscle morphometry, gene expression and metabolic signal transduction measurements. The effect of diet was examined using Student *t*-test with  $\alpha = 0.05$ . Results revealed that CR heifers had smaller ( $P < 0.05$ ) LM fiber cross-sectional area and a greater percentage of oxidative type I and IIA fibers. The oxidative fiber type of the CR LM was not associated with elevated expression of *PGC1 $\alpha$* , a mitochondria biogenesis factor. Reverse transcription-qPCR revealed *PGC1 $\alpha$*  mRNA levels were 40% lower in CR than CON LM. Western blot analysis detected reduced ( $P < 0.05$ ) amounts of PGC1 $\alpha$ 4, a PGC1 $\alpha$  splice variant, in CR LM. The PGC1 $\alpha$ 4 target gene, *IGF-I*, was 40% lower ( $P < 0.05$ ) in CR than CON. Downstream mediators of autocrine IGF-I signaling are attenuated in CR by comparison to CON. The amount of phosphorylated AKT was lower ( $P < 0.05$ ) in CR than CON. However, activated components of the AKT/mTOR signaling axis, p70 S6 kinase, ERK1/2 and 4E-BP1, were equivalent or unaffected ( $P > 0.05$ ) by CR. Activation of the energy sensor, AMP kinase, was not detected in either CON or CR lysates indicating that a block to mTOR-controlled protein synthesis is absent. Protein lysates from CR LM contained less ( $P < 0.05$ ) total and phosphorylated, inactive GSK3 $\beta$  than CON LM. Elevated GSK3 $\beta$  activity is associated with inhibition of protein synthesis as well as induction of atrogin. Reverse transcription-qPCR demonstrated no differences ( $P < 0.05$ ) in atrogin expression levels between CON and CR. These results indicate that CR suppresses a hypertrophic PGC1 $\alpha$ 4/IGF-I/AKT pathway in young heifer calves through an undiscovered mechanism.

**Key Words:** skeletal muscle, metabolism, IGF-I

**T230 Use of ultrasound for assessment of muscle area and depth in postmortem preweaned Holstein calves.** Jessica H. Davis\*<sup>1</sup>, Heidi A. Rossow<sup>1</sup>, and Chris M. Barker<sup>2</sup>, <sup>1</sup>*Veterinary Medicine Teaching and Research Center, University of California, Davis, Tulare, CA*, <sup>2</sup>*Center for Vectorborne Diseases, University of California, Davis, Davis, CA*.

The prewean period is one of the most important stages of dairy calf development. A tool is needed to more accurately assess calf growth, specifically muscle development. The objective was to determine if ultrasound can be used to predict *longissimus dorsi* (ribeye) linear depth and *external carpi radialis* (front) and *semitendinosus* (hind) area in postmortem preweaned Holstein calves. Postmortem bull and heifer calves (n = 191, age 17.3  $\pm$  20.67 d, body weight 37.9  $\pm$  19.07 kg) were obtained from 2 calf ranches between April and July 2013. Ultrasound images of the ribeye, front, and hind muscles were collected using an Aloka 500V equipped with a 5-cm 7.5-MHz linear transducer. Ultrasound ribeye linear depth and front and hind areas were calculated using the Ultrasound Image Capture System. The ribeye was dissected and measured for linear depth. The front and hind muscles were dissected and the cross-sectional planes were traced onto transparency paper. The transparency paper was photocopied and individual paper muscle tracings were cut out and weighed. The weights of the paper muscle tracings were then converted to areas using the known area of a standard 8.5  $\times$  11 inch paper. Means were calculated using PROC GLM in SAS (version 9.2). Mean dissected values for the ribeye, front, and

hind muscles (1.65  $\pm$  0.44 cm, 6.23  $\pm$  1.83 cm<sup>2</sup>, 9.05  $\pm$  2.37 cm<sup>2</sup>) were greater than the respective mean ultrasound values (1.46  $\pm$  0.37 cm, 5.41  $\pm$  1.49 cm<sup>2</sup>, 8.60  $\pm$  2.40 cm<sup>2</sup>) indicating ultrasound underestimated the true linear depth and area values consistently. The relationship between the dissected and ultrasound measurements was tested using Pearson correlation coefficient (PROC CORR). Overall, there was a strong, positive relationship between both the dissected and ultrasound measurements for the ribeye ( $r = 0.55$ ,  $P < 0.01$ ), front ( $r = 0.65$ ,  $P < 0.01$ ), and hind muscle ( $r = 0.80$ ,  $P < 0.01$ ). The weight, age, and sex of the calf and the operator of the ultrasound may explain some of the variability not accounted for by the correlation coefficient. The *semitendinosus* muscle displayed the highest correlation coefficient and may be used in future studies to assess calf muscle growth and guide implementation of dynamic feeding changes on both dairy farms and calf ranches.

**Key Words:** ultrasound, dairy calf, muscle

**T231 Increasing the metabolizable protein supply enhanced growth performance and humoral immune response of preconditioning beef steers.** Philippe Moriel<sup>1</sup>, Luis Felipe Arelaro\*<sup>1</sup>, Matt Poore<sup>2</sup>, Rodrigo Marques<sup>3</sup>, and Reinaldo Cooke<sup>3</sup>, <sup>1</sup>*North Carolina State University, Waynesville, NC*, <sup>2</sup>*North Carolina State University, Raleigh, NC*, <sup>3</sup>*Oregon State University, Burns, OR*.

We evaluated the effects of metabolizable protein (MP) supply on growth performance, and post-vaccination measurements of humoral immune response of beef steers. Angus steers (n = 36; 231  $\pm$  21 kg; 184  $\pm$  18 d) were weaned on d 0, stratified by weight and age on d 7, and randomly assigned into 1 of 18 drylot pens (2 steers/pen). Treatments were assigned to pens (6 pens/treatment), and consisted of isocaloric, corn silage-based diets formulated to provide 85, 100 or 115% of the daily MP requirements. Diets were offered at 2.1% of BW (DM basis) from d 7 to 49. Steers were vaccinated on d 21 and 35 with modified-live and killed vaccines (Select-Vac, Zoetis). Blood samples were collected from jugular vein on d 0, 21, 35 and 49 to determine the plasma cortisol concentrations, and serum titers against bovine viral diarrhea virus-1b (BVDV1b), infectious bovine rhinotracheitis (IBR) and *Mannheimia haemolytica* leukotoxin. Data were analyzed using GLIMMIX procedure of SAS, and pen(treatment) as random effect. Body weight differed only on d 49, and was greatest ( $P \leq 0.05$ ) for 115MP, least for 85MP and intermediate ( $P \geq 0.21$ ) for 100MP steers (297, 278 and 290  $\pm$  7 kg, respectively). From d 7 to 49, 100MP and 115MP steers had similar ADG ( $P = 0.13$ ), but both had greater ( $P < 0.001$ ) ADG than 85MP steers (1.20, 1.35 and 0.83  $\pm$  0.068 kg/d, respectively). Plasma cortisol concentrations were greater ( $P \leq 0.03$ ) for 85MP vs. 100MP and 115MP steers on d 21 and 35. Serum BVDV1b titers were greatest ( $P \leq 0.02$ ) for 115MP, and similar ( $P = 0.37$ ) between 85 and 100MP steers (5.8, 3.7 and 3.0  $\pm$  0.60 base 2 log, respectively). Mean serum *M. haemolytica* leukotoxin titers were greater ( $P = 0.05$ ) for 85MP vs. 100MP and 115MP steers (3.12, 2.39 and 2.49  $\pm$  0.21 ng/antibody bound, respectively). Hence, a 15% MP deficiency increased plasma cortisol, and both factors may have suppressed innate immune system leading to greater multiplication of endogenous *M. haemolytica* and a compensatory greater production of serum *M. haemolytica* leukotoxin titers. In contrast, 15% MP excess enhanced growth performance and increased serum BVDV1b titers of beef steers.

**Key Words:** metabolizable protein, steer, immune



**T232 Characterization of key factors constraining lamb survival in marginal environments.** Francisco A. Sales\*<sup>1</sup>, Sue A. McCoard<sup>2</sup>, and Raúl J. Lira<sup>1</sup>, <sup>1</sup>*Instituto de Investigaciones Agropecuarias, Punta Arenas, Chile*, <sup>2</sup>*AgResearch, Palmerston North, New Zealand*.

Sheep farming normally occurs in areas with limited vegetation. Climate change is placing more pressure on farming in these challenging environments. There is limited research on factors limiting lamb survival in marginal environments. The objective of this study was to characterize fetal and maternal factors associated with low (<80%) lamb survival in Patagonia. Corriedale ewes, synchronized, superovulated and mated to Suffolk rams and managed under commercial grazing conditions were used. Single- (n = 8) and twin-bearing (n = 8) ewes were euthanized at d 140 of gestation and mass of the fetal and placental components and ewe carcass traits were obtained. The effect of litter size (single vs twin) on maternal and fetal characteristics was analyzed using ANOVA. Maternal live weight was similar between twin- and single-bearing ewes ( $60.4 \pm 1.6$  vs  $61.4 \pm 1.6$  kg,  $P = 0.7$ ), while twin-bearing ewes had lower carcass weight compared with single-bearing ewes ( $18.9$

$\pm 0.6$  vs  $20.7 \pm 0.6$  kg,  $P = 0.05$ ), associated with a trend for reduced loin area ( $11.8 \pm 0.7$  vs  $13.8 \pm 0.7$  kg,  $P = 0.07$ ), but similar carcass fat depth (Fat GR:  $4.5 \pm 1.1$  vs  $4.12 \pm 1.1$  mm,  $P = 0.8$ ; Fat C:  $3.8 \pm 0.6$  vs  $3.8 \pm 0.6$  mm,  $P = 1.00$ ). Twin fetuses were lighter ( $3.24 \pm 0.11$  vs  $4.18 \pm 0.14$  kg,  $P < 0.01$ ), with proportionally lighter organ and leg muscle weight ( $P < 0.05$ ), a tendency for less perirenal fat ( $9.15 \pm 0.64$  vs.  $10.99 \pm 0.78$ ,  $P = 0.07$ ) and disproportionately lighter adrenal glands ( $P = 0.05$ ), *M. adductor* ( $P = 0.02$ ) and *M. gluteus* ( $P = 0.03$ ) compared with singletons. Total placentome weight ( $256.2 \pm 10.6$  vs  $391.5 \pm 14.9$  g,  $P < 0.01$ ) and number ( $49.6 \pm 4.0$  vs  $75.6 \pm 5.7$ ,  $P < 0.01$ ) were lower in twin- compared to single-bearing ewes. Reduced placental mass and reduced fetal weight in twins compared with singletons is consistent with prior studies. The greater mobilization of skeletal muscle but not fat in twin-bearing ewes is likely a mechanism to enhance protein supply to the fetuses. Compared with studies in less marginal environments, the fetuses (both singles and twins) in this study had a higher percentage of perirenal fat (0.3% vs 0.2%), which may indicate an adaptive response, to enhance fetal survival.

**Key Words:** sheep, fetal, growth

## Horse Species

**T233 Velocity-related changes in stride variables of the intermediate gait of the Irish Draught horse of North America.** Molly Nicodemus\*<sup>1</sup>, Rachel Fletcher<sup>1</sup>, and Jeannette Beranger<sup>2</sup>, <sup>1</sup>Mississippi State University, Mississippi State, MS, <sup>2</sup>Livestock Conservancy, Pittsboro, NC.

Although originating as a farming breed having a gait derived from the bloodlines of the extinct ambling Irish Hobby horse, the Irish Draught horse today is bred to excel in the sport-horse industry. With around 500 in North America, the breed is on the Livestock Conservancy's (LC) watch list. Study objectives were to determine the relationship between trotting velocities and stride variables. Ten Irish Draught horses selected by the LC and Irish Draught Horse Society of North America were worked at a slow velocity (SV) and fast velocity (FV) at the trot (SV = 4.5 ± 0.2 m/s; FV = 5.2 ± 0.1 m/s). Frame-by-frame analysis using the Ariel Performance Analysis System was performed documenting hoof contact and lift-off for 10 strides for each horse at each velocity with stride variables given as a % of stride duration. Means (SD) were calculated and student's paired *t*-tests were performed (*P* = 0.05). Both velocities demonstrated a leaping diagonal footfall sequence with diagonal limb pairs at hoof contact and periods of bipedal support (FV = 79 ± 4, SV = 87 ± 3%). While neither velocity demonstrated a 4-beat rhythm at hoof contact, the diagonal limbs disassociated at lift-off (4 ± 1%) at the SV with the hind lifting first creating a period of forelimb unipedal support (9 ± 1%). Along with limb support and coupling of diagonal limbs, fore stance duration (FV = 41 ± 3, SV = 48 ± 1%), stride length (FV = 3.8 ± 0.1, SV = 3.2 ± 0.1 m/s), and length of suspension (FV = 21 ± 4, SV = 4 ± 2%) distinguished between velocities (*P* < 0.05). Stride duration (FV = 0.70 ± 0.01, SV = 0.74 ± 0.02 s) and rate (FV = 1.41 ± 0.02, SV = 1.37 ± 0.01 strides/sec) and hind stance duration (FV = 41 ± 4, SV = 43 ± 2%) remained consistent between velocities. The North American Irish Draught horse did not demonstrate an ambling gait at velocities measured. Nevertheless, pattern and timing of the disassociated limbs and resulting limb support are unique compared with breeds that are today used in the breeding of the Irish Draught horse such as the Thoroughbred and Warmblood, and thus, potentially reflecting gait characteristics of the extinct Irish Hobby horse.

**Key Words:** stride variable, trot, velocity

**T234 Influence of fibrolytic enzymes and yeast addition in horse's diet on digestibility, blood chemistry and fecal coliform.** M. M. Y. Elghandour<sup>1</sup>, A. E. Kholif<sup>2</sup>, A. Z. M. Salem\*<sup>1</sup>, J. C. Vázquez Chagoyán<sup>1</sup>, J. S. Martínez Castañeda<sup>1</sup>, L. M. Camacho<sup>3</sup>, R. Montes de Oca<sup>1</sup>, and T. A. Morsy<sup>2</sup>, <sup>1</sup>Facultad de Medicina Veterinaria y Zootecnia, Universidad Autónoma del Estado de México, Toluca, Estado De México, Mexico, <sup>2</sup>Dairy Science Department, National Research Centre, Giza, Egypt, <sup>3</sup>Unidad Académica de Medicina Veterinaria y Zootecnia, Universidad Autónoma de Guerrero, Altamirano, México.

Improving fibrous feeds utilization ensures low consumption of high-starch grains and may reduce various pathologies. Fibrolytic enzymes (FE) like cellulase and xylanase, and yeast addition can improve fibrous feeds utilization. Therefore, the current study aimed to study the effect of FE (Exp. 1) and yeast (Exp. 2) on the utilization of diet with 11.2% CP and 51.1% NDF. Thirty-two mares of Quarter Horse (450-500 kg BW) were used in a complete randomize design for 15 d, with 10 d for adaptation and 5 d for samples collection. For Exp. 1, mares (n=16)

were distributed onto 4 treatments to be fed on the basal diet without FE (control), or plus cellulase at 10 mL/mare/d (CELL), plus xylanase at 10 mL/mare/d (XYL), or plus mixture cellulase and xylanase at 5 ml of each enzyme/mare/d (CX). For Exp. 2, mares (n=16) were distributed onto 4 treatments to be fed on the basal diet without yeast (control), or fed the control diets plus Procreatin 7 (1.5 × 10<sup>10</sup> cfu/g *S. cerevisiae*) at 15 g/mare/d (P7), plus Biocell F53 (2 × 10<sup>10</sup> cfu/g *S. cerevisiae*) at 11 g/mare/d (F53), or plus Biosaf SC47 (1.5 × 10<sup>10</sup> cfu/g *S. cerevisiae*) at 15 g/mare/d (SC47); yeast products were in powder form. Both of enzyme and yeast doses were mix with the 1 kg of concentrate diet at 0400 h. Mares were fed the concentrates twice daily at 0400 and 1600 h, while the forage of oat straw was offered ad libitum at 0500 and 1700 h. Acid insoluble ash concentrations in feed and fecal samples were used for digestibility determination. No effects for FE and yeast were obtained in blood alanine transaminase, aspartate aminotransferase, urea, creatinine, total protein and glucose. Addition of CELL, XYL and CX increased (*P* = 0.001) nutrient intakes from oat straw versus control. Moreover, CELL, XYL and CX increased (*P* < 0.05) digestibilities of DM, OM, NDF and ADF. Enzymes decreased (*P* < 0.05) concentration of fecal coliform. In the contrary, F53 increased nutrients intake of oat and nutrients digestibility (*P* < 0.05) without difference compared with other treatments. Yeast had no effect on fecal coliform concentration. It could be concluded that addition of FE at 10 mL/mare/d or addition of Biocell F53 at 11 g/mare/d improved feed intake and nutrients digestibility without affecting mare's blood parameters.

**Key Words:** enzyme, horse, yeast

**T235 Changes in salivary IgA and nasopharyngeal leukocyte populations in response to prolonged head elevation.** Jill M. Bobel\*, Megan R. Di-Lernia, Jeffrey R. Abbott, Maureen T. Long, and Lori K. Warren, University of Florida, Gainesville, FL.

Prolonged head elevation is thought to be a major contributor to the increased risk of respiratory disease associated with transportation in horses. Prior investigations have focused on immunological changes in the lower respiratory tract. The aim of this study was to characterize the response to head elevation in the upper respiratory tract. Twelve horses (mean ± SEM, 552 ± 10 kg; 11.5 ± 1.4 y) were tethered for 12 h with their heads elevated at a height of 1.5 m to induce physiological stress. While tied, horses had unlimited access to bermudagrass hay and were offered water every 2 h. Each horse underwent head elevation on 4 occasions, each separated by 30 d. When not tied, horses were maintained on pasture forage. Nasopharyngeal flush (NPF) and saliva samples were obtained before head elevation, immediately after (0 h), and 12, 24, and 72 h post head elevation. Mucus content and leukocyte populations were quantified in NPF and IgA was measured in saliva. Data were compared using mixed model ANOVA with repeated measures. NPF samples contained more mucus at 0 h post (*P* < 0.02) compared with pre-head tie samples. Percentage and number of neutrophils in NPF increased at 0 h post (*P* < 0.0001) and the number remained elevated through 72 h (*P* = 0.04). Lymphocytes, monocytes, and eosinophils in NPF increased in number (*P* < 0.05) but decreased in percent (*P* < 0.05) in response to head elevation. While the proportion of these cells normalized by 72 h, the numbers declined to levels lower than pre-head tie values. Percentage of CD8+ T cells and B cells in NPF were lower at 0 h post (*P* < 0.05) and returned to baseline by 12 h. Percentage of CD4+ and the ratio of CD4+ to CD8+ cells increased at 0 h post (*P* < 0.01) and remained elevated through 72 h (*P* < 0.05). Salivary IgA

increased at 0 h post ( $P < 0.0001$ ), then decreased below baseline at 12 h post ( $P < 0.0001$ ) and remained lower than pre-head elevation values through 72 h ( $P < 0.05$ ). The immunological changes observed in the upper respiratory tract agree with those reported for lower tract, and highlight increased risk of disease following prolonged head elevation.

**Key Words:** immunosuppression, upper respiratory disease, horse

**T236 Utilization of the equine SNP 70 beadchip in monitoring inbreeding and describing the genetic background in an Arab horse herd.** Mohammed Al Abri<sup>\*1</sup>, Samantha Brooks<sup>2</sup>, and König von Borstel<sup>3</sup>, <sup>1</sup>Cornell University, Ithaca, NY, <sup>2</sup>University of Florida, Gainesville, FL, <sup>3</sup>Göttingen University, Göttingen, Germany.

Horse breeders rely heavily on the accurate identification of individual ancestry through pedigrees. Errors in such pedigrees may inaccurately assign horses to false lineages or breed memberships, and can result in inaccurate estimates of inbreeding. Discrepancies in pedigree records can lead horse owners into making misguided purchasing and breeding decisions. Genome-wide SNP data provide a robust and precisely quantitative tool to resolve many lineage assignments and provide genomic measures of inbreeding values. The aim of this project was to pilot a comparison between pedigree and genomic relatedness measures. Here, we describe a herd of 36 pedigreed Egyptian Arabian horses genotyped using the Equine SNP70 (Geneseek, Inc.) Genomic inbreeding values and pair-wise relatedness between horses within the herd were estimated from the genotypic data. Pedigree derived inbreeding values were significantly correlated with the genomic inbreeding values across the population ( $r = 0.44$ ,  $P = 0.007$ ), although within an individual the 2 values could differ substantially. Genomic inbreeding values were also positively correlated with the year of birth ( $r = 0.29$ ,  $P = 0.086$ ), demonstrating a trend in inbreeding over the years. A multi-dimensional scaling analysis (MDS), a phylogenetic analysis and a clustering analysis were performed to compute the relationships between the horses and the results were compared with the pedigree information. These same analyses were also conducted for the herd among US, Polish, and Egyptian Arabian horses to examine their Arabian ancestry. The within herd analysis was successful in recapturing much of first-degree relationships, although more distant relationships were not entirely reconstructed. The herds' Egyptian lineage was successfully assigned among Arabian horse sub-groups. It can be concluded that genome-wide genotypes are good alternatives to compute inbreeding and verify the integrity of pedigrees in horses in cases where records are unavailable or in doubt. Although costly, application of this tool in a breeding program can enable informed mating decisions and assess inbreeding in lines of horse already at risk of losing genomic diversity.

**Key Words:** Arabian horse, pedigree inbreeding, genomic inbreeding

**T237 Evaluation of single nucleotide polymorphisms effects on injury predisposition in a population of multi-discipline athletically trained horses.** Sarah Mercer<sup>1</sup>, Neely Walker<sup>2</sup>, and Matthew Garcia<sup>\*1,2</sup>, <sup>1</sup>Louisiana State University, Baton Rouge, LA, <sup>2</sup>LSU AgCenter, Baton Rouge, LA.

Tendon and ligament injuries (TLIs) in the performance horse represent a significant burden to the equine industry. Furthermore, treatment is often unsuccessful and re-injury common, prevention of TLIs is a major goal. The objective of the current study was to evaluate a population of athletically trained horses from multiple disciplines for SNP located 5 candidate genes in association with increased injury predisposition or injury resistance. A total population of 63 performance horses with

documented injury history or injury resistance was utilized for the current study. Specifically, the study was comprised of 25 horses of various ages with at least one injury and 33 horses of various ages with no injury history. A 5mL blood sample collected from all horses via jugular venipuncture and DNA was subsequently extracted from white blood cell buffy coats for SNP genotyping. The 5 candidate genes selected for evaluation included the Angiotensin 1 converter enzyme gene (ACE), the ATPase  $\alpha$  2 peptide gene (ATP1A2) the Bradykinin receptor B2 gene (BDKRB2), the Collagen type 1  $\alpha$  1 gene (COL1A1) and the Collagen type 5  $\alpha$  1 gene (COL5A1). Candidate genes in the current study have been previously reported to be associated with jumping ability (ACE), racing ability (ATP1A2), and ligament and tendon injuries (BDKRB2, COL1A1 and COL5A1). A total of 64 single nucleotide polymorphisms (SNP) were selected across all candidate genes (ACE = 11, ATP1A2 = 14, BDKRB2 = 9, COL1A1 = 14, COL5A1 = 16). A mixed model analysis was utilized for the current data set with independent variables of sex, breed, age, discipline, training start age, first competition age, first age of injury, and number of years in competition. Independent effects included injury status (injured or not injured) and inherited SNP genotype for each unique SNP located on the previously described candidate genes. Although multiple SNP were identified in the current study as being associated with injury susceptibility/resistance, these SNP and a greater number of candidate genes must be evaluated in larger more diverse populations before implementation into selection strategies.

**Key Words:** equine, injury, SNP

**T238 Effect of oil supplementation on milk IgG, serum insulin, glucose, placental efficiency, and immune status of foals.** Lauren B. Hodge<sup>\*</sup>, Brian J. Rude, Caleb O. Lemley, and Torea L. Bova, *Mississippi State University, Starkville, MS.*

The objective of this research was to evaluate the effects of supplementing pregnant mares with omega-3 fatty acids and how this may affect the suckling foal. Pregnant mares ( $n = 18$ ) were randomly assigned to 1 of 3 diets beginning 28 d before their expected foaling date until 84 d after foaling. Diet 1 was a commercial feed fed to meet NRC requirements, based on forage analysis; diet 2 was diet 1 plus a blended fish oil; and diet 3 was diet 1 plus a blend of fish and soybean oil. Blood samples were collected 28 d before their expected foaling date, and at 14 d increments. Placental efficiency was calculated as a ratio of placenta weight:foal weight. Milk samples were obtained at foaling and on remaining blood collection days. Body weights were recorded the same day blood samples were collected. Placentas were weighed and ~5 g sample taken. No differences were found for mare plasma IgG ( $P = 0.1318$ ), serum insulin ( $P = 0.3886$ ), plasma glucose ( $P = 0.2407$ ), or milk IgG ( $P = 0.1262$ ) concentrations for treatment or time period of sampling. Mare packed cell were not different ( $P = 0.0885$ ) among treatments; however, decreased ( $P < 0.0001$ ) as the trial progressed. Mare body weight and body weight change were not different ( $P = 0.5704$ ;  $P = 0.08265$ , respectively) among treatments, or relative to time of sampling. Mare body weight change did differ ( $P < 0.0001$ ) relative to time. Foal plasma IgG ( $P = 0.2767$ ), serum insulin ( $P = 0.4843$ ), or plasma glucose ( $P = 0.1204$ ) were not affected by treatment or time of collection. Foal packed cell was not different ( $P = 0.6275$ ) among treatments, however, there was a difference ( $P = 0.0005$ ) relative to time of sampling. Foal body weight change and total gain were not different among treatments ( $P = 0.6825$ ;  $P = 0.8220$ , respectively); however foal body weight were least ( $P = 0.0041$ ) for foals consuming diet 1 and greatest for foals consuming diet 3, with foals consuming diet 2 being intermediate. Foal body weight change decreased as the trial progressed ( $P < 0.0001$ ). Placental efficiency and nitrites were not different ( $P =$



0.1631;  $P = 0.5604$ , respectively) among treatments. Research should be conducted to evaluate supplementation earlier in gestation.

**Key Words:** horse nutrition, fat, supplementation

**T239 Testis tissue explant culture supports the viability of equine spermatogonial stem cells.** Kyle C. Caires\*<sup>1</sup>, Louie Y. Chen<sup>2</sup>, Rachel A. Lemcke<sup>1</sup>, and Laurie A. Seigler<sup>1</sup>, <sup>1</sup>Berry College, Department of Animal Sciences, Mount Berry, GA, <sup>2</sup>RDBL, National Institute of Environmental Health Sciences, National Institutes of Health, Research Triangle Park, NC.

Spermatogenesis is a stem-cell dependent process that supplies an indefinite supply of spermatozoa during adult male life. Sperm production is well characterized in rodents and depends on a milieu of endocrine and growth factors, but little is known regarding the regulation of male fertility and spermatogonial stem cells (SSC) in other domestic animals. The objective of the present study was to develop an organ culture system to investigate spermatogenesis in horses. To accomplish this aim we obtained testicular parenchyma from pre- and post-pubertal stallions ( $n = 3$  for each age group) and cultured those tissues upon 0.4- $\mu\text{m}$  membranes in XC medium (DMEM supplemented with 1-, 5- and 10% fetal bovine serum) over a weeklong period with alternate-day media changes. All data sets were evaluated using ANOVA and differences between means were considered significant at  $P < 0.05$ . Immunohistochemical analysis of *PLZF*, *GFRAL*, and *DDX4* expression were used to confirm the identity of undifferentiated spermatogonia, differentiated spermatogonia and meiotic germ cells, whereas *GATA4* was used as a marker for Sertoli cell and Leydig cell populations. Histological analysis of fresh and cultured testis tissue indicates the survival of germ-line stem cells and somatic cells populations within morphologically normal seminiferous tubules at 24, 48, and 72 h during the culture period with different levels of serum. Sertoli cell and Leydig cell populations remained stable ( $P > 0.05$ ) throughout the culture period, but the appearance of pyknotic nuclei in germ and somatic cells observed following 120 h of culture indicates limitations of our approach for long-term tissue maintenance, and this notion is confirmed by TUNEL analysis of apoptosis. Investigating the effect of specific growth factor and hormone treatment combinations on the biological activity and stem cell behavior of undifferentiated spermatogonia is ongoing. Collectively, these results demonstrate successful culture of equine spermatogonial stem cells, and provide a useful ex-vivo model for investigating spermatogenesis in stallions without the costs typically associated with whole-animal experimentation.

**Key Words:** equine testis, stem cells, spermatogonial stem cells

**T240 Valerianic acid detection in equine urine after administration of calming supplement.** Celina M. Checure\*<sup>1</sup>, Nikki McGreevey<sup>1</sup>, Travis J. De Wolfe<sup>1</sup>, Simon F. Peek<sup>1</sup>, Greg A. Barrett-Wilt<sup>1</sup>, Richard G. Godbee<sup>2</sup>, and Benjamin J. Darien<sup>1</sup>, <sup>1</sup>University WI-Madison, Madison, WI, <sup>2</sup>Central Garden & Pet, Phoenix, AZ.

Valerian root (*Valeriana officinalis*) is an herbal tranquilizer used in horses. However, many plants and herbs are classified as forbidden by the USEF Equine Drugs and Medications Rule due to their potential to affect performance. The aim of this study was to establish a reliable withdrawal period before competition for trainers considering valerianic acid supplements as a training aid. Ten mares between 10 and 20 years of age were administered an oral paste of valerian root extract (36 mg) daily for 5 consecutive days. Urine samples were collected by sterile catheterization before the first and all subsequent treatments. Urine was also collected at 4, 8, 24, 48, and 72 h after the last treatment. Urine

samples were coded for blinding and stored at 4°C until testing. Valerianic acid in urine was analyzed using a triple-quadrupole mass spectrometer coupled to an Agilent 1100 capillary HPLC system. Quantitation of urine valerianic acid was done by generating a calibration curve using the peak areas of 3 replicate injections for each calibration point. Two different calibration curves for quantitation of valerianic acid in urine were created. One curve was generated by spiking valerianic acid at 0 (blank), 25, 50, 100, 200, and 400 ng/mL into known blank urine before protein precipitation and solid-phase extraction. A second curve was generated for later samples by pooling 2 mL from each of 4 blank urine samples and then adding valerianic acid as described. Regression analysis of a representative calibration curve generated from these standards yielded a linearity of  $R^2 = 0.9969$ . Based on the signal-to-noise ratio of the 25 ng/mL calibration standard, the limit of quantitation was estimated at 20 ng/mL. From a total of 90 post-valerianic acid treatment urine samples, 3 samples were positive. Urine samples obtained from one horse was positive for valerianic acid on d 2 (21 ng/mL) and 3 (29.5 ng/mL) of treatment. A different mare tested positive (67.8 ng/mL) 4 h after the last dosage (d 5). Based on our results, horses should be withdrawn from competition for at least 5 d after the oral administration of 36 mg valerianic root extract.

**Key Words:** valerian, herbal, tranquilizer

**T241 Influence of  $\alpha$ -linolenic acid supplementation in mature horses at maintenance: Body composition.** Jessica L. Leatherwood\*<sup>1</sup>, Emily D. Lamprecht<sup>2</sup>, Mark J. Anderson<sup>1</sup>, Kyle J. Stutts<sup>1</sup>, Marcy M. Beverly<sup>1</sup>, and Stanley F. Kelley<sup>1</sup>, <sup>1</sup>Sam Houston State University, Huntsville, TX, <sup>2</sup>Cargill Incorporated, Elk River, MN.

Twenty mature horses (455 to 457 kg and 5 to 10 yr) were utilized in a randomized complete block design to evaluate incorporation of  $\alpha$ -linolenic acid into plasma and the effects of supplementation on performance variables in mature horses. Horses were blocked by BW, age, and sex and randomly assigned to treatment within block for a 112 d trial. Dietary treatments included control (no  $\alpha$ -linolenic acid; CON) or 150 mg/kg BW/d  $\alpha$ -linolenic acid (ALA) derived from a flaxseed oil (TRT; Clear Valley Omega 3 Oil; Cargill, Inc., Eddyville, IA). Diets consisted of CON horses ( $n = 10$ ) fed 0.25% BW (as-fed) concentrate only or TRT horses fed ( $n = 10$ ) the same concentrate with additional ALA offered at 12 h intervals. Horses were housed by block and maintained in adjacent dry lots with ad libitum access to coastal Bermudagrass (*Cynodon dactylon*) hay. Body weight and BCS were obtained every 14 d and concentrate was adjusted accordingly. Rump fat (RF), longissimus dorsi area (LD), and longissimus dorsi fat thickness (LDF) were obtained every 28 d via ultrasonography. Blood samples were collected on d 0, 56, and 112 to determine plasma fatty acid concentrations by gas chromatography. Data were analyzed using the PROC MIXED procedure of SAS. Fatty acid levels in the oil supplement remained stable over the course of the trial. Plasma concentrations of ALA were greater ( $P \leq 0.01$ ), and plasma levels of arachidonic acid were lower ( $P \leq 0.01$ ) in TRT compared with CON beginning at d 56 to d 112 of the study. Body weight and BCS were not influenced ( $P = 0.96$  and  $P = 0.14$ , respectively) by dietary treatment; however, all horses gained BW and BCS throughout the trial ( $P \leq 0.01$ ). Rump fat, LD and LDF were not influenced ( $P \geq 0.54$ ) by dietary supplementation although, performance variables increased ( $P \leq 0.01$ ) over time across treatments. These results indicate that this source of ALA is stable and will incorporate into circulation through targeted supplementation; however, further studies are needed to fully elucidate effects of dietary ALA supplementation to mature horses.

**Key Words:** equine,  $\alpha$ -linolenic acid, longissimus dorsi

**T242 In vitro evaluation of protein content on forage digestion using equine fecal inocula.** Tayler L. Hansen\*, Brooke M. Eubanks, Emily K. Rizzo, and Lori K. Warren, *University of Florida, Gainesville, FL.*

Previous research has indicated crude protein (CP) and neutral detergent fiber (NDF) concentrations are related to forage digestibility by horses. However, it is unclear if greater CP concentrations are simply correlated with higher quality forage or if CP amounts influence digestion and microbial fermentation in the hindgut by increasing nitrogen substrates to cellulolytic bacteria. The objective of this study was to determine the influence of varying amounts of CP on forage digestibility using an in vitro hindgut fermentation model. We hypothesized increasing CP presence would increase in vitro dry matter digestibility (IVDMD), NDF digestibility (NDFD), and acid detergent fiber digestibility (ADFD). Quadruplicate 0.5 g forage samples (alfalfa, bermudagrass, and orchardgrass) and cellulose (Sigma-Aldrich) were incubated at 37.5°C for 48 h in an ANKOM DaisyII Incubator. Casein (Sigma-Aldrich) was added to the digestion jars at 1 of 4 levels (no added casein, 66.1, 125.9, and 188.8 g) to represent control, NRC CP recommendations, industry-typical diets, and 3× NRC recommendations, after adjusting for 51% prececal CP digestibility. Freshly voided fecal samples were collected on 4 separate days from mature horses with ad libitum access to bermudagrass hay to serve as microbial inoculum for DaisyII runs (n = 4). Data were analyzed using ANOVA (SAS, v 9.3) as a split-plot design with the main plot as casein level and subplot as forage type with an error term of casein level × block. Means were separated using LSD comparisons. In vitro DMD decreased ( $P < 0.0001$ ) as added casein increased. Control IVDMD across all forage types ( $47.5 \pm 9.6\%$ , mean  $\pm$  SE) was greater than ( $P < 0.05$ ) IVDMD for all added casein treatments ( $30.7 \pm 13.0$ ,  $32.3 \pm 13.6$ ,  $28.0 \pm 11.2\%$ , as casein level increased). Both NDFD and ADFD were greater ( $P < 0.0001$ ) in control compared with casein-added treatments. In vitro DMD, NDFD, and ADFD differed ( $P < 0.0001$ ) by forage type. Casein as a protein source may have affected results with this closed-system in vitro model. Further evaluations are needed to determine the relationship between protein in the equine hindgut and fiber digestion in vivo.

**Key Words:** DaisyII, fiber, horse

**T243 Changes in plasma calcium and phosphorus concentrations in mares fed decreasing amounts of dietary Ca and P just prior to weaning.** Ashley L. Fowler\*, Brittany E. Harlow, Morgan B. Pyles, Susan H. Hayes, Andrea D. Crum, and Laurie M. Lawrence, *University of Kentucky, Lexington, KY.*

During lactation, the concentrations of Ca and P in milk are highly conserved, frequently at the expense of bone mineral. Replacement of bone mobilized during lactation and dietary changes in the post-weaning period may further alter Ca and P metabolism, but it is unknown if these changes influence the ability to achieve Ca and P homeostasis. The objective of this study was to examine changes in plasma Ca and P in lactating mares fed decreasing amounts of Ca and P just before weaning. Four mares in late lactation ( $12 \pm 5.7$  yr;  $527 \pm 26$  kg) were transitioned to a diet containing Ca and P in amounts appropriate for nonlactating mares for 14 d, weaned and then maintained on the same diet for 7 d. Four nonlactating mares ( $11 \pm 4$  yr;  $552 \pm 33$  kg) received the same diet and served as the controls. We hypothesized that low mineral intake during late lactation would stimulate bone resorption (increasing blood Ca and P) and that in the post-weaning period there would be bone deposition (decreasing blood Ca and P). Blood samples for P and Ca analysis were obtained 14 d before weaning (dietary Ca and P equal to

or exceeding requirements), at 2 d before, and 7 d after weaning (dietary Ca and P adequate for nonlactating mares). Saliva samples were taken 7 d post-weaning for P analysis. Data were analyzed using ANOVA with repeated measures with group (lactating or nonlactating) and sample date as main effects. Plasma P increased from baseline to pre-weaning ( $P < 0.05$ ) across groups and did not change further post-weaning. The increase in plasma P at 2 d pre-weaning might reflect an increase in bone resorption as a result of decreasing amounts of dietary P, but a decrease in plasma P 7 d post-weaning was not observed. Salivary P was not different between groups post-weaning. Plasma Ca decreased from baseline to pre-weaning ( $P = 0.053$ ) across groups, but did not change further post-weaning. The Ca:P ratio in the blood decreased from 3.7:1 at baseline to 2.8:1 at pre-weaning ( $P < 0.05$ ). From these data, it appears that changes in Ca and P intakes influence blood Ca and P more than the physiological change of lactation cessation.

**Key Words:** horse, lactation, mineral

**T244 Relationship between training difficulty and aggression in horses.** M. J. Anderson\*, J. L. Leatherwood, K. Jones, K. J. Stutts, M. M. Beverly, and S. F. Kelley, *Sam Houston State University, Huntsville, TX.*

Training of horses involves a balance of trust and dominance between animal and trainer. Yet in a herd, horses will establish a social hierarchy, which lends to antagonistic behaviors. The objective of this research was to characterize behavior and interactions between horses and determine the implications on the trainability of the horse by trainers of varying skill levels. To accomplish this test, horses (n = 10) from the Sam Houston State University behavior and training course were randomly selected and individually introduced in 9.14-m round pens to control horses (n = 5) over a 2-wk period. The interaction between the horses was recorded by 3 independent observers and scored on a 15 cm line scale ranging from submissive (1) to aggressive (15). Horses were individually worked by one of 5 trainers of varying skill levels. Activities included ground work only, and maneuvers began with haltering and leading to advanced maneuvering through obstacle courses. Horses were blindly scored by the same 3 observers using a 15 cm line scale ranging from obedient (1) to resistant (15). Horses were then placed into 2 groups (Difficult, Easy) based on average difficulty of training, and the aggression scores were compared across the 2 groups using the PROC GLM procedure in SAS 9.2. Correlations were calculated between the average aggression and training scores for all test horses. Results from the training scores showed a difference ( $P < 0.01$ ) between the 2 selected groups, indicating that the separation of the groups was valid. However, no difference was detected ( $P = 0.22$ ) in aggression scores between the Difficult and Easy training groups. This illustrates that the aggression of a horse in a herd or in establishing a social order may not be a good indicator of the difficulty to subsequently train the horse. This is further demonstrated in the weak correlation ( $r = 0.408$ ) between the average aggression and training scores for the test horses. While in establishing social order aggression may be observed, but it is not the sole factor in determining dominance in a herd. Most likely the trait of dominance may be more important in terms of trainability than aggression and should be the focus of future behavior research.

**Key Words:** horse, behavior, training

**T245 The occurrence of different mycotoxins (aflatoxins, fumonisins, zearalenone, ochratoxin, deoxynivalenol, ergot alkaloids) in horse feed.** Nicole Reisinger\*<sup>1</sup>, Paula Kovalsky<sup>2</sup>, Verena Starkl<sup>2</sup>, Simone Schaumberger<sup>2</sup>, Michael Sulyok<sup>3</sup>, and Ursula

Hofstetter<sup>2</sup>, <sup>1</sup>*Biomin Research Center, Tulln, Austria*, <sup>2</sup>*Biomin Holding GmbH, Herzogenburg, Austria*, <sup>3</sup>*Center for Analytical Chemistry, Department for Agrobiotechnology (IFA-Tulln), University of Natural Resources and Life Sciences (BOKU), Tulln, Austria*.

Mycotoxins are toxic secondary metabolites of fungi, which can be found in cereal grains and animal feeds. The occurrence of mycotoxins in feed of livestock animals is well investigated. In contrast, only limited information is available on the occurrence of mycotoxins in horse feed as well as on their effects on horse health. The aim of this study investigate the occurrence of the mycotoxins aflatoxins (Afla), fumonisins (FUM), zearalenone (ZEN), ochratoxin A (OTA), deoxynivalenol (DON) and ergot alkaloids in horse feed in Europe. Hay, straw and concentrate feed samples (n = 135) were collected from different countries of Europe in 2014. Samples were analyzed by liquid chromatography – tandem mass spectrometry (Streit et al., 2013). In total, mycotoxins could be found in 83% of all tested samples. Ergot alkaloids (e.g., ergotamine, festuclavine) were detected in 63% of the samples and therefore represented the most frequently occurring mycotoxin in our survey. In the individual samples, concentrations up to 1,230 µg/kg were determined. In addition, a high prevalence of DON (60%) and ZEN (54%) was observed. Maximum concentrations of 3,800 µg/kg and 200 µg/kg were measured for DON and ZEN, respectively. FUM was detected in 16% of the feed samples, maximum concentration measured was 330 µg/kg. In contrast, Afla (1.5%) and OTA (3%) were detectable in only a few of the feed samples. Results of this study clearly show that mycotoxins are frequent contaminants of horse feed, although usually only high quality feed is given to horses. There are only scattered reports on the effects of mycotoxins on horses. However, it is known that horses are extremely sensitive to FUM (Caloni et al., 2011) and also negative effects of ZEN have been reported (Minervini et al., 2006). Furthermore, ergot alkaloids are described to induce dystocia, agalactia and lameness in horses (Cross et al., 1995; Douthit et al., 2012). Therefore, an effective mycotoxin risk management program can help to protect horses against negative effects of mycotoxins.

**Key Words:** horse, equine, mycotoxins

**T246 Effectiveness of a brewer's yeast supplement with or without fat for performance horses.** Jeneva R. Seidl<sup>\*1</sup>, Torea L. Bova<sup>1</sup>, J. Latham Brister<sup>1</sup>, Lauren B. Hodge<sup>1</sup>, Angela R. Mays<sup>2</sup>, and Brian J. Rude<sup>1</sup>, <sup>1</sup>*Mississippi State University, Starkville, MS*, <sup>2</sup>*F.L. Emmert Company, Cincinnati, OH*.

The objective of the current trial was to evaluate the effect of additional fat to a brewer's yeast supplement on hoof, coat, and body condition of performance horses. Twelve performance geldings randomly allotted to one of 3 dietary treatments: 1) a commercially available horse feed (10% CP, 4.5% fat) at 0.9% BW/d; 2) diet 1 plus a brewer's yeast product at 113g/d; 3) diet 2 plus vegetable oil at 5% of the diet. Diet 1 was given to meet basic nutrient requirements of 10% CP and 0.6% LYS. All geldings were fed half of their diet treatment twice a day for 84 d. Geldings had ad libitum access to bermudagrass pasture and hay throughout the trial. Body weight (BW) measurements and body evaluations were collected at initiation of the trial and every subsequent 28 d until 84 d. Body evaluations included coat condition, body condition score (BCS), and hoof condition. Coat and hoof condition were evaluated on scale ranging from 1 to 5 (1 reflecting poor or damaged and 5 reflecting glossy) accounting for condition, texture and appearance. Body condition was based on the standard BCS scale of 1 to 9. Four total evaluations were taken during the experiment, an initial evaluation and then every 28 d. Data were subjected to ANOVA using the GLM procedures of SAS. No effect of diet was found for hoof (3.5, 3.4, and

3.1;  $P = 0.6207$ ), coat (3.3, 3.3, and 3.3;  $P = 0.0826$ ), or BCS (5.3, 5.6, and 5.6;  $P = 0.9967$ ) for diets 1, 2, and 3, respectively. Body weights were not different (509, 524, and 560 kg;  $P = 0.4602$ ) among diets 1, 2, and 3, respectively, nor was the change in body weight during the trial different (1.9, -6.8, and 4.5 kg, respectively;  $P = 0.6815$ ). Addition of fat to brewer's yeast supplement did not enhance body scores or weight change. Diet 1 (basal diet fed to all treatments) was a concentrate based supplement containing a large amount of available energy. Feeding diet 1 at 0.9% BW/d may have masked the effects of increased energy from fat. Further research should be conducted to evaluate brewer's yeast product and fat with horses while being fed a less nutritious basal diet.

**Key Words:** equine, brewer's yeast, fat supplementation

**T247 Effectiveness of a brewer's yeast supplement with or without fat for weanling horses.** J. Latham Brister<sup>\*1</sup>, Torea L. Bova<sup>1</sup>, Jeneva R. Seidl<sup>1</sup>, Lauren B. Hodge<sup>1</sup>, Angela R. Mays<sup>2</sup>, and Brian J. Rude<sup>1</sup>, <sup>1</sup>*Mississippi State University, Starkville, MS*, <sup>2</sup>*F.L. Emmert Company, Cincinnati, OH*.

The objective of this study was to evaluate the effect of additional fat to a brewer's yeast supplement on hoof, coat, and body condition of weanling horses. Twelve Quarter Horse weanlings (7 colts and 5 fillies) were utilized in an 84 d feeding trial using 3 dietary treatments: 1) a commercially available horse feed (10% CP, 4.5% fat) at 0.9% BW; 2) diet 1 plus a brewer's yeast product at 113g/d; 3) diet 2 plus vegetable oil at 5% of the diet. Weanlings were fed half of their ration twice daily (0600 and 1800 h) with all 3 treatments receiving the diet 1 portion of their treatment at 0.9% BW/d, to meet NRC requirements based on forage analysis. Body weight (BW) measurements and body evaluations were collected at initiation of the trial and every subsequent 28 d until 84 d. Body evaluations included coat condition, BCS, and hoof condition. Coat and hoof condition were evaluated on a scale ranging from 1 to 5 (1 reflecting poor or damaged and 5 reflecting glossy), with coat evaluation scale accounting for condition, texture, and appearance. Body condition score was based on the standard BCS scale of 1 to 9. During the first 7 d post weaning, weanlings were divided into groups of 3 (1 weanling randomly assigned to each treatment) and housed together in open lot to reduce stress. Weaned groups were offered ad libitum access to hay and water while introduced to the diet 1 portion of their dietary treatment. Diets were determined as a percent of the weanling's BW, which was adjusted every 28 d. Weanlings were also provided ad libitum access to grass, bermudagrass hay, and water. Data were analyzed through ANOVA using the GLM procedures of SAS. As expected, BW increased throughout the trial for all 3 treatments. After 28 d foals on diet 2 and 3 had increased BW ( $P = 0.0102$ ; 211 and 207 kg, respectively) compared with foals on diet 1 (194 kg). There were no differences for BCS ( $P = 0.2768$ ), coat score ( $P = 0.3243$ ), or hoof score ( $P = 0.5014$ ). Supplementation with brewer's yeast increased BW gains, but additional fat did not affect horses on the concentrate diet. In conclusion, added fat did not increase brewer's yeast affects in weanling horses.

**Key Words:** equine, brewer's yeast, fat supplementation



# International Animal Agriculture

**T248 Proposal for standard methods and procedure for guinea pig carcass evaluation, jointing, and tissue separation.** Davinia Sánchez-Macias\*<sup>1</sup>, Noemi Castro<sup>2</sup>, Miguel Rivero<sup>3</sup>, Anastasio Argüello<sup>2</sup>, and Antonio Morales-delaNuez<sup>4</sup>, <sup>1</sup>*Department of Agro-industrial Engineering, Universidad Nacional de Chimborazo, Riobamba, Chimborazo, Ecuador*, <sup>2</sup>*Animal Science Department, Universidad de Las Palmas de Gran Canaria, Arucas, Las Palmas, Spain*, <sup>3</sup>*Department of Veterinary Histology and Pathology, Universidad de Las Palmas de Gran Canaria, Arucas, Las Palmas, Spain*, <sup>4</sup>*Facultad de Ciencia Pecuarias, Escuela Superior Politécnica de Chimborazo, Riobamba, Chimborazo, Ecuador*.

The South American guinea pig rodent has become a livestock animal acceptable for human consumption in different parts of the world. Its white meat has a great potential as a new protein source, and its social and economic importance for different human populations is considered key for development. Scarce data are found in the literature when the statistical livestock information is checked, and few researches have been done about morphological characteristics of guinea pigs carcasses. These works do not follow the same procedures, using different criteria, overall the jointing, making it difficult to compare different studies. The aim of the present study is to suggest a practical and normalized method to analyze the guinea pig carcass characteristics allowing their evaluation. It describes the main traits to be considered from the birth of the animal to the carcass analysis. This work concerns (1) growth, feeding, pre-slaughter and slaughter processing, (2) method for the definition, hanging and presentation of the carcass, (3) carcass morphological characteristics, (4) jointing procedure based on 4 anatomically defined regions, (5) methods for 20 evaluating meat pH and color and (6) method for tissue separation. This proposal could be useful to compare data of these animals under different conditions.

**Key Words:** guinea pig, carcass, jointing

**T249 Cow-calf production performance under different management systems in Thailand.** Jirayut Khemsawat<sup>1</sup>, Skorn Koonawootrittriron\*<sup>1</sup>, Thanathip Suwanasopee<sup>1</sup>, and Mauricio A. Elzo<sup>2</sup>, <sup>1</sup>*Kasetsart University, Bangkok, Thailand*, <sup>2</sup>*University of Florida, Gainesville, FL*.

The numbers of beef cattle and cow-calf producers in Thailand are decreasing every year due to a sharp increase in demand and high prices offered for all types of cattle by neighboring countries. To help evaluate the extent of this problem, an assessment of the current status of cow-calf production in Thailand is needed. Thus, the objective of this study was to compare the cow-calf production performance and profitability in 3 regions of Thailand (UN = upper Northeast region; LN = lower Northeast region, CT = Central region). The factors considered were number of cows per farm, paddock size, number of laborers, birth weight, weaning weight, sale age, calving interval, costs and profits. Data for these factors were gathered from 501 cow-calf producers (130 producers in UN, 341 producers in LN, and 30 producers in CT) using questionnaires, interviews, visits, and seminars. Means and SD were used to describe factors. Least squares means (LSM) for all factors in each region were computed using a linear model that included region (UN, LN, CT) and farm size (small: less than 10 cows; medium: 11 to 20 cows; large: more than 20 cows) as fixed effects, and residual as a random effect. Cow-calf producers had from 7.7 (UN) to 12.9 (CT) cows per farm, paddock size ranged from 1.3 (UN) to 2.1 (CN)

ha, and hired 1.9 (LN) to 2.2 (UN) laborers for their operations. Calf birth weights ranged from 24.3 (UN) to 29.9 (CT) kg, weaning weights ranged from 151.9 (LN) to 193.4 (CT) kg, weaning ages from 7.8 (CT) to 8.6 (UN) mo, and sale ages ranged from 11.7 (CT) to 13.5 (LN) mo. Calving intervals were from 12.6 (LN) to 13.7 (UN) mo. Factor LSM differed among regions ( $P < 0.05$ ), except for weaning age, sale age and calving interval. Cow-calf producers in CT had higher profits and lower costs than UN and LN ( $P < 0.05$ ). These results suggested the need for different cow-calf production strategies would need to be implemented in each Thai region improve productivity and profitability in a sustainable manner.

**Key Words:** cattle, cow-calf production, cost

**T250 Chemical composition, physical parameters and cholesterol status of fertile and unfertile eggs from chicken and quail birds.** Patience Olusola Fakolade\*, *Osun State University, Ejigbo, Osun State, Nigeria*.

A study to compare the chemical composition, physical parameters and cholesterol status of fertile and unfertile eggs from chicken and quail bird was evaluated. One hundred twenty Harco black and 120 Japanese quail birds were used for the study. Birds were fed with compounded feeds and divided into 2 groups, group A had 60 birds each from chicken and quail to produce unfertile eggs while group B had 55 hen and 5 cocks each from chicken and quail, to produce fertile eggs. All the birds were subjected to appropriate vaccination and medication procedures. Both eggs (fertile and unfertile) were evaluated for their variations in chemical composition, physical status and cholesterol levels in a  $3 \times 3$  factorial design. Results for chemical composition, showed that, unfertile chicken eggs had highest significant values ( $P < 0.05$ ) of (Ca, P, Na, Zn, Fe and K), than for fertile chicken eggs, unfertile quail eggs, and fertile quail eggs. The fertile quail eggs had significant higher value for (Na, Zn and Fe) but lower value for (Ca, P, and K) than the unfertile eggs. In this study, the physical characteristics measured followed the same trend as that of the chemical composition except for the shell weight and yolk color. Quail eggs for both fertile and unfertile eggs had lowest significant value ( $P < 0.05$ ) for cholesterol (10 mg/100 g and 36.49 mg/100 g) and viscosity level (16.20 mL/s and 28.90 mL/s), than for chicken eggs with (1150 mg/100 g and 1291.20 mg/100 g) and the viscosity level had (55.60 mL/s and 69.20 mL/s). Quail eggs had lower cholesterol level than chicken eggs and as such could be incorporated into human diet to increase protein intake and reduces cholesterol accumulation in the diet, through consumption of egg.

**Key Words:** chicken egg, quail egg, fertile egg

**T251 Correlation between the guinea pig tissue composition and carcass measurements.** Iván Barba<sup>1</sup>, Lida Barba\*<sup>1,2</sup>, César Hernández<sup>1</sup>, Julio Palmay<sup>1</sup>, Nibaldo Rodríguez<sup>2</sup>, and Davinia Sánchez-Macias<sup>1</sup>, <sup>1</sup>*Department of Agroindustrial Engineering, Universidad Nacional de Chimborazo, Riobamba, Chimborazo, Ecuador*, <sup>2</sup>*School of Informatics Engineering, Pontificia Universidad Católica de Valparaíso, Valparaíso, Chile*.

Combining carcass measurements, such as weights, conformation and fat classes and carcass dimensions would lead to moderate predictions of muscle, fat or bone content in carcass. A correlation analysis of these parameters is necessary to build up potential prediction equations. This

study was performed to establish correlations between guinea pig carcass composition and measurements. The carcasses of 40 guinea pigs, males and females of 3 and 12 mo age (fattening and discarded animals, respectively) with similar management previous to slaughter were used. Seventeen measurements consistent in weight, yield, and the length of carcass pieces were analyzed to identify the major correlation with the 9 variables of tissue composition. The tissue composition is represented as subcutaneous fat (SF), intermuscular fat (IF), muscle (M), bone (B), skin (S), waste (W), and 3 combined variables SF-IF, B-W, and muscle plus freeze loss (M-FL). In all the cases, an important positive correlation with live weight at slaughter (LWS), empty body weight (EBW), hot carcass weight (HCW), and cold carcass weight (CCW) was found. The variables related to fat were highly correlated with the measurement perirenal and pelvic fat (PPF). The linear measurements internal carcass length (L), external carcass length (ECL), lumbar circumference (LC), and thorax circumference (ThC) showed high correlation with the tissue composition variables that are not related to fat. The major correlation was found in the relation M-LWS, M-EBW, M-HCW, M-CCW with a Pearson Coefficient of  $R = 95\%$ , followed by the relation S-LWS, S-EBW, S-HCW, S-CCW with  $R = 94\%$ . Some measurements that are not correlated with any variable are hind limb length (F), buttock width (BW), and thorax width (ThW).

**Key Words:** guinea pig, carcass composition, correlation

**T252 Prediction of guinea pig carcass tissue composition from weight and linear normalized measurements.** Lida Barba\*<sup>1,2</sup>, Iván Barba<sup>1</sup>, Julio Palmay<sup>1</sup>, César Hernández<sup>1</sup>, Nibaldo Rodríguez<sup>2</sup>, and Davinia Sánchez Macías<sup>1</sup>, <sup>1</sup>Department of Agroindustrial Engineering, Universidad Nacional de Chimborazo, Riobamba, Chimborazo, Ecuador; <sup>2</sup>School of Informatics Engineering, Pontificia Universidad Católica de Valparaíso, Valparaíso, Chile.

The evaluation of carcass composition makes possible to assign a value for the carcass. It is possible to predict carcass composition by measurements taken on the carcass: weights, carcass yield, fatness, conformation and other non-invasive methods. The use of these measurements offers an advantage over joint dissection, as they are faster and easier to make and do not involve any loss of carcass tissue. The aim of this work is to develop equations to predict the tissue composition of guinea pig carcasses, by using weight, yield and linear measurements. The data were obtained after a practical and normalized method to measure the guinea pig carcass, based on anatomical guidelines. The carcass measurements of 40 South American Guinea pigs, males and females of 3 and 12 mo age (fattening and discarded animals, respectively) were used for multiple regression analysis; which was implemented from the correlation matrix of dependent and independent variables and the significance test. The independent variables or predictors used were live weight at slaughter (LWS), hot carcass weight (HCW), perirenal and pelvic fat (PPF), external carcass length (ECL), and thorax circumference (ThC). The accuracy of the predictions was evaluated with root mean square error (RMSE) and coefficient of determination ( $R^2$ ). The prediction equations for carcass composition in grams were more accurate than those related to composition proportion. In the prediction of some variables in grams such as total muscle (TM), skin (S) and muscle + freeze-thawing loss (MpFL) were obtained values of  $R^2 = 95\%$ , for total bone (TB) was obtained an  $R^2 = 90\%$ , and for subcutaneous fat (SF) and total fat (TF) were obtained  $R^2 = 78\%$  and  $76\%$ , respectively. Lean (or muscle) and fatness are both the most important commercial components of a

carcass. In this case, PPF was a good predictor for guinea pig carcass fatness, whereas HCW and ThC were good predictors of lean content.

**Key Words:** guinea pig, carcass composition, prediction

**T253 Comparison between a method based upon the anatomical structure and a method based upon standardized butchering practice of guinea pig carcass.** Julio Palmay<sup>1</sup>, César Hernández<sup>1</sup>, Iván Barba<sup>1</sup>, Roberto Remache<sup>1</sup>, Antonio Morales de la Nuez<sup>2</sup>, Anastasio Argüello<sup>3</sup>, Noemí Castro<sup>3</sup>, and Davinia Sánchez Macías\*<sup>1</sup>, <sup>1</sup>Department of Agroindustrial Engineering, Universidad Nacional de Chimborazo, Riobamba, Chimborazo, Ecuador; <sup>2</sup>Facultad de Ciencia Pecuarías, Escuela Superior Politécnica de Chimborazo, Riobamba, Chimborazo, Ecuador; <sup>3</sup>Animal Science Department, Universidad de Las Palmas de Gran Canaria, Arucas, Las Palmas, Spain.

A diversity of methods for jointing meat animal carcasses exists in the literature and in the technical regulations in different countries. Standardized methods with good repeatability are needed for researching works. The objective of this work is to determine a representative cut for tissue carcass composition of guinea pig from 2 different jointing methods. Forty guinea pig carcasses were selected: 20 from 3-mo-age fattening animals (10 males and 10 females) and 20 from 12-mo-age breeding discarded animals (10 males and 10 females). Each carcass was divided into 2 half carcasses; the right half carcass was jointed in 2 cuts following a technical standard commercial regulation from Peru (forequarter and hindquarter), and the left half carcass was jointed in 4 cuts following anatomical points (neck, shoulder, hind leg and ribs). Each cut was dissected in skin, muscle, sub-cutaneous fat, inter-muscular fat, waste and bone. Total fat was calculated as the sum of sub-cutaneous and inter-muscular fat, inedible section as the sum of bones and remainder, and muscle + freeze-thawing loss. All results are presented in proportion to the cut. A factorial ANOVA procedure was used with the statistical program SAS (v. 11), for each tissue comparing the different cuts and total tissue in the carcass, and second, the effect of the animal group. In some animal groups, the neck differed from the carcass in terms of proportion for sub-cutaneous and inter-muscular fat tissues, total fat and bones. On the other hand, the muscle proportion in the hindquarter cut in 12 mo-age female guinea pigs was higher respect to the muscle proportion in the whole carcass. The other studied cuts were similar in terms of proportions of the different tissues respect to the whole carcass composition. However, it is noted that the closest cut to the tissue composition of the guinea pig carcass, in percentage values, was the hind leg cut. We conclude that the most representative cut of guinea pig carcass is the hind leg, and thus the jointing method proposed in 4 cuts is the most appropriated for these animals.

**Key Words:** carcass tissue composition, guinea pig, jointing

**T254 Carcass tissue composition in fattening or discarded guinea pigs.** Julio Palmay<sup>1</sup>, Iván Barba<sup>1</sup>, César Hernández<sup>1</sup>, Erick Ureña<sup>1</sup>, Antonio Morales de la Nuez<sup>2</sup>, Noemí Castro<sup>3</sup>, Anastasio Argüello<sup>3</sup>, and Davinia Sánchez Macías\*<sup>1</sup>, <sup>1</sup>Department of Agroindustrial Engineering, Universidad Nacional de Chimborazo, Riobamba, Chimborazo, Ecuador; <sup>2</sup>Facultad de Ciencia Pecuarías, Escuela Superior Politécnica de Chimborazo, Riobamba, Chimborazo, Ecuador; <sup>3</sup>Animal Science Department, Universidad de las Palmas de Gran Canaria, Arucas, Las Palmas, Spain.

The development of tissue composition during growth may be reflected in the carcass tissue composition due to age at slaughter and sex. For

this reason, the aim of this work is to analyze the carcass composition in 6 different carcass cuts, in males and females fattening or discarded guinea pigs. Forty guinea pig carcasses were selected: 20 from 3-mo-age fattening animals (10 males and 10 females) and 20 from 12-mo-age breeding discarded animals (10 males and 10 females). Each carcass was divided into 2 half carcasses; the right half carcass was jointed in 2 cuts following a technical standard commercial regulation from Peru (forequarter and hindquarter), and the left half carcass was jointed in 4 cuts following anatomical points (neck, shoulder, hind leg and ribs). Each cut was dissected in skin, muscle, sub-cutaneous fat, inter-muscular fat, waste and bone. Total fat was calculated as the sum of sub-cutaneous and inter-muscular fat, inedible section as the sum of bones and remainder, and muscle + freeze-thawing loss. All results are presented in tissue weights of the cut or whole carcass. A factorial ANOVA procedure was used with the statistical program SAS (v. 11), comparing among the different cuts, and analyzing the effect of age and sex. For the whole carcass composition, 3 mo-age guinea pigs showed the lowest fat content and female 12 mo-age animals the highest value. For muscle, bones, skin and waste, 12 mo-age guinea pigs had higher values in carcass than the younger animals, and no differences were found due to the sex. Into the 4 cuts jointing method, long leg and ribs cuts had the higher values of fat, muscle, bones and skin, while ribs presented the highest waste content. And into the commercial jointing method, the forequarter showed the highest contents of all the analyzed tissues. We conclude that tissue composition in the whole carcass is different in fattening or discarded guinea pigs, except for the fat content, in this case, 3 mo-age females have similar values than discarded guinea pigs.

**Key Words:** carcass tissue composition, guinea pig, jointing

**T255 Substitution of commercial concentrate by mulberry forage (*Morus alba* ‘Linn’) in the ration of fattening tropical Pelibuey lambs.** Andrés Alpízar-Naranjo<sup>1</sup>, Javier Arece-García<sup>2</sup>, Marcos Esperance<sup>2</sup>, Yoel López<sup>1</sup>, Michael Molina<sup>2</sup>, and Eliel González-García<sup>3</sup>, <sup>1</sup>Escuela de Ciencias Agrarias, Facultad de Ciencias de la Tierra y el Mar, Universidad Nacional de Costa Rica, Heredia, Costa Rica, <sup>2</sup>Estación Experimental de Pastos y Forrajes “Indio Hatuey,” Matanzas, Cuba, <sup>3</sup>INRA UMR868, Systèmes d’Élevage Méditerranéens et Tropicaux (SELMET), Montpellier Cedex 2, France.

The effects of substituting concentrate by chopped and fresh home-ground mulberry forage were evaluated in an experiment with fattening lambs lasting 126 d from weaning. The basal forage diet was composed by *Pennisetum purpureum*, *Panicum maximum* ‘Likoni’ and sugarcane. Forty-eight entire Pelibuey lambs (initial BW 20.6 kg) were randomly allocated in 4 experimental groups according to treatments which consisted on the level of supplementing (or not) the basal diet with mulberry forage: M-1: mulberry at 1% of BW (DM basis); M-0.75: mulberry at 0.75% plus 0.1 kg/lamb/d of concentrate; M-0.50: mulberry at 0.50% plus 0.2 kg of concentrate; and M-0: supplementing basal diet with 0.3 kg of concentrate (Control). Average daily gain (ADG, g/d) was significantly affected ( $P < 0.01$ ) by the supplementation regimen with M-1 lambs yielding the lowest ADG (100.2 g/d). No differences in ADG among the other groups were observed (124.7; 125.4 and 128.9 g/d for the treatments M-0.75, M-0.50 and M-0, respectively). Feed conversion was 11.6, 9.9, 10.3 and 9.7 kg DM/ kg of BW gain for M-1, M-0.75, M-0.50 and M-0 lambs, respectively. Differences in final BW at slaughtering and hot or cold carcass yields were coherent with those found in growth performance. Supplementing with mulberry forage at 0.75% of BW was the feeding alternative showing the best productive response. Results are shown in Table 1.

**Table 1 (Abstr. T255).** Performance data

Item	Diet				SEM
	M-1	M-0.75	M-0.50	M-0	
Initial BW, kg	20.5	20.7	20.7	20.7	0.38
Final BW, kg	29.6 <sup>b</sup>	32.2 <sup>a</sup>	32.3 <sup>a</sup>	32.8 <sup>a</sup>	0.57
BW change, kg	9.1 <sup>b</sup>	11.5 <sup>a</sup>	11.6 <sup>a</sup>	12.1 <sup>a</sup>	—
ADG, g/lamb/d	100.2 <sup>b</sup>	124.7 <sup>a</sup>	125.4 <sup>a</sup>	128.9 <sup>a</sup>	3.08
Total feed intake, g DM/lamb/d	835.8 <sup>b</sup>	908.2 <sup>a</sup>	953.7 <sup>a</sup>	930.0 <sup>a</sup>	7.0
Average feed intake, g DM/kg BW	33.4	34.3	36.0	34.8	—
Average feed intake, g DM/kg BW <sup>0.75</sup>	74.6	77.9	81.7	79.1	—
Feed conversion, kg DM/kg BW gain	11.6 <sup>a</sup>	9.9 <sup>c</sup>	10.28 <sup>b</sup>	9.70 <sup>c</sup>	—
Hot carcass weight, kg	11.5 <sup>b</sup>	13.3 <sup>ab</sup>	13.5 <sup>a</sup>	13.7 <sup>a</sup>	0.30
Cold carcass weight, kg	11.2 <sup>b</sup>	13.0 <sup>ab</sup>	13.2 <sup>a</sup>	12.8 <sup>ab</sup>	0.28
Gastrointestinal tract weight, kg	8.5	9.4	7.7	7.9	0.25
Offal’s weight, kg	0.8	1.0	1.0	0.9	0.03
Perirenal and mesenteric fat, kg	0.95	1.2	0.93	1.12	0.09
Hot carcass yield, %	40.2 <sup>b</sup>	41.0 <sup>ab</sup>	43.2 <sup>a</sup>	43.4 <sup>a</sup>	0.45
Cold carcass yield, %	39.3	40.2	42.2	40.8	0.57

<sup>a,b</sup>Values with different superscripts in the same row differ at  $P < 0.05$  (Tukey test).

**Key Words:** fattening tropical lamb, concentrate substitution, mulberry forage supplementation

**T256 Carcass quality of guinea pigs: Sex effect on fattening or discarded animals.** César Hernández<sup>1</sup>, Iván Barba<sup>1</sup>, Julio Palmay<sup>1</sup>, Lisbeth Medina<sup>1</sup>, Antonio Morales de la Nuez<sup>2</sup>, Noemí Castro<sup>3</sup>, Anastasio Argüello<sup>3</sup>, and Davinia Sánchez Macías<sup>\*1</sup>, <sup>1</sup>Department of Agroindustrial Engineering, Universidad Nacional de Chimborazo, Riobamba, Chimborazo, Ecuador, <sup>2</sup>Facultad de Ciencia Pecuarías, Escuela Superior Politécnica de Chimborazo, Riobamba, Chimborazo, Ecuador, <sup>3</sup>Animal Science Department, Universidad de las Palmas de Gran Canaria, Arucas, Las Palmas, Spain.

Guinea pigs are used for meat production in South America, Africa, and Asia. The increased interest in this product is due to the low production price, and because of their relatively rapid reproduction and the large litter size. The objective of the present study was to evaluate the effect of age and sex on weight, yield, drip loss and pH evolution in guinea pig carcasses. Forty guinea pigs (10 male and 10 female of 3 mo age, and 10 male and 10 female of 12 mo age discarded animals) were used. The animals were fasted during 14 h before slaughter. Live weight at slaughter (LWS), empty body weight (EBW), and hot and cold carcass weights (HCW and CCW) were recorded. pH evolution was measured at 15, 45 min and 24 h after slaughter in *Longissimus dorsi* (LD) and *Psoas major* (PM) muscles. Four different carcass yields and drip loss (in grams and percentage) were calculated. Twelve mo-age animals showed higher LWS, EBW, HCW and CCW than 3 mo-age guinea pigs. The lowest values of hot or cold carcass yields (HCY, CCY) were observed in 3 mo-age male guinea pigs and no differences were found among the other groups. CCY was lower than HCY in all cases, due to the drip loss during the chilling time (24h at 4°C). When yields were presented related to empty body weight, the values were higher than



the yields that used live weight at slaughter, as expected. Discarded animals had higher drip losses, but expressed in percentage, 3 mo-age females had the highest losses, while the 12 mo guinea pigs showed the lowest losses. pH declined significantly during 24h in both muscles and all the animals, with the exception of 12 mo-age female guinea pigs. At 15 and 45 min LD did not show differences due to sex or age. After 24h, 12 mo-age females had the highest pH values (6.37) and 3 mo-age females had the lowest (5.54). When pH in PM was analyzed, the results revealed different behavior than LD. At 15 min and 24 h, 12 mo females presented the lowest pH (6.29 and 6.30, 15 min and 24 h, respectively), while the in the other groups pH ranged from 6.70 to 6.44 at 15 min, and was near to 6 after 24h postmortem.

**Key Words:** carcass quality, sex, guinea pig

**T257 Comparison of linear carcass measurements in male or female fattening and discarded guinea pigs.** César Hernández<sup>1</sup>, Julio Palmay<sup>1</sup>, Iván Barba<sup>1</sup>, Jairo Espinoza<sup>1</sup>, Antonio Morales de la Nuez<sup>2</sup>, Noemí Castro<sup>3</sup>, Anastasio Argüello<sup>3</sup>, and Davinia Sánchez Macías<sup>\*1</sup>, <sup>1</sup>Department of Agroindustrial Engineering, Universidad Nacional de Chimborazo, Riobamba, Chimborazo, Ecuador; <sup>2</sup>Facultad de Ciencia Pecuarias, Escuela Superior Politécnica de Chimborazo, Riobamba, Chimborazo, Ecuador; <sup>3</sup>Animal Science Department, Universidad de las Palmas de Gran Canaria, Arucas, Las Palmas, Spain.

Linear carcass measurements (LCM) are important variable of carcass quality. Several studies describe a relationship between LCMs and body live weight and other parameters of carcass and meat quality in different species. Guinea pigs are an important source of protein in numerous undeveloped countries and currently they are introducing in developed countries. The present study analyzes the effect of sex and fattening or discarded guinea pigs on LCMs carcass. Forty guinea pig carcasses after 24 h of chilling were selected: 20 from 3months-age fattening animals (10 males and 10 females) and 20 from 12 mo-age breeding discarded animals (10 males and 10 females). The following measurements in carcasses after chilling for 24 h at 4°C were recorded: carcass length (L), loin length (Lo), hind limb length (F), width of the buttocks (G), lumbar circumference (LC), thorax circumference (ThC) and thorax width (ThW). Furthermore carcass compactness (CarC) and leg compactness (LegC) were calculated. L, Lo, LC and ThC were higher in 12 mo-age than 3 mo-age guinea pigs. Sex did not affect the described measurements, and G values were similar (7 cm) in all studied animals. F was higher in 12 mo-age males (12 cm), while 3 mo-age females showed the smaller value (9 cm). There was a sex effect on F for 12 mo animals, being higher in males than in females (12 vs 10 cm, respectively). When CarC was analyzed an age effect was evident in 12 mo-age animals, which showed 40g cm<sup>-1</sup> of carcass weight vs 25–30g cm<sup>-1</sup> for the 3 mo-age guinea pigs. So, after 3 mo the animals not only increased their length, but they also increased their body mass. Sex had an important effect on LegC of fattening guinea pigs: males had higher values than females (67% and 55%, respectively). However, in discarded guinea pigs, males showed smaller compactness than females (50% and 60%, respectively).

**Key Words:** linear carcass measurement, carcass quality, guinea pig

**T258 Fatty acid composition of fats from female and male muskoxen (*Ovibos moschatus*) living in western Greenland.** Susana P. Alves<sup>1</sup>, Angelo Cabo<sup>1</sup>, Katrine Raundrup<sup>2</sup>, Rui J. B. Bessa<sup>1</sup>, and André M. de Almeida<sup>\*3</sup>, <sup>1</sup>CIISA/FMV - Centro Interdisciplinar de Investigação em Sanidade Animal, Faculdade de Medicina

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Muskoxen (*Ovibos moschatus*) are large ruminants living in Arctic habitats, and their diet is usually dominated by grasses, sedges and dicots. Muskoxen have an exceptional ability to fatten during spring and summer to support their energy needs during the arctic winter. However, little is known about the fatty acid (FA) composition of muskox meat, adipose tissue and liver. The objective of this study is to characterize the FA composition in samples from muskox females and males from Western Greenland. Muskox samples (muscle, adipose tissue and liver) from adult female (n = 12) and male (n = 8) were collected around Kangerlussuaq (West Greenland) during the winter hunting season of 2014. Samples were freeze-dried and lipids were extracted using dichloromethane and methanol (2:1, v/v). Total lipids were measured gravimetrically. Fatty acid methyl esters (FAME) were prepared from the lipid extracts with sodium methoxide in methanol followed by hydrochloric acid in methanol. FAME were quantified by GC-FID using a Shimadzu GC-2010 Plus (Shimadzu, Kyoto, Japan) equipped with a TR-CN100 capillary column (100 m, 0.25 mm i.d., 0.20 µm film thickness, Teknokroma, Spain). FAME were expressed as g/100 g of total peaks and data were analyzed using the PROC MIXED of SAS (SAS Institute Inc., Cary, NC) with a model that included the sex (female vs. male) as the single effect. Muskox muscle, adipose tissue and liver contained about 396, 916 and 150 mg/g DM of total lipids, respectively. Females showed the highest fat content ( $P = 0.046$ ) compared with males, which might be an additional energy reserve for the late-winter lactation. More than 60 FA were detected in muskoxen samples, however, the 18:1cis-9 was the dominant FA in muscle and adipose tissue, presenting more than 43% and 32% of total FA, respectively. The great oleic acid concentration in muskoxen fats might indicate a higher lipogenic activity in these animals. Differences between sexes on the FA composition were minor in liver and more pronounced in adipose tissues. In general, males presented more levels of branched-chain FA, while female presented more oleic acid.

**Key Words:** musk ox, fatty acid profile

**T259 Body condition score, subcutaneous fat and reproductive performance relationship in *Bos indicus* cows in a tropical region of Mexico.** Ivette Rubio, Victoria Blas, Manuel D. Corro\*, Clara Murcia, and Carlos S. Galina, Faculty of Veterinary Medicine and Zootechnics National Autonomous University of Mexico, Mexico, DF, Mexico.

With the aim of evaluate the relationship between body condition, subcutaneous fat and reproductive performance in *Bos indicus* cattle in the Mexican tropics. Sixty multiparous cows were selected, with an average of 186.5 ± 90.8 d postpartum, synchronized with intravaginal device with natural progesterone. Body condition (BCS) scale 1–9, subcutaneous body fat (BF) by ultrasonography and live weight (LW) were measurement at 3 different times during a 90 d breeding period in all cows. The BF was defined as the subcutaneous layer of fat between the skin and fascia of the *longissimus dorsi* muscle and measured between the 2nd and 3rd spinous of the lumbar vertebrae. We assessed the level of progesterone blood at the beginning of the breeding season. The cows were grouped, according to the BCS, group1 (n = 25) cows with less than 4.5 and group 2 (n = 35) more than 4.5. An ANOVA was used to determine the differences between BCS, BF and LW. A linear correlation was performed to assess the association between BCS, BF and LW. At the beginning of the breeding season a larger ( $P < 0.05$ ) proportion of cows in group 2 were cycling, and responded to synchronization than

those in group 1. BF was higher ( $P < 0.05$ ) in group 2 than group 1,  $0.995 \pm 0.091$  mm and  $0.847 \pm 0.114$  mm, respectively. Although, there was a moderate positive relationship between BCS and BF, BCS can be a predictor of fat thickness as an indicator of body energy stored to maintain reproductive function. Because of this, Bos indicus cows with higher BF, at beginning of breeding season, would have a greater chance to become pregnant by the end of breeding season.

**Key Words:** body condition, subcutaneous fat, reproduction

**T260 Effects of diet type and a yeast product in performance, nutrient digestibility, intestinal morphology, carcass composition and visceral organ mass by guinea pigs.** Andres E. Idrobo, Karina M. Boada, Patricia X. Falconi, and Christian H. Ponce\*, *Departamento de Ciencias de la Vida y Agricultura, Universidad de las Fuerzas Armadas ESPE, Sangolquí, Ecuador.*

An experiment was conducted to evaluate the effects of a yeast culture/enzymatically hydrolyzed yeast product (Celmanax; Varied Industries Corp. Inc., Mason City, IA) and a diet type on performance, nutrient digestibility, intestinal morphology, carcass composition and visceral organ mass by guinea pigs. Eighty-eight guinea pigs (initial BW =  $453.21 \text{ g} \pm 21.90 \text{ g}$ ) were group fed in 24 pens during a 69 d feeding period. Treatments were arranged in a  $2 \times 2$  factorial and included a concentrate diet (C) or a mixture of forage and concentrate diet (M) and supplementation of Celmanax (Ce, 0 or 0.1% of diet DM). No type of diet  $\times$  addition of Ce interactions were detected in any variable measured ( $P \geq 0.157$ ). Animals fed the M treatment had greater CMS, ADG, and improved G:F during the feeding period ( $P < 0.001$ ). Supplementation of Ce had greater CMS and improved G:F during the first 49 d of the experiment ( $P \leq 0.048$ ). However, overall performance was not altered by Ce supplementation ( $P \geq 0.247$ ). Fat digestibility was greater for C ( $P < 0.001$ ), and crude fiber digestibility was decreased ( $P = 0.004$ ) compared with M. Nutrient digestibilities were not altered by Ce supplementation ( $P \geq 0.285$ ). Feeding M increased dressing percent, empty body weight, fractional mass of: liver, large intestine, portal drained viscera, and total splanchnic tissue ( $P \leq 0.019$ ) compared with C. Carcass characteristics and visceral organ mass were not altered by Ce supplementation. Fatty acid profile from adipose tissue was not different across treatments ( $P \geq 0.053$ ). The relation between n-3:n-6 fatty acids was greater for animal fed M compared with C ( $P = 0.019$ ). Villus height and crypt depth were increased by Ce supplementation ( $P \leq 0.019$ ). Animals fed M diet had greater crypt depth ( $P = 0.05$ ). Results indicate that feeding a mixture of forage and a concentrate diet results in positive effects in performance and carcass characteristics, and supplementation of Celmanax may have positive effects on performance and intestinal characteristics under both feeding regimens.

**Key Words:** diet type, guinea pig, yeast product

**T261 Energy flow analysis on dairy farms in North Carolina and Malawi.** Shalimbala Chizonda\* and Jonathan C. Allen, *North Carolina State University, Raleigh, NC.*

Milk production in developing and tropical countries is subject to different constraints than in the US. Even large, advanced dairies in Malawi have different inputs of energy and materials to optimize their milk output compared with moderate sized dairy farms in the US. The objective of this study was to compare energetic inputs, outputs, and efficiencies of dairy farms in Malawi (southeast Africa) and North Carolina to assess sustainable milk production. Feed consumption and milk production data were collected from Katete Dairy herd in Lilon-

gwe, Malawi, consisting of 408 cattle with 108 lactating cows, and the NCSU Dairy Research and Teaching Farm, housing 245 Holstein and 55 Jersey cows and 170 lactating cows. Both are teaching farms that employ extensive record keeping. Milk production averaged 34 and 19.5 kg/cow/day at NCSU and Katete, respectively. Feed energy intake was 85.8 Mcal/cow/d at NCSU and 135 Mcal/cow/d at Katete. Thus, return of feed energy as milk energy was 24.7% efficient at NCSU and 11.3% efficient in the lower producing cows at Katete. Manure is recycled for crop production at Katete, and is processed with a solids separation and lagoon system at NCSU. Additional factors that impact the overall farm efficiencies include inputs of petroleum fuels and electricity, and the number of calves, heifers and dry cows fed. The project identifies factors in dairy production that should be considered to improve efficiency, sustainability, and milk supply. Both farms had a substantial portion of the energy fed to the cows lost to the environment as composted manure in soils or lagoons that did not capture the heat or methane generated.

**T262 Nestlé China Dairy Farming Institute: Development of collaborative, science-based, practical, sustainable courses.** Karen Nielsen\*, Pamela Ruegg, and David Combs, *University of Wisconsin, Madison, WI.*

The Chinese dairy industry has expanded rapidly as the growing middle class demands higher protein diets. However, because there were not many dairy farms in China's traditional agricultural system, few farmers and farm managers have the knowledge and experience to run dairy farms using modern, science-based practices. To improve dairy science knowledge, Nestlé opened a comprehensive, collaborative, science-based Dairy Farming Institute (DFI) in 2014 that includes farms, laboratories, classrooms, living quarters, offices, and cafeteria in Shuangcheng, Heilongjiang Province, in northeastern China. The University of Wisconsin-Madison was chosen to lead DFI course development. Faculty, staff and affiliates of the UW-Madison work on all elements of curriculum design, program planning, establishment of learning objectives and core competencies, and evaluation of training programs and trainers. Courses are developed by teams of UW subject matter experts, industry partners and DFI trainers, and include instruction at basic, intermediate, and advanced levels for farm workers, supervisors and managers, and for dairy professionals such as veterinarians and nutritionists. Courses developed are practical, hands-on, and science-based with a goal of consistency in content, evaluation, and excellence in course delivery. Courses are offered many times and the trainers delivering the lectures, and leading the group case study work and hands-on activities in the barns or laboratories may be different from one course to the next. Therefore, materials must include clear instructions for future trainers and train the trainer sessions are provided before every new course. New trainers are evaluated, suggestions and advice are provided, and those who receive favorable evaluations continue to teach the DFI courses, making the program sustainable. Participating DFI partners include Alltech, Alta Genetics, Avery Weigh-Tronix, Boehringer Ingelheim, Eastrock, Elanco, Foester Technik, GEA, Goke/Storti, IFCN, Land O'Lakes, SCR, Zoetis, and Northeast Agricultural University in Harbin.

**Key Words:** dairy course development, university-industry partnerships

## Lactation Biology II

**T263 Risk factors for undergoing lactations >15 months in high-producing Holstein cows in a hot environment.** Jessica María Flores-Salas\*<sup>1</sup> and Miguel Mellado<sup>2</sup>, <sup>1</sup>Universidad Autónoma Agraria Antonio Narro Unidad Laguna, Torreón, Coahuila, México, <sup>2</sup>Universidad Autónoma Agraria Antonio Narro, Saltillo, Coahuila, México.

An epidemiological study of risk factors for involuntary extended lactations (15 to 40 mo) using a multiple variable logistic regression was carried out on 3278 high-yielding dairy cows in an intensive well-managed Holstein herd, milked 3 times daily in northern Mexico. Additional objectives were to assess the association of lactation length (15 to 40 mo) with milk yield and to assess the effect of multiple services (4 to  $\geq 14$ ) on pregnancy per artificial insemination. Also, a survival analysis was performed using the Cox proportional hazards model to test how the occurrence of pregnancy affects lactation length. Retained placenta (odds ratio (OR) = 1.3), metritis (OR = 1.8), ketosis (OR = 1.4), peak milk yield (<50 vs > 50 kg, OR = 1.4), temperature-humidity index at 60 d postpartum (<82 vs > 82 units, OR = 1.4), and 305-d milk yield (<11,000 vs > 11,000 kg, OR = 1.6) significantly increased the risk for lactations > 15 mo. Primiparous cows had less than half the risk of extended lactations (OR = 0.3) compared with multiparous cows. Once a cow had conceived, her risk of having a prolonged lactation dropped sharply ( $P < 0.01$ ). A strong linear association was found between lactation length and total milk yield for primiparous (450 to 1349 d in milk, maximum milk yield 37,852 kg;  $r = 0.71$ ) and pluriparous (450 to 1221 d in milk, maximum milk yield 38,021 kg;  $r = 0.75$ ). Pregnancy per artificial insemination (P/AI) in cows with extended lactation decreased linearly as number of services increased (P/AI = 50.5% for 4 services and 12.8% for  $\geq 14$  services). The data showed that well-managed Holstein cows milked 3 times daily were capable of lactating up to 40 mo with remarkable high persistency and with high milk yield at dry-off. Additionally, this study supports previous findings indicating that reproductive and metabolic disorders associated with calving are important risk factors for extended lactations, derived from the link of periparturient diseases with depressed reproduction in dairy cows.

**Key Words:** milk production, hot environment, extended lactations

**T264 Enhanced pre-weaning nutrition stimulates mammary gland development in dairy heifer calves.** Adam J. Geiger\*<sup>1</sup>, Robert E. James<sup>1</sup>, Catherine L. Parsons<sup>1</sup>, Anthony V. Capuco<sup>2</sup>, and Robert M. Akers<sup>1</sup>, <sup>1</sup>Department of Dairy Science, Virginia Polytechnic Institute and State University, Blacksburg, VA, <sup>2</sup>United States Department of Agriculture-Agricultural Research Service, Beltsville, MD.

It is established that feeding heifers a high plane of nutrition post-weaning but before puberty negatively affects mammary gland (MG) development and milk yield. The mass of the MG parenchyma (PAR) increases up to 60-fold from birth until 3 mo of age. Interestingly, enhanced nutrition during this time (pre-weaning) does not appear to negatively affect MG development and may in fact be beneficial. Therefore, the objectives of this study were to clarify the effects of feeding a higher plane of nutrition pre-weaning to Holstein heifer calves on MG growth, and particularly PAR and fat pad (MFP) development. Thirty-six Holstein heifer calves (<1 wk old) were reared on 1 of 2 dietary treatments to create 2 physiologically diverse groups of animals. Diets included (1) a control milk replacer (MR) fed at 454 g powder

per day (CON; 20% crude protein [CP], 20% fat), or (2) an accelerated MR fed at 1,135 g powder per day (ACC; 28% CP, 25% fat). Milk replacer mixed at 15% solids was fed as indicated until the 8th week when feeding was reduced 50% to induce weaning. Calves were housed individually with ad libitum access to water. Starter feeding was initiated at wk 5 and balanced between treatments. Udders were examined visually and by palpation with teats measured weekly. At weaning, a subset of calves were killed ( $n = 6/\text{diet}$ ). Whole MG were removed, dissected, and weighed. ACC calves consumed more MR liquid pre-weaning (4575 vs 3199 mL MR/day;  $P < 0.01$ ). At wk 8, ACC calves had longer front (1.3 vs 0.9 cm;  $P < 0.01$ ) and rear (1.2 vs 0.8 cm;  $P < 0.01$ ) teats. Whole untrimmed udders of ACC-fed calves were heavier (255 vs 66 g;  $P < 0.01$ ). Differences were more pronounced after skin was trimmed (198 vs 38 g;  $P < 0.01$ ). Dissected PAR and MFP were both greater for ACC calves (10.5 vs 1.4 g and 173 vs 29 g, respectively;  $P < 0.01$ ). Overall differences remained if expressed on a body weight basis ( $P \leq 0.05$ ). Results provide compelling evidence that pre-weaning nutrition markedly affects MG development in dairy calves. Efforts in our lab are underway to uncover the cellular and molecular mechanisms responsible for these differences.

**Key Words:** mammary gland, milk replacer, parenchyma

**T265 Bovine, caprine and ovine serotonin receptors expression in the mammary gland during lactating and dry off by immunohistochemistry.** Aridany Suarez-Trujillo\*<sup>1</sup>, Miguel A. Rivero<sup>2</sup>, Anastasio Argüello<sup>1</sup>, Juan Capote<sup>3</sup>, and Noemi Castro<sup>1</sup>, <sup>1</sup>Department of Animal Science, Universidad de Las Palmas de Gran Canaria, Arucas, Las Palmas, Spain, <sup>2</sup>Department of Morphology, Universidad de Las Palmas de Gran Canaria, Arucas, Las Palmas, Spain, <sup>3</sup>Canarian Agronomic Science Institute, La Laguna, Tenerife, Spain.

Serotonin (5-HT) functions as a feedback inhibitor of lactation. This action is mediated by 5-HT receptors (5-HTR). In situ hybridization studies showed 5-HTR subtypes are expressed in bovine mammary epithelial and myoepithelial cells as well as blood vessels, and vary by receptor subtype. The aim of this study was to determine the distribution of the 5-HTR subtypes (1B, 1E, 2A, 2B, 4 and 7) in cattle, goat and sheep mammary glands using immunohistochemistry, and to compare distribution among lactating and dry udders. Hypothalamus and mammary gland samples were taken at slaughter from 3 lactating and 3 nonlactating animals of each species. Hypothalamic tissue was used as positive control. Tissues were fixed, paraffin embedded and 5- $\mu\text{m}$  sections were placed on slides. Immunohistochemical staining was performed using rabbit primary antibodies against 5-HTR for the 1B, 1E, 2A, 2B, 4 and 7 subtypes. Anti-rabbit secondary antibody was conjugated with streptavidin peroxidase, and visualization of binding was realized using diaminobenzidine substrate. Tissues were counterstained with hematoxylin. Myoepithelial cells were identified in serial sections by positive immunohistochemical staining for calponin. All 6 receptor subtypes were expressed in mammary epithelial cells in the 3 species studied. Furthermore, 5-HTR 1E was expressed in the myoepithelial cells in all 3 species, and in the blood vessels of cows. Receptor subtype 4 was found in myoepithelial cells of cows and sheep. In all 3 species blood vessels were positively stained for receptors 1B, 2A and 2B. In the lactating animals, receptor distribution in the mammary epithelial cells was cytoplasmatic. However, in mammary tissue from nonlactating animals, distribution changed to the apical membrane in all species. In conclusion, 5-HTR are expressed in epithelial, myoepi-



thelial and blood vessels within mammary tissue of cattle, sheep and goats, with distribution of subtype varying by cell-type and species. Furthermore, distribution of the receptors on mammary epithelial cells changes between lactating and nonlactating states.

**Key Words:** serotonin receptors, mammary gland, immunohistochemistry

### **T266 Effect of the supplementation of a functional additive in prepartal cows of two breeds on the level of IgG in colostrum.**

Angela Moreno and Claudia Ariza-Nieto\*, *Corporacion Colombiana de Investigacion Agropecuaria CORPOICA, Bogota, Colombia.*

The purpose of this study was to estimate the effect of supplementation of a functional additive during the last third of gestation in cows on the level of IgG in colostrum. Twenty-eight (28) Holstein and White Orejinegro (WON) cows were assigned to a randomized complete block design with 2 factors (breed and supplementation) with repeated measures (i.e., days of lactation). The supplementation factor included 3 treatments: CTL) Control without supplementation, S0) Supplement without additive, S1) Supplement with additive. Colostrum samples were obtained during the first 3 d of lactation for the determination of crude protein (CP), immunoglobulin G (IgG) and Brix grades. A correlation analysis between Brix grades and IgG was performed. Concentrations of IgG determined by the ELISA technique were strongly correlated with the percentages of Brix grades ( $n = 82$ ,  $r = 0.954$ ,  $P < 0.0001$ ), indicating that it can be used as an estimate of the concentration of IgG. The concentration of crude protein in colostrum was affected by the breed  $\times$  day interaction ( $P < 0.05$ ), it was lower in WON cows (10.3%) than Holstein cows (11.5%) on d 1, whereas on d 2 and 3 WON cows had greater concentrations of protein (5.9% and 6.5%, respectively) compared with Holstein cows (4.8% and 4.1%, respectively). There was a breed  $\times$  day  $\times$  supplementation interaction on colostrum IgG ( $P < 0.05$ ). On d 1, S1 cows of both Holstein (55.1 mg/mL) and WON breeds (47.3 mg/mL) had greater concentrations of colostrum IgG than CTL Holstein (44.4 mg/mL) and WON (32.5 mg/mL) cows. The decrease in colostrum IgG from d 1 to d 3 was greater in Holstein (95%) than in WON (86%) cows. It can be concluded that dietary supplementation with functional additives increased the colostrum concentration of IgG in both cow breeds.

**Key Words:** feed additive, cow, colostrum

### **T267 The effects of cabergoline administration at dry-off of lactating cows on udder engorgement, milk leakages, lying behavior, and udder health at calving.**

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Cabergoline is an ergot-derivative with high affinity for the D<sub>2</sub> dopamine receptors whose dopaminergic effects cause inhibition of prolactin (PRL) secretion, and thus it could be considered a molecule that acts as a potential dry-off facilitator. One hundred ninety-nine Holstein cows (102 primiparous; 97 multiparous) producing  $\geq 18$  kg/d at dry-off were split in 2 treatments with the objective of evaluating the effects of cabergoline at dry-off (between 70 and 50 d before the expecting calving date) on PRL secretion, udder engorgement, milk leakages, udder health at calving, and cow wellbeing after dry-off. Treatments consisted of a single i.m. injection of 5 mL of a solution containing 5.6 mg of cabergoline (CAB) or 5 mL of saline solution as a placebo (CTRL). Each animal was evaluated for presence or absence of milk leakages during the 4 d following

drying-off and udder engorgement determined using a digital algometer. Lying behavior was recorded during 10 d after drying-off. Twenty-five cows from each treatment were randomly chosen and blood sampled at 3 and 15 d after dry-off and at 5 and 3 d before the expected calving date to determine serum PRL concentrations. Data were analyzed using a mixed-effects model for repeated measures. Cows on CTRL had greater ( $P < 0.05$ ) serum PRL concentrations than cows on CAB at 3 and 15 d after dry-off. Cows on CTRL had a greater ( $P < 0.01$ ) udder engorgement ( $24.0 \pm 0.33$  Newtons) than cows on CAB ( $22.4 \pm 33$  Newtons) throughout the 4 d following dry-off, and it decreased ( $P < 0.05$ ) as days since dry-off increased. The overall incidence of milk leakage in cows on CAB ( $3.1 \pm 0.88\%$ ) was 73.5% of that obtained in cows on CTRL ( $11.7 \pm 1.64\%$ ); and cows on CAB had 0.2 lesser odds ( $P < 0.001$ ) to incur in milk leakage than cows on CTRL. The day following dry-off, CTRL cows lied about 1.5 h/d less ( $P < 0.05$ ) than cows on CAB. It is concluded that an i.m. administration of 5.6 mg of cabergoline at dry-off effectively reduces PRL secretion, udder engorgement, milk leakages, and improves lying time the day following dry-off.

**Key Words:** involution, mammary gland, prolactin

### **T268 Dry-off facilitator cabergoline hastened the GLUT-1 decrease and lactoferrin increase in the mammary tissue during drying-off in dairy cows.**

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In ruminants, the early phase of drying-off is a period of mammary gland involution where lactose secretion is inhibited and lactoferrin is stimulated. GLUT-1 is a glucose transporter that has a key role in supplying substrate for lactose synthesis. The analysis of the changes in lactose and lactoferrin contents in mammary secretions and in GLUT-1 and lactoferrin contents in the udder can provide valuable information about the speed of the mammary involution. To assess the effect of prolactin inhibition by cabergoline on udder involution, 14 Holstein dairy cows were injected with a single i.m. administration of 5.6 mg cabergoline ( $n = 7$ ) or placebo ( $n = 7$ ) within 4 h after the last milking the day of drying off (D0). After D0, hay and water ad libitum was supplied to the cows for 10 d. Mammary secretion samples, collected during lactation (D-6) and at D1, D2, D3, D4, D8 and D14 after the drying-off, were used for lactose and lactoferrin analysis. Mammary biopsy samples, collected at D-6, D1 and D8, were used for GLUT-1 mRNA and lactoferrin analyses. Lactose content of mammary secretions progressively decreased during involution, whereas their lactoferrin content increased. The change in lactose content was associated with paralleled change in GLUT-1 mRNA level in the udder. These decreases were faster in cabergoline treated cows compared with controls with lower lactose content in cabergoline treated cows already by D1 than in controls ( $P < 0.05$ ) and significant decrease in GLUT-1 mRNA levels at D1 and D8 respectively for cabergoline and control treatments compared with D-6 ( $P \leq 0.05$ ). The rise of lactoferrin content in mammary secretions was significant starting at D4 in the cabergoline treated cows ( $P \leq 0.05$ ) whereas it only happened at D8 in controls ( $P < 0.05$ ). Overall, cabergoline treatment decreased GLUT-1 mRNA level ( $P < 0.05$ ) and increased lactoferrin content ( $P = 0.10$ ). Similarly, lactoferrin immunostaining intensity in the mammary tissue was higher at D1 than at D-6 in cabergoline treated cows ( $P \leq 0.05$ ), whereas it tended to be higher only at D8 in controls ( $P < 0.10$ ). Our results indicate that cabergoline treatment was efficient to hasten the udder involution and therefore facilitates the dry-off.

**Key Words:** cow, drying-off, prolactin

**T269 Transcriptome analysis of the mammary gland reveals new insights for the role of serotonin in lactation.** Jimena

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Serotonin (5-HT) in the mammary gland is known to regulate processes such as calcium homeostasis, tight junction permeability, and milk protein gene expression. The rate-limiting enzyme in the synthesis of non-neuronal 5-HT is tryptophan-hydroxylase (TPH1). Our objective was to further explore the roles of 5-HT in the mammary gland during lactation. We used whole body TPH1 knockout dams (KO; 5-HT deficient, n = 4) and compare them to wild-type (WT; n = 4) and rescue (RC; KO + 100 mg/kg 5-hydroxytryptophan injected daily, n = 4) dams. Mammary tissues were collected on d 10 of lactation. Total RNA extraction, amplification, library preparation, and sequencing were performed following the Illumina mRNA-seq protocol. Sequencing reads were mapped to the mouse reference genome using Tophat. The resulting alignments were used to reconstruct transcript models by Cufflinks. Differential gene expression was analyzed using Cuffdiff. Overall, 97 and 204 genes (false discovery rate, FDR ≤ 0.01) showed at least a 2-fold expression difference between WT and KO and between WT and RC, respectively. Most of the differentially expressed genes (DEG) are related to calcium homeostasis, regulation of apoptosis, cell cycle and cell differentiation, proliferation, and the immune response, among others. Additionally, enrichment analysis using Gene Ontology (GO) and Medical Subject Headings (MeSH) databases revealed the alteration of several biological processes (FRD ≤ 0.01) including fat cell differentiation and lipid metabolism, regulation of extracellular signal-related kinases and mitogen-activated kinase cascades, insulin resistance, nuclear transport, regulation of membrane potential, release of calcium from the endoplasmic reticulum into the cytosol. The majority of the biological processes and metabolic pathways altered in the KO dams are required for mammary gland homeostasis. Our study reveals the importance of non-neuronal 5-HT for normal mammary gland function and lactation. The potential modes of regulation of the bovine mammary gland during lactation by 5-HT should be further investigated.

**Key Words:** RNA sequencing, tryptophan-hydroxylase 1, lactation

**T270 Lactation performance of dairy cows milked 3 times per day and supplemented with two different formulations of bovine somatotropin.** Jozivaldo P. G. Morais\*, Andressa P. da S. Cruz, Natalia S. Minami, Luiz P. Veronese, Tiago A. Del Valle, and Simone D. Sartorio, Universidade Federal de São Carlos, UFSCAR, Araras, SP, Brazil.

The aim of this randomized field study was to evaluate the lactation performance of dairy cows supplemented with 2 different 500-mg bovine somatotropin (bST) formulations. Sixty Holstein cows were allocated to a control, 90 Holstein cows to a somatotropin fast release formulation (FR-bST, Boostin-S, LG Life Science), and 90 Holstein cows to a somatotropin slow release formulation (SR-bST, Lactotropin, Elanco Animal Health). Treatments (TRT) were delivered every 14 d starting at 57–70 d post calving until the end of lactation, or 17 injections. Before TRT assignment, animals were paired by milk yield, lactation number, and randomly assigned to one of the 3 TRT. Evaluated parameters were milk production, milk protein, milk fat, milk SCC, and BCS. Somatotropin treatment increased milk yield ( $P < 0.001$ ). SR-bST treated cows, yielded 1.6 kg/day more milk than FR-bST treated cows ( $P = 0.008$ ). Somatotropin increased milk protein % ( $P = 0.002$ ), decreased BCS ( $P < 0.001$ ), and had no effect on milk fat % ( $P = 0.120$ ) and SCC ( $P = 0.964$ ). Results are shown in Table 1.

**Table 1 (Abstr. T270).** Production variables of cows milked 3 times per day without bST and with 2 different bST formulations<sup>1</sup>

Item	Treatment			SEM	P-value <sup>2</sup>	
	No bST	FR-bST	SR-bST		C1	C2
Pre-TRT period						
Milk, kg/d	40.04	40.38	39.96	0.464	0.880	0.635
Fat, %	3.36	3.17	3.28	0.063	0.349	0.469
Protein, %	2.68	2.73	2.69	0.014	0.448	0.216
SCC, ×10 <sup>3</sup> /mL	38.74	39.99	40.92	2.459	0.350	0.885
BCS	2.94	2.94	2.95	0.016	0.851	0.975
TRT period						
Milk, kg/d	34.63	38.03	39.60	0.131	<0.001	0.008
Fat, %	3.18	3.06	3.10	0.001	0.120	0.576
Protein, %	3.01	3.11	3.07	0.001	0.002	0.086
SCC, ×10 <sup>3</sup> /mL	242.6	181.1	166.8	11.50	0.964	0.865
BCS	3.04	2.95	2.93	0.003	<0.001	0.263

<sup>1</sup>FR-bST = Boostin-S, LG Life Science; SR-bST = Lactotropin, Elanco Animal Health. Starting on d 63 bovine somatotropin was administered at 14-d intervals to cows on the groups to receive bST.

<sup>2</sup>C1 = control vs. bST; C2 = FR-bST vs. SR-bST.

**Key Words:** milk composition, milk yield, recombinant bST

**T271 Effects of milking frequency and prolactin on milk production and expression of prolactin receptors in the mammary gland of dairy cows.** Izabella Thompson\*<sup>1</sup>, Severine Ollier<sup>1</sup>, Xin Zhao<sup>2</sup>, and Pierre Lacasse<sup>1</sup>, <sup>1</sup>AAFC-Dairy and Swine Research and Development Centre, Sherbrooke, QC, Canada, <sup>2</sup>Department of Animal Science, McGill University, Sainte-Anne-de-Bellevue, QC, Canada.

A better understanding of how biological processes affect milk production in dairy cows is important to improve lactation persistence. The present study investigated the sensitivity of the mammary gland to prolactin. Eleven mid-lactation cows were milked twice daily during the first 2 experimental weeks. During wk 3–10 cows were differentially milked; the right quarters being milked thrice daily (3×), while the left were milked once daily (1×). During wk 11–14, all quarters were milked twice daily. After 4 weeks of differential milking, cows received daily i.m. injections of the dopamine antagonist domperidone (300 mg; DOMP; n = 6) or DMSO (CTL; n = 5) for 8 weeks (wk 7–14). Blood and milk samples were collected weekly for measurement of prolactin (PRL) as well as milk components for the latter. Milk was also collected from front quarters every 2 weeks and RNA was extracted from milk fat fraction. Expression of PRL receptors (long; PRLRL and short; PRLRS) were measured by real-time RT-PCR. Mammary gland biopsies were taken from rear quarters on wk 2, 6, 10 and 14 for subsequent gene and protein analyses. Differential milking improved milk production in the 3× quarters compared with the 1× quarters ( $P < 0.01$ ). Increases in fat ( $P = 0.04$ ), protein ( $P = 0.05$ ) urea ( $P = 0.01$ ) and lactose ( $P < 0.01$ ) were observed in the 1× quarters, while greater SSC ( $P < 0.01$ ) was observed in the 3× quarters. Blood PRL concentrations were greater for DOMP than CTL cows ( $24.8 \pm 2.3$  vs  $13.9 \pm 2.5$  ng/mL;  $P < 0.01$ ). There was no effect of injections on milk production, but milk from DOMP cows had greater concentrations of lactose ( $P = 0.02$ ) than CTL. Interestingly, DOMP cows had greater expression of both forms of PRLR ( $P < 0.01$ ) in milk fat compared with CTL. Additionally, greater PRLR expression was observed in the 3× than 1× quarters ( $P < 0.01$ ). Results show that milk frequency improves milk production and alters composition and expression of PRLR in the mammary gland. Moreover, administration

of domperidone increased PRL concentration and is related to greater expression of both forms of PRLR in the mammary gland.

**Key Words:** milk production, mammary gland, prolactin

**T272 Effects of inhibiting the lactogenic signal at calving on milk production and metabolic perturbations in cows.** Noémie Vanacker\*<sup>1</sup>, Ollier Séverine<sup>1</sup>, Blouin Richard<sup>2</sup>, and Lacasse Pierre<sup>1</sup>, <sup>1</sup>AAFC-Dairy and Swine Research and Development Centre, Sherbrooke, Québec, Canada, <sup>2</sup>Département de Biologie, Université de Sherbrooke, Sherbrooke, Québec, Canada.

During the periparturient period, the abrupt increase in energy demand for milk production induces metabolic disorders. Our previous work has shown that reducing milk output by milking once a day or partially in the first days of lactation reduces these disorders. The aim of this study was to reduce metabolic disorders by limiting milk production during the first week of lactation through inhibition of the lactogenic signal driven by prolactin. Twenty-two late gestation cows were assigned into 2 groups based on parity and milk production of the first 100 d of the previous lactation. Both groups received 8 i.m. injections of either quinagolide (Quin; 2mg of an inhibitor of prolactin release) or water (CTL) just after calving, and the subsequent 7 injections were given every 12h. Milk production was measured until d 28 post-calving. Blood samples were taken from d 1 (calving) to 5 and then on d 7, 10, 14, 21 and 28 to measure concentrations of urea, phosphorus, calcium, glucose, non-esterified fatty acids (NEFA),  $\beta$ -hydroxybutyrate and prolactin (PRL). Prolactin concentrations were lower ( $P < 0.01$ ) in Quin cows from d 2 to d 5. Interestingly, between d 10 and d 28, PRL concentrations were greater ( $P < 0.05$ ) in Quin than CTL cows. Milk production was lower in Quin cows from d 2 to d 6 than in control ( $24.3 \pm 6.4$  kg/d vs  $34.8 \pm 4.1$  kg/d;  $P < 0.05$ ). There was no residual effect of quinagolide on milk production after d 6. The blood glucose and calcium concentrations were greater ( $P < 0.05$ ) in Quin cows whereas the concentration of  $\beta$ -hydroxybutyrate was greater ( $P < 0.05$ ) in CTL cows during the first week of lactation. Blood NEFA, urea and phosphorus were not affected by treatment. In conclusion, reducing the prolactin peak at calving is effective to reduce milk production during the first week of lactation without compromising the global productivity of the dairy cow. This reduction in milk production allows a reduction of the metabolic stress during this period.

**Key Words:** prolactin, quinagolide, energy balance

**T273 Effect of increased milking frequency in early lactation on milk production, proliferation and apoptosis of mammary cells in dairy cow.** Juliana Mergh Leão\*<sup>1</sup>, Juliana Aparecida Mello Lima<sup>1</sup>, Sandra Gesteira Coelho<sup>1</sup>, José Reinaldo Mendes Ruas<sup>2</sup>, Anilton César Vasconcelos<sup>1</sup>, Ângela Maria Quintão Lana<sup>1</sup>, Ronaldo Braga Reis<sup>1</sup>, and Helton Mattana Saturnino<sup>1</sup>, <sup>1</sup>Universidade Federal de Minas Gerais-UFMG, Belo Horizonte, Minas Gerais, Brazil, <sup>2</sup>Empresa de Pesquisa Agropecuária de Minas Gerais-EPAMIG, Felixlândia, Minas Gerais, Brazil.

The objectives of the present study were to evaluate the effect of increased milking frequency (IMF) during early lactation on milk yield, proliferation and apoptosis in mammary epithelial cells of F<sub>1</sub> Holstein  $\times$  Zebu cows. Fourteen cows F<sub>1</sub> Holstein  $\times$  Zebu cows were randomly distributed into 2 groups. The control group (2 $\times$ ) was milked twice a day up to 210 d in milking (DIM), and the group subjected to IMF (4 $\times$ ) was milked 4 times a day from 2 to 21 DIM, and twice a day from 22 to 210 DIM. Milk production was measured daily from 4 to 30 DIM and then

each 15 d until 210 d. Mammary biopsies were performed on d 2, 7, 14, 21, and 28 postpartum. Biopsies were obtained using a biopsy needle (12 g  $\times$  12). Mammary tissue was used to measure rates of cell proliferation and apoptosis using Ki-67 nuclear proliferation antigen localization and terminal deoxynucleotidyl transferase nick-end labeling (TUNEL), respectively. Data were analyzed with a repeated measures design and means were compared by Tukey's test ( $P < 0.05$ ). During IMF, the 4 $\times$  produced  $2.6 \pm 0.2$  kg/d more milk than the 2 $\times$  ( $P < 0.05$ ). After IMF phase, milk production from the 4 $\times$  decreased and was similar to 2 $\times$  up to 210 DIM ( $P > 0.05$ ). Mammary cell proliferation and apoptosis were not affected by milking frequency ( $P > 0.05$ ). Rates of proliferation and apoptosis were influenced by days of lactation ( $P < 0.05$ ). On the second day of lactation, the highest apoptosis rate (0.3%) was observed. Days 7 (0.16%), 14 (0.13%), 21 (0.11%) and 28 (0.06%) had similar and lower rates in comparison with the second day of lactation. There was a higher percentage of proliferation in mammary epithelial cells on d 2 (2.12%). On d 7 (0.99%) and 14 (0.87%), the proliferation rate dropped to about half the value observed on d 2 and in the following week (0.47%) it was observed another reduction which remained until the 28th of lactation (0.43%). Changes in milking frequency during early lactation did not alter milk yield after IMF and mammary cell population dynamics suggesting that maybe the mammary biopsy procedure used in the current study influenced these responses negatively.

**Key Words:** biopsy, Ki-67, terminal deoxynucleotidyl transferase nick-end labeling (TUNEL)

**T274 Effect of cortisol on mammary epithelial cell turnover: milk synthesis, proliferation and apoptosis.** J. A. Negro\*<sup>1</sup>, G. M. Krempel, S. A. Oliveira, G. F. Bomfim, H. Z. Polato, and F. C. Lahr, *Basic Science Department, FZEA/USP, Pirassununga, SP, Brazil.*

Cortisol is essential for copious synthesis of milk components, and cortisol administration delayed the decline of milk yield in late lactation. However, the effect of cortisol in proliferation, activity, and survival of the epithelial cells remains controversial. The objective of this study was to evaluate the effect of cortisol on the survival and proliferation of epithelial cells. Twenty-four Saanen goats were submitted to administration of ACTH (Cortisol group) or placebo (Control group). Milk yield and milk quality (fat, protein, lactose, and CCS) were measured daily and weekly, respectively. Cortisol concentrations in plasma were measured before and after ACTH or Placebo administration at 60, 90, and 120 d of lactation. All mammary biopsies were conducted 1 h after ACTH or placebo administration. Biopsies were taken of 4 goats submitted to ACTH and 4 goats submitted to placebo at 60, 90 and 120 d of lactation. Apoptosis was determined using terminal deoxynucleotidyl transferase dUTP nick-end labeling (TUNEL) kit, and proliferating cell was determined using a proliferating cell nuclear antigen (PCNA) kit. The mRNA levels of Bax and Bcl-2 were analyzed by RT-PCR. At the same time, Bax and Bcl-2 synthesis were measured by ELISA kit. Statistical analysis was performed by ANOVA using mixed model, and the level of significance was set at  $P < 0.05$  for main effects and interactions. Although, cortisol increased significantly after ACTH administration milk yield, milk quality, casein and lactose in milk were similar for both groups. Furthermore, percentage of apoptotic cells measured by TUNEL, percentage of proliferation measured by PCNA, and the expression and synthesis of Bax (promoter of apoptosis) were similar in ACTH and placebo groups (control). However, the expression and synthesis of Bcl-2 (inhibitor of apoptosis) were significantly higher after ACTH administration when compared with placebo administration. Furthermore, the relationship between Bax/Bcl-2 was significantly lower after ACTH administration when compared with placebo administration.



These results support the hypothesis that cortisol can delay apoptosis in the mammary gland.

**Key Words:** mammary gland, Bax, Bcl-2

**T275 Effects of growth hormone and insulin-like growth factor on synthesis and secretion of  $\beta$ -casein,  $\beta$ -lactalbumin and lactoferrin in mammary epithelial cells.** J. A. Negro\*<sup>1</sup>, G. M. Krempel, S. A. Oliveira, G. F. Bomfim, F. C. Lahr, and H. Z. Polato, *Basic Science Department, FZEA/USP, Pirassununga, SP, Brazil.*

Growth hormone (GH) and insulin-like growth factor (IGF-I) are associated to milk production traits, however the mechanism how GH and IGF-I increase the milk synthesis remains unclear. The objective of this study was to evaluate the effect of GH and IGF-I on the synthesis and secretion of  $\beta$ -casein,  $\beta$ -lactalbumin, and lactoferrin by mammary epithelial cells. Twenty-four Saanen goats were used, and 5 goats were biopsied. Milk yield and milk quality (fat, protein, lactose and CCS) were measured daily and weekly, respectively. Growth hormone and insulin-like growth factor-I concentrations in plasma were measured once a day, at 30 and 60 d of lactation. All mammary biopsies were conducted just after blood samples. Biopsies were taken of 5 goats at 30 and 60 d of lactation. Epithelial cells were isolated and cell culture was used to study expression and synthesis of  $\beta$ -casein,  $\beta$ -lactalbumin, and lactoferrin. Four treatments were imposed to epithelial cell culture: (1) control (basal medium); (2) GH (10 ng/mL); (3) IGF-I (10 ng/mL); (4) GH + IGF-I (10 ng/mL + 10 ng/mL). The mRNA levels of  $\beta$ -casein,  $\beta$ -lactalbumin, and lactoferrin were analyzed by RT-PCR. At the same time, the synthesis of  $\beta$ -casein,  $\beta$ -lactalbumin, lactoferrin, and lactose in were measured by ELISA kit in culture medium. Statistical analysis was performed by ANOVA using mixed model, and the level of significance was set at  $P < 0.05$  for main effects and interactions. Growth hormone (GH) and insulin-like growth factor (IGF-I) were associated with milk yield, and goats classified as higher producing (4 from 24 experimental goats) presented high GH and IGF-I levels.  $\beta$ -Casein expression and synthesis were significantly higher for IGF-I and GH + IGF-I treatments when compared with control. Lactoferrin expression and synthesis were significantly higher for GH and GH + IGF-I treatments when compared with control. However,  $\beta$ -lactalbumin expression and synthesis were not change by hormone treatments when compared with control. These results support the hypothesis that GH and IGF-I can modulate directly the synthesis of milk.

**Key Words:** growth hormone (GH), insulin-like growth factor (IGF-I), milk synthesis

**T276 The barrier integrity of bovine mammary epithelial cells in vitro in response to lipopolysaccharide (LPS) and lipoteichoic acid (LTA) treatment.** Christina Zbinden\*<sup>1,2</sup>, Rupert M. Bruckmaier<sup>1</sup>, and Olga Wellnitz<sup>1</sup>, <sup>1</sup>*Veterinary Physiology, Vetsuisse Faculty University of Bern, Bern, Switzerland*, <sup>2</sup>*Graduate School for Cellular and Biomedical Sciences, University of Bern, Bern, Switzerland.*

The blood-milk barrier of the bovine mammary gland prevents an intermixture of blood and milk. During mastitis, the permeability of the blood milk barrier is increased, which is reflected by an exchange of blood constituents into milk, and vice versa. The aim of this study was to investigate the role of mammary epithelial cells in the regulation of blood-milk barrier alteration during mastitis induced by cell wall components of *Escherichia coli* (LPS) and *Staphylococcus aureus* (LTA). Low passage primary bovine mammary epithelial cells (bMEC) from 3 different cows were grown separately on Transwell inserts. The

level of integrity of the epithelial barrier was measured by means of transepithelial electrical resistance (TEER), as well as by diffusion of the fluorophore Lucifer Yellow (LY) across the cell layer. The formation of tight junctions between adjacent epithelial cells was examined by immunofluorescence staining of zona occludens-1 and by transmission electron microscopy. The cultured cells formed tight cell layers sealed by tight junctions. The barrier integrity was reduced after 3 h ( $P < 0.05$ ; paired *t*-test relative to unchallenged cells) in response to 500  $\mu$ g/mL LPS from *E. coli*, and after 7 h in response to 20 mg/mL LTA from *S. aureus*. At these dosages, the fluorescence values of LY in the apical compartment of the Transwell insert dropped within 24 h from 5386 to  $560 \pm 235$  RFU in response to LPS, and from 5386 to  $1416 \pm 144$  RFU in response to LTA. No significant changes in barrier permeability were observed in response to 200  $\mu$ g/mL LPS, or to 2 mg/mL LTA. Although LPS and LTA affected the barrier permeability most likely due to an opening of the tight junctions, LPS additionally caused considerable cell damage reflected by increased LDH concentrations in cell culture medium ( $P < 0.05$ ). These results confirm a pathogen-specific impairment of the blood-milk barrier during mastitis, which involves differences in cell degradation.

**Key Words:** bovine mastitis, blood-milk barrier, tight junction

**T277 Intravenous challenge with lipopolysaccharide does not induce a mammary immune response in dairy cows and does not affect the blood-milk barrier.** Olga Wellnitz\*<sup>1,2</sup>, Emmanouil Kalaitzakis<sup>1,2</sup>, Heinrich Bollwein<sup>2</sup>, and Rupert M. Bruckmaier<sup>1</sup>, <sup>1</sup>*Veterinary Physiology, Vetsuisse Faculty University of Bern, Bern, Switzerland*, <sup>2</sup>*Clinic of Reproductive Medicine, Vetsuisse Faculty University of Zurich, Zurich Switzerland.*

An intramammary challenge with lipopolysaccharide (LPS) from *Escherichia coli* is known to induce a considerable immune response of the mammary gland and an impairment of the blood-milk barrier. The aim of the current study was to investigate a potential immune reaction in the mammary gland and potential changes of the integrity of the blood-milk barrier in response to an intravenous, i.e., systemic challenge, with LPS. Ten lactating dairy cows in wk 3 of lactation were challenged intravenously (V. jugularis) with 0.5  $\mu$ g/kg BW *E. coli* LPS (O26:B6). Rectal temperature was measured hourly. Milk samples of one udder quarter were taken immediately before and then every 30 min until 5 h after challenge. Mammary gland biopsies of one quarter were taken immediately before and 8 h after LPS challenge for RT-qPCR of immunorelevant factors. Rectal temperature increased ( $P < 0.001$ ) within 1 h of LPS administration from  $38.3 \pm 0.1$  to  $39.2 \pm 0.1$ °C and stayed elevated throughout the 5 h of experiment. The milk somatic cell count was  $83.3 \pm 19.1 \times 10^3$ /mL immediately before challenge and did not change throughout the experiment. Lactate-dehydrogenase concentrations in milk as a marker for blood-milk barrier impairment was  $48.1 \pm 6.0$  U/L immediately before challenge and did not change throughout the experiment. Relative mRNA expression of immunorelevant factors; that is, the cytokines TNF- $\alpha$  and interleukin-1 $\beta$  in mammary gland tissue did not change in response to intravenous LPS injection. In conclusion, in contrast to intramammary injections, an intravenous injection of 0.5  $\mu$ g/kg BW LPS induces a systemic immune response shown by an increase in rectal temperature, but has obviously no effects on factors of mammary gland immune response which typically change during LPS-induced mastitis. Also the blood-milk barrier integrity does not appear to be influenced by systemic LPS.

**Key Words:** intravenous lipopolysaccharide, mammary gland, blood-milk barrier

**T278 Characterization of mammary glucose metabolism during milk fat depression.** Natalie L. Urrutia\*<sup>1</sup>, Kevin J. Harvathine<sup>1</sup>, and Dale E. Bauman<sup>2</sup>, <sup>1</sup>The Pennsylvania State University, University Park, PA, <sup>2</sup>Cornell University, Ithaca, NY.

Milk fat synthesis requires butyrate, acetate, and reducing equivalents (NADPH) as substrates. NADPH is provided from glucose through the pentose phosphate pathway and isocitrate dehydrogenase (IDH1). During milk fat depression (MFD), reduced milk fat synthesis may result in decreased uptake of nutrients by the mammary gland, including glucose. The objective of this study was to characterize expression of glucose metabolism and NADPH synthesis enzymes in mammary tissue during MFD using tissue from a previously published experiment. Cows were arranged in a 3 × 3 Latin square design with 14-d experimental periods. Treatments were control (CON), CLA-induced MFD (CLA; 3 d i.v infusion of 10 g/d of *trans*-10,*cis*-12 CLA in an intralipid emulsion), and a low forage, high oil diet (LF/HO; 45.9% forage, 3.0% soybean oil, and 1.5% fish oil). Milk fat yield was decreased 24% by CLA and 38% by LF/HO. Mammary biopsies were performed 1 to 3 h after milking at the end of each treatment. Gene expression of enzymes involved in glucose metabolism and NADPH synthesis were determined by Real-Time PCR and analyzed relative to the geometric mean of housekeeping genes (18S ribosomal subunit, ribosomal protein S9 and  $\beta$ 2-microglobulin). Data were analyzed using the fit model procedure of JMP Pro and the model included fixed effects of treatment and geometric mean of housekeeping genes and random effects of cow and period. Glucose transporter 1 (GLUT1) and IDH1 were significantly reduced by the LF/HO treatment when compared with CON (28 and 36%, respectively;  $P < 0.05$ ). Phosphogluconate dehydrogenase expression was lower in the LF/HO treatment when compared with CLA ( $P < 0.05$ ), but not different compared with CON. Other genes involved in glucose metabolism such as HK2, ACLY, G6PDH and GLUT8 were not affected by treatments. These results suggest that during diet-induced MFD the reduction in glucose uptake coincides with decreased expression of GLUT1 and that the decrease in use of NADPH corresponds to a downregulation of the isocitrate pathway. The smaller decrease in milk fat in the CLA treatment may have limited the ability to observe a treatment effect during CLA-induced MFD.

**Key Words:** glucose, NADPH, isocitrate

**T279 Transcriptome adaptation of the bovine mammary gland to a diet supplemented with linseed oil.** Eveline M. Ibeagha-Awemu<sup>1</sup>, Ran Li<sup>1</sup>, Adolf A. Ammah<sup>1</sup>, Nathalie Bissonnette\*<sup>1</sup>, Chaouki Benchaar<sup>1</sup>, and Xin Zhao<sup>2</sup>, <sup>1</sup>Agriculture and Agri-Food Canada, Dairy and Swine Research and Development Centre, Sherbrooke, Quebec, Canada, <sup>2</sup>Department of Animal Science, McGill University, Ste-Anne-De-Bellevue, Quebec, Canada.

Diets rich in unsaturated fatty acids (UFA) have been shown to increase the contents of some milk beneficial fatty acids (FA) including conjugated linoleic acid (CLA). The mechanism by which UFAs modulate

cow's genetics resulting in increased milk CLA content is not clear. This study examined the transcriptome of the bovine mammary gland following dietary supplementation with linseed oil. Twelve Holstein cows (35±10 kg milk; 150±50 DIM) were used in a complete randomized block design and fed a total mixed ration (control diet) for 28 d (d 1-28; control period, CP) followed by a treatment period (TP; d 29-56) consisting of control diet + 5% linseed oil (57%  $\alpha$ -linolenic acid). Milk samples were collected weekly for fat, protein and individual FA determination. Mammary gland biopsies were performed on 6 cows on d 14 (CP) and d 35 and 56 (TP). Global transcriptome was analyzed using RNA-sequencing. Milk fat percent decreased ( $P < 0.0001$ ) from 3.62% (CP) to 2.52% (TP) while protein percent was unchanged by treatment. The proportions of C4:0, C8:0, C14:0, C16:0 and C14:1 decreased ( $P \leq 0.0003$ ) while C18:1n-11t, C20:3n-3, C20:5n-3, C22:5n-3, CLA10t12c and CLA9c11t increased ( $P \leq 0.035$ ) during the TP. RNA-sequencing generated 459 million reads out of which 89% mapped to unique positions on the bovine genome. Eight genes (*CSN2*, *CSN1S1*, *CSN1S2*, *LGB*, *CSN3*, *LALBA*, *COX1*, and *GLYCAM1*) out of 11121 expressed constituted 73.70% of mapped reads. One hundred seventy-nine genes were significantly regulated (79 up- and 100 down-regulated) after FDR correction. Most affected genes were *FBP2* (11-fold up-regulated) and *MROH2B* (4-fold down-regulated). Further regulated genes with roles in FA synthesis/uptake included *LPIN1*, *SREBF1*, *INSIG1*, *FASN*, *BDH1*, *ACSS1*, *ACADVL*, *SLC25A34*, *SLC39A11*, *TIEG2*, and *CYP2B6*. Differentially expressed genes were significantly enriched in several KEGG pathways including PPAR and insulin signaling pathways. This study has provided a broader picture of the transcriptomics events that are involved in mammary gland adaptation to a diet rich in UFA. Our study provides further knowledge on mammary lipogenesis and data that can be used to develop new nutritional strategies for a better management of milk increased beneficial FAs.

**Key Words:** bovine mammary gland transcriptome, linseed oil, lipogenesis

## Nonruminant Nutrition: General II

**T280 Chemical treatment of corn stover with calcium hydroxide increases its energy value for swine.** Victor G. Perez\*, Lester Pordesimo, Amanda Knorr, and Terry Radke, *ADM Animal Nutrition, Quincy, IL.*

The objective was to estimate DE and ME content in corn stover (CS) and CS treated (CST) with Ca(OH)<sub>2</sub> for swine. The Ca(OH)<sub>2</sub> was added to CS in the amount of 6.6% of the DM in CS, with enough water to raise moisture content in CST to 50% (wet basis). After 7 d, CST was sun dried and both CS and CST were pelleted (9 mm). Dietary treatments were a basal diet (97.28% corn), and the basal diet with 30% of either CS or CST added at the expense of corn. The experiment was a RCBD based on initial BW, and each treatment had 8 block replicates (barrows; 90 ± 0.7 kg BW). Feed was offered to provide 2.5 times the energy maintenance requirement. It was assumed that CST had 10% more energy than CS, based on preliminary data from an in vitro organic matter digestibility (Boisen and Fernandez, 1997). Pigs were placed in metabolic crates and allowed 7 d of adaptation, followed by 4 d of total collection of feces and urine. The DE and ME values in ingredients were calculated by the difference procedure. Data were analyzed using the PROC GLM procedure of SAS. Pairwise comparisons were used for treatment means separation. Measured DE and ME values in corn (Table 1) were 2% smaller or less than published values (NRC, 2012). Feed intake was not different between CS and CST. Chemical treatment of CS increased ( $P \leq 0.06$ ) its DE and ME by 43 and 34%, respectively (Table 1), perhaps because it increased ( $P < 0.05$ ) digestibility of dietary NDF (17.7 vs. 29.2%; SEM = 3.22) and ADF (17.5 vs. 35.7%; SEM = 2.20). In conclusion, chemical treatment of CS with Ca(OH)<sub>2</sub> increases its energy value in swine.

**Table 1 (Abstr. T280).** Energy value (kcal/kg DM) of corn, corn stover (CS), and CS treated with Ca(OH)<sub>2</sub> (CST) in pigs (90 ± 0.7 kg BW)

Item	Corn	CS	CST	SEM	CS vs. CST, kcal (%)
DE	3,864 <sup>a</sup>	914 <sup>b</sup>	1,306 <sup>c</sup>	106	392 (43%)
ME	3,758 <sup>a</sup>	917 <sup>b</sup>	1,229 <sup>c</sup>	105	312 (34%)

<sup>a-c</sup>Within rows, means with different superscript differ ( $P \leq 0.06$ )

**Key Words:** corn stover, sow, metabolizable energy

**T281 Hammer status in hammer mill affects feed particle size and piglet performance after weaning.** David Solà-Oriol, Laia Blavi\*, and Roser Sala, *Animal Nutrition and Welfare Service, Department of Animal and Food Sciences, Universitat Autònoma de Barcelona, Bellaterra, Spain.*

Hammers of hammer mill are changed when working edges are worn but, often this is not decided according to the quality standards of feed and their possible effects on performance. The aim of the present work was to study the effect of different hammer status (new; N or worn; W) on particle size distribution of ground diets and the performance of weaned piglets. One hundred sixty 28-d-old piglets [Pietrain × (Landrace × Large White)] were distributed into 16 pens according to their initial BW following a RCBD. A pre-starter (PS; 0 to 14d) and a starter (ST; 14 to 35d) diet was formulated to contain 2.63 Mcal/kg NE, 20.2% CP, 1.37 Lys and 2.48 Mcal/kg NE, 15.1% CP, 1.20 Lys, respectively. Two experimental treatments were performed according to the hammer status N or

W. PS and ST diets were ground at Ø = 2 and Ø = 3 mm, respectively. Feed was offered ad libitum in mash form. Individual animal weight and feed disappearance were recorded at 0, 14, and 35 d post-weaning to calculate average daily feed intake (ADFI) and average daily gain (ADG). Data were analyzed with ANOVA taking into account block of BW and hammer status as main factors. Feed particle size distribution was determined as % of particles ≥1 mm, ≥0.75 mm, ≥0.5 mm and <0.5 mm. Different size distribution was mainly observed for ST. The W diets showed 21% and 3.7% more particles of ≥1 mm and ≥0.5 mm, but 8.5% and 9.3% less of ≥0.75 mm and <0.5 mm, respectively than N diet. For PS period, no different BW, ADG and ADFI was observed ( $P > 0.05$ ). However, higher ADFI (666 vs 619 g/d;  $P = 0.03$ ) and a tendency to higher ADG (418 vs 387 g/d;  $P = 0.10$ ) were observed for the animals fed the N diet for ST period. Considering the entire period (0 to 35d), higher BW (18.9 vs 18.1 kg;  $P = 0.03$ ), ADFI (505 vs 471 g/d;  $P = 0.01$ ) and ADG (318 vs 295 g/d;  $P = 0.03$ ) were observed for the animals fed the N diets. It is concluded that hammer status affects particle size and piglet performance in weanling diets in a larger extend in ST than PS diets.

**Key Words:** hammer mill, particle size, piglet

**T282 Effect of salt particle size and extended mixing times on the mixing uniformity of a corn-soy swine diet.** Marut Saensukja-roenphon\*<sup>1</sup>, Cassandra K. Jones<sup>1</sup>, Charles H. Fahrenholz<sup>2</sup>, Kessinee Chitakasempornkul<sup>1</sup>, and Charles R. Stark<sup>1</sup>, <sup>1</sup>Kansas State University, Manhattan, Kansas, <sup>2</sup>Phibro Animal Health Corporation, Manhattan, Kansas.

The uniformity of a feed mixture is determined from the coefficient of variation (CV) of 10 samples in a single batch of feed. The feed industry standard is a CV of <10% using a single source tracer such as salt, trace minerals, or iron filings. The uniformity of mix can be affected by many factors, including mixer design, particle size of the ingredients, and mixing time. Previous research has determined the minimum mix time to maximize the mixing efficiency, but some hypothesize that over-mixing may lead to ingredient segregation. However, there is limited data regarding the effects of extended mixing or of the analytical marker appropriate for maximum precision of the assay. The objectives of this experiment were to determine (1) the effects of extended mix time, and (2) particle size of the marker on the CV in a corn-soy swine diet. Treatments were arranged in a 3 × 7 factorial design with 3 salt particle sizes: fine (350 µm), medium (464 µm), and coarse (728 µm) and 7 mix times: 2, 3, 5, 15, 30, 45 and 60 min. There were 3 replicates per treatment and 10 samples per replicate. Salt concentrations were determined using a Quantab Chloride Titrator. Data were analyzed using the GLIMMIX procedure of SAS. There was no interaction between mix time and salt particle size ( $P > 0.60$ ). The extended mix time did not result in segregation ( $P > 0.30$ ; 9.6, 11.5, 11.8, 11.2, 12.50, 10.6, and 10.1% CV for 2, 3, 5, 15, 30, 45, and 60 min, respectively). Particle size of the salt significantly affected the uniformity of mix ( $P < 0.01$ ; 20.7, 8.4, and 7.7% CV for the coarse, medium, and fine salt, respectively). These results indicate that feed did not segregate after mixing for one hour and that greater number of particles per gram increased the precision of the analysis, likely due to an increased probability that the tracer was present in proportionate quantities in the sample tested. More research is needed to determine if sample size or number of samples



per batch should be increased, as well as the effect of diet particle size on uniformity of mix.

**Key Words:** mixing uniformity, mixing time, particle size

**T283 Optimization of different probiotics on improving the quality of cottonseed meal fermentation using response surface methodology.**

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Central composite design of response surface methodology (RSM) was employed to optimize *Lactobacillus* content ( $X_1$ :  $4.5 \times 10^6$  to  $5.5 \times 10^6$  cfu/g), *Bacillus subtilis* content ( $X_2$ :  $9.0 \times 10^6$  to  $1.1 \times 10^7$  cfu/g) and yeast content ( $X_3$ :  $5.50 \times 10^6$  to  $7.0 \times 10^6$  cfu/g) of solid-state fermentation (SSF) cottonseed meal with low pH value, high dry matter recovery (DMR) and high the reducing-sugar content. Results indicated that the data were adequately fitted into 3 s-order polynomial models. The *Lactobacillus* content, *Bacillus subtilis* content, and yeast content were found to have significant linear, quadratic and interaction effects on pH value, the DMR and the reducing-sugar. The optimal extraction conditions were predicted to be *Lactobacillus* content of  $5.50 \times 10^6$  cfu/g, *Bacillus subtilis* content of  $1.08 \times 10^7$  cfu/g and yeast content of  $6.08 \times 10^5$  cfu/g. The pH value, DMR and the reducing sugar predicted by RSA were 5.01, 91.8% and 1.69%, respectively. The detection index obtained experimentally was close to its predicted values. The establishment of such model provides a good experimental basis employing RSM for optimizing the quantity of inoculation of *Lactobacillus*, *Bacillus subtilis*, and yeast on fermentation.

**Key Words:** quantity of inoculation, pH, dry matter recovery

**T284 Effects of ginger root, star anise, and *Salvia miltiorrhiza* on growth performance, antioxidant status and serum metabolites in growing pigs.**

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To compare the effects of ginger root (*Zingiber officinale*), star anise (*Illicium verum*) and dan-shen root (*Salvia miltiorrhiza*) that were processed to particle sizes of 300  $\mu$ m on growth performance, serum antioxidant status and serum metabolites of growing pigs, 24 Laiwu pigs at 70-d old were randomly allocated to 4 treatments with 6 replicates in a complete randomized design. The 4 treatments (TRT) were (1) TRT 1, pigs were fed corn-soybean meal based diets; (2) TRT 1 + 10 mg/kg ginger; (3) TRT 1 + 5 g/kg star anise; (4) TRT 1 + 10 g/kg dan-shen. ADG, ADFI and feed conversion rate of each replicate were measured weekly. At d35 of the experiment, blood samples were obtained from 6 pigs per treatment to determine antioxidant enzymatic activities and metabolites in the serum. All dates were subjected to ANOVA using the one-way of SAS program software (version 9.2). Differences among treatments were tested using Duncan's multiple-range test. All pigs had similar ADFI, ADG, feed conversion rate and concentrations of malondialdehyde in serum over the entire experimental period. However, comparing to TRT 1, supplementation of ginger, star anise and dan-shen increased activities of total antioxidant capacity and superoxide dismutase ( $P = 0.003$ ,  $P < 0.001$ , respectively). The activity of superoxide dismutase in TRT 2 was higher ( $P < 0.05$ ) than TRT 4. Concentration of glutathione in serum was higher ( $P < 0.05$ ) with dietary

supplementation of star anise and dan-shen than TRT 1 and TRT 2. As compared with that of TRT 1, supplementation of 3 botanical additives reduced ( $P < 0.05$ ) concentrations of total cholesterol but had same concentrations of total protein, globulin and low density lipoprotein in serum of growing pigs. Supplementation of star anise increased ( $P < 0.05$ ) concentrations of albumin and high-density lipoprotein comparing to TRT 1. Concentration of high-density lipoprotein in serum of ginger-supplemented pigs was higher ( $P < 0.05$ ) compared with that of TRT1. Dietary supplementation of ginger, star anise and dan-shen may improve serum antioxidant status and serum metabolites of growing pigs in a certain degree.

**Key Words:** ginger, star anise, dan-shen root (*Salvia miltiorrhiza*)

**T285 Effect of oregano essential oil supplementation on oxidative stability of eggs enriched with polyunsaturated fatty acids during storage.**

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This study evaluated the inclusion of oregano essential oil (OEO) *Lippia origanoides* Kunth in diets enriched with polyunsaturated fatty acids (PUFA) on oxidative stability of eggs during storage. A total of 144 Babcock Brown hens 48-week-old were assigned to a completely randomized factorial design  $2 \times 2$ ; Factor A oil type (palm and fish), Factor B OEO level (0 and 100 g/Ton) with the repeated measures of days of storage (0, 30, 60 d). The statistical model included the fixed effects of oil type, OEO level, day of storage and their interactions. At the end of the period (8 weeks) 6 eggs per replicate were collected; 2 eggs were analyzed as fresh, the remaining 4 were storage at 4°C; 2 for 30days and 2 for 60 d. Egg yolks were used for the analysis of PUFA and thiobarbituric acid reactive substances (TBARS) that measures malonaldehyde concentration (MDA). Results showed no significant interactions effect ( $P > 0.05$ ). In fish oil groups, the concentration of PUFA and docosahexaenoic acid (DHA) increased (+16.8 and +1.4%, respectively) ( $P < 0.05$ ). The concentration of MDA was higher in fish oil groups (41.6 ng/g yolk) compared with palm oil groups (32.6 ng/g yolk). The groups supplemented with OEO improved oxidative stability during storage (MDA 31.1 ng/g yolk) compared with the groups without supplementation (MDA 34.1 ng/g yolk). Concentration of MDA showed an effect on storage time ( $P < 0.05$ ); on day zero egg yolks had lower MDA (28.3 ng/g yolk), whereas on d 30 and 60 of storage MDA increased (31.4 and 38 ng/g yolk, respectively). The OEO showed potential as a natural antioxidant in the diet of layers hens improving the oxidative stability of eggs stored at 4°C up to 60 d.

**Key Words:** layer hen, lipid peroxidation, egg yolk

**T286 Effects of dietary inclusion of *Lactobacillus acidophilus* on growth performance, health, and carcass traits of growing-finishing pigs.**

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The objective of this study was to determine the effect of a single strain direct-fed microbial (DFM, *Lactobacillus acidophilus*  $1 \times 10^9$  cfu/g) on growth performance and carcass traits of growing-finishing pigs. Sixty pigs (30 gilts and 30 barrows) with initial BW of  $44.4 \text{ kg} \pm 1.8 \text{ kg}$  were allotted to 2 dietary treatments in a randomized complete block design with sex and initial BW as blocks. Experimental diets met the NRC 2012

nutrient requirements. The inclusion levels of DFM were 0.28, 0.24, and 0.20% during phase 5 (d 0 to 28), phase 6 (d 28 to 49), and phase 7 (d 49 to 70), respectively. Growth performance was measured for 10 wk. Blood samples were collected at wk 9 to measure TNF $\alpha$  and blood urea nitrogen (BUN). At wk 10, pigs were slaughtered at a local abattoir to evaluate carcass and loin quality including HCW, CCW, backfat thickness (1st, 10th, and last rib), loin weight, loin color (Minolta colorimeter), loin marbling score (1 to 10, NPPC scale), drip loss (48 h), loin eye area, and chemical composition. Data were analyzed using Proc Mixed of SAS with treatment and sex as fixed effects and initial BW block as a random effect. P value less than 0.05 was considered significant and less than 0.10 tendency. The ADG, ADFI, and G:F were not different between treatments. Backfat thickness of pigs with DFM was smaller ( $P < 0.05$ , 24.5 vs. 29.6 mm) at the last rib and tended to be smaller ( $P = 0.088$ , 30.1 vs 34.4 mm) at the 1st rib than pigs without DFM. Loin of pigs with DFM tended to be redder ( $P = 0.084$ , a\* value for redness: 7.22 vs. 6.35) than pigs without DFM. Chemical composition, drip loss, TNF $\alpha$ , BUN, and intestinal morphology did not differ between treatments. Collectively, dietary supplementation of a single strain DFM, *Lactobacillus acidophilus* to grower-finisher feeds reduced backfat thickness of pigs without affecting growth performance and loin quality.

**Key Words:** direct-fed microbial, growing-finisher pig, carcass trait

**T287 Nutrient digestibility of high oleic soybean meal by broilers.** Guilherme Hosotani\*, Monty S. Kerley, and Marcia C. Shannon, *University of Missouri, Columbia, MO.*

Two 21-d experiments were conducted to compare soybean meal (SBM) from high oleic parent soybean cultivars on growth performance, ileal AA (IAA) digestibility, and apparent metabolizable energy (AME). Soybean meal was prepared by laboratory-scale mechanical extraction. In Exp. 1, growth performance was evaluated. Fifty 1-d-old male broilers (Ross 308) were randomly placed in battery cages and allocated to 2 dietary treatments with 5 replicates and 5 birds per replicate. The SBM sources consisted of cold-pressed conventional SBM (CON) and cold-pressed high oleic SBM (HO). All SBM sources were heated in a forced-air oven at 120°C for 20 min. Diets were formulated to meet or exceed NRC (1994) requirements. Broilers were weighed and feed disappearance measured on d 7, 14, and 21. In Exp. 2, IAA digestibility and AME were estimated using 100 one-d-old broilers randomly allotted to 3 dietary treatments with 10 replicates and 5 birds per replicate. Chicks were fed a corn-SBM based diet for 17 d, and a diet was developed by substituting the soybean cultivars at 48% in a corn starch-dextrose basal diet and fed from d 18 to 21. A nitrogen-free diet was fed to determine ileal endogenous AA losses. Titanium dioxide (0.5%) was included in all diets as a digesta flow marker. Excreta were collected from d 19 to 21 and ileal content at d 21. Statistical analyses were performed as a randomized complete block design using PROC GLM of SAS with significance level set at  $P \leq 0.05$ . Chicks fed diets containing HO had decreased ADG ( $P < 0.05$ ; 25.3 vs. 29.4 g) and increased feed conversion ratio ( $P < 0.05$ ) from d 0 to 14 (1.71 vs. 1.56) and d 0 to 21 (1.54 vs. 1.47), compared with CON. Amino acid digestibility did not differ among all essential AA. There were no differences in AME (kcal per g) from SBM indirectly calculated between CON and HO, with values of 2,330 and 2,161, respectively. In conclusion, although there were no differences in nutrient digestibility, broilers fed HO had impaired performance compared with CON.

**Key Words:** broiler, soybean, high oleic acid

**T288 Effect of dietary quercetin and oregano essential oil on growth performance, carcass characteristics, meat quality and antioxidant properties in pigs under transport stress conditions.** Yi Zou, Yuanfei Zhou\*, Hongkui Wei, and Jian Peng, *Department of Animal Nutrition and Feed Science, College of Animal Science and Technology, Huazhong Agricultural University, Wuhan, Hubei, China.*

The current study was conducted to investigate the effects of dietary oregano essential oil and quercetin supplementation on growth performance, meat quality and antioxidant property in transported pigs. A total of 336 finishing pigs (Large White  $\times$  Landrace) with an initial body weight of  $72 \pm 4.0$  kg were randomly allotted to 4 groups (5 replicate pens per treatment, 17 pigs per pen). Animals in these 4 groups consumed basal diet (control), basal diet supplemented with 100 mg/kg of vitamin E (VE), or 25 mg/kg of oregano essential oil (OEO) or 25 mg/kg quercetin for 4 wk, respectively. After 4 wk feeding period, 144 pigs were transported for 7 h by high stocking density (275kg/m<sup>2</sup>). Pigs were slaughtered to determine meat quality at 45 min and 24 h postmortem from the M. longissimus dorsi muscles. Blood samples were also collected for measurement of glutathione peroxidase (GSH-Px), total superoxide dismutase (T-SOD), malondialdehyde (MDA) and reactive oxygen species (ROS) levels. Statistical analysis was performed using the GLM procedure (SAS Inst. Inc., Cary, NC). Pen was the experimental unit, and the mean differences were determined using Fisher's test of LSD. The level of statistical significance was taken at  $P < 0.05$  or  $P < 0.01$ . Compared with control group, both OEO and quercetin groups had significant higher average daily gain (ADG), whereas only OEO group had higher final body weight and lower feed intake/gain ( $P < 0.05$ ). Hot carcass weights and dressing percentage was higher in OEO group than those in control and VE groups ( $P < 0.05$ ). The OEO and quercetin groups had significant ( $P < 0.05$ ) lower drip loss and MDA levels but higher ( $P < 0.01$ ) pH value and lightness value in meat compared with control group. In serum, compared with the control group, OEO and quercetin group had increased activation of GSH-Px and T-SOD ( $P < 0.01$ ), and decreased MDA and ROS levels ( $P < 0.01$ ). Moreover, compared with quercetin and VE groups, OEO group had a higher GSH-Px activity in serum. In general, dietary OEO and quercetin may protect against transport stress and improve redox status in transported pigs.

**Key Words:** finishing pig, transport stress, meat quality

**T289 Effect of vitamin E supplementation on performance of male broiler chickens—A meta-analytic approach.** M. A. Pompeu\*, L. F. L. Cavalcanti<sup>2</sup>, and F. L. B. Toral<sup>1</sup>, <sup>1</sup>*Universidade Federal de Minas Gerais, Belo Horizonte, MG, Brazil,* <sup>2</sup>*CNPq, RHAe, Seva Engenharia, Projeto Intergado, Contagem, MG, Brazil.*

Vitamin E (VE) is broadly recognized by its effects on immune response and meat quality of broiler chickens. However, studies have presented controversial results of VE effects on broiler performance. A meta-analysis was conducted to estimate the linear effect of VE supplementation on body weight (BW, 39–49 d of age), daily weight gain (DWG), daily feed intake (DFI) and feed conversion ratio (FCR) of male broiler chickens, using a data set composed by 33 peer-reviewed journal articles from 2010 to 2014. Data were analyzed by linear mixed models approach, where studies (s) were assumed as random effect whereas VE total amount on diet as fixed effect. VE levels in the database were: mean =  $91.89 \pm 106.6$  mg/kg, min = 0 mg/kg, max = 400 mg/kg, while the response variables presented the following averages and observations: BW =  $2.16 \pm 0.66$  kg, n = 41, s = 12; DWG =  $61.6 \pm 16.02$  g, n = 44, s = 13; DFI =  $91.7 \pm 19.6$  g, n = 49, s = 13; FCR =  $1.78 \pm 0.18$  g/g, n = 56, s = 16. The neces-

sity to model error heteroscedasticity was evaluated based on Akaike's information criterion and Schwarz criterion. Significance was declared at  $P \leq 0.05$ . None of the analyzed variables was linearly influenced by VE supplementation, where the estimated slopes were not different from zero with  $P$ -values equal to: 0.99, 0.80, 0.22, 0.40, for BW, DWG, DFI and FCR, respectively. The absolute value of correlation between random terms (i.e., intercept and slope) was never greater than 0.27, what indicates that VE effect is independent of variables' values range, with the exception of DFI ( $r = -0.86$ ). Although 33 studies were evaluated in this research, few of them presented all performance variables simultaneously, what could have reduced the meta-regression power of analysis. Moreover, most studies presented a small range and few VE levels (i.e., less than 4 levels). More standardized and integrated research should be conducted to investigate possible effects of VE on broilers performance; nevertheless, there is not yet, in the current literature, any evidence indicating that the non-effect hypothesis should be rejected.

**Key Words:** meta-regression, nutrition, poultry

**T290 The duration required to detect differences in bone mass accumulation in young pigs fed diets with varied vitamin D, Ca, and P concentrations.** Lynzie M. Miller\*, Laura A. Amundson, and Thomas D. Crenshaw, *University of Wisconsin, Madison, Wisconsin.*

The time course for accumulation of bone mass was established in 49 pigs (~28 d of age) fed diets with either low (LCaP; 75% Ca; 95% P) or high (HCaP; 150% Ca; 120% P) Ca and P (expressed as a percentage of requirements) each formulated without (-D; 0 IU D/kg) or with (+D; 280 IU D/kg) supplemental vitamin D<sub>3</sub>. Bone mass was measured by dual-energy x-ray absorptiometry (DXA). Accumulation of skeletal mass was calculated from DXA scans at weekly intervals for the first 3-wk period, then at 2-wk intervals for the next 4 wk. Animals were fed a recovery diet (+D HCaP) during the last 4 wk. Growth and skeletal mass traits were analyzed as randomized block design for each weekly interval with inferences about diet differences based on orthogonal contrasts for main effects of CaP or vitamin D and the CaP × D interaction. Differences ( $P < 0.01$ ) in skeletal mass due to dietary Ca and P were detected after 1 wk, but differences ( $P < 0.01$ ) due to vitamin D effects were not detected until 2 wk. However, during the recovery phase the initial dietary vitamin D treatments affected recovery responses, not dietary Ca and P. Pigs fed diets without vitamin D failed ( $P < 0.01$ ) to regain skeletal mass over the 4-wk recovery period, but pigs previously fed diets supplemented with vitamin D recovered skeletal mass equal to that of control pigs. In conclusion, DXA provides an effective method to detect differences in bone mineral content gain (gBMC, g/d). Young pigs responded to dietary depletions of Ca and P earlier than to vitamin D, but recovery from a vitamin D depletion was not detected after a 4-wk period.

**Table 1 (Abstr. T290).** Whole-body bone mineral content gain, g/d

Days	-D LCaP	-D HCaP	+D LCaP	+D HCaP	SEM
0 to 8 <sup>a</sup>	0.72	2.43	1.03	2.74	0.33
0 to 16 <sup>abc</sup>	1.14	2.84	3.01	6.20	0.30
0 to 24 <sup>abc</sup>	1.15	2.58	5.22	8.48	0.30
0 to 55 <sup>b</sup>	11.98	12.39	17.92	18.22	0.66

a = CaP effect,  $P < 0.01$ ; b = D effect,  $P < 0.01$ ; c = CaP × D effect,  $P < 0.02$ .

**Key Words:** bone mineral, DXA, recovery

**T291 Comparison of response criteria used to assess dietary vitamin D<sub>3</sub> requirements in young pigs.** Laura A. Amundson\* and Thomas D. Crenshaw, *University of Wisconsin, Madison, WI.*

Recent concerns for increased mortality of young pigs attributed to hypovitaminosis D have focused attention on response criteria used to assess vitamin D requirements. The objective of the current experiment was to determine the dose-response relationships between dietary vitamin D<sub>3</sub> and skeleton mineral accumulation and serum 25-OH D<sub>3</sub> in growing pigs. All pigs were fed diets with 95% of the recommended Ca and P, but no supplemental vitamin D for 1 wk post-weaning. At 1 wk after weaning (~25 d), 84 crossbred pigs (n = 4 pigs/pen, 3 pens/diet) were randomly assigned to 1 of 7 diets formulated to supply 95% of the recommended Ca and P concentrations with either 0, 50, 100, 200, 400, 800 or 1,600 IU D<sub>3</sub>/kg. Sows that produced the litters were fed the UW Swine Research and Teaching Center standard gestation and lactation diets, both formulated to supply 325 IU vitamin D<sub>3</sub>/kg. A segmented 2-phase linear model was applied to regress variables to vitamin D intake. Growth (ADG, kg/d), whole body bone mineral content (BMC, g) and bone mineral density (BMD, g/cm<sup>2</sup>) via dual energy x-ray absorptiometry (DXA) scans, femur mechanical properties (bending force, yBM, kg-cm; stress, kg/cm<sup>2</sup>) responses at the end of the 28-d trial (n = 6 pigs/diet, 2 pigs/pen) peaked at diet concentrations between 50 to 100 IU vitamin D<sub>3</sub> ( $P < 0.05$ ), but serum 25-OH D<sub>3</sub> concentrations continued to increase as concentrations of dietary vitamin D<sub>3</sub> increased to 1,600 IU. Intersections of the 2 regression slopes were 81, 84, and 77 IU vitamin D<sub>3</sub>/d for ADG, BMD, and yBM respectively. Slopes for the upper range were not significantly different. No plateau was detected in serum 25-OH D<sub>3</sub> concentrations. Although limited by the number of groups in the lower range, diets with 100 IU vitamin D<sub>3</sub>/kg appear adequate to meet requirements of young pigs for growth and skeletal traits. No benefit was attributed to the increased serum 25-OH D<sub>3</sub> concentrations.

**Table 1 (Abstr. T291).** Effects of supplemental vitamin D<sub>3</sub> on performance

Trait	Vitamin D <sub>3</sub> (IU/kg diet)						SEM	
	0	50	100	200	400	800		1,600
ADG, kg/d	0.338	0.385	0.452	0.472	0.485	0.434	0.486	0.024
BMD, g/cm <sup>2</sup>	0.289	0.327	0.371	0.380	0.397	0.366	0.370	0.014
yBM, kg-cm	278	318	463	434	473	451	405	50
25-OH D <sub>3</sub> , ng/mL	2.1	1.9	2.1	5.3	9.5	13.4	18.4	1.0

**Key Words:** hypovitaminosis D, bone mineral, mechanical test



## Nonruminant Nutrition: Sows and piglets

**T292 Meta-analysis of the influence of live yeast addition on feed intake in lactating sows.** Eric Chevaux<sup>\*1</sup>, David Guillou<sup>1</sup>, and Ernest Keith<sup>2</sup>, <sup>1</sup>Lallemand Animal Nutrition, Blagnac, France, <sup>2</sup>Lallemand Animal Nutrition, Milwaukee, WI.

Increasing the lactating sow feed intake is generally considered beneficial, but high inter-individual variability of the measurements makes difficult designing the trials on this subject. A meta-analysis of 10 trials (1111 sows, 8 genetics lines, parity 1–8, lactation duration 19–31 d) was undertaken, comparing “control” diets (n = 12) to diets supplemented with the same strain of live yeast *Saccharomyces cerevisiae* boulardii CNCM I-1079 (LY) at 1.0 or 2.0 × 10<sup>9</sup> cfu/kg (n = 12), to increase the chance to detect feed intake differences. In these trials, individual feed intake of sows (ranging 3.9 to 8.5 kg/d) was measured; trials were performed in commercial farms or close situation, in Europe or North America. Measurements of litter weight at weaning and number of piglets weaned could be retrieved from all 10 trials, but not changes of sow weight or P2 backfat thickness. Data were analyzed by ANOVA (SPSS 19.0) incorporating a random effect of trial, the treatment effect being considered weighted or not for experimental power or measurement variability. Effect of location of the experiment, year (1999 to 2013), lactation length, or duration of pre-treatment with live yeast were incorporated in the model, then excluded because of lack of significance (P > 0.20). Litter size, litter weight at weaning, parity or backfat thickness were included as covariate, and then discarded from final model as neither significance nor correlations were found. Live yeast added to sows diets stimulated appetite overall (Table 1), leading to a significant increase of feed intake ranging between +200 (weighted by number of replicates) and +300 g/d (un-weighted). Feed intake difference higher than the mean standard error of measurement was found in 6 trials out of 10. Nonetheless, factors affecting the magnitude of yeast effect could not be elucidated with only 10 trials in the compilation. Subsequently, the present results deserve being confirmed with new studies including accurate measurements of sows DMI and body condition.

**Table 1 (Abstr. T292).** DMI (kg) per treatment according to the statistical model

Weighted	Control	LY	SEM	P-value
None	5.714	5.997	0.346	0.03
1/Var	5.725	5.985	0.340	<0.01
No. of replicates	5.608	5.805	0.345	0.01

**Key Words:** sow, DMI, live yeast

**T293 Effects of palm kernel expellers on productive performance, nutrient digestibility, and white blood cells of lactating sows.** J. Kim<sup>\*1</sup>, Y. Jang<sup>1</sup>, S. Kim<sup>1</sup>, W. Kim<sup>1</sup>, K. Jang<sup>1</sup>, K. Kim<sup>1</sup>, B. Kim<sup>1</sup>, S. Park<sup>1</sup>, I. Park<sup>1</sup>, Y. H. Kim<sup>2</sup>, J. C. Park<sup>2</sup>, J. Seo<sup>3</sup>, Y. Kim<sup>4</sup>, S. Seo<sup>1</sup>, M. Song<sup>1</sup>, <sup>1</sup>Chungnam National University, Daejeon, Republic of Korea, <sup>2</sup>National Institute of Animal Science, Cheonan, Republic of Korea, <sup>3</sup>Pusan National University, Miryang, Republic of Korea, <sup>4</sup>Chonbuk National University, Jeonju, Republic of Korea.

A preliminary study was conducted to investigate the effects of palm kernel expellers on productive performance, nutrient digestibility, and changes in white blood cells (WBC) of lactating sows. A total of 14 sows (200 ± 12 kg BW; 2.5 parity) were randomly assigned to 2 dietary treatments: a diet based on corn and soybean meal (CON) and CON + 20% palm kernel expellers (PKE). Sows were fed the treatments for 28

d (weaning) after farrowing. Blood was collected from each sow and randomly selected 4 piglets in each sow before farrowing or on d 3, 7, or 14 of lactation. Sows were fed respective treatments containing 0.2% chromic oxide from d 15 to 21 of lactation. Fecal samples were collected daily for the last 3 d after the 4-d adjustment period. Measurements were performances and WBC changes of sows and litter, nutrient digestibility of sows, and daily diarrhea of litter. Data were analyzed using the PROC GLM procedure of SAS. The statistical model for every measurement except frequency of diarrhea included dietary effect and parity as a covariate. The chi-squared test was used for the frequency of diarrhea. Sows fed PKE had greater ADFI (7.38 vs. 7.10 ± 0.06 kg/d; P < 0.05) and lost less BW (–6.85 vs. –8.54 ± 0.51 kg; P < 0.05) and backfat depth (–0.42 vs. –0.71 ± 0.09 mm; P < 0.05) than those fed CON. However, there were no differences on digestibility of dry matter, nitrogen, and energy and weaning to estrus interval of sows fed either CON or PKE. Piglets from sows fed PKE gained more BW (203 vs. 181 ± 7.62 g/d; P = 0.08) and had less frequency of diarrhea (6.80 vs. 8.56%; P = 0.07) than those from sows fed CON. However, no difference was found on preweaning mortality of piglets from sows fed either CON or PKE. Sows fed PKE had lower number of WBC (9.57 vs. 11.82 ± 0.83 × 10<sup>3</sup>/μL; P = 0.09) before farrowing than those fed CON, but no difference on d 3 and 7. Similarly, piglets from sows fed PKE had lower number of WBC (7.86 vs. 9.80 ± 0.62 × 10<sup>3</sup>/μL; P < 0.05) on d 14 of lactation than those fed CON, but no difference on d 3 and 7. In conclusion, PKE increased ADFI and decreased changes of BW and backfat depth of lactating sows compared with CON, but no differences were found on other measurements.

**Key Words:** lactating sow, palm kernel expellers, performance

**T294 Effects of the level of bovine plasma in pregnant gilts on the birth weight of the litter.** Bárbara V. Freitas<sup>\*</sup>, Simone M. M. K. Martins, Jose A. R. Ulloa, Gustavo A. Campos, and Lúcio F. Araújo, University of São Paulo, Pirassununga, São Paulo, Brazil.

Plasma is considered an important ingredient in animal feed for high quality, functional protein. In piglets, it has been associated with causing a reduction in inflammation and immune stimulation. The use of the ingredient is growing in sows during the pregnancy to improve weight and homogeneity at birth and the body condition of the sow at the end of lactation, reducing the number of “days open” before conception. The study aimed to evaluate the effect of different plasma levels in the diet of gilts on the birth weight of piglets. Twenty-four pregnant, primiparous females (Topigs Norsvin) aged 215 ± 9.46 d and weighing 127 ± 8.0 kg were used. The compositions of the diets were based on corn and soybean meal, in accordance with NRC recommendations (2012). The experimental design was completely randomized, with 4 treatments: 0, 0.5, 1.0 and 2.0% of plasma added to the feed. Six repetitions per treatment were used and the experimental unit was one primiparous pregnant sow. During the gestational period, from 1 to 3 d after insemination, the gilts consumed a mean 1.200 kg of feed per day, from 4 to 79 d, a mean of 1.800 kg per day and from 80 to 110 d, a mean of 2.800 kg of feed per day. Blood plasma was added to the diet formulation according to treatment. After birth, all the live piglets, placentas, stillbirths, and mummies were weighed. The mean weight of the complete litter, including mummies and stillbirths, was 16.85 kg, 17.36 kg, 17.63 kg and 18.52 kg for the treatments 0, 0.5, 1.0 and 2.0% of added plasma, respectively. The characteristics of total litter weight, live births weight, fetal membrane weight and number of live-born, stillborn and mummy-

fied piglets were not affected by the inclusion of plasma in the diet of sows. The characteristics of the total litter weight, live births weight, fetal membrane weight and number of live-born piglets were analyzed using PROC REG (SAS, 2012), while stillborn and mummified piglets were analyzed using PROC GLIMMIX, Poisson distribution (SAS, 2012). Level of significance  $P < 0.05$ .

**Key Words:** primiparous pregnancy, gestation, blood plasma.

**T295 Effect of dietary supplementation of oregano essential oils to sows on oxidative stress status, lactation feed intake and reproductive performance.** Chengquan Tan<sup>1</sup>, Jiangtao Ao<sup>1</sup>, Guang Long<sup>1</sup>, Haiqing Sun<sup>1,2</sup>, and Jian Peng<sup>\*1</sup>, <sup>1</sup>*Department of Animal Nutrition and Feed Science, College of Animal Science and Technology, Huazhong Agricultural University, Wuhan, Hubei, China,* <sup>2</sup>*YangXiang Joint Stock Company, Guigang, Guangxi, China.*

The aim of the current study was to evaluate the effects of supplementing sow diets with oregano essential oils (OEO) during gestation and lactation on oxidative stress status, lactation feed intake and reproductive performance of sows. Sixty multiparous large white sows were randomly assigned to 1 of 2 dietary treatments within 24h after service to weaning: control (C) or C+ 300mg/kg of OEO (OEO). Serum samples collected from sows on d 10, 60, 90 and 109 of gestation and d 1, 3, 7 and 21 of lactation were used to determine reactive oxygen species (ROS), glutathione peroxidase (GSH-Px) and oxidative stress markers [malondialdehyde (MDA), 8-hydroxy-deoxyguanosine (8-OHdG)] levels. Data were analyzed using GLM procedure. The average daily gain (ADG) of suckling piglets was subjected to analysis of covariance with the piglet weight after cross-foster as the covariate. Variations of oxidative stress parameters were using the procedure for repeated measurements, which included the effects of treatment, physiological stage and replicate. Differences between means were significant at  $P < 0.05$  and trends identified when  $P > 0.05$  but  $\leq 0.10$ . The results showed that serum levels of ROS, MDA and 8-OHdG were higher ( $P < 0.05$ ) during gestation (d 90, 109) and lactation (d 1, 3) than early gestation (d 10). Sows fed OEO diet elevated serum concentration of GSH-Px on d 60 of gestation ( $P = 0.08$ ), and d 1 ( $P = 0.07$ ) of lactation than C group, while serum concentrations of MDA on d 1 of lactation ( $P < 0.01$ ), 8-OHdG on d 109 of gestation ( $P = 0.09$ ), d 1 ( $P = 0.06$ ) and d 3 ( $P = 0.10$ ) of lactation and ROS on d 1 ( $P = 0.10$ ) and d 3 ( $P = 0.10$ ) of lactation were lower in sows treated with OEO dietary than C group. Dietary OEO treatment increased sow feed intake in the third week of lactation (6.46 vs 6.03 kg/d,  $P = 0.07$ ), accordingly, had higher ADG (252.36 vs 233.61 kg/d,  $P < 0.01$ ) of suckling piglets of entire lactation than C group. In conclusion, our results demonstrated that there was an increased systemic oxidative stress during late gestation and early lactation of sows. Nevertheless, OEO dietary supplementation attenuated oxidative stress in sows.

**Key Words:** oxidative stress, oregano essential oil, sow

**T296 Sow productivity at farrowing was not affected when 50% of a co-product combination was included in the gestation diet.** Elizabeth Magowan<sup>1</sup>, Paul McMullen<sup>1</sup>, Aishling O'Connell<sup>2</sup>, Rosanna Wregor<sup>2</sup>, and Wallace Henry<sup>\*3</sup>, <sup>1</sup>*Agri-Food and Biosciences Institute, Hillsborough, United Kingdom,* <sup>2</sup>*JMW Farms, Tynan, United Kingdom,* <sup>3</sup>*Rektify Limited, Gilford, United Kingdom.*

This study, aimed to compare the performance of sows offered a diet containing a high level of co-products during gestation (d5–112). A control (cereal/soy) gestating diet was compared with one containing 50%

of a co product (a blend of potato skins, whey permeate, and residual grains and effluent from the production of an alcoholic beverage). A total of 176 sows (88/treatment), penned in groups of 8, were group fed over 11 time periods (each group was balanced for weight, parity and condition score). Individual sows were used as the experimental unit when assessing effects on litter performance. Lactation feed intake was measured for 10 pairs of sows per treatment. The DE, CP, lysine and CF content of the diets were formulated to be 2.77 and 2.96 Mcal/kg, 17.7 and 14.4%, 0.7 and 0.7%, 9.1 and 7.4% for the co product and control diets respectively. Sows were offered feed once a day during gestation using a feed curve that offered sows 8.60 Mcal/d from d 7–34, 6.69 Mcal/d from d 35–83 and 8.60 Mcal/d from d 84–112 of gestation. Sows were fed ad lib during lactation they farrowed. Changes in sow condition score and weight and litter performance were measured. Sow dirtiness was scored on a 0–3 point scale. Statistical analysis used ANOVA with period as a blocking factor. There was no difference in sow parity (3.85), weight (204 kg) or condition scoring (2.9) on d 5 of gestation. Due to the lower energy content of the co product diet, total sow intake was numerically higher when it was offered (245 vs. 229 kg). There was no effect of diet on sow weight at farrowing (266 kg), condition score (3.2), litter size (13.6), number born alive (12.7), dead (0.73) or mummified (0.17). When the co-product diet was offered, sows were dirtier ( $P < 0.001$ ) (2.09) compared with when the control diet was offered (1.03). The average daily lactation feed intake of sows was statistically similar when the co product diet (5.97 kg/d) was offered compared with when the control diet was offered (6.62 kg/d). In conclusion, the inclusion of 50% of the blended co product had no significant impact on sow weight, condition score or productivity at farrowing.

**Key Words:** dry sow, co-product, performance

**T297 Effect of supplying a nucleotide product (Harv-con NT) to sow diets on the reproductive performance and the health status of the offspring.** I-Fen Hung<sup>\*1</sup>, Fuguei Li<sup>2</sup>, Shigeng Zou<sup>3</sup>, and Merlin D. Lindemann<sup>1</sup>, <sup>1</sup>*University of Kentucky, Lexington, KY,* <sup>2</sup>*Interflavor Ltd., Zhongshan City, Guangdong, China,* <sup>3</sup>*WENS Group, Yunfu City, Guangdong, China.*

Nucleotides have demonstrated their ability to improve several biological responses during stress periods, including parturition. This experiment was conducted in a commercial farm (WENS Groups) in China. Around d 87 of gestation, sows ( $n = 316$ ) were assigned to 2 dietary treatments based on their housing unit: 1) the standard farm gestation diets provided by WENS Group or 2) the standard diets top dressed with 3.5 g/sow/d of a nucleotide product (Harv-con NT, NT; Interflavor Ltd., China). Approximately 1 week before expected parturition, sows were moved to farrowing facilities and assigned to 2 dietary treatments: (1) the standard farm lactation feed; or (2) the standard diet top dressed with 8 g/sow/d (on the day of moving to 7 d post-farrowing) and 12 g/sow/d (7 d post-farrowing till weaning) NT in the lactation diet, which resulted in a  $2 \times 2$  factorial arrangement. The data were analyzed using GLM program of SAS with the housing unit as the experimental unit, 4 units per treatment. Supplying NT to sow diets during late gestation had no effect on the total litter size at birth as expected (11.5 vs. 11.9,  $P = 0.19$ ), but reduced the “healthy” pig litter size (9.9 vs. 10.8,  $P = 0.01$ ) as determined by the farm. Litter weight of the healthy pigs at birth was not affected by the treatments (14.4 vs. 14.4,  $P = 0.88$ ), thus the individual birth weight was increased in NT sows (1.4 vs. 1.3kg,  $P = 0.01$ ). In the units with diarrhea, less diarrhea was observed in litters from sows fed NT in gestation compared with those from Control sows (25 vs. 69%,  $P = 0.07$ ). Also, more sows in the gestation NT treatment returned to heat within 7 d post weaning compared with control sows

(95 vs. 82%,  $P = 0.01$ ). Neither NT treatment in gestation or lactation had an effect on colostrum and d13–17 milk nutrient composition (i.e., fat, protein, and lactose) or immunoglobulin profile. In conclusion, adding NT to the sow diets during late gestation increased healthy pig birth weight and ratio of returning heat within 7d post weaned. It also reduced diarrhea incidence during the suckling period in the units that experienced diarrhea. However, supplying NT to the sow diets during lactation had no effects on these observations. Supplying NT to sow diet during gestation or lactation had no effects on milk composition as well.

**Key Words:** nucleotide, sow, piglet

**T298 The effect of coated sodium butyrate supplementation in sow and nursery diets on reproductive performance and nursery pig performance.** Young Dal Jang\*, Merlin D. Lindemann, H. James Monegue, and James S. Monegue, *University of Kentucky, Lexington, KY*.

The experiment was conducted to evaluate the effect of coated sodium butyrate (CSB) supplementation to peripartal and lactating sows and nursery pigs on reproductive performance and nursery growth performance. In Exp. 1, a total of 43 gestating sows (d 81–90 of gestation) were allotted to 0 ( $n = 15$ ), 500 ( $n = 16$ ), and 1,000 ( $n = 12$ ) ppm of CSB (CM3000, Hangzhou King Techina Feed Co., Ltd., China) based on breed, parity and BW, and then fed treatment diets until weaning. In Exp. 2, a total of 72 weanling pigs were selected from the 0 ( $n = 5$  sows) and 1,000 ( $n = 4$  sows) ppm CSB treatments in Exp. 1 and allotted within the sow treatment to 0, 500, and 1,000 ppm of CSB in nursery diets for 3 replicates with 4 pigs per pen as a split plot design (SPD) for a 35-d growth study. All pigs were injected with ovalbumin at weaning and d 14 postweaning as an immune challenge. All data were analyzed by ANOVA using GLM procedure of SAS as a completely randomized design in Exp. 1 and a SPD in Exp. 2. In Exp. 1, there was no difference on sow BW, litter size, litter weight, lactation feed intake or milk composition. However, colostral IgG ( $P = 0.06$ ; 2,447, 3,337, and 2,723 mg/dl for 0, 500, and 1,000 ppm of CSB treatments, respectively) and IgA ( $P = 0.09$ ; 406, 540, and 464 mg/dl) concentrations tended to increase quadratically as CSB supplementation levels increased. In Exp. 2, pigs from the 1,000 ppm CSB sow treatment had greater BW ( $P < 0.01$ ; 23.1 vs. 21.6 kg), ADG ( $P < 0.01$ ; 0.462 vs. 0.418 kg/d), ADFI ( $P < 0.01$ ; 0.776 vs. 0.684 kg/d) and F:G ratio ( $P = 0.07$ ; 1.67 vs. 1.64) than those from the 0 ppm CSB sow treatment at d 35 postweaning. For the nursery treatments, ADG ( $P < 0.05$ ; 0.428, 0.428, and 0.463 kg/d for 0, 500, and 1,000 ppm CSB, respectively) and ADFI ( $P = 0.06$ ; 0.707, 0.717, and 0.767 kg/d) during the 35-d period increased linearly as CSB supplementation levels increased whereas F:G ratio had a quadratic response ( $P = 0.10$ ; 1.64, 1.68, and 1.64). In summary, CSB supplementation increased colostral IgG and IgA concentrations of sows when supplemented in the sow diet and improved growth performance of nursery pigs under an immune challenge when supplemented in the nursery diet.

**Key Words:** coated sodium butyrate, reproductive performance, sow

**T299 Nucleotide supplementation improves growth performance of weaned piglets.** Gabriela de Mello Miassi\*<sup>1</sup>, Luan Sousa Santos<sup>2</sup>, Livea Maria Gomes<sup>1</sup>, Patrícia Nardin Berto<sup>1</sup>, Mayra Dib Saleh<sup>1</sup>, Alessandro Borges Amorin<sup>3</sup>, Marcos Livio Panhoza Tse<sup>1</sup>, and Dirlei Antonio Berto<sup>1</sup>, <sup>1</sup>Faculdade de Medicina Veterinária e Zootecnia, FMVZ/UNESP, Botucatu, São Paulo, Brazil, <sup>2</sup>Faculdade de Ciências Agrárias e Veterinárias, FCAV/UNESP, Jaboticabal, São Paulo, Brazil, <sup>3</sup>Instituto de Ciências Agrárias e Tecnológicas, ICAT/UFMT, Rondonópolis, Mato Grosso, Brazil.

This study was conducted to evaluate effects of dietary supplementation of nucleotide (disodium 5 guanylate and disodium 5 inosinate) on the growth performance and plasmatic lipid profile of weaned piglets. Eighty-four weaned piglets of 21 d of age ( $6.04 \pm 0.25$  kg average initial BW) were allotted to 4 dietary treatments with 7 pens per treatment and 3 pigs per pen, in a randomized complete block design. The treatments were dietary supplementation with 0% (control); 0.2%; 0.4% or 0.6% nucleotide of diet during 34 d. Performance was determined from 21 to 36, 21 to 48 and 21 to 55 d of age. Blood samples were collected to determinate lipid profile (triglycerides, total cholesterol, HDL cholesterol and LDL+VLDL cholesterol) on 21 and 55 d of age. Data were submitted to ANOVA using PROC GLM of SAS and the effects of supplementation levels of nucleotide were studied through regression analysis. During the first period (21 to 36 d of age) no differences were detected on the performance. During the second period (21 to 48 d of age) a quadratic effect ( $P < 0.01$ ) of the levels of nucleotide was verified on average daily feed intake and average daily weight gain, with optimal response to the levels of 0.31% and 0.32%, respectively. During the whole experimental period (21 to 55 d of age) there was a quadratic effect of the levels of nucleotide ( $P < 0.01$ ) on average daily weight gain with optimal response to the level of 0.31%. There was no significant effect on triglycerides, total cholesterol, HDL cholesterol and LDL+VLDL cholesterol. The overall results suggest that dietary nucleotide supplementation at 0.32% improves the performance of weaned piglets.

**Key Words:** disodium 5 guanylate, disodium 5 inosinate, pig

**T300 Effect of nucleotide supplementation in high soybean meal inclusion diets on weight gain and inflammatory process in weanling pigs.** David Solà-Oriol<sup>1</sup>, Wellington Coloma<sup>1</sup>, Elisabet Borda\*<sup>2</sup>, and José Francisco Pérez<sup>1</sup>, <sup>1</sup>Animal Nutrition and Welfare Service, Department of Animal and Food Sciences, Universitat Autònoma de Barcelona, Bellaterra, Spain, <sup>2</sup>R&D Animal Nutrition, Bioibérica, Palafròlles, Spain.

Soybean protein ingredients (SB as soybean meal; SBM and soya protein concentrates; SBP) are widely used in pig diets; however they may also promote acute intestinal inflammation and affect gut mucosal integrity and performance. Several beneficial effects of nucleotide supplementation on gut immune function, inflammation and diarrhea has been reported, which could reduce the negative effects SB in piglets. The aim of the present work was to study the effect of including nucleotides (Nucleoforce Piglets, Bioibérica, SA) in diets containing different levels of SB on acute inflammation process and early performance of weaned piglets. Two hundred forty 28-d-old piglets [Pt  $\times$  (LD  $\times$  LW)] were distributed into 24 pens (10 pigs/ pen) according to the initial BW following a RCBD. Two pre-starter diets differing in the SB content as protein source (Low or High) formulated to contain 10.3 MJ/kg of NE, 19.5% CP and 1.28% Lys were supplemented with or without nucleotides (500 mg/kg) following a  $2 \times 2$  factorial arrangement. High SB diet contained 15% of extruded soybeans, 6.82% SBM and 5.62% SBP; while Low SB diet was obtained by partly replacing SB ingredients with fish meal (8.44%) and animal plasma (2.5%) according to their protein content. Feed was offered ad libitum in mash form. Individual animal weight and feed disappearance were recorded at d 28 and 42 of live. Blood samples were collected on d 7 to determine TNF $\alpha$  as inflammation marker. Data was analyzed with ANOVA following a  $2 \times 2$  factorial arrangement with GLM procedure of SAS. Higher BW (9.59 vs 9.09 kg;  $P = 0.03$ ), ADFI (205 vs 167 g/d;  $P = 0.002$ ) and ADG (140 vs 103 g/d;  $P = 0.04$ ) was observed for Low SB diet than high SB diet, independently of the nucleotide supplementation. An interaction



between SB inclusion in the diet and nucleotide supplementation was observed ( $P = 0.04$ ) reflecting that nucleotides promote a decrease of TNF $\alpha$  in High SB group (95.1 vs 67.4 pg/mL) but not in Low SB group (60.3 vs 70.1 pg/mL) without and with nucleotides, respectively. It is concluded that nucleotide supplementation in diets containing high inclusion of soybean meal and soybean protein products reduces acute inflammation process.

**Key Words:** soybean meal, nucleotide, inflammation

**T301 Effects of palm kernel expellers on growth performance, nutrient digestibility, and blood profiles of weaned pigs.** Y. Jang<sup>\*1</sup>, J. Kim<sup>1</sup>, S. Kim<sup>1</sup>, W. Kim<sup>1</sup>, K. Jang<sup>1</sup>, K. Kim<sup>1</sup>, B. Kim<sup>1</sup>, S. Park<sup>1</sup>, I. Park<sup>1</sup>, Y. H. Kim<sup>2</sup>, J. C. Park<sup>2</sup>, J. Seo<sup>3</sup>, Y. Kim<sup>4</sup>, S. Seo<sup>1</sup>, M. Song<sup>1</sup>,  
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A study was conducted to investigate the effects of palm kernel expellers on growth performance, nutrient digestibility, and blood profiles of weaned pigs. A total of 88 weaned pigs (6.94  $\pm$  0.76 kg BW; 28 d old) were randomly allotted to 2 dietary treatments (4 pigs/pen; 11 replicates/treatment): a typical nursery diet based on corn and soybean meal (CON) and CON + 20% of palm kernel expellers (PKE). Pigs were fed for 6 wk using a 3-phase feeding program with declining diet complexity and with phases of 1, 2, and 3 wk, respectively. Blood was collected from randomly selected 2 pigs in each pen before weaning and on d 7 after weaning. Pigs were fed dietary treatments containing 0.2% chromic oxide from d 29 to 35 after weaning. Fecal samples were collected from randomly selected 2 pigs in each pen daily for the last 3 d after the 4-d adjustment period. Measurements were growth performances, nutrient digestibility, blood profiles, and incidence of diarrhea. Data were analyzed using the PROC MIXED procedure of SAS. The statistical model for every measurement except frequency of diarrhea included dietary effect as a fixed effect and sex as a random effect. The chi-squared test was used for the frequency of diarrhea. The PKE tended to increase ADG (246 vs. 215 g/d;  $P = 0.06$ ), increased ADFI (470 vs. 343 g/d;  $P < 0.05$ ), and decreased G:F (0.522 vs. 0.628 g/g;  $P < 0.05$ ) during phase 2 compared with CON, but did not affect growth performance during phase 1 and 3. During overall experimental period, PKE tended to increase ADG (383 vs. 362 g/d;  $P = 0.05$ ) and increased ADFI (549 vs. 496 g/d;  $P < 0.05$ ) compared with CON, but did not affect G:F. However, no differences were found on digestibility of dry matter, nitrogen, and energy between CON and PKE. The PKE tended to reduce frequency of diarrhea (15 vs. 25%;  $P = 0.08$ ) for the first 2 wk after weaning compared with CON. Similarly, PKE tended to decrease white blood cells (8.19 vs.  $9.56 \times 10^3/\mu\text{L}$ ;  $P = 0.07$ ), red blood cells (2.92 vs.  $3.25 \times 10^6/\mu\text{L}$ ;  $P = 0.09$ ), and packed cell volume (11.1 vs. 12.6%;  $P = 0.06$ ) on d 7 after weaning compared with CON. In conclusion, PKE had no negative effects on growth performance, nutrient digestibility, and blood profiles of weaned pigs.

**Key Words:** growth performance, palm kernel expellers, weaned pig

**T302 Beneficial effects of a prebiotic supplement on growth and performance of weaned piglet.** Edi Vianello, Anne-Kathrin Blässe\*, and Bernhard Eckel, Dr. Eckel GmbH, Niederzissen, Germany.

Weaning is a crucial phase in piglet rearing as the change from milk to solid feed in combination with external stressors such as regrouping may be related to an impaired gut and animal health. Villus atrophy and

a limited activity of digestive enzymes can lead to an accumulation of undigested feed in the lower intestinal segments followed by increases in the count of pathogenic bacteria such as *E. coli* and subsequent diarrhea. Prebiotics are non-digestible feed components that selectively stimulate the growth of positive bacteria such as lactobacilli and may thus reduce pathogens. The present study investigated the potential of a specific prebiotic acid and salt mixture including formic and lactic acid and their salts as well as butyrate and gluconate (PreAcid, Dr. Eckel GmbH, Niederzissen, Germany) in increasing piglet growth and performance. Twenty weaned crossbred piglets each [Pietrain x (Large White x German Landrace)] with an initial BW of 8  $\pm$  1 kg were randomly allocated to a control group and a treatment group. The animals were individually fed a diet based on wheat, barley and soybean meal for a period of 6 wk and the treatment group received the basal diet supplemented with 1% PreAcid. Statistical analyses were performed using SAS (fixed factors: mother, sex, treatment, group). Energy efficiency (6,139 kcal/kg weight gain vs 6,330 kcal/kg weight gain,  $P < 0.05$ ) and energy conversion (39 g/kcal vs 38 g/kcal,  $P < 0.05$ ) were significantly improved by the treatment. In conclusion, the present trial demonstrated that the inclusion of the specific prebiotic acidifier in diets for weaned piglets increased feed efficiency indicating an improved energy utilization.

**Key Words:** prebiotic, piglet, growth performance

**T303 Effect of dietary melatonin supplementation on growth performance and behavior, and their correlations in weaned pigs.** Kyeongsu Chae<sup>\*1</sup>, Junseung Choi<sup>2</sup>, Jonggun Kim<sup>1</sup>, and Kwang-Youn Whang<sup>1</sup>,  
<sup>1</sup>Department of Biotechnology, Graduate School, Korea University, Seoul, Korea, <sup>2</sup>Dodram Swine Service, Gyeonggi, Korea.

Some behaviors such as roaming and fighting observed in weaned pigs have been considered to be unhelpful to growth performance. Melatonin (MEL) secreted from the pineal gland has a sedative effect on human behavior. In this study, MEL was supplemented in the weaned pig diets to determine the effect of MEL on growth performance and behaviors, and correlation between them. Two hundred twenty-five 21-d-old pigs were allotted into 4 treatments (9 or 10 pigs per pen) with 6 replications and fed diets containing MEL at doses of 0, 25, 50 and 100 mg/kg of diet (M0, M25, M50 and M100) for 35 d. By using closed-circuit television cameras, the behavior of pigs was recorded to score resting (RT), roaming (RM), feeding (FD) and fighting (FT) states for 24 h on d 3, 10, 17, 24 and 31. The BW and feed intake were measured every week to calculate ADG and feed efficiency (FE). The data were analyzed by ANOVA, followed by the Duncan's new multiple range test. Correlations between growth performance and the behaviors were analyzed with reference to the Pearson correlation coefficients. The BW was higher in M25 group (not different from M0) than M50 and M100 groups on d 28 ( $P < 0.05$ ) but there was no difference at the end of experiment. The ADG was higher in M25 group than the other groups during d 21–28 ( $P < 0.05$ ). There were no differences in ADFI and FE throughout the experiment. The RT was lower and RM was higher in M50 group (not different from M0) than M25 and M100 groups on d 10 ( $P < 0.05$ ) but there were no differences on the other days. The MEL did not alter FD and FT throughout the experiment. Correlations between growth performance and behavior were found in M0 group but not in MEL-treated groups (M25, M50 and M100) during d 7–14. Therefore, it is concluded that MEL at dose of 25 mg/kg of diet may affect BW in weaned pigs on d 28 according to the improved ADG during d 21–28 but MEL does not affect ADFI and FE. This study also suggests that MEL affects RT and RM within 14 d. However, the behaviors affected by MEL are not correlated with growth performance.

**Key Words:** pig, melatonin, behavior

# Physiology and Endocrinology: Environment, metabolism and physiological processes

**T304 Effect of the environmental conditions over the vaginal temperature and respiration rate on wild type and slick-haired Puerto Rican Jersey cows.** Amneris M. Castro-Ramos\*, Gladycia C. Muñiz-Colón, Jaime E. Curbelo-Rodríguez, Melvin Pagán-Morales, Alexander Mesonero-Morales, Adalberto de Jesus-de Jesus, Neftali Lluch-García, and Héctor L. Sánchez-Rodríguez, *University of Puerto Rico at Mayagüez Campus, Mayagüez, Puerto Rico.*

The slick hair phenotype is associated with enhanced thermoregulation in Holstein cows; however, no such data are available in slick-haired Jersey cows. The relationship between hair coat type (phenotypically determined), vaginal temperature (VT; as an index of core body temperature), respiration rate (RR), and thermal humidity index (THI) was evaluated in 10 Puerto Rican Jersey cows [5 wild type-haired (WT;  $111.2 \pm 0.8$  d in milk (DIM); 1.2 lactations) and 5 slick-haired (SLICK;  $171.6 \pm 1.5$  DIM; 1.2 lactations)]. Cows were exposed to solar radiation from 1030 to 1130 h and to artificial shade from 1130 to 1030 h during 5 consecutive days. Data loggers recorded VT (Tidbit v2), air temperature, and relative humidity (HOBO Pro v2) every 5 min and the THI was determined. Daily THI ranged from 63.47–81.77. The RR was recorded at 0600, 0800, 0900, 1000, 1100, 1200, 1300, 1400, and 1500 h daily. Data were analyzed using the GLIMMIX and CORR procedures of SAS. There were no differences in VT between hair type groups either under sun or shade exposure ( $P = 0.43$ ). From 1030 to 1130 h (direct solar radiation exposure) mean VT values of  $39.08 \pm 0.02$  and  $39.26 \pm 0.02^\circ\text{C}$  were observed in SLICK and WT cows, respectively. During the 1130–1030 h (shade period), SLICK and WT cows presented mean VT values of  $38.80 \pm 0.01$  and  $38.89 \pm 0.01^\circ\text{C}$ , respectively. However, SLICK cows presented lower RR than their WT counterparts ( $P = 0.004$ ). Mean daily RR values of  $56.0 \pm 5.2$  and  $77.6 \pm 5.0$  breaths per minute were observed in the SLICK and the WT cows, respectively. Correlation coefficients of 0.68 ( $P < 0.0001$ ) and 0.47 ( $P < 0.0001$ ) were observed between the VT and the RR in WT and SLICK cows, respectively. However, VT was less associated with THI in WT than in SLICK cows [ $r = 0.24$  ( $P < 0.0001$ ) and  $r = 0.50$  ( $P < 0.0001$ ), respectively]. In conclusion, slick-haired Jersey cows were able to maintain similar vaginal temperature than their wild type-haired counterparts, but with a lower respiration rate. These findings suggest that wild type-haired Jersey cows' body temperature regulation requires greater heat dissipation through evaporation than their slick-haired counterparts.

**Key Words:** slick-haired Jersey cow, thermoregulation, solar radiation

**T305 Effects of the thermal humidity index on vaginal temperature of slick- and wild type-haired Puerto Rican Holstein cows.** Héctor L. Sánchez-Rodríguez\*, Amneris M. Castro-Ramos, Melvin Pagán-Morales, Jaime E. Curbelo-Rodríguez, Alexander Mesonero-Morales, and Gladycia C. Muñiz-Colón, *University of Puerto Rico at Mayagüez Campus, Mayagüez, Puerto Rico.*

The slick hair phenotype has been suggested as an alternative for dairy production in tropical and subtropical countries. The present study evaluated the relationship between hair coat type (phenotypically determined), vaginal temperature (VT) and thermal humidity index (THI) in lactating Puerto Rican Holstein cows. In trial 1, 4 wild type-haired (WT;  $169.8 \pm 3.8$  d in milk (DIM) and  $3.8 \pm 2.2$  lactations) and 5 slick-haired (SLICK;  $146.6 \pm 63$  DIM and  $3.0 \pm 2.1$  lactations) cows were used. In trial 2, 5 WT ( $173.2 \pm 42.3$  DIM and  $3.0 \pm 2.1$  lactations) and 5 SLICK

( $163.8 \pm 43.0$  DIM and  $2.8 \pm 1.7$  lactations) cows were evaluated. Data loggers recorded VT (Tidbit v2), air temperature, and relative humidity (HOBO Pro v2) every 5 min for 8 consecutive days and the THI was determined. The THI presented daily ranges from 63.72 to 80.63 and 61.79–81.47 on trial 1 and trial 2, respectively. Data were analyzed by the GLIMMIX and CORR procedures of SAS. In trial 1 there were no differences in VT between SLICK and WT cows ( $38.71 \pm 0.08$  and  $38.71 \pm 0.09^\circ\text{C}$ , respectively;  $P = 0.96$ ). Daily VT in SLICK and WT cows began to increase ( $P < 0.001$ ) at similar THI values (68.49 and 65.02, respectively;  $P = 0.82$ ). In this first trial, correlation coefficients of 0.27 ( $P < 0.0001$ ) and 0.36 ( $P < 0.0001$ ) were found between the VT and THI in SLICK and WT cows, respectively. In trial 2, hair coat type and time of the day interacted to affect VT ( $P < 0.001$ ). During the 1900–2100 h WT cows presented VT values  $0.25^\circ\text{C}$  greater than SLICK cows ( $P = 0.02$ ). From 2200 to 1800 h WT and SLICK cows presented similar VT values ( $38.68 \pm 0.10$  and  $38.60 \pm 0.07^\circ\text{C}$ , respectively;  $P = 0.39$ ). Also in trial 2, daily VT began to increase ( $P < 0.001$ ) in both hair type groups when THI reached 70.08. In this second trial, correlation coefficients of 0.32 ( $P < 0.001$ ) and 0.42 ( $P < 0.001$ ) were found between the VT and THI in SLICK and WT cows, respectively. In general, environmental conditions had a greater association with the vaginal temperature in wild type cows than in their slick-haired counterparts, and in trial 2 than in trial 1. However, vaginal temperature only differed between hair type groups during a limited period.

**Key Words:** slick-haired Holstein cow, thermoregulation, thermal humidity index

**T306 Relationship between daily milk production and core-body temperature of lactating Holstein cows.** E. O. S. Batista<sup>\*1,2</sup>, C. Collar<sup>1</sup>, P. D. Carvalho<sup>3</sup>, N. Silva-Del-Rio<sup>1</sup>, P. S. Baruselli<sup>2</sup>, and A.H. Souza<sup>1</sup>, <sup>1</sup>University of California, Tulare, CA, <sup>2</sup>University of Sao Paulo, Sao Paulo, SP, Brazil, <sup>3</sup>University of Wisconsin, Madison, WI.

Our objective was to study the daily variation in the core-body temperature (CBT) of lactating dairy cows under heat stress. Holstein cows (8 multiparous and 5 primiparous) producing  $37.1 \pm 2.9$  kg/d and at  $234 \pm 15$  DIM were used in the study. Cows were milked twice daily and located in a commercial dry-lot facility in California. The CBT of all cows was synchronously measured at every 10 min throughout 7 d from August 7 to August 13, 2014, using a temperature data logger (iButton, Maxim Integrated) attached to an intravaginal CIDR device. Body condition scores were recorded at device insertion. Environmental temperature was collected at 10 min intervals throughout the experimental period (Average =  $29.0^\circ\text{C}$ ; Max =  $38.9^\circ\text{C}$ ; Min =  $18.3^\circ\text{C}$ ). Individual milk production was recorded daily and extracted from the herd's management software (Dairy Comp-305). Two cows lost their device and records were not used. Data were analyzed by repeated measures using the GLIMMIX procedure of SAS. In some analysis, cows were categorized into 2 milk production categories (high and low) based on milk production average. Parity, days in milk, and body condition score were not associated with CBT ( $P > 0.10$ ). In contrast, time of the day and milk production influenced CBT ( $P < 0.01$ ). Although large individual CBT variations were observed, further analysis comparing cows with high vs. low milk production indicated that maximal CBT was reached later in the afternoon and was greater for high compared

with low producing cows ( $P = 0.04$ ). More importantly, high producing cows remained for a longer period (45.1% of their daily time budget,  $P < 0.01$ ) with their CBT above the 39.9°C threshold compared with the low producing cows (27.1% of their daily time budget). In conclusion, cows with greater daily milk production seem to be more susceptible to heat stress. Thus, cooling systems should be designed to match milk production requirements.

**Key Words:** milk production, heat stress, body temperature

**T307 Effect of castration and multi alleviation treatment on growth and physiological responses in Korean cattle.** Seung Ju Park, Min Yu Piao, Hyun Jin Kim, Hyeok Joong Kang, and Myunggi Baik\*, *Seoul National University, Gwanak-gu, Seoul, Republic of Korea.*

This study was performed to determine the effects of castration and multi alleviation treatment (MAT) on growth and physiology in Korean cattle male calves. Forty Korean cattle calves (BW = 197.0 kg; 188 d of age) were allocated to 4 treatments ( $n = 10/\text{group}$ ): castration with MAT (12 ml of 2% lidocaine hydrochloride injection for local anesthesia in the scrotum and 2 mg/kg body weight of 50 mg/ml flunixin meglumine injection for antiinflammation in the muscle of the buttock immediately before castration), castration without MAT, no castration with MAT, and no castration without MAT. Surgical castration was performed using a Newberry knife and Henderson castrating tool. The 0.9% NaCl solution was used for non-drug groups instead of MAT. Blood was collected immediately before castration and drug injection (0 h) and 0.5 h, 6 h, 1 d, 3 d, 7 d, and 14 d after castration, body temperature was measured at same time points. Food intake was recorded daily, and body weight was measured at 0 d before castration and 14 d. Average daily gain was analyzed using 2-way ANOVA, using mixed model procedure of SAS. Other variables were analyzed in a repeated measures model using PROC GLIMMIX of SAS. Castration tended to decrease ( $P = 0.07$ ) average daily gain (castration group 0.58 kg/d vs. non-castration group 0.79 kg/d), but MAT did not affect weight gain. Castration increased ( $P < 0.001$ ) body temperature only at 1 d after castration, but not at other time points. Castration increased ( $P < 0.001$ ) plasma cortisol concentrations (castration group 102 ng/mL vs. non-castration group 49 ng/mL) only at 0.5 h after castration, but not at other time points. MAT did not affect cortisol concentrations. Castration increased ( $P < 0.05$ ) plasma nonesterified fatty acid (NEFA) concentrations at 0.5 h, 1 d, and 14 d after castration, but MAT did not affect NEFA concentrations. In conclusion, castration of bulls increased temporally circulating cortisol and NEFA concentrations and tended to decrease forage intake and retard animal growth, but neither increased cortisol concentration nor retarded growth caused by castration were alleviated by MAT.

**Key Words:** alleviation treatment, castration, cortisol

**T308 The effect of exercise in pregnant Holstein heifers on fitness and heat tolerance.** Jessica Winkler\* and Timothy G. Rozell, *Kansas State University, Manhattan, KS.*

Heat stress negatively affects milk production and feed intake of dairy cows resulting in efforts to find ways of reducing heat stress. A possible avenue could be implementing an exercise regimen to improve thermoregulatory capacity. Our objective was to determine if exercising late gestation heifers in a warm climate could physiologically improve thermoregulation. Pregnant Holstein heifers ( $n = 25$ ) were randomly assigned to 3 treatment groups: exercise (EX;  $n = 8$ ), exercise control (EC;  $n = 8$ ; walked with exercise heifers to exerciser but held in a hold-

ing pen), and sedentary control (SC;  $n = 9$ ). Exercise regimens were implemented 4–5 d per wk in the morning for approximately 30–45 min for 8 wk using a motorized panel walker to control duration and speed. Data were collected on fitness test d 0, 28, and 56 of the experiment in which heifers were pushed to reach their maximum speed and time before exhaustion. Pre- and post-exercise blood samples, intra-vaginal temperature, duration, final speed, and heart rate were collected. During wk 2 and wk 6 of the experiment, intra-vaginal temperature devices recorded temperature for 2–4 d and all data were analyzed using PROC GLIMMIX. Duration (min) and final speed (kph) were significantly greater ( $P = 0.04$ ,  $P = 0.009$ ) at d 28 for the EX compared with the SC (duration = 19.31 vs. 13.81; final speed = 6.65 vs. 5.20), implying fitness was achieved. During wk 6, EX heifers spent a smaller percentage of time in Zone 3 (temperature > 39.17°C) than EC during the hottest h of a d (7% vs. 76%;  $P = 0.045$ ), and both EX and EC heifers also spent less time in Zone 3 compared with the SC during the cooler part of a d ( $4.16e^{-17}\%$  vs. 20%;  $P = 0.02$ ). During the first wk of lactation, milk protein tended ( $P = 0.06$ ) to be greater in EX than in SC heifers (3.88 vs. 3.57). There was no significant difference between pre- and post-serum samples ( $\text{Na}^+$ , base excess, ionized Ca, glucose, lactate,  $\text{pCO}_2$ ,  $\text{pO}_2$ ) and heart rate collected on fitness days. Based on results from our pilot study, it appears that heifers undergoing an exercise regimen may be able to improve their ability to regulate a homeostatic body temperature.

**Key Words:** heat acclimation, cattle exercise

**T309 Comparative efficacy of dexamethasone or corticotropin releasing hormone and vasopressin administration as a model to induce chronic physiological stress in beef cattle.** Nathan D. May\*<sup>1</sup>, Jeff A. Carroll<sup>3</sup>, Nicole C. Burdick Sanchez<sup>3</sup>, Shelby L. Roberts<sup>1</sup>, Heather D. Hughes<sup>1</sup>, Paul R. Broadway<sup>3</sup>, Kate P. Sharon<sup>2,3</sup>, Michael A. Ballou<sup>2</sup>, and John T. Richeson<sup>1</sup>, <sup>1</sup>*Department of Agricultural Sciences, West Texas A&M University, Canyon, TX*, <sup>2</sup>*Department of Food and Animal Sciences, Texas Tech University, Lubbock, TX*, <sup>3</sup>*USDA-ARS, Livestock Issues Research Unit, Lubbock, TX.*

The objective of this study was to delineate a model for chronic stress by evaluating physiological and hematological alterations in cattle administered: 1) 0.5 mg/kg BW dexamethasone (DEX) once daily at 10am for 3 d consecutively, or 2) 0.3 µg/kg BW corticotropin releasing hormone (CRH) and 1 µg/kg BW vasopressin (VP) twice daily at 10am and 10pm for 3 d consecutively. Twelve ( $n = 6$ ) beef steers (BW = 389 ± 11 kg) were stratified by BW, assigned randomly to treatment, and fitted with indwelling rectal temperature (RT) devices and jugular catheters, before relocation into individual stanchions in an environmentally-controlled facility. Blood samples were collected at 0.5-h intervals from -2 to 6 h relative to each 10am challenge. Serum was analyzed for cortisol concentration and whole blood samples were evaluated to profile hematological responses via automated hemocytometer. All data were analyzed using PROC MIXED with repeated measures and steer was the subject. A  $\text{trt} \times \text{time}$  interaction ( $P < 0.001$ ) was observed for RT, which increased transiently in both treatments following the 10am challenge time each d. However, the magnitude of the febrile response was greater ( $P < 0.001$ ) for DEX. Likewise, serum cortisol increased transiently in response to both challenges, yet the cortisol increase was more pronounced for CRH-VP ( $\text{trt} \times \text{time}$ ;  $P < 0.001$ ). On d 1, cortisol was 24.8 and 30.4 ng/mL for DEX and CRH-VP, respectively ( $\text{trt} \times \text{d}$ ;  $P < 0.001$ ). Total peripheral blood leukocytes (PBL) were increased in both treatments; however, the PBL concentration was greater for DEX vs. CRH-VP from h 5 to 54 ( $\text{trt} \times \text{d}$ ;  $P < 0.001$ ). The total PBL was associated with a corresponding increase in circulating neutrophils observed for DEX, but not CRH-VP on d 2 (12.2 vs.  $7.2 \times 10^3/\mu\text{L}$ ) and 3 (11.7



vs.  $6.6 \times 10^3/\mu\text{L}$ ;  $\text{trt} \times \text{d}$ ;  $P < 0.001$ ). In contrast, eosinophils decreased ( $P < 0.001$ ) sharply for DEX on d 1 and remained undetectable through d 3. These data suggest that exogenous administration of both DEX and CRH-VP alter RT and cortisol variables indicative of stress, while DEX may more strongly inhibit neutrophil translocation from peripheral blood, a key component of stress-induced immunosuppression.

**Key Words:** cattle, immunosuppression, stress

**T310 Intraperitoneal administration of lipopolysaccharide induces differential expression of mRNA encoding inflammatory mediators in the oviducts of mice.** Katheryn L. Cerny\* and Phillip J. Bridges, *University of Kentucky, Lexington, KY.*

Infection with gram-negative bacteria is a major cause of aberrant inflammation in the oviduct; consequences can include tubal infertility and/or ectopic pregnancy. Understanding inflammatory responses due to bacterial infection is necessary for the development of novel treatment options that specifically target inflammatory responses. Our objective was to test the hypothesis that intraperitoneal (IP) administration of *E. coli*-derived lipopolysaccharide (LPS) induces the expression of inflammatory mRNAs in the mouse oviduct. On the day of estrus, 6–8 week old CD1 mice ( $n = 4/\text{treatment}$ ) were treated IP with 0 (control), 2  $\mu\text{g}$  (low dose) or 10  $\mu\text{g}$  (high dose) of LPS from *E. coli* serotype 055:B5 in 100 mL of PBS. Mice were killed 24 h later and the oviducts collected for determination of inflammatory gene expression by a targeted nanostring approach using the nCounter GX Mouse Inflammation Kit (Nanostring Technologies, Seattle, WA). Real-time PCR was used to validate selected mRNAs. The effect of LPS was evaluated by one-way ANOVA and treatment means of differentially expressed mRNA ( $P < 0.05$ ) were separated using a post-hoc LSD test. In total, 56/179 targeted genes were affected by treatment ( $P < 0.05$ ). Pairwise comparison revealed 8 mRNA differentially expressed in control vs. low dose, 50 mRNAs in control vs. high dose and 43 mRNAs in low vs. high dose ( $P < 0.05$ ). These results indicate that systemic treatment with LPS induces inflammation in the oviducts of mice; this study provides evidence of a new model to investigate the regulation of oviductal inflammation in the future.

**Key Words:** oviduct, inflammation, immune response

**T311 Effects of lipopolysaccharide (LPS)-induced inflammatory response on early embryo survival in ewes.** M. R. Graham\*<sup>1</sup>, E. C. Bowdridge<sup>1</sup>, S. A. Bowdridge<sup>1</sup>, I. Holásková<sup>1</sup>, T. H. Elsasser<sup>2</sup>, and R. A. Dailey<sup>1</sup>, <sup>1</sup>*West Virginia University, Morgantown, WV*, <sup>2</sup>*United States Department of Agriculture-Agricultural Research Service (USDA-ARS), Beltsville Agricultural Research Center (BARC), Beltsville, MD.*

The effects of proinflammatory pathway triggers on pregnancy success was assessed in early pregnant ewes. Effects of endogenous cytokine activation (via LPS), direct exogenous tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) and TNF- $\alpha$ +Dexamethasone (TNF- $\alpha$ +Dex) administration were compared in regard to pregnancy rate. Dorset  $\times$  Texel ewes ( $n = 38$ ) were synchronized for estrus and bred to fertile rams (d 0). On d 6, ewes were assigned to receive via the jugular 2.5 mL containing either PBS ( $n = 9$ ), 2.5  $\mu\text{g}/\text{kg}$  LPS ( $n = 9$ ), 1  $\mu\text{g}/\text{kg}$  TNF- $\alpha$  ( $n = 10$ ) at 0 and 30min, or 2.5  $\mu\text{g}/\text{kg}$  of LPS after 3.5 mL containing 0.14 mg/kg BW im Dex ( $n = 10$ ) at -12h and 0h. Behavioral changes and rectal temperatures (BT) were recorded before challenge and hourly for 12 h. Jugular blood was collected before challenge, every 30 min for 3 h, hourly until 12 h, at 24, 36, and 48 h, and on d 10 and d 26 for progesterone. At d 26, pregnancy

was examined using ultrasonography. The proportion of ewes pregnant was analyzed by Chi-squared and Fisher's exact test with preplanned contrasts. Other data were analyzed by repeated measures ANOVA. Treatment with LPS resulted in peak BT at 4 h (40°C; control 38°C;  $P < 0.05$ ), increased lethargy, mucosal response, white blood cell (WBC) count ( $9.8 \times 10^6$ ; control  $9.1 \times 10^6$ ;  $P < 0.05$ ), serum TNF- $\alpha$  (0.9 ng/mL; control 0.2 ng/mL;  $P < 0.05$ ). Treatment with LPS+Dex resulted in peak BT at 4 h (39°C), but was blunted by Dex. Additionally, Dex blocked mucosal and lethargic responses and attenuated LPS-induced increases in TNF- $\alpha$  and did not improve pregnancy rate compared with LPS alone ( $P > 0.05$ ). TNF- $\alpha$  treatment increased BT at 1 h (39°C), did not induce lethargy; 5/10 ewes showed a mucosal response; and WBC counts did not change but high concentrations of TNF- $\alpha$  were not sustained. More control and TNF- $\alpha$  ewes (15/19) were pregnant than LPS or LPS+Dex ewes (9/19;  $P = 0.045$ ). In summary, an inflammatory response was elicited by LPS and resulted in reduced proportion of ewes pregnant regardless of Dex administration. Treatment with TNF- $\alpha$  did not affect proportion of ewes pregnant, perhaps due to low serum TNF- $\alpha$  levels or different pathway components stimulated by LPS. These data show that a sustained inflammatory response to LPS decreased pregnancy rate and may involve a complex cascade of inflammatory mediators elicited by LPS.

**Key Words:** LPS, embryonic loss, sheep

**T312 Intramammary lipopolysaccharide induces increased luteal mRNA abundance of tumor necrosis factor alpha and toll-like receptor 2 but not luteolysis in dairy cows.** Johannes Lüttgenau<sup>1</sup>, Olga Wellnitz<sup>1,2</sup>, David Kradolfer<sup>3</sup>, Christina Zbinden<sup>2</sup>, Susanne E. Ulbrich<sup>3</sup>, Rupert M. Bruckmaier\*<sup>2</sup>, and Heiner Bollwein<sup>1</sup>, <sup>1</sup>*Clinic of Reproductive Medicine, Vetsuisse Faculty, University of Zurich, Zurich, Switzerland*, <sup>2</sup>*Veterinary Physiology, Vetsuisse Faculty, University of Bern, Bern, Switzerland*, <sup>3</sup>*ETH Zurich, Animal Physiology, Institute of Agricultural Sciences, Zurich, Switzerland.*

Recently, we observed that the application of *Escherichia coli* lipopolysaccharide (LPS) into the mammary gland induce severe clinical symptoms (pyrexia, increased cardiac and respiratory frequencies), an intense stress, and systemic immune response (increased plasma cortisol and haptoglobin concentrations). However, no obvious suppression of luteal size, blood flow, and progesterone secretion in lactating dairy cows were detectable. To investigate possible effects of intramammary LPS challenge on gene expression in luteal tissue, biopsies of the corpus luteum (CL) were analyzed. On d 9 of the estrous cycle (d 1 = ovulation), 8 lactating dairy cows received once 200  $\mu\text{g}$  LPS (dissolved in 10 mL NaCl) and 6 cows received once 10 mL saline (control) into one quarter of the mammary gland. Luteal tissue was collected for biopsy 24 h before and 6 h after treatment, using an ultrasound-guided semi-automatic high-speed biopsy needle. RT-qPCR was applied to assess the mRNA expression of toll-like receptors (*TLR2*, *TLR4*), tumor necrosis factor  $\alpha$  (*TNF $\alpha$* ), steroidogenic factors (*STAR*, *3 $\beta$ HSD*), and factors related to apoptosis (*CASP3*) and prostaglandin synthesis (*PGES*, *PGFS*, *FP*). Data were normally distributed (Shapiro-Wilk-test) and Student's *t*-test was used for dependent comparisons of repeated measures. LPS challenge increased ( $P \leq 0.05$ ) mRNA abundance of *TNF $\alpha$* , *TLR2* and *CASP3*, whereas *TLR4*, *STAR*, *3 $\beta$ HSD*, and prostaglandin-related factors did not change significantly. In control cows, only mRNA expression of *CASP3* was increased ( $P \leq 0.05$ ) at 6 h after compared with 24 h before treatment. Our results indicate that LPS-induced mammary immune

response increases luteal TNF $\alpha$  and TLR2 synthesis but do not induce luteolysis in the bovine CL.

**Key Words:** lipopolysaccharide, mammary gland, corpus luteum

**T313 Different weaning age changes piglets blood parameters related to stress prior anxiety test.** Patricia M. Ramos<sup>\*1,2</sup>, Maicon Sbardella<sup>1,2</sup>, Marcelo A. S. Coutinho<sup>1,2</sup>, Valdomiro S. Miyada<sup>1,2</sup>, and Eduardo F. Delgado<sup>1,2</sup>, <sup>1</sup>"Luiz de Queiroz" College of Agriculture, Piracicaba, SP, Brazil, <sup>2</sup>University of São Paulo, São Paulo, Brazil

This study aimed to identify behavior and metabolites changes in piglets after different weaning when submitted to anxiety test. Thirty-two piglets were separated in 2 homogeneous groups, regarding sex and weight, within 2 pairs of sows based on same parturition date. Half of each sow litter was submitted to different weaning ages (days): 23 (early weaning - EW) and 30 (normal weaning - NW). One week after the NW date, piglets at the same age were submitted to an anxiety test (ATest) in elevate plus maze. Piglets were weighed and blood sampled on the day before and at weaning and also before and after the maze. Blood glucose (mg/dL), lactate (mg/dL) and protein (g/dL) concentrations were measured. ATest was recorded for 3 min and analyzed. GLM was run and means compared by Tukey's test at 0.05. Average daily gain during the week after early weaning were greater ( $P < 0.01$ ) for NW animals, that were still suckling, compared with EW animals. This scenario was inverted in the week after NW date. The blood glucose before weaning was greater ( $P < 0.01$ ; SEM  $\pm 2.26$ ) for the EW (127.62) than for NW (114.28) piglets. Those differences were still present ( $P < 0.01$ ;  $\pm 3.11$ ) in the blood before ATest for EW and NW at 115.92 and 101.54, respectively. Differences were not observed neither after weaning nor after ATest. Likewise, lactate was greater ( $P < 0.05$ ;  $\pm 4.48$ ) in EW vs NW animals before weaning and ATest, with 53.55 vs 38.48 and 66.21 vs 35.09 (SEM  $\pm 6.32$ ), respectively. Inverted after weaning, where NW (52.85) animals presented greater ( $P < 0.05$ ;  $\pm 4.43$ ) lactate compared with EW (39.87). Lactate did not change between groups after ATest. Total protein was different ( $P < 0.05$ ;  $\pm 0.12$ ) between EW and NW animals only after weaning, with 5.48 and 5.06, respectively. There was no difference between the EW and NW on the variables recorded in ATest. After anxiety test blood glucose was negatively correlated ( $-0.35$ ;  $P < 0.01$ ) to the entry in open arms. Early weaning changes blood parameters at weaning and when animals are submitted to novel environmental challenges. Blood glucose concentration is negatively associated with more active and less anxious animals in maze test.

**Key Words:** early weaning, metabolites, maze

**T314 Conjugated linoleic acid (CLA) isomers strongly improves the redox status of bovine mammary epithelial cells (BME-UV1).** Loredana Basiricò<sup>1</sup>, Arnouf Troscher<sup>\*2</sup>, Daniele Dipasquale<sup>1</sup>, Patrizia Morera<sup>1</sup>, Andrea Serra<sup>3</sup>, Marcello Mele<sup>3</sup>, and Umberto Bernabucci<sup>1</sup>, <sup>1</sup>Dipartimento di scienze e tecnologie per l'Agricoltura, le Foreste, la Natura e l'Energia, Università degli Studi della Tuscia, Viterbo, Italy <sup>2</sup>BASF-SE, Ludwigshafen, Germany, <sup>3</sup>Dipartimento di Scienze Agrarie, Alimentari e Agro-ambientali, Università di Pisa, Pisa, Italy.

To date, there is no information available concerning the relationship between administration of CLA and changes in oxidative status of bovine mammary gland. The aim of this study was to assess in vitro the role of CLA in cell protection against the oxidative stress of bovine mammary cells. The uptake rates of CLA in BME-UV1 cells at 3 h and 48 h were

tested using DAD HPLC analysis. BME-UV1 cells were treated with complete medium containing 50  $\mu$ M of *cis-9,trans-11* CLA (c9,t11), *trans-10,cis-12* CLA (t10,c12) and CLA Mixture (50% c9,t11 and 50% t10,c12). After 48 h from addition of CLA, cell samples were collected for determining oxidative markers such as nicotinamide adenine dinucleotide phosphate (NADPH), glutathione (GSH/GSSG), thiobarbituric acid reactive substances (TBARS), cytoplasmic superoxide dismutase (SOD), glutathione peroxidase (GPx1), glutathione S-transferase (GST) and glutathione reductase (GR) using commercial kits. The mRNAs quantification of antioxidant enzymes (SOD, GPx1, GST and GR) was performed by rt-PCR. The potential protection of CLA against H<sub>2</sub>O<sub>2</sub>-induced oxidative stress was assessed by XTT assay after 48h of pre-treatment with CLA and 3h with H<sub>2</sub>O<sub>2</sub>. The data were analyzed by ANOVA and differences were declared significant at  $P < 0.05$ . The results showed that the uptake rates of CLA in cells increased from 3 (26.54%) to 48h (73.95%). CLA increased ( $P < 0.01$ ) the concentration of reduced GSH (CLA 17.25 vs control 11.73  $\mu$ M) and NADPH (CLA 0.28 vs control 0.20  $\mu$ M), mostly in cells treated with t10,c12 (0.50  $\mu$ M). The enzymatic antioxidant activity of GR (CLA 7.00 vs control 11.82 U/L) was decreased ( $P < 0.01$ ) whereas GSPx1 ( $P < 0.01$ ), GST ( $P < 0.05$ ) and SOD ( $P < 0.01$ ) activities (CLA 19.85, 0.70, 0.05 vs control 7.07 U/L, 0.61 U/mL, 0.02 U/mL, respectively) were increased in cells treated with CLA. Lower levels of TBARS (CLA 1.50 vs control 1.96  $\mu$ M/MDA equivalent) were observed in cells treated with CLA. CLA had not any substantial effect on gene expression of antioxidant enzymes. All CLA isomers were able to enhance ( $P < 0.01$ ) cell resistance against oxidative stress (CLA 2.85 vs control 2.65 O.D.). Findings of the present study corroborate the antioxidant role of CLA, in particular of t10,c12 isomer, by improving of redox status in cells, and might be of help during physiological oxidative stress situations such as the periparturient period in dairy cow. This work was funded by BASF-SE.

**Key Words:** conjugated linoleic acid, bovine mammary cell, oxidative stress

**T315 Antioxidant potential in the gut of juvenile fish fed with lyophilized bovine colostrum.** Debora B. Moretti<sup>\*</sup>, Wiolene M. Nordi, Thaline M. P. Cruz, Jose Eurico P. Cyrino, and Raul Machado-Neto, University of São Paulo, Piracicaba, São Paulo, Brazil.

Bovine colostrum is a rich source of immunological factors and contains a large concentration of antioxidant components that can affect gut physiology. Thus, the antioxidant potential was evaluated in the gut of juvenile pacu (*Piaractus mesopotamicus*, Holmberg, 1887), an omnivorous fish, and dourado (*Salminus brasiliensis*, Cuvier, 1816), a carnivorous fish, fed with diets containing 0, 10 and 20% of lyophilized bovine colostrum (LBC) for either 30 or 60 d. The antioxidant capacity in the intestinal tissue was assessed by the determination of superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and by the oxygen radical absorbance capacity (ORAC value expressed as equivalent trolox in  $\mu$ M/mg of protein). Experimental design was completely randomized with treatments arranged as a 3  $\times$  2 factorial, considering the 3 diets and the 2 periods as main effects. If F value was significant, Tukey test was used for comparisons between means ( $P < 0.05$ ). In pacu, interaction between diet and period was observed to SOD ( $P < 0.05$ ), juveniles fed 0 and 10% LBC did not change enzyme activity at 30 (423  $\pm$  27 and 692  $\pm$  42 U/mg of protein, respectively) and 60 d (504  $\pm$  14 and 890  $\pm$  57 U/mg of protein, respectively), whereas juvenile fed 20% LBC showed higher enzyme activity at 60 d (1122  $\pm$  71 U/mg of protein) compared with 30 d (595  $\pm$  61 U/mg of protein). GPx was not influenced by diet and period ( $P > 0.05$ ) and CAT was

affected only by period ( $P < 0.05$ ), showing higher activity at 60 d ( $891 \pm 86$  U/mg of protein) compared with 30 d ( $437 \pm 99$  U/mg of protein). The ORAC was affected by diet and period ( $P < 0.05$ ); the juveniles fed 10% LBC ( $505 \pm 100$   $\mu$ M/mg) had greater oxygen absorbance capacity than the juveniles fed 20% LBC ( $348 \pm 104$   $\mu$ M/mg), and the value was greater at 30 ( $628 \pm 61$   $\mu$ M/mg) than at 60 d ( $234 \pm 31$   $\mu$ M/mg). In dourado, GPx was affected by period ( $P < 0.05$ ), showing greater activity at 30 ( $224 \pm 30$  U/mg of protein) than at 60 d ( $94 \pm 19$  U/mg

of protein). SOD, CAT and ORAC value were not affected by diet and period ( $P > 0.05$ ). Inclusion of lyophilized bovine colostrum improved SOD activity in the gut of juvenile pacu, an omnivorous fish, indicating a possible protective action of this lacteal secretion.

**Key Words:** colostrum, antioxidant enzyme, oxygen radical absorbance capacity (ORAC)



# Physiology and Endocrinology: Reproductive tissues, gametes, and embryo development

**T316 Stem cell factor (SCF) activates AKT-p70RSK-S6 signaling in porcine trophoblast cells.** Yurina Choi\*, Wooyoung Jeong, Heejo Bang, Yujin Sung, and Jinyoung Kim, *Dankook University, Cheonan, Korea.*

During early pregnancy, a well-coordinated network between the conceptus and maternal uterus is especially crucial in pigs which involve a prolonged pre-attachment phase. This network is regulated by an astonishing amount of molecules such as growth factors. SCF (Stem Cell Factor) is a multipotent growth factor that elicits diverse biological actions on various types of tissues. In pig, SCF and their receptors are expressed in the uterine endometrium and conceptus during early pregnancy, but little is known about the biological role of SCF in the conceptus. Therefore, the aim of present study was to access SCF-induced intracellular signaling and cellular activities in porcine trophoblast (pTr) cells. The effects of SCF were studied using pTr cells isolated from Day 12 pig conceptuses. Abundance of phosphorylated (p)-proteins relative to total proteins were determined by Western blot analyses of whole cell extracts 3 times and subjected to least squares using SAS software. In vitro cultured pTr cells were incubated with different concentrations of recombinant SCF (0–50 ng/mL). SCF dose dependently increased AKT phosphorylation, reaching 3.3-fold at 20 ng/mL. Within 30 min after 20 ng/mL of SCF treatment, levels of p-AKT, p-p70RSK and p-S6 proteins increased by 3.0-, 3.1- and 4.0-fold, respectively, and then returned to basal levels by 120 min. To ensure stimulatory effect of SCF on AKT signaling, cells were pre-incubated with AKT blocker (LY294002) 1 h before SCF treatment. Twenty  $\mu$ M of LY294002 decreased SCF-induced p-AKT, p-p70RSK and p-S6 proteins. Also immunofluorescence analyses found that p-S6 were localized abundantly within the cytoplasm of SCF-treated cells, but p-S6 was present at basal levels in the presence of LY294002. Furthermore, SCF increased pTr cell migration by 200%, but LY294002 significantly reduced this stimulatory effect to basal level. In conclusion, results of the present study suggest that SCF activates migration of trophoblast through AKT signaling and supports the hypothesis that SCF is a critical regulatory factor of conceptus development during early pregnancy in pigs.

**Key Words:** SCF, early pregnancy, trophoblast

**T317 In vitro fertilization (IVF) from low or high antral follicle count pubertal beef heifers using semi-defined culture conditions.** C. C. Chase, Jr.\*<sup>1</sup>, R. A. Cushman<sup>1</sup>, A. K. McNeel<sup>1</sup>, E. C. Wright-Johnson<sup>1</sup>, O. L. Amundson<sup>2</sup>, E. L. Larimore<sup>2</sup>, B. N. Richardson<sup>2</sup>, G. A. Perry<sup>2</sup>, S. C. Tenley<sup>3</sup>, J. R. Wood<sup>3</sup>, A. S. Cupp<sup>3</sup>, J. L. Vallet<sup>1</sup>, D. D. Sypher<sup>1</sup>, and J. L. Miles<sup>1</sup>, <sup>1</sup>USDA, ARS, US Meat Animal Research Center, Clay Center, NE, <sup>2</sup>Dept. of Animal Science, South Dakota State Univ., Brookings, SD, <sup>3</sup>Dept. of Animal Science, University of Nebraska, Lincoln, Lincoln, NE.

Our objective was to compare the in vitro maturation and fertilization of oocytes collected from low and high antral follicle count (AFC) heifers. Trans rectal ultrasonography was performed on 120 heifers to determine AFC and presence of a corpus luteum (i.e., pubertal). Those 10 heifers with the lowest AFC (avg. 14.2) and those 10 heifers with the highest AFC (avg. 29.9) all with evidence of estrous cyclicity (i.e., pubertal) were synchronized with 2 injections of PGF<sub>2 $\alpha$</sub>  and killed over 4 d; on d 5 to 6 of the estrous cycle. Nineteen heifers (n = 9 low and n = 10 high AFC) were at the appropriate stage of the estrous cycle. The

IVF procedures and media were as described (P. J. Hansen's Laboratory, IVP Protocol). Cumulus-oocyte complexes (COCs) from follicles less than 8 mm in diameter were cultured in maturation medium (5% CO<sub>2</sub>; 38.5°C) for 24 h. Matured COCs were fertilized using thawed frozen semen from a crossbred bull that was purified using Percoll separation procedures. Motile spermatozoa were added to COCs in fertilization medium at a final concentration of 1x10<sup>6</sup> spermatozoa per mL. About 24 h later, presumptive zygotes were placed in micro drops of development medium under oil, and cultured (5% CO<sub>2</sub>; 5% O<sub>2</sub>; 38.5°C). On d 3 and 8 after fertilization, cleavage and blastocyst development, respectively, were assessed. Data were analyzed using the MIXED procedure of SAS and the model included the effects of collection d, group, and their interaction. Percentage data were analyzed using the GLIMMIX procedure with a binomial distribution and a logit link. Neither collection d nor the interaction differed ( $P \geq 0.13$ ). High compared with low AFC heifers had greater numbers of COCs ( $P < 0.01$ ; 27.0  $\pm$  3.79 vs. 9.6  $\pm$  3.97 per heifer), oocytes that cleaved ( $P < 0.04$ ; 15.2  $\pm$  2.63 vs. 6.1  $\pm$  2.76 per heifer), and developed to blastocysts ( $P < 0.007$ ; 4.08  $\pm$  0.662 vs. 0.83  $\pm$  0.695 per heifer). There was no difference ( $P > 0.9$ ) in the percentage of COCs that cleaved (low = 52.3  $\pm$  8.37%, high = 53.6  $\pm$  7.98%) or in the percentage of COCs that developed to blastocysts ( $P < 0.13$ ; low = 6.6  $\pm$  2.79% vs. high = 13.7  $\pm$  2.66). These results agree with our previous findings, high AFC heifers had greater numbers of COCs, oocytes that cleaved, and blastocysts compared with low AFC heifers; however, AFC does not appear to affect oocyte development and maturation through the blastocyst stage.

**Key Words:** heifer, IVF, antral follicle count

**T318 Effect of L-carnitine in serum-supplemented IVM medium on mitochondrial membrane potential, ROS levels and subsequent embryo development of bovine oocytes.** Beatriz C. S. Leao\*, Nathália A. S. Rocha Frigoni, Priscila C. Dall'Acqua, and Gisele Z. Mingoti, *Laboratory of Physiology of Reproduction, School of Veterinary Medicine, Sao Paulo State University, Araçatuba, Sao Paulo, Brazil.*

This study aimed to evaluate the effects of supplementation with different concentrations of L-carnitine (L-car) during in vitro maturation (IVM) of bovine oocytes on their mitochondrial membrane potential (MMP), reactive oxygen species (ROS) levels, and subsequent embryonic development. Cumulus-oocyte complexes (COC) were matured for 22 h at 38.5°C, 5% CO<sub>2</sub> in air, on IVM medium (TCM-199 with bicarbonate, hormones and 10% FCS) supplemented with 0 (Control), 1, 5 or 10 mM of L-car. Matured and immatured oocytes (0 h) were stained with 500 nM of MitoTracker Red (Molecular Probes, Invitrogen), or with 5 mM of H<sub>2</sub>DCFDA (Molecular Probes, Invitrogen). Stained oocytes (MMP: n = 191; and ROS: n = 250) were evaluated under an epifluorescence inverted microscope (excitation 579/495nm and emission 599/404nm, respectively) and the images analyzed by Q-Capture Pro image software to measure the arbitrary fluorescence units. The fluorescence intensity values were subtracted from mean values of "background" in the images. The 0h group was chosen as the calibrator, and each treatment value was divided by the mean of the 0h. In a second trial, COC (n = 1875) were IVM as above and then fertilized. The presumptive zygotes were cultured in SOF medium at 38.5°C, 5% CO<sub>2</sub> in air, for 7 d (Day 0 = IVF), when the blastocysts (BI) rates were

evaluated. The averages were compared by ANOVA followed by the Tukey's test and data are presented as mean  $\pm$  SEM. It was found a reduction ( $P < 0.05$ ) on MMP after IVM with L-car 10mM ( $0.4 \pm 0.0^c$ ) in comparison to 1 mM ( $1.1 \pm 0.1^{ab}$ ) and 5 mM ( $1.0 \pm 0.0^{ab}$ ), as well as to 0h ( $1.0 \pm 0.1^a$ ); however, no treatment differed from the Control ( $0.8 \pm 0.0^{abc}$ ). In respect to ROS levels, we found an increase ( $P < 0.05$ ) in oocytes matured with 10 mM ( $2.3 \pm 0.1^c$ ), in comparison to Control ( $1.8 \pm 0.1^b$ ), 1 mM ( $1.9 \pm 0.1^b$ ), 5 mM ( $2.0 \pm 0.1^b$ ) and 0 h ( $1.0 \pm 0.0^a$ ). BI rates were similar ( $P > 0.05$ ) in all treatments ( $25.2\% \pm 3.7$  to  $37.1\% \pm 2.7$ ). In conclusion, the reduction on MMP after IVM of bovine oocytes with L-car 10 mM was followed by an increase of ROS level. However, there was no influence on their acquisition of capacity to BI development. Financial support: FAPESP (#2012/10084-4 and #2013/07382-6)

**Key Words:** L-carnitine, mitochondrial membrane potential, ROS level

### T319 Cell proliferation in ovarian follicles in nonpregnant ewes: Effects of plane of nutrition and arginine.

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The aim of this study was to investigate the role of the NO system in ovarian function by determining if Arg supplementation affects cell proliferation in ovarian follicles in nutritionally compromised ewes. Ewes were stratified by weight and randomly assigned into either control (C;  $n = 35$ ), overfed (O; 2xC;  $n = 37$ ), or underfed (U; 0.6xC;  $n = 36$ ) groups 8 weeks before Arg-treatment. Ewes from each nutritional group were randomly assigned to one of 2 i.v. treatments: saline (~10 mL) or Arg (L-Arg-HCl, 155  $\mu$ mol Arg/kg of BW) which was initiated on d 0 of the estrous cycle and administered 3 times per day (0700, 1400, 2100 h) until ovary collection at the late-luteal stage of the first estrous cycle, or early or mid-luteal stages of the second estrous cycle. Ovaries were fixed in formalin solution followed by immunohistochemical localization of Ki67 (a marker of proliferating cells), and image analysis of granulosa (G) and thecal (T) layers. Data were analyzed by ANOVA using SAS. During nutritional treatment, C maintained BW, O gained  $6 \pm 1.2$  kg, and U lost  $14 \pm 1.3$  kg. Ki67 was detected in G and T layers, and other ovarian compartments. Cell proliferation in G and T of healthy follicles was affected by stage of the estrous cycle, but not plane of nutrition or Arg-treatment. Cell proliferation was 1.3–1.5-fold greater ( $P < 0.001$ ) at early and mid than late luteal phase. For G and T of healthy follicles, interactions ( $P < 0.05$ ) between plane of nutrition, Arg-treatment and/or stage were detected, demonstrating stimulatory effects of Arg-treatment on cell proliferation at the early luteal phase in O and U, and inhibitory Arg-effects at the mid-luteal phase in O ewes. Cell proliferation was greater ( $P < 0.001$ ) in healthy antral than atretic follicles in all groups ( $14.4 \pm 0.4$  vs.  $1.9 \pm 0.1\%$ ). These data show that cell proliferation in G and T layers is affected by plane of nutrition, Arg-treatment and/or stage of the estrous cycle that likely affects follicular function. The mechanism of regulation of ovarian cell proliferation by diet and Arg remains to be elucidated. Supported by USDA-AFRI grant 2011–67016–30174, and Hatch Projects ND01748 and ND01754 to ATGB and DAR.

**Key Words:** plane of nutrition, arginine, ovary

### T320 Comparison of mRNA expression of dominant follicle and follicular cyst in lactating dairy cows.

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Follicular cysts are an important disorder affecting 5 to 30% of dairy cows that leads to abnormal estrous cycle behavior and economic losses due to infertility. The aim of this study was to compare the mRNA expression in follicular cells of cows classified either as having a follicular cyst or cycling with dominant follicle (DF). Lactating dairy cows ( $n = 22$ ) were examined weekly by ultrasound and 11 cows (Holstein,  $n = 7$ ; Jersey  $n = 3$ ; and Shorthorn  $n = 1$ ) were classified as having a follicular cystic (CYS, follicle  $> 25$  mm with absence of corpus luteum) whereas 11 cows (Holstein,  $n = 11$ ) were classified as cycling with a dominant follicle (CON, presence of corpus luteum and dominant follicle,  $n = 11$ ). Cows were at 94.4 (30–382) DIM, milk yield  $37.9 \pm 12$  kg/d, and parity  $2.6 \pm 1.3$ . Cows in CON had follicle diameter of  $14.10 \pm 3$  mm whereas cows in CYS had follicle diameter of  $35.73 \pm 10.2$  mm. Follicular fluid from each cow was aspirated and follicular cells were retrieved immediately by centrifugation, frozen in liquid nitrogen and stored at  $-80^\circ\text{C}$  until RNA extraction. The mRNA expression for *LHCGR*, *STAR*, *3 $\beta$ -HSD*, *P450 $\text{sc}$* , *CYP19A1*, *IRS1*, *IGF*, *TLR4*, *TNF*, *IL1- $\beta$* , *IL8* and *IL6* was measured by real-time PCR. Statistical analysis was performed using the MIXED procedure of SAS. *LHCGR*, *3 $\beta$ -HSD*, *CYP19A1*, *IRS1* mRNA expression was higher ( $P < 0.05$ ) in CON ( $1.37 \pm 0.4$ ,  $5.58 \pm 1.2$ ,  $1.33 \pm 0.2$ ,  $1.41 \pm 0.3$ , respectively) than CYS ( $0.69 \pm 0.3$ ,  $3.24 \pm 0.9$ ,  $0.24 \pm 0.01$ ,  $0.74 \pm 0.2$ , respectively) with breed effect ( $P < 0.07$ ). Expression of mRNA for *IGF*, *TLR4*, *TNF* and *IL8* was lower ( $P < 0.05$ ) in CON ( $0.55 \pm 0.07$ ,  $0.61 \pm 0.2$ ,  $0.54 \pm 0.3$ ,  $0.12 \pm 0.01$ , respectively) than in CYS ( $4.62 \pm 0.7$ ,  $3.40 \pm 0.5$ ,  $1.58 \pm 0.3$ ,  $0.52 \pm 0.04$ , respectively), with no breed effect ( $P > 0.20$ ). There was no difference ( $P > 0.05$ ) for *STAR*, *P450 $\text{sc}$* , *IL6* and *IL1- $\beta$*  mRNA expression between CYS and CON. In conclusion, cows in CYS had lower expression of genes related to steroidogenesis and energy metabolism and greater expression of genes related to inflammation than CON. It seems that an inflammatory response may be involved in the follicular cyst etiology.

**Key Words:** follicle, cystic, inflammation.

### T321 Colony-stimulating factor 2 affects development of the bovine preimplantation embryo differently for females than males.

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Colony-stimulating factor 2 (CSF2) regulates early embryonic development by modifying the epigenome, reducing apoptosis, and altering ratio of cells in the trophectoderm (TE) and inner cell mass (ICM) of the blastocyst. Previously, CSF2 reduced trophoblast elongation in female embryos but increased elongation in males. Here it was tested whether sexual dimorphism in response to CSF2 can be observed as early as the blastocyst stage. Embryos were produced in vitro using X- or Y-sorted sexed semen ( $n = 1612$  putative zygotes). On d 5 of culture, droplets were supplemented with 5  $\mu$ L vehicle (control) or 10 ng/mL bovine CSF2. Blastocysts ( $n = 210$ ) were collected at Day 7 and labeled with a nuclear dye (Hoescht 33342; total cells) and a TE cell marker (CDX2). Number of ICM cells was calculated by subtraction. Statistical analysis was performed using the Proc Mixed procedure of SAS; data represent least squares means  $\pm$  SEM. Treatment of female embryos with CSF2 increased the proportion of zygotes ( $P = 0.0213$ ) and cleaved embryos ( $P = 0.0252$ ) to become a blastocyst but there were no effects in males ( $P > 0.10$ ). The percent of zygotes becoming blastocysts on Day 7 was  $14.7 \pm 2.1$  vs  $21.5 \pm 2.1\%$  for control and CSF2 in females and  $16.2 \pm 2.0$  vs  $16.3 \pm 2.0\%$  in males. There was no effect of CSF2 treatment, sex, or the interaction on the total cell number or number of TE ( $P > 0.10$ ). There was a tendency ( $P = 0.0934$ ) for ICM number to be less in females ( $56.2 \pm 3.1$  vs  $61.0 \pm 2.9$ ) and the TE:ICM ratio was greater ( $P$

= 0.0217) for females ( $1.64 \pm 0.91$ ) compared with males ( $1.45 \pm 0.09$ ). Numerically (but not significantly), CSF2 tended to decrease ICM in females ( $53.9 \pm 3.6$  vs  $58.6 \pm 3.7$ ) but not in males ( $60.4 \pm 3.5$  vs  $61.5 \pm 3.4$ ). There was a tendency for a CSF2 by sex interaction ( $P = 0.0955$ ) for TE:ICM ratio. In females CSF2 increased ratio ( $1.73 \pm 0.11$  vs  $1.55 \pm 0.11$ ), but no effect was observed in males ( $1.41 \pm 0.10$  vs  $1.50 \pm 0.10$ ). In conclusion, CSF2 exerts different responses on development of female and male preimplantation embryos. Consequences of actions of CSF2 on ICM and TE cell differentiation require further investigation. Support: NIH HD080855.

**Key Words:** embryo, colony-stimulating factor 2, sex

**T322 Effect of fertility stressors on transcriptome of peripheral blood leukocytes (PBL) in dairy cows at the onset of conceptus implantation.** Eduardo S. Ribeiro<sup>1,2</sup>, Rafael S. Bisinotto<sup>1,2</sup>, Fabio S. Lima<sup>1,2</sup>, Natalia P. Martinez<sup>1,2</sup>, Leandro F. Greco<sup>1,2</sup>, William W. Thatcher<sup>1,2</sup>, and José E. Santos<sup>1,2</sup>, <sup>1</sup>Department of Animal Sciences, University of Florida, Gainesville, FL, <sup>2</sup>D.H. Barron Reproductive and Perinatal Biology Research Program, University of Florida, Gainesville, FL.

Objectives were to investigate changes in transcriptome of PBL occurring at the onset of implantation and how they are affected by fertility stressors. Lactating cows ( $n = 481$ ) had estrous cycle and ovulation synchronized to receive their first insemination (AI) postpartum. On d 19 after AI, PBL were isolated and mRNA extracted. A subsample of PBL mRNA from 36 cows was subjected to transcriptome analysis using the Affymetrix platform. Pregnancy was diagnosed on d 34 after AI. Two fertility stressors were evaluated, progesterone concentration during development of the ovulatory follicle and clinical uterine diseases (UTD). At the onset of synchronization, experimental design was established to have cows ovulating follicles that grow under low (LP) or high (HP) concentrations of progesterone. In addition, cows were evaluated daily on the first 10 d postpartum for diagnosis of UTD. Statistics was performed using Limma on R and FDR adjustment was used. LP during development of the ovulatory follicle reduced pregnancy per AI (P/AI; 34 vs 53%) and altered the transcriptome of PBL. In the HP group, pregnancy resulted in upregulation of classical interferon stimulated genes (e.g., *IFI6*, *ISG15*, *OAS1Y*); whereas in the LP group, pregnancy resulted in downregulation of a large number of inflammatory response genes (e.g., *HP*, *JUN*, *MYD88*). Particularly distinct transcriptome was observed in LP cows that failed to become pregnant, which indicated an inflammatory state. Cows that suffered from UTD also presented reduced P/AI (33 vs 50%) but only subtle differences in transcriptome, although potentially important. In pregnant cows previously diagnosed with UTD, expression of *OAS1X* was downregulated whereas *CD244* was upregulated compared with pregnant cows not diagnosed with UTD. Fertility stressors were associated with altered PBL transcript profiles at the onset of implantation. Differences observed might represent either a primary cause of subfertility or a consequence of impaired developmental potential of their conceptus and its ability to secrete signaling molecules to modulate the maternal immune system.

**Key Words:** cow, fertility, leukocyte

**T323 Circulating concentrations of bovine pregnancy associated glycoproteins and late embryonic mortality in lactating dairy herds.** Ky G. Pohler<sup>1</sup>, Marcos H. Pereira<sup>2</sup>, Francisco R. Lopes<sup>2</sup>, Jose L. M. Vasconcelos<sup>2</sup>, Michael F. Smith<sup>1</sup>, and Jon A. Green<sup>1</sup>, <sup>1</sup>Division of Animal Sciences, University of Missouri, Columbia, MO, <sup>2</sup>FMVZ-UNESP, Botucatu, SP, Brazil.

In cattle, the incidence of late embryonic mortality (EM) has been reported to range from 3.2 to 42.7%. In some cases, the economic consequences of late EM are reported to be greater than that of early EM, because late EM can cause a significant delay in conception date. Although considerable effort has been directed toward understanding the causes of early EM, relatively little is known about the causes or mechanisms associated with late EM. The objectives of this experiment were as follows: (1) to determine the association between circulating concentrations of pregnancy associated glycoproteins (PAGs) and late EM in lactating dairy cattle following fixed-timed artificial insemination (TAI) on d 0 or fixed-timed embryo transfer (TET) on d 7, and (2) to identify a circulating concentration of PAGs on d 30 below which late EM would be likely to occur. Cows were diagnosed pregnant on d 30 of gestation based on presence of a fetal heartbeat and reconfirmed on d 60 of gestation. Late EM occurred when a cow had a viable embryo on d 30 of gestation but not on d 60 following TAI or TET. Only pregnant cows were included in the analysis (TAI-maintained,  $n = 413$ ; TAI-EM  $n = 77$ ; TET-maintained,  $n = 238$ ; TET-EM,  $n = 47$ ) which was subjected to Proc Glimix procedures of SAS. Cows that were pregnant at d 30 of gestation and maintained the pregnancy until d 60 had significantly greater ( $P < 0.05$ ) circulating concentrations of PAGs at d 30 of gestation compared with cows that experienced late EM between d 30 and 60 of gestation in both TAI and TET. A receiver-operating characteristic curve was generated using MedCal Software to determine circulating concentration of PAGs on d 30 that should predict EM with  $\geq 95$  accuracy in both TAI and TET. Based on positive and negative predicative value analysis, a circulating concentration of PAGs below 1.4 ng/mL (TAI) and 1.85 ng/mL (TET) was 95% accurate in predicting EM (between d 30 - d 60) at d 30 of gestation. In summary, PAGs may provide a good marker for predicting EM between d 30 to 60 of gestation and may be able to accurately predict cows that will undergo late EM for the purpose of investigating mechanisms leading to late EM.

**T324 Etiology of early pregnancy losses in Holstein dairy cows based on serum pregnancy-associated glycoprotein and progesterone concentrations.** Maria J. Fuenzalida\*, Paulo D. Carvalho, Milo C. Wiltbank, Pamela L. Ruegg, and Paul M. Fricke, *University of Wisconsin-Madison, Madison, WI.*

Our objective was to describe the mechanism and timing of pregnancy losses (PL) in cows after the first timed artificial insemination (TAI). A total of 136 cows that experienced PL were included in a matched case-control study. Cases were obtained from 3,164 cows from 4 dairy farms enrolled in a prospective cohort study. Cows with pregnancy-specific protein B (PSPB)  $\geq 0.3$  ng/mL on d 25 after TAI and were open based on transrectal ultrasonography 27 to 32 d (PG1) were defined as early PL ( $n = 49$  cows). Cows that were pregnant at PG1 but open at subsequent evaluations were considered later PL ( $n = 87$  cows). Cows that remained pregnant during the study period (from TAI to 90 d after TAI) were defined as Controls ( $n = 266$  cows). Two Control cows were matched to each PL cow based on days in milk and parity. Progesterone (P4) and PSPB were measured weekly from 10 d before TAI until a cow was diagnosed open or remained pregnant and reached 90 d after TAI. Week of PL and cause of PL (embryonic death [ED] or corpus luteum regression [CLR]) was determined from weekly PSPB using a cutoff of the lowest PSPB concentrations of Controls (from 25 to 88 d after TAI) and weekly P4 concentrations. Data were analyzed by *t*-test, chi-squared and linear regression. For early PL, 30.6% (15/49), 16.3% (8/23) and 53.1% (26/49) were due to ED, CLR and undefined causes, respectively. For cows with later PL, 37.9% (33/87), 48.3% (42/87), and 13.8% (12/87) were due to ED, CLR and undefined causes ( $P = 0.04$ ). Mean average days for occurrence of PL based on PSPB concentration



cutoff for ED was 48.6 (ranging from 28 to 88) and for CLR was 45.3 (ranging from 24 to 86) ( $P = 0.29$ ). For early PL, P4 concentrations on d 25 were  $5.8 \pm 3.4$  ng/mL and on d 32 were  $6.6 \pm 2.3$  ng/mL ( $P > 0.05$ ). PSPB concentrations on d 25 were  $0.6 \pm 0.6$  ng/mL and on d 32 were  $1.9 \pm 1.5$  ng/mL ( $P < 0.05$ ). In conclusion, the occurrence of pregnancy loss due to ED occurred with more frequency in early PL near 32 d, whereas later PL were more likely due to CLR. Supported by AFRI Competitive Grant no. 2010–85122–20612.

**Key Words:** pregnancy loss, progesterone, pregnancy-associated glycoprotein

**T325 Supplementation with insulin-like growth factor-1 during in vitro culture protects bovine embryos from deleterious actions of menadione.** Nathália A. S. Rocha-Frigoni\*, Beatriz C. S. Leão, Priscila C. Dall'Acqua, and Gisele Z. Mingoti, *Laboratory of Physiology of Reproduction, School of Veterinary Medicine, University of Sao Paulo State (UNESP), Araçatuba, Sao Paulo, Brazil.*

The objective of this study was to evaluate the protective effect of insulin-like growth factor (IGF-1) under oxidative stress condition induced by menadione (MD) on bovine embryos in vitro cultured (IVC). Cumulus-oocyte complexes were matured in TCM-199 with bicarbonate, hormones and 10% FCS for 22 h at 38.5°C in 5% CO<sub>2</sub> in air. After fertilization, the presumptive zygotes were IVC in SOF medium supplemented with 100 μM IGF-1, at 38.5°C in 5% CO<sub>2</sub> in air, for 7 d. On Day 6 the culture medium was supplemented with 5 μM MD. The cleavage rates and embryonic development were evaluated at Day 3 and Day 7, respectively (IVF = d 0). At d 7 blastocysts were stained to quantify the reactive oxygen species (ROS) levels with 5 μM H<sub>2</sub>DCFDA (Molecular Probes, Invitrogen), the caspase activity (Image iT LIVE Red Caspase-3 and -7 Detection Kit, Molecular Probes) or the apoptotic index using TUNEL (In Situ Cell Death Detection Kit, Fluorescein, Roche Applied Science). Stained embryos were evaluated under an epifluorescence inverted microscope (excitation 495/550–510–550nm and emission 404/595/590 nm, respectively for ROS, caspase and TUNEL). The images were analyzed by Q-Capture Pro image software for determining the fluorescent intensity. The results were compared by ANOVA followed by Student *t*-test ( $P < 0.05$ ) and are presented as mean ± SEM. The cleavage rates did not differ ( $P > 0.05$ ) among groups ( $77.1\% \pm 1.9$  to  $82.7.5\% \pm 2.2$ ). The blastocyst rates were  $38.1\% \pm 2.50^{ab}$  (Control),  $38.9\% \pm 1.97^a$  (IGF),  $21.39\% \pm 2.93^c$  (MD) and  $25.7\% \pm 3.32^{bc}$  (IGF+MD). The intracellular levels of ROS were  $111.1 \pm 1.7^b$  (Control),  $118.3 \pm 2.1^a$  (IGF),  $118.8 \pm 2.1^a$  (MD) and  $112.1 \pm 3.6^{ab}$  (IGF+MD). The caspase activity did not differ ( $P > 0.05$ ) among groups ( $28.2 \pm 2.7$  to  $41.1 \pm 3.2$ ). Although, the rates of apoptosis were  $12.8\% \pm 1.0^b$  (Control),  $9.1\% \pm 0.75^c$  (IGF),  $22.3\% \pm 2.3^a$  (MD) and  $15.6\% \pm 1.6^b$  (IGF+MD). In conclusion, the supplementation with IGF-1 during IVC reversed the detrimental effects of MD on embryonic levels of ROS and apoptosis, as well as improved the embryo development.

**Key Words:** IGF-1, reactive oxygen species, apoptosis

**T326 Menadione induces oxidative stress and reduces embryo development.** Priscila C. Dall'Acqua\*, Nathália A. S. Rocha-Frigoni, Beatriz C. S. Leão, and Gisele Z. Mingoti, *Laboratory of Physiology of Reproduction, School of Veterinary Medicine, University of Sao Paulo State (UNESP), Araçatuba, São Paulo, Brazil.*

Oxidative stress during the in vitro production of embryos culminates with a rise in reactive oxygen species (ROS) levels and apoptosis. Menadione (MD) induces ROS generation and can be used as a tool

to understand the actions of oxidative stress on embryo development. This study evaluated the effects of different MD concentrations during in vitro culture on bovine embryo development, intracellular levels of ROS and apoptosis rate. Cumulus-oocyte complexes were matured in vitro in TCM-199 with bicarbonate, hormones and 10% FCS during 22 h at 38.5°C in 5% CO<sub>2</sub> in air. After fertilization, zygotes were cultured in SOF medium at 38.5°C in 5% CO<sub>2</sub> in air, for 7 d (IVF = Day 0). On Day 6, SOF was supplemented with 0 (Control group), 2.5 μM MD (MD 2.5) or 5.0 μM MD (MD 5.0). The cleavage and blastocyst rates were evaluated at Days 3 and 7, respectively. Day-7 blastocysts were stained with H<sub>2</sub>DCFDA (Molecular Probes, Invitrogen) or with TUNEL (in situ cell death detection kit, Roche Life Science) to evaluate the ROS levels and the apoptotic rates, respectively. Stained embryos were evaluated under epifluorescence (excitation 495/510–550 nm and emission 520/590 nm, respectively for H<sub>2</sub>DCFDA and TUNEL) and the images were analyzed by Q-Capture Pro Image Software to determine the fluorescence intensity. Data were analyzed by ANOVA followed by Tukey's test ( $P < 0.05$ ) and are presented as Mean ± SEM. The cleavage rates were similar ( $P > 0.05$ ) among groups ( $78.2\% \pm 1.14$  to  $80.5\% \pm 0.88$ ). The blastocyst rates were lower ( $P < 0.05$ ) in MD 5.0 ( $25.9\% \pm 2.06^b$ ) compared with Control ( $36.4\% \pm 1.51^a$ ), and both were similar ( $P > 0.05$ ) to MD 2.5 ( $32.5\% \pm 1.94^{ab}$ ). The ROS levels were higher in MD 2.5 ( $0.75 \pm 0.05^b$ ) and MD 5.0 ( $1.11 \pm 0.07^b$ ) compared with Control ( $0.72 \pm 0.04^a$ ). The rates of apoptosis were higher ( $P < 0.05$ ) in MD 5.0 ( $19.24\% \pm 0.69^c$ ) compared with MD 2.5 ( $13.28\% \pm 0.71^b$ ) and Control ( $10.0\% \pm 0.45^a$ ). In conclusion, MD concentrations of 2.5 and 5.0 μM were effective in inducing oxidative stress in bovine embryos produced in vitro and the detrimental effects were dose-dependent. The higher the oxidative stress, the more detrimental were the effects, causing reduction in the embryonic development, increasing on the intracellular ROS levels and apoptosis.

**Key Words:** menadione, oxidative stress, apoptosis

**T327 Antioxidants improve membrane integrity and acrosome and sperm mitochondrial activity in ram sperm after cryopreservation.** Elenice A. Moraes\*<sup>1</sup>, Wildelfrancys L. Souza<sup>1</sup>, Jonathan M. S. Costa<sup>1</sup>, and James K. Graham<sup>2</sup>, <sup>1</sup>Federal University of San Francisco Valley, Petrolina, PE, Brazil, <sup>2</sup>Colorado State University, Fort Collins, CO.

There has been a renewed interest in the effects of oxidative damage to human sperm, as this damage to mitochondrial DNA and membrane architecture may explain the impaired fertility of the cryopreserved sperm. Different antioxidants have been tested to improve sperm quality but distinct and consistent beneficial effects are lacking. The objective was to determine if adding a combination different antioxidants improved ram sperm quality after cryopreservation. Thirty ejaculates, from 3 rams, were split and diluted into an egg-yolk-tris diluent containing different antioxidant treatments: control; 100 μM melatonin (MEL) plus 0.05% ascorbic acid (MEL+AA); 100 μM MEL plus 90 μL Trolox-C (MEL+TRO); 90 μL TRO plus 0.05% AA (TRO+AA); and 100 μM MEL plus 0.05% AA plus 90 μL of TRO (MEL+AA+TRO); to final concentration  $200 \times 10^6$  sperm/mL. The samples were cooled to 5°C/2 h, packaged into 0.5-mL straws, and frozen in static liquid nitrogen vapor for 15 min before being plunged into liquid nitrogen. Straws were thawed (37°C/30 s). The motility was determine using CASA, and to plasma membrane integrity the samples were stained with 2 L of Hoechst 33342 and 2 μL PI and incubated at 37°C/8min. Acrosomal integrity was determined, visually, after staining the sperm with 50 μL FITC-PNA at 37°C/20min. The sperm mitochondrial function was assessed using Rhodamine and PI (samples were incubated at 37°C/8

min with 2  $\mu$ L Rhodamine and 2  $\mu$ L PI) and the samples assessed for the proportion of viable spermatozoa with high mitochondrial activity. Variables were determined by ANOVA using a Tukey test. Sperm treated with MEL+AA (29.1%) and MEL+AA+TRO (28.9%) maintained higher percentages of cells with intact plasma membranes after thawing ( $P < 0.05$ ) than other treatments. The antioxidant combination (MEL+AA+TRO) resulted in the highest percentages of sperm with intact acrosomes (84.5%) and mitochondrial activity (96.4%) compared with other treatments ( $P < 0.05$ ). All antioxidant treatments exhibited higher motility (61.4% versus 53%), acrosome integrity (78.1% versus 68.2%) and mitochondrial activity (92.7% versus 88.6%), than control ( $P < 0.05$ ). Therefore, adding a combination of MEL+AA+TRO to ram sperm improved cell cryosurvival. Supported by FACEPE/CAPES.

**Key Words:** melatonin, Trolox-C

### **T328 Testosterone enhances basal, FSH- and IGF-I-stimulated aromatase gene expression in porcine granulosa cells in vitro.**

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Ovarian follicular development is regulated by systemic as well as autocrine and paracrine factors such as estradiol (E2). E2 is produced by growing follicles via aromatase conversion of androgens into estrogens. Both FSH and IGF-I induce aromatase activity in granulosa cells (GC) of several species. To further evaluate the effects of FSH, IGF-I and testosterone on abundance of aromatase (*CYP19A1*) mRNA, GC from small porcine follicles (1 to 5 mm) were cultured for 2 d in medium containing 5% fetal bovine serum and 5% porcine serum followed by 48 h in serum-free medium containing control (no addition), FSH (30 ng/mL), or FSH plus IGF-I (30 ng/mL) in the absence or presence of 500 ng/mL of testosterone in a 2  $\times$  3 factorial design. Cellular RNA was isolated and relative *CYP19A1* mRNA abundance was measured using real-time PCR with *18S* rRNA as a housekeeping gene. Data were analyzed via GLM procedure of SAS. Alone, FSH increased ( $P < 0.05$ ) *CYP19A1* mRNA abundance 63-fold. Addition of IGF-I to FSH-treated cells increased *CYP19A1* mRNA abundance 6.5-fold above that seen with FSH alone. Testosterone alone increased ( $P < 0.05$ ) *CYP19A1* mRNA abundance 30-fold and further amplified both FSH- and FSH plus IGF-I-induced *CYP19A1* mRNA abundance by 3- to 4-fold. In a second experiment, GC were cultured as in the first experiment except that medium (containing 500 ng/mL of testosterone as E2 precursor) was collected after 1 and 2 d of treatment for E2 measurement via RIA. FSH increased ( $P < 0.05$ ) E2 production by 1.3- and 1.5-fold after 1 d and 2 d of treatment, respectively. IGF-I increased ( $P < 0.05$ ) FSH-induced E2 production by 4.7- and 9.2-fold after 1 d ( $0.19$  vs.  $0.94 \pm 0.06$  ng/ $10^5$  cells) and 2 d ( $0.21$  vs.  $1.94 \pm 0.09$  ng/ $10^5$  cells) of treatment,

respectively. In conclusion, testosterone-amplified basal and hormone-induced aromatase gene expression indicates that androgens produced by theca cells may act as a paracrine factor to increase aromatase activity in GC in addition to serving as E2 precursors for GC. This intra-ovarian regulatory mechanism may be vital for optimal follicular growth in pigs.

**Key Words:** aromatase, granulosa cells, pig

### **T329 Ovarian follicular dynamics in early- and late-maturing**

**Brahman cows.** Rui A. d'Orey Branco\*<sup>1,2</sup>, Thomaz H. Welsh<sup>2</sup>, Don Neuendorff<sup>1</sup>, and Ron D. Randel<sup>1</sup>, <sup>1</sup>Texas A&M AgriLife Research, Overton, TX, <sup>2</sup>Department of Animal Science, Texas A&M University, College Station, TX.

Age at first calving is greater in *Bos indicus* than in *Bos taurus* cattle. Two different populations of Brahman cows exist based on age at first calving. Specifically, cows that are capable of calving by 26 mo of age are defined as early maturing (EM). Conversely, those cows that must be over 33 mo of age to calve are considered late maturing (LM). The objective of this trial was to compare follicular dynamics of EM and LM cows. This study's herd (located near Overton, Texas) was comprised of 120 pure bred Brahman cows. The EM group (n = 7) calved before 26 mo of age and the LM group (n = 7) calved at 33 mo of age or later. Cow BCS was between 4.5 and 7.5 and all of the cows calved at least 60 d before the trial began. Follicular evaluation was performed by transrectal ultrasound (US; Medison Co. SA-600; 7.5 MHz probe) and began after a naturally occurring estrus detected by a heat check bull. Cows were examined daily by US until signs of a new estrus were detected. During this second estrus the cow was examined by US every 4 h. Data were analyzed using PROC GLM of SAS. The EM and LM groups did not differ ( $P > 0.1$ ) with respect to the diameter of the pre-ovulatory follicle or the time elapsed from the onset of estrus to ovulation. There were 3 follicular waves in 85.7% (6/7) of the LM and 14.3% (1/7) in EM cows (chi-squared;  $P < 0.05$ ). The size of the largest subordinate follicle on the day before estrus was greater ( $P < 0.01$ ) in EM ( $10.42 \pm 2.38$  mm) than in LM ( $5.23 \pm 2.31$  mm) cows. The number of subordinate follicles (>4mm) one day before estrus in EM cows ( $4.28 \pm 2.55$ ) was greater than in the LM cows ( $1 \pm 0.76$ ;  $P < 0.01$ ). The total of follicular diameters one day before estrus was greater ( $P < 0.01$ ) in EM ( $32.43 \pm 5.63$  mm) than in LM ( $7.43 \pm 5.63$  mm) cows. These results suggest that early and late maturing cows have differing aspects of follicular dynamics such as the number of follicular waves, the number of subordinate follicles, and the diameter of the second largest follicle. Further studies to determine whether this phenotypic difference is present in cows of other breeds are warranted.

**Key Words:** follicular dynamics, early maturing, late maturing

## Production, Management and the Environment II

**T330 Influence of egg holding time on hatchability of Hubbard broiler eggs.** Modupe Orunmuyi\*<sup>1</sup> and Chidiebere Livinus Akanwa<sup>2</sup>, <sup>1</sup>Federal University, Oye-Ekiti, Ikole-Ekiti, Ekiti State, Nigeria, <sup>2</sup>Ahmadu Bello University, Zaria, Kaduna State, Nigeria.

In a study to determine the influence of egg storage time on hatchability in Zaria, northern guinea savannah of Nigeria, 1,634 Hubbard broiler breeder eggs stored for 1 to 7 d at 16°C were set in 8 batches when the birds were 42–49 weeks of age to determine the maximum egg holding time. Birds were raised on deep litter at a mating ratio of 1 rooster to 6 hens in a pedigree hatching involving 12 sire families. Data collected were subjected to ANOVA (GLM PROCEDURE) using SAS 2008. There were significant ( $P < 0.05$ ) differences in all parameters studied namely; set weight, transfer weight, fertility%, hatchability of all eggs set (%hatch), hatchability of fertile eggs (% hatchability) and chick weight except egg weight at collection. Average initial egg weight before storage ranged from 68.08 to 69.57g. The range of values for set weight, transfer weight, %fertility, %hatch, %hatchability and chick weight at hatch were 64.88–68.08g, 55.19–58.87g, 54.63–70.92%, 26.50–39.81%, 41.58–62.92% and 41.13–44.77% respectively. Set weight was highest at 1 d storage and lowest at 7 d storage. Transfer weights were similar from 1 d to 5 and were different from d 6 and 7. Although fertility % was lowest for 1 d storage, it was not significantly different from d 3 and 7. Percentage hatch was lowest for 7 d storage (26.50%) while % hatchability was highest for 1 d storage (62.92%). Chick weight at hatch was similar on all days except d 4. Fertility % was lowest (54.63 ± 0.06%) for eggs stored for 7 d. Eggs meant for hatching may not be stored for more than 5 d in this environment.

**Key Words:** Hubbard broiler, egg storage, hatchability

**T331 Performance of dual-purpose cows supplemented with two sources of energy in subtropical conditions.** Isela Salvador-Loreto<sup>1</sup>, Benito Albarran-Portillo\*<sup>1</sup>, Fernando Vicente-Mainar<sup>2</sup>, Isela G. Salas-Reyes<sup>1</sup>, Carlos M. Arriaga-Jordan<sup>1</sup>, and Julieta G. Estrada-Flores<sup>1</sup>, <sup>1</sup>Universidad Autonoma del Estado de Mexico, Toluca, Mexico, Mexico, <sup>2</sup>Servicio Regional de Investigación y Desarrollo Agroalimentario, Villaviciosa, Asturias, España.

The aim of this study was to assess the performance of lactating dual purpose cows (milk and calves), supplemented with 2 sources of energy: molasses (Mo) or ground corn (Gc) during the dry season under subtropical conditions in Mexico. In the subtropical regions of Mexico, during the dry season there is a lack of quality and quantity of fodders to support lactating cows and their suckling calves under grazing systems. Therefore, farmers supplement their cows to maintain milk production and calves weight gain. Supplements are based on cracked corn, cob and husk (CCH), soybean meal and urea (70 g/cow/d). The CCH processing method reduced ruminal starch digestibility, limiting the potential energy cows could obtain from the supplement. Treatments (TX) consisted of molasses or ground corn at 6% inclusion in CCH supplement (5 kg of DM cow/d). Response variables were recorded every 2 weeks, considered as experimental periods (EP). Milk, milk components, body weight, calves weight and body condition score, were subjected to a mixed model procedure from SAS 9.0 for a complete randomized experimental design. Cow was defined as a random effect. There were no significant differences among response variables due to TX (milk yield 7.1 vs 7.0 kg/cow/d, molasses and ground corn TX, respectively), but there were differences ( $P < 0.05$ ) for EP on milk yields which increased

by the end of the experiment with milk yields of 6.5 and 8.6 (kg/cow/d, EP 1 and 5, respectively). There were no differences on animal response variables when molasses or ground corn at 6% inclusion were added to supplements as an extra source of energy.

**Key Words:** supplement, molasses, ground corn

**T332 Evaluation of the feedlot performance, carcass yield, and production costs of finishing beef cattle supplemented with  $\beta$ -agonists.** Silvia Larios-Cueto, Gilberto Aranda-Osorio\*, Rodolfo Ramirez-Valverde, Hermilo Suarez-Dominguez, and Jose M. Monzon-Armenta, Universidad Autonoma Chapingo, Chapingo, Texcoco, Mexico.

The objective of this study was to evaluate the effect of  $\beta$ -agonists (zilpaterol and ractopamine hydrochloride) supplementation on finishing beef cattle performance and carcass quality. These agonists are approved in Mexico. There were used 507 commercial crossbred (*Bos taurus* × *Bos indicus*) young bulls, initially weighing 446.3 ± 9.0 kg, from the tropical region (Veracruz State), and allotted to 3 pens (19 each) of the Experimental Feedlot Facility of the University of Chapingo (Mexico State a temperate region), each pen was randomly assigned to the following treatments: 1) Control (no supplement added), 2) Ractopamine (0.91 mg/kg of body weight (BW) per day) and 3) Zilpaterol (0.15 mg/kg of BW per day). All animals received the same finishing diet, which has the following calculated nutritional composition: 14.48% CP; 1.79 and 1.16 Mcal/kg of NEm and NEg, respectively, 11.16% CF, and 2.2 Ca:P. At the moment that cattle arrived to the feedlot they received a prophylactic management (identified, weighed, vaccinated, dewormed, vitaminized and implanted). The  $\beta$ -agonists were supplemented at least 35 d at the end of the finishing period. The variables evaluated were: dry matter intake (DMI), average daily gain (ADG), feed:gain (F:G), gain:feed (G:F), hot carcass yield (HCY), trimmed carcass fat (TCF) and benefit/cost (B/C) ratio. Data were analyzed under a completely randomized design with the GLM procedure of SAS, and to compare means the Tukey Test was performed. The only variable that was not statistically analyzed was DMI because it was calculated collectively. Cattle performed very similarly and no differences ( $P > 0.05$ ) were found among treatments, the mean values being 11.88 kg/animal per day for DMI, 1.80 kg for ADG, 7.35 kg for F:G, 143 g/kg for G:F, 57.7% for HCY and 2.2% for TCF. Although, the B/C ratio was better for ractopamine treatment (1.13 vs. 1.05 for control and zilpaterol, respectively). Under the conditions of this study, the supplementation of  $\beta$ -agonists (ractopamine or zilpaterol hydrochloride) during the finishing period of young bulls showed a tendency to increase animal performance and carcass traits, but only ractopamine demonstrated to be profitable.

**Key Words:** bullock, ractopamine, zilpaterol

**T333 Physiological responses in Santa Ines ewes submitted to stressful conditions of management.** Monalisa de Melo Stradiotto\*<sup>1,2</sup>, Alice Deléo Rodrigues<sup>1,2</sup>, and João Alberto Negrão<sup>1,2</sup>, <sup>1</sup>University of Sao Paulo–USP, Pirassumunga, SP, Brazil, <sup>2</sup>University of Sao Paulo State–UNESP, Jaboticabal, SP, Brazil.

Literature data suggest that although there is a great variability of stress responses, animals maintain a pattern of response to different stressful factors. In this context, the objective of this experiment was to verify the different physiological responses of Santa Ines ewes to management



conditions considered stressful to the animals. For that, 50 Santa Ines ewes were submitted to 3 different impositions of stress (1: weaning, 2: first milking, 3: change of milker). Cortisol levels of experimental ewes were monitored along lactation and after weaning, 60 d after birth. Plasma samples were collected through jugular puncture in 5 times at -20 min (20 min before the stressful stimulus), 0 min (at the moment of the stressful stimulus) and 60, 120 and 300 min after the stimulus. Hormonal analyses were done using immunoenzymatic kits with readings at 450nm in ELISA equipment at the Laboratory of Animal Physiology of FZEA/USP. Hormonal levels were analyzed in subdivided parcels, which means as repeated measures in time, through MIXED procedure. Means were compared by Tukey test with a significance level of 0.05. For the change of milker stimulus, it were observed greater values in cortisol levels in 0, 60 and 120 min after stress imposition (31.557 ng/mL  $\pm$  26.153; 30.106 ng/mL  $\pm$  18.138 and 23.989 ng/mL  $\pm$  15.982, respectively;  $P = 0.0069$ ), statistically different from the other times of measurement. In the stress of the first milking, it were observed greater values of cortisol levels 60 min after milking (48.274 ng/mL  $\pm$  60.07;  $P = 0.001$ ) and almost 5 h later, the basal levels were reestablished. Therefore, the pattern of cortisol release in the first milking and weaning differed where in the last, it remained elevated until 300min after stress imposition (40.334 ng/mL  $\pm$  59.971), showing that this type of stress can influence the productive life of Santa Ines ewes.

**Key Words:** cortisol, milking, stress

**T334 Goal structure and reasons for selecting a goat enterprise by US meat goat producers.** Narayan P. Nyaupane\*<sup>2</sup>, Jeffrey M. Gillespie<sup>1</sup>, and Kenneth W. McMillin<sup>1</sup>, <sup>1</sup>Louisiana State University, Agricultural Center, Baton Rouge, LA, <sup>2</sup>The Samuel Roberts Noble Foundation Inc., Ardmore, OK.

The US meat goat industry has expanded rapidly in recent years; however, the US continues to rely on goat meat imports due to a rise in demand associated with increased immigration. US meat goat industry leaders need to know the current direction and drivers of production so that necessary measures can be taken for establishing a sustainable and competitive industry. We seek to determine US meat goat producers' goal structure and their perceived reasons of farming. Survey data from 584 US meat goat producers was used for this study. Using the fuzzy pair-wise comparison method, the hierarchy of 7 major goals that a meat goat producer could have were determined and ordinary least squares regressions were conducted to analyze the factors affecting producer goal structure. To determine the producer's reasons of farming, the following question was asked with a list of 14 possible reasons in a 5 point Likert-scale: "To what extent do you agree or disagree that your selection of a goat enterprise as opposed to other agricultural enterprises is because of the following reasons"? Ordered probit regressions were conducted for factors affecting reasons of farming. Results show that *Maximize Profit* was the primary goal of US meat goat producers with the mean score of 0.51, followed by *Have Family Involved in Agriculture* (0.50), *Avoid Years of Loss/Low Profit* (0.49) and *Have Time for Other Activities* (0.48), *Increase Net Worth* (0.46), *Control Weeds/Vegetation* (0.40), and *Increase Farm Size* (0.33). Means of producer agreements showed that the most frequently cited reasons for producers selecting goat enterprise were: *I enjoy working with goats* (4.59), *goat production fits well into my land management plan* (4.30), *I can raise goats on a relatively small acreage* (4.25), and *goat grazing preferences are different from other species* (4.17). Regression results show that producer demographics, farm characteristics, regional variables, and other economic indicators affect producer goal structure and enterprise selection.

**Key Words:** goat, goal structure, reasons for farming

**T335 Managing variability in decision making in swine growing-finishing units.** P. S. Agostini<sup>1</sup>, E. G. Manzanilla<sup>2</sup>, C. de Blas<sup>3</sup>, A. G. Fahey<sup>4</sup>, C. A. Silva\*<sup>5</sup>, and J. Gasa<sup>1</sup>, <sup>1</sup>Universitat Autònoma de Barcelona, Bellaterra, Spain, <sup>2</sup>Animal and Grassland Research and Innovation Centre, Teagasc, Fermoy, Ireland, <sup>3</sup>Universidad Politécnica de Madrid, Madrid, Spain, <sup>4</sup>University College Dublin, Dublin, Ireland, <sup>5</sup>Universidade Estadual de Londrina, Londrina, Brazil.

Analysis of data from pig producing companies may be useful for the optimization of farm management. However, obtaining these data and drawing conclusions from them presents several challenges. In the present study, information from 686 batches of growing-finishing (GF) pigs from 404 GF farms integrated in 7 Spanish companies was obtained. Management and facility factors affecting feed conversion ratio (FCR) and mortality (MORT) were studied by multivariate linear mixed regression analysis in each single company (A to G) and in an overall database (OD). Factors studied were geographic location of the farm, trimester the pigs entered the farm, genetics, sex, use of circovirus vaccine, number of pig origins the pigs were obtained from, age of the farm, percentage of slatted floor, type of feeder, drinker and ventilation, number of phases and form of feed, antibiotic administration system, water source, and number and initial weight of pigs. The variability among companies was much higher than within companies and some factors presented no variability within companies. In most companies and in the OD batches had better FCR and/or lower MORT ( $P \leq 0.05$ ) when animals: 1) were placed between April and September, 2) came from a unique sows farm origin and 3) had lower initial body weight, 4) were originated from a Pietrain pig-sire, presence of males and segregated in pens (only for FCR), 5) were housed in farms less than 10 years old (only for FCR), 6) were housed in farms where the drinking water was properly treated (only for MORT) and 7) were raised in farms with less than 800 pigs (only for MORT). Age of the farm, antibiotic administration system, and water source were provided by only some companies and were not included in the OD model. However, when analyzed in particular companies, these 3 variables had an important effect and may be of interest even for the companies that do not record them. In general, models developed for each company were more accurate than those obtained from the OD.

**Key Words:** grow-finishing pig, facilities, management

**T336 The production performance of egg layers in different rearing systems.** Bilgehan Yılmaz Dikmen\*, Aydin Ipek, Ümran Sahan, and Arda Sözcü, Department of Animal Science, Faculty of Agriculture, University of Uludağ Bursa, Turkey.

Today alternative rearing systems for caged hen egg production has gained importance. The objective of this study was to determine the production performance of laying hens kept in different rearing systems (conventional, enriched cages and free-range system). A total of 480 egg layer strain (*Lohmann Brown*) were weighed and randomly allocated into 3 rearing system (n = 160, 4 replicates of 40 hen per group) as conventional cage, enriched cage and free range pen at 17 wk of age. The 3 rearing systems were located on a single farm and experienced identical seasonal temperature fluctuations and photoperiod (16L:8D). Feed and water were given ad libitum until 66 wk of age. Live weight was measured at the beginning and at the end of the study. Egg production as hen day, number of damaged eggs, dirty eggs and mortality were monitored daily. Feed intake and egg weight were recorded on weekly basis. The data were analyzed with using ANOVA, and significant means were separated using Duncan Multiple Range Test. The final live weight of hens were found similar in conventional and enriched cage system (1.95 vs. 1.94  $\pm$  0.03 kg, respectively), but was higher in free-range

system ( $2.09 \pm 0.02$  kg;  $P < 0.001$ ). The earliest 5% egg production age was found in enriched cage system ( $138.3 \pm 1.75$  d;  $P < 0.05$ ). The latest 50% egg production age was found in free-range system ( $159.25 \pm 1.75$  d;  $P < 0.01$ ). The highest mean egg production ratio was found in free range rearing system ( $89.27 \pm 0.87\%$ ;  $P < 0.05$ ). The egg mass was higher in free-range system, but was similar in conventional and enriched cage systems ( $59.76, 56.80$  vs.  $56.66 \pm 0.34$  g/d/hen, respectively;  $P < 0.001$ ). The feed intake was found to be higher in free-range system, but similar between conventional and enriched cage systems ( $124.58, 117.06$  and  $118.06 \pm 0.56$  g/d/hen, respectively;  $P < 0.001$ ). The damaged egg ratio was low but dirty egg ratio was higher in free-range systems ( $0.35 \pm 0.07\%$  vs.  $3.30 \pm 0.15\%$ , respectively;  $P < 0.001$ ). The mortality ratio was higher in enriched cage system ( $6.25\%$ ;  $P < 0.05$ ). It was concluded that free-range system has a positive effect on egg production and egg weight, but feed intake and dirty egg ratio were higher than the caged system.

**Key Words:** rearing system, egg production, egg layer

**T337 Effect of a single dose of injectable trace minerals on measurements of performance, innate and humoral immune responses of beef heifers.** Luana S. Caramalac<sup>\*1</sup>, Philippe Moriel<sup>2</sup>, Luis F. A. Artioli<sup>2</sup>, and John D. Arthington<sup>3</sup>, <sup>1</sup>Mato Grosso do Sul State University, Aquidauana, MS, Brazil, <sup>2</sup>North Carolina State University, Waynesville, NC, <sup>3</sup>University of Florida, Ona, FL.

Injectable trace minerals (ITM) administered simultaneously to novel antigens of porcine red blood cells (PRBC) affects trace mineral status, innate and humoral immune responses of weaned beef heifers. Angus heifers ( $n = 22$ ;  $234 \pm 39$  kg of BW;  $225 \pm 19$  d of age) were stratified by BW and age, and randomly assigned to receive a 2.5-mL s.c. injection of sterile saline (SAL; 0.9% NaCl), ITM (MultiMin 90; Multimin USA, Inc. Fort Collins, CO; 60, 10, 15 and 5 mg/mL of Zn, Mn, Cu and Se), or a squalene-based oil-in-water adjuvant (ADJ; AddaVax; InvivoGen, Inc. San Diego, CA) that promotes humoral immune response. Thereafter (d 0), heifers were immunologically challenged with a 10-mL i.m. injection of PRBC solution (25% PRBC; 75% sterile PBS), allocated to drylot pens (2 pens/treatment) and limit-fed a fescue hay-based diet at 2.2% of BW for 28 d. Shrunken BW was recorded on d 0 and 28, blood samples collected on d 0, 3, 7, 15, 21 and 28, and liver samples collected on d 14. Body weight on d 28 ( $263, 262$  and  $258 \pm 19$  kg) and mean ADG ( $0.81, 0.98$  and  $0.96 \pm 0.180$  kg/d for SAL, MM and ADJ heifers) did not differ among treatments ( $P \geq 0.72$ ). But, treatment effects were detected ( $P \leq 0.05$ ) for concentrations of liver Se, plasma ceruloplasmin (Cp) and serum PRBC titers. Liver Se was greater for MM vs. ADJ and SAL heifers ( $2.21, 1.36$  and  $1.45 \pm 0.177$  mg/kg, respectively), whereas plasma Cp was greater for ADJ vs. SAL ( $26$  vs.  $20 \pm 1.6$  mg/dL;  $P = 0.02$ ), and intermediate for ITM heifers ( $23 \pm 1.6$  mg/dL;  $P \geq 0.17$ ). Likewise, serum PRBC titers were greater for ADJ vs. SAL ( $1.6$  vs.  $0.7 \pm 0.28$  log<sub>2</sub> base;  $P = 0.02$ ), and intermediate for ITM heifers ( $1.1 \pm 0.28$  log<sub>2</sub> base;  $P \geq 0.13$ ). Treatment x time effect was detected for plasma haptoglobin (Hp) concentrations ( $P < 0.0001$ ), which did not differ from d 0 to 28 ( $P \geq 0.18$ ), but were greater for ADJ vs. SAL and ITM heifers on d 3 ( $0.71, 0.05$  and  $0.07 \pm 0.032$  mg/mL). Hence, although injectable trace mineral injection enhanced liver Se concentrations, the magnitude of the innate and humoral immune responses to novel antigen were less compared with a water-in-oil adjuvant.

**Key Words:** trace minerals, heifer, humoral

**T338 Pre-weaning injections of bovine somatotropin enhanced post-weaning growth performance and puberty attainment of beef heifers.** Philippe Moriel<sup>1</sup>, Luis F. A. Artioli<sup>\*1</sup>, Phillip Lancaster<sup>3</sup>, John D. Arthington<sup>3</sup>, and Reinaldo F. Cooke<sup>2</sup>, <sup>1</sup>North Carolina State University, Waynesville, NC, <sup>2</sup>Oregon State University, Burns, OR, <sup>3</sup>University of Florida, Ona, FL.

Effects of injections of bovine somatotropin (bST) to suckling beef heifers on pre- and post-weaning growth performance and puberty achievement. Angus x Brangus heifers ( $n = 30$ ; BW =  $140 \pm 20$  kg; age =  $132 \pm 15$  d) were stratified by BW and age, and randomly assigned to receive s.c. injections of saline (SAL; 5 mL; 0.9% saline) or half-dose of bST (250 mg of somatotropin zinc; Posilac, Elanco, Greenfield, IN) on d 0, 14 and 28 (15 heifers/treatment). Then, cow-calf pairs were grazed on 4 bahiagrass pastures (7 to 8 pairs/pasture) from d 0 until weaning (d 126). Unshrunk BW and blood samples were collected on d 0, 14, 28 and 126. From d 126 to 350, heifers were grazed on bahiagrass pastures (1 pasture/treatment) and fed blackstrap molasses-based concentrate at 1.1% BW (DM basis). Unshrunk post-weaning BW was obtained every 28 d, and blood samples every 10 d to determine plasma progesterone (P4) concentrations. Heifers were considered pubertal when 2 consecutive plasma samples had  $P4 \geq 1.5$  ng/mL. During the pre-weaning phase, bST injections increased ADG from d 0 to 42 ( $1.2$  vs.  $1.1 \pm 0.04$  kg/d;  $P = 0.03$ ), but not ADG from d 42 to 126 or weaning BW ( $0.89$  vs.  $0.84 \pm 0.028$  kg/d and  $266$  vs.  $261 \pm 7$  kg;  $P \geq 0.28$ ). Also, bST injections increased pre-weaning mean plasma IGF-1 ( $97$  vs.  $85 \pm 3.7$  ng/mL;  $P = 0.02$ ), and tended to decrease plasma leptin on d 42 ( $5.2$  vs.  $5.9 \pm 0.24$  ng/mL;  $P = 0.06$ ). Pre-weaning bST injections increased post-weaning mean ADG and BW on d 350 ( $0.30$  vs.  $0.22 \pm 0.02$  kg/d and  $329$  vs.  $311 \pm 3$  kg;  $P < 0.01$ ), tended to increase overall puberty achievement ( $49$  vs.  $30 \pm 6.9\%$  of total heifers;  $P = 0.06$ ) and decrease age at puberty ( $402$  vs.  $435 \pm 13$  d;  $P = 0.08$ ), but did not affect BW at puberty ( $287$  vs.  $288 \pm 7$  kg;  $P = 0.95$ ). Hence, 3 half-dose injections of bST administered to suckling beef heifers at 14-d intervals, between 132 and 160 d of age, did not affect body weight at weaning or puberty, but enhanced post-weaning ADG, tended to increase puberty attainment and decrease age at puberty.

**Key Words:** somatotropin, heifer, puberty

**T339 Feed intake and feed conversion ratio in integrated pig fattening farms are affected by management practices and barn conditions.** Caio Abécio Silva<sup>\*1</sup>, Piero da Silva Agostini<sup>2</sup>, and Josep Gasa<sup>2</sup>, <sup>1</sup>Universidade Estadual de Londrina. Departamento de Zootecnia, Londrina, Paraná, Brazil, <sup>2</sup>Universidad Autónoma de Barcelona, SNIiBA, Depto. de Ciencia Animal y Alimentos, Barcelona, Cataluña, Spain.

This study aims to investigate the effect of some in-farm management and facility factors on the average daily feed intake (ADFI) and feed conversion ratio (FCR) of grow-finishing pigs. Information from 1,532 batches from 216 grow-finishing farms consisting of 1,025,385 pigs ( $22.6$  to  $116.7$  kg of live weight) belonging to 4 integrated pig companies (located in southwest of Parana State, Brazil) were collected by survey during 2010 and 2012. Risk factors obtained included 13 management practices and 9 facilities factors. A multivariate linear mixed regression model was used including factors that presented a  $P < 0.20$  in a univariate model for ADFI and FCR. The “company” and “farms within the company” were considered as random factors whereas “batches within the farm and company” as the residual factor. Risk factors in the multivariable model were considered to have a significant effect if  $P \leq 0.05$  and a trend effect if  $0.05 < P \leq 0.10$ . The mean ADFI was  $2.14 \pm 0.10$  kg and FCR was  $2.45 \pm 0.12$  kg/kg. Lower ADFI and/or better

FCR ( $P < 0.01$ ) were found in batches with (a)  $\leq 20$  pigs per pen vs  $>20$  pigs (respectively 3.4 and 4.1 points), (b) linear feeder vs plastic conic (2.6 points, both), (c) females vs barrows or mixed (4.1 and 4.9 points), (d) pigs coming from other farms vs those coming from own integrated farms (1.3 point, both), (e) absence of water depth area in pens vs presence of water depth (1.8 point, just ADFI,  $P = 0.06$ ) and (f) barns built in masonry vs barns built in wood or mixed (2.5 points, just FCR). For each 1-kg of increase of initial and final body weight the ADFI was, respectively 0.007 and 0.010 kg higher ( $P < 0.001$ ) and FCR 0.03 poorer and 0.012 better, respectively ( $P < 0.001$ ). FCR was also 0.015 poorer per additional day of the fattening duration. Total variability explained by the models was 41% for ADFI and 55% for FCR. This study indicates that ADFI and FCR were influenced by some farm facilities and management practices that could be modified considering that some of them do not demand great investments.

**Key Words:** grow-finishing pig, management, performance

**T340 Modeling and simulation for beef cattle stocker-finishing systems analysis.** Julio Otavio Barcellos\*<sup>1</sup>, Leonardo C. Canellas<sup>1</sup>, Vinicius N. Lampert<sup>2</sup>, Fernanda G. Moojen<sup>1</sup>, Marcela Kuczynski Rocha<sup>1</sup>, Eduardo T. Azevedo<sup>1</sup>, Gabriel R. Pereira<sup>1</sup>, Silvio R. O. Menegassi<sup>1</sup>, and Eduardo Antunes Dias<sup>1</sup>, <sup>1</sup>NESPRO/UFRGS, Porto Alegre, RS, Brazil, <sup>2</sup>EMBRAPA/CPPSUL, Bagé, RS, Brazil.

The purpose of this study was to present a management model for stocker-finishing cattle systems through modeling and simulation results from intervening the different levels of intensification, serving as a Decision Support System (DSS) for ranchers. Model was developed and operated in Microsoft Excel spreadsheets, with data collected in the field and literature reviews. The system allows to individually manipulated the animal feeding in the herd throughout the production cycle. This approach will measure the results quantitatively, through the existing stocking rate, and qualitative, through the daily weight gain from each diet administered to animals. DDS spreadsheets support the individual management for up to 500 animals in a period of analysis of maximum 53 mo. The model is operated in a single spreadsheet, divided into 3 components: Feeding Systems; Animal Performance; and Summary & Economic Analysis, being all levels interconnected. Model validation was simulated based on the Pasture-Based System (PBS) within 2 situations: Pasture/Feedlot-Based System for Finishing Cattle (FBSF) and Pasture/Feedlot-Based System for Stocker Cattle (FBSS). There was an increase in productivity for FBSF and FBSS (321 and 314 kg/hectare/year, respectively) and a decrease in slaughter age (21 and 23 mo, respectively) compared with PBS, 274 kg/hectare/year and 26 mo ( $P < 0.05$ ). In the economic analysis, PBS (119.23 U\$) showed higher annual gross margin per hectare, followed by FBSF (79.65 U\$) and FBSS (57.25 U\$) ( $P < 0.05$ ). The model provides an overview of integrated management, showing major results during herd's feeding, purchase and selling prices, and animal performance of stocker-finishing systems in beef cattle production. The intensification observed in our study decreases the economic return in the evaluated system. Intensification is not indicated regarding biological and economic parameters used in this simulation. Scenario studies simulating purchase and selling prices, costs and another management interventions are necessary to better support the decision making process.

**Key Words:** intervention assessment, decision-making, beef-cattle production

**T341 Performance and profitability on beef cattle in intensive tropical pasture in Amazon biome.** Fabiano Alvim Barbosa\*<sup>1</sup>, Vando Telles Oliveira<sup>2</sup>, Filipe Lage Bicalho<sup>3</sup>, Luciano Bastos Lopes<sup>4</sup>, Juliana Mergh Leão<sup>1</sup>, and Lucas Luz Emerick<sup>1</sup>, <sup>1</sup>Universidade Federal de Minas Gerais, UFMG, Belo Horizonte, Minas Gerais, Brazil, <sup>2</sup>Instituto Centro de Vida, ICV, Alta Floresta, Mato Grosso, Brazil, <sup>3</sup>Soluções Integradas ao Agronegócio, SIGA, Alta Floresta, Mato Grosso, Brazil, <sup>4</sup>Empresa Brasileira de Pesquisa Agropecuária, Embrapa, Sinop, Mato Grosso, Brazil.

To avoid deforestation in the Amazon rainforest is necessary to increase productivity and profitability in beef cattle. So the aim of this study was to evaluate economic efficiency and productivity of 5 intensive pasture systems (IPS) on beef cattle production in Mato Grosso, Amazon biome, January 2013 to December 2014, part of the Livestock Low Carbon Integrated Project-ICV. Production and economic data were collected using control software (Prodap Professional GP) and analyzed by electronic spreadsheets (Microsoft Excel). The total of IPS was 177.7 ha (ha) and 1,877 cattle. The average of pasture was 35.5 ha/farm and 125 animals/IPS. The IPS was established with *Panicum maximum* 'Mombaça', with limestone, potassium chloride, triple phosphate and urea as soil analysis. During the rainy season were placed urea according stocking rate. The systems was developed exclusively on a grazing system with a base herd comprised of Zebu (Nelore) and crossbred *Bos taurus* × *Bos indicus* cattle. The zootechnical and economic indexes were compared in 3 periods, April–September 2013, October–2013 to March 2014, April–September 2013, by Duncan test, besides Person correlations, with significance level of 5%. During rainy season, October–2013 to March 2014, stocking rates (5.15 animals/ha), live weight/ha (1,553 kg), body weight produced (404 kg/ha) and total operating costs (US\$ 248.18/ha) were higher than other periods ( $P < 0.05$ ). There was no statistical difference to net margin of 155.49 US\$/ha ( $P > 0.05$ ). High correlations were found (0.76) between body weight produced (kg/ha) and total operating costs (US\$/ha) ( $P < 0.05$ ). High correlations were found (0.898) between body weight produced (kg/ha) and net margin (US\$/ha) ( $P < 0.05$ ) indicating that higher productivity is related to higher net margin, despite the higher cost per area. The period had influence on production of beef cattle and on costs in intensive pasture system, but the net margin indicated that the activity of beef cattle in Amazon biome can be profitable and had high correlation with animal production.

**Key Words:** cost, productivity, sustainability

**T342 Carcass quality of grain-finished first-calf heifers.** Jose A. Arce\*<sup>1</sup>, Shawn L. Archibeque<sup>1</sup>, Jason K. Ahola<sup>1</sup>, Richard K. Peel<sup>1</sup>, Dale R. Woerner<sup>1</sup>, Jack C. Whittier<sup>2</sup>, and George E. Seidel<sup>3</sup>, <sup>1</sup>Department of Animal Sciences, Colorado State University, Fort Collins, CO <sup>2</sup>Department of Animal Science and Panhandle Research and Extension Center, University of Nebraska, Scottsbluff, NE, <sup>3</sup>Department of Biomedical Sciences, Colorado State University, Fort Collins, CO.

Single-calf beef production systems require harvesting females after early weaning their first calf; these systems reduce the share of energy destined for maintenance of adult cows. However, pregnancy hormones accelerate bone ossification, which may negatively affect carcass value. The objective of this study was to evaluate preliminary data on estimated lean maturity (LE), bone maturity (BM), overall maturity (OM), marbling (MA) and Warner Bratzler shear force (WBSF) of carcasses from primiparous females approximately 30 mo of age. A set of 53 Angus-based yearling heifers (BW = 352 ± 38.9 kg) were synchronized and inseminated with sexed semen to calve at approximately 24 mo of age; calves were early weaned at 106 ± 21 d postpartum. First calf heifer-



ers (n = 42) were taken to a feedlot to be fed acclimating, set up, and finishing diets for 6, 22, and 60 d, respectively. Diets were 12.7, 13.8 and 12.5% CP; and 0.84, 1.15 and 1.39 Mcal·kg<sup>-1</sup> NE<sub>g</sub>, respectively. At harvest, carcasses were scored for LE and BM to estimate OM in which A, B and C maturities corresponded to scores of 100, 200 and 300, respectively. Also MA was evaluated (slight = 300, small = 400, modest = 500 and moderate = 600) and a LM sample was removed for WBSF measurement. Given the large decrease in quality grade with OM ≥ 300 and that according to approximate age few heifers were supposed to fall into that category, carcasses were sorted by OM as <300 or ≥300, and the resulting means for LE, BM, MA and WBSF were compared with a *t*-test. Means ± SD for the 64% of carcasses classified as < 300 OM were: 170 ± 14 LE, 248 ± 29 BM 220 ± 15 OM, 470 ± 80 MA and 4.87 ± 1.10 kg WBSF. Remaining carcasses were classified as ≥ 300 OM with 169 ± 15 LE, 342 ± 37 BM, 301 ± 3 OM, 485 ± 70 MA and 4.98 ± 0.64 kg WBSF. No differences were detected between the 2 maturity groups for LE (P = 0.76), MA (P = 0.53) or WBSF (P = 0.67); meanwhile significant differences were found for BM and OM (P < 0.001). Therefore, differences in ossification did not affect characteristics related to meat palatability of carcasses of previously pregnant heifers slaughtered at about 30 months of age.

**Key Words:** bone ossification, carcass maturity, shear force

**T343 Occurrence of mycotoxins in corn samples collected in the United States in 2014.** Simone Schaumberger\*<sup>1</sup>, Paula Kovalsky<sup>1</sup>, Michael Sulyok<sup>2</sup>, and Ursula Hofstetter<sup>1</sup>, <sup>1</sup>*Biomim Holding GmbH, Herzogenburg, Austria*, <sup>2</sup>*University of Natural Resources and Life Sciences, Department IFA Tulln, Vienna, Austria*.

Mycotoxin occurrence in agricultural commodities is a worldwide phenomenon and poses a risk to human and animal health. A mycotoxin survey was conducted to raise awareness of the incidence of mycotoxins in cereals intended for animal feed. The focus of this study is to compare the extent of mycotoxin contamination in corn samples from 27 states in the United States in 2014. During harvest 507 corn samples were collected and analyzed for aflatoxins (Afla), zearalenone (ZEN), deoxynivalenol (DON), fumonisins (FUM), T-2 toxin (T-2) and ochratoxin A (OTA). Samples were analyzed using liquid chromatography-tandem mass spectrometry, high performance liquid chromatography and enzyme-linked immunosorbent assay. In total, 87% of samples tested were positive for the major mycotoxins mentioned above and out of those 36% were co-contaminated with more than one mycotoxin. The most frequently occurring mycotoxin was DON with a prevalence of 62% and an average value of 1,441 ppb. DON concentrations exceeding the respective FDA threshold (1,000 ppb; 8%) could be predominantly found in samples deriving from several northern states, California and Florida. FUM occurred in 55% of all samples tested with average value of 4,512 ppb. The FDA threshold for FUM (5,000 ppb) was exceeded in 4% of the samples. Afla and ZEN were detected in about 20% of the samples with average contamination levels of 35 and 484 ppb, respectively. Compared with the other mycotoxins, prevalence of T-2 toxin was low (13%), whereas OTA was not detected at all. Our results demonstrate a significant mycotoxin contamination of US corn harvested in 2014. Overall, the most frequently occurring mycotoxin was DON. FUM ranked second among the 6 mycotoxins analyzed. Owing to the fact that both DON and FUM have the potential to cause subclinical health effects in animals even at comparably low levels in feed (Antonissen et al., 2014), these 2 mycotoxins can be regarded as relevant threat for US livestock productivity. Occurrence of more than one mycotoxin was observed in around one third of the samples. Hence, due to potential

additive and synergistic effects, special attention should be paid to co-occurrence of mycotoxins in animal feed (Grenier and Oswald, 2011).

**Key Words:** mycotoxin, survey, corn

**T344 Antibiotic resistance gene abundance in feces of calves fed pirlimycin-containing whole milk.** Tiffany Casteel\*, Heather Littler, Partha Ray, Amy Pruden, and Katharine Knowlton, *Virginia Tech, Blacksburg, VA*.

Non-saleable (waste) milk containing antibiotics is commonly fed to dairy calves. Effects of ingestion of antibiotics at an early age on the gut microbiome and the development of antibiotic resistance genes (ARG) in the naïve gut are unknown. Pirlimycin, a lincosamide active against gram-positive bacteria, is commonly used as mastitis therapy. Lincosamides are considered highly important in human medicine, with applications in *S. aureus* and *C. difficile* infections. Resistance to pirlimycin is emerging and is of concern for both animal and human health. The objective was to determine the effect of early antibiotic exposure on the abundance of ARG in feces of milk-fed calves. Eight female Holstein calves were assigned to pasteurized whole milk (control; n = 4) or milk containing 0.2 mg/L of pirlimycin (treatment; n = 4). Calves were enrolled after receiving 2 colostrum feedings, were blocked by age, and paired by block. Calves were fed 5.68 L of pasteurized whole milk, treatment or control, divided into 2 daily feedings, from d 1 to d 50 of age. After weaning at the age of 50 d, calves were fed *ad libitum* non-medicated starter grain. Fecal samples were collected weekly until 85 d of age and freeze-dried. DNA was extracted from all samples using QiaAmp Fast DNA Stool Mini Kit and qPCR was used to quantify the abundance (gene copies/g of wet feces) of *TetW* and 16S rRNA. Data were analyzed using PROC GLIMMIX in SAS. Differences were declared significant at P < 0.10, and trends at P < 0.15. Abundance of 16S rRNA was not different between control and pirlimycin-fed calves (mean = 9.81 log gene copies/g wet feces). Abundance of *TetW* tended to be higher (9.51 log gene copies/g wet feces; P = 0.13) in pirlimycin-fed calves compared with control calves. The relative abundance of *TetW* (0.49 gene copies/16S rRNA) in feces of calves was not influenced by treatment. Feeding pirlimycin-containing non-saleable milk to growing calves may increase environmental loading of *TetW*. Additional research is needed on effects on other ARG and on the post-excretion fate of these genes following application of manure from calves fed waste milk.

**Key Words:** antibiotic resistance, waste milk, dairy calf

**T345 Endocrine profiles during early gestation are affected by breed: Suffolk versus Cheviot dams.** Ana Meikle\*<sup>1</sup>, Milena Sequeira<sup>1</sup>, L. M. Fermin<sup>2</sup>, Sarah Pain<sup>2</sup>, Paul Kenyon<sup>2</sup>, and H. T. Blair<sup>2</sup>, <sup>1</sup>*Veterinary Faculty, Montevideo, Uruguay*, <sup>2</sup>*University of Massey, New Zealand*.

Previous studies have shown that Cheviot embryos are shorter than Suffolk embryos at d 19 of gestation, even when uterine capacity is not a limiting factor for embryo growth, evidence of the role the maternal uterine environment plays in embryo development. We determined if there were differences in the circulating concentrations of progesterone (P4), IGF-1, insulin and adiponectin between Cheviot and Suffolk dams during early gestation. Ewes (Cheviot, n = 8 and Suffolk, n = 6; 58.6 vs. 78.2 kg live weight, P < 0.05, with no differences in body condition score) were inseminated (d 0) laparoscopically with fresh semen from rams of the same breed. Blood samples were taken from the ewes every other day from d 0 to 21. Hormone concentrations were analyzed by repeated measures including using the fixed effects of maternal breed,

day of sampling and their interaction; the covariance structure was first-order autoregressive AR(1), and for P4 concentrations the number of corpora lutea was used as a covariable. Cheviot dams had a higher P4 concentration than Suffolk dams from d 6 to 21 of pregnancy ( $6.82 \pm 0.31$  vs.  $5.84 \pm 0.33$  ng/mL,  $P < 0.05$ ) and P4 was also affected by ovulation rate ( $P < 0.001$ ) as animals presenting with 2 corpora lutea had greater P4 than those with a single ovulation. No effect of ovulation rate was found for the rest of the hormones. Insulin concentrations were also greater in Cheviot dams compared with Suffolk dams ( $27.0 \pm 2.0$  vs.  $20.4 \pm 2.3$   $\mu$ UI/ mL,  $P < 0.05$ ), while IGF-I concentrations did not differ among breeds. Suffolk ewes tended to present greater adiponectin concentrations than Cheviot ewes ( $4.4 \pm 1.0$  vs.  $2.1 \pm 0.8$  ng/mL,  $P = 0.099$ ). The greater P4 concentration found in Cheviot dams is consistent with their higher insulin concentration, as insulin stimulates granulosa cell P4 production. The opposite profile was found for adiponectin concentration, with the higher levels found in Suffolk dams potentially associated with adiponectin inhibition of insulin-induced progesterone secretion by granulosa cells, as has been found in other species. The differences in hormone concentrations according to breed could explain the distinct embryo growth found during early gestation.

**Key Words:** sheep, gestation, hormone

**T346 Gene expression in the kidneys of broilers fed dietary ochratoxin A for different time periods.** C. P. Zeferino\*<sup>1</sup>, K. D. Wells<sup>1</sup>, A. S. A. M. T. Moura<sup>2</sup>, G. E. Rottinghaus<sup>1</sup>, and D. R. Ledoux<sup>1</sup>, <sup>1</sup>University of Missouri, Columbia, MO, <sup>2</sup>São Paulo State University, Botucatu, São Paulo, Brazil.

Consumption of ochratoxin A (OTA) contaminated diets results in economic losses to the poultry industry. The objective of the current study was to evaluate the effects of quantity and time of exposure to dietary OTA on performance, organ weights, serum biochemistry, and gene expression in the kidneys of chicks. One hundred eighty day-old male broiler chicks (Ross 308) were randomly assigned to a  $3 \times 3$  factorial arrangement of treatments (3 levels of OTA: 0, 1 and 2 mg OTA/kg diet and 3 time periods: 7, 14 and 21 d) with 4 replicate pens of 5 birds each per treatment. Birds fed 2 mg OTA/kg diet had decreased feed intake, body weight gain and increased serum uric acid levels at 14 and 21 d, and also poorer feed conversion and increased relative kidney weight. Birds fed 1 mg OTA/kg diet had decreased serum total protein. At 21 d, birds fed 1 mg OTA/kg diet had decreased albumin, and aspartate aminotransferase concentrations. For RNA-Sequencing analysis (RNA-Seq), kidney samples were collected weekly from 3 controls and 3 chicks fed 1 mg OTA/kg. NextGENe software was used for read alignment and transcript quantification. Reads per kilobase of target per million tiled reads (RPKM) were used in the Student's *t* test ( $P < 0.05$ , fold change  $> 2$ ). A total of 27,638,976 50-bp RNA-Seq reads were produced over the 3 time periods. Transcripts (40,782) were assembled de novo and annotated by homology to either *G. gallus* or *H. sapiens*. Genes associated with carbohydrate and amino acid metabolism were downregulated and genes associated with the immune system were upregulated at 7 and 14 d. Genes associated with lipid metabolism and xenobiotics biodegradation and metabolism were also downregulated at 14 d. These metabolic changes disappeared at 21 d, suggesting that damage to the kidney and other related organs was repaired or the damage was contained. In conclusion, the decreased performance and increased kidney weight and serum uric acid levels in birds fed 2 mg OTA/kg confirmed the nephrotoxic effects of OTA. The supplementation of 1 mg OTA/kg diet caused time-dependent alterations in renal gene expression in chicks.

**Key Words:** chicken, mycotoxin, RNA-sequencing

**T347 Maintenance energy requirements of gestating beef cows and effects on cow reproduction and performance of calves.** Brit H. Boehmer\* and Robert P. Wettemann, Oklahoma Agricultural Experiment Station, Stillwater, OK.

Spring calving, Angus cows were used to evaluate the repeatability of maintenance energy requirements (MR) during subsequent gestations, and the effects of MR on reproduction in cows and performance of calves. A total of 62 cows (5 to 11 yr of age,  $150 \pm 2$  d of gestation) were individually fed a complete diet to meet MR (Model 1, NRC 1996) during 1 or more yr. Body weights were obtained twice weekly and daily feed intake was adjusted every 2 wk until constant BW was achieved for 30 d (regression analyses). Cows were classified by MR as low (L;  $< 0.5$  SD less than yearly mean), moderate (M;  $\pm 0.5$  SD of mean) or high (H;  $> 0.5$  SD more than mean). Cows ( $n = 26$ ) were selected for repeatability analyses when MR was determined on 2 sequential years. The influence of MR on metabolic body weight, DMI, and pregnancy rate of cows and WW and ADG of calves was determined by ANOVA (PROC MIXED; SAS Inst. Inc.). Repeatability of individual cow MR in sequential years was determined using Pearson correlations with year in the model (PROC CORR; SAS). Pregnancy rate was analyzed by Chi-squared test (PROC FREQ; SAS). Mean MR of cows was  $88.79 \pm 0.89$  kcal/kg BW<sup>0.75</sup>/d. Mean yearly difference between cows with the greatest and least MR was 26.9%. Year influenced MR of cows ( $P < 0.001$ ) but cow age did not affect MR ( $P = 0.99$ ). Maintenance energy requirement of cows was repeatable ( $r = 0.30$ ,  $P = 0.02$ ) between sequential years. Repeatability of MR was determined when cows were fed to maintain a constant body weight rather than estimates of MR. Metabolic body weight at maintenance was greater in L cows ( $P = 0.01$ ;  $117.1 \pm 1.1$  kg<sup>0.75</sup>) compared with H cows ( $111.8 \pm 1.1$  kg<sup>0.75</sup>) and M cows tended to be intermediate ( $P < 0.09$ ;  $114.45 \pm 1.1$  kg<sup>0.75</sup>). At maintenance, DMI was greater in H cows ( $P < 0.001$ ) compared with M and L cows. Subsequent pregnancy rate of cows conceiving to AI was not influenced by MR ( $P = 0.11$ ). Maintenance energy requirement of cows did not influence performance of calves before weaning ( $P = 0.92$ ). Production efficiency of beef cows may be improved by identifying cows that require less energy input and maintain performance.

**Key Words:** beef cow, maintenance, cow-calf performance

**T348 Activation and deactivation of renal genes of chicken associated with induced ochratoxicosis at different exposure times.** C. P. Zeferino\*<sup>1</sup>, K. D. Wells<sup>1</sup>, A. S. A. M. T. Moura<sup>2</sup>, G. E. Rottinghaus<sup>1</sup>, and D. R. Ledoux<sup>1</sup>, <sup>1</sup>University of Missouri, Columbia, MO, <sup>2</sup>São Paulo State University, Botucatu, São Paulo, Brazil.

This study investigated the expression of genes that were turned on or off in renal cells of chicks as a result of different exposure times to ochratoxin A (OTA). One hundred and 80 d-old male broiler chicks (Ross 308) were randomly assigned to a  $3 \times 3$  factorial arrangement of treatments (3 levels of OTA: 0, 1 and 2 mg OTA/kg diet and 3 time periods: 7, 14 and 21 d) with 4 replicate pens of 5 birds each per treatment. For RNA-Sequencing analysis (RNA-Seq), kidney samples were collected weekly from 3 controls and 3 chicks fed 1 mg OTA/kg. Birds fed 2 mg OTA/kg diet were not used because their reduced feed intake could affect gene expression. The libraries were constructed by Illumina's TruSeq RNA protocol. NextGENe software was used for alignment and transcript quantification. Reads per kilobase of target per million tiled reads (RPKM) were used in the Binary test analysis ( $P < 0.05$ ). A total of 27,638,976 50-bp RNA-Seq reads were produced over the 3 time periods. Transcripts (40,782) were assembled de novo and annotated by homology to either *G. gallus* or *H. sapiens*. The interleukin 9 (*IL9*) and tubby like protein 1 (*TULP1*) genes were activated at 7 d, the growth

hormone secretagogue receptor (*GHSR*) gene was activated at 14 d, and the G protein-coupled receptor kinase 6 (*GRK6*) and glyceraldehyde-3-phosphate dehydrogenase (*GAPDH*) genes were activated at 21 d. The preprogastrin (*LOC396365*) was activated during all time periods. The sperm associated antigen 4 (*SPAG4*), sperm-associated antigen 4 protein-like (*LOC100857131*) and V-region-like B-G antigen, and MHC class IV antigen (*B-G*) were deactivated at 7 d, the zinc finger B-box domain-containing protein 1-like (*LOC771469*), NK2 homeobox 1, variant 2, and NK2 homeobox 8 (*NKX2-1* and *NKX2-8*), forkhead box O1 (*FOXO1*), myosin heavy chain (*MyHC*) and claudin 18 (*CLDN18*) genes were deactivated at 14 d and finally, the V-region-like B-G antigen (*B-G*) and xeroderma pigmentosum, complementation group C (*XPC*) genes were deactivated at 21 d. The turning on and off of the genes may be a response to the carcinogenic and tumorigenic effects of OTA in birds.

**Key Words:** broiler, nephrocarcinogenicity, ochratoxin A

**T349 Evaluation of models for predicting acidosis risk of barley grain in finishing beef cattle.** Uchenna Y. Anele<sup>1</sup>, Marylou Swift<sup>2</sup>, Tim A. McAllister<sup>1</sup>, and Wenzhu Yang\*<sup>1</sup>, <sup>1</sup>Lethbridge Research Centre, Agriculture and Agri-Food Canada, Lethbridge, AB, Canada, <sup>2</sup>Alberta Agriculture & Rural Development, Lethbridge, Lethbridge, AB, Canada.

A model to predict acidotic risk of barley grain was developed from a batch culture consisting of 50 barley samples varying in bulk density (BD), processing method (standard vs. precision processing: sieving grains into large versus small kernels and rolling based on kernel size), processing index (BD after rolling/BD before rolling), processing (ground vs. dry-rolled), geographical and agronomic origin. The objective of this study was to develop a model that can predict the relative 'hotness' of individual barley samples. Of all the independent variables (pH, starch content, dry matter disappearance (DMD), neutral detergent fiber, acid detergent fiber, in vitro gas kinetics, total and molar proportions of individual short-chain fatty acids at different incubation times) considered, dry matter disappearance at 6 h of incubation (DMD6) accounted for 90.5% of the variation in acidosis index with a root mean square error (RMSE) of 4.46%. When the new model ( $-0.7826 + 2.5536 \times \text{DMD6}$ ) was applied to 3 independent data sets to predict acidosis, it accounted for 33.4, 90.9 and 25.6% of the variation in calculated acidosis index. Significant ( $P < 0.01$ ) mean bias was evident in 2 of the data sets and it under-predicted acidosis index by 26.1 and 5.35%. There were marked similarities in the acidosis index ranking of barley samples by the models as shown by the result of a correlation analysis between calculated and predicted acidosis index ( $R^2 = 0.67$ ,  $P < 0.01$ ). We observed variations in the acidosis index ranking of samples that were processed differently (processing index of 75 versus 85% and precision processing versus control). Results suggest that our model

which is based on DMD6 has the potential to predict acidosis risk and can rank different barley samples based on their acidotic risk; however, the model would benefit from further refinement.

**Key Words:** acidosis, barley, prediction

**T350 Influence of growth-promoting implants on endocrine factors and efficiency of replacement beef heifers.** Laura R. Meyer\*<sup>1</sup>, Thomas L. Devine<sup>1</sup>, Michael L. Looper<sup>1</sup>, Dirk Philipp<sup>1</sup>, Donald S. Hubbell III<sup>2</sup>, Rick W. Rorie<sup>1</sup>, and Charles F. Rosenkrans Jr.<sup>1</sup>, <sup>1</sup>Department of Animal Science, Division of Agriculture, University of Arkansas, Fayetteville, AR, <sup>2</sup>Livestock and Forestry Research Station, Division of Agriculture, University of Arkansas, Batesville, AR.

Anabolic steroid implants are commonly used in the beef industry to increase BW gain in feedyard cattle. However, there are a limited number of implants approved for use in replacement heifers. Objectives were to use growth-promoting implants to add value to low-BW heifers, and to determine the relationships among hormones and cow efficiency. Charolais  $\times$  Balancer heifers ( $n = 65$ ;  $179 \pm 30$  kg;  $255 \pm 12$  d of age) were used to determine the influence of androgenic and/or estrogenic implants on serum concentrations of insulin-like growth factor-I (IGF-I), prolactin (PRL), cortisol (CORT), and reproduction. Heifers were blocked by BW and assigned to 1 of 4 implant treatment groups: 1) control, no implant (CON;  $n = 16$ ); 2) trenbolone acetate (TBA; 200 mg of TBA;  $n = 15$ ); 3) trenbolone acetate plus estradiol (TBA+E2; 40 mg TBA and 8 mg E2;  $n = 17$ ); or 4) zeranol (ZER; 36 mg ZER;  $n = 17$ ). Heifers were implanted on d 0, and blood samples collected on d 0 (15 Nov), 106 (1 Mar), and 195 (29 May) of the experiment. Heifers were AI on d 220 and exposed to an Angus bull for 28 d starting 12 d after AI. Calf birth date, and birth weight were recorded. At weaning, dam and calf weights were determined and cow efficiency was calculated by dividing calf adjusted 205 d weight by dam weight at weaning. Treatment did not influence ( $P > 0.10$ ) calving rate (41.5%), Julian calving date ( $96.8 \pm 2.5$  d), cow efficiency ( $0.47 \pm 0.23$ ), concentrations of IGF-I ( $78.4 \pm 3.9$  ng/mL), PRL ( $83.5 \pm 5.81$  ng/mL), CORT ( $36.3 \pm 9.9$  ng/mL), or PRL:CORT (ratio =  $2.19 \pm 0.24$ ). Serum PRL and IGF-I increased ( $P < 0.01$ ) from d 0 to 195. Serum cortisol increased ( $P < 0.01$ ) from d 0 to 106, and concentrations were similar from d 106 to 195. Multiple regression analyses indicated PRL and IGF-I at d 195 described 30% of variation in adjusted 205 d calf weights; 35% of variation in cow weight at weaning was explained by PRL at d 0 and 106; and PRL:CORT at d 0 and IGF-I at d 195 accounted for 44% of variation in cow efficiency. Anabolic implants did not affect heifer reproduction. Serum PRL, CORT, and IGF-I of heifers at weaning, yearling, and pre-breeding may serve as biomarkers of cow efficiency.

**Key Words:** cortisol, IGF-I, prolactin



## Ruminant Nutrition: Beef II

**T351 Effect of monensin inclusion on intake and digestion in *Bos indicus* and *Bos taurus* steers consuming bermudagrass hay.** Natasha L. Bell\*<sup>1,2</sup>, Todd R. Callaway<sup>3</sup>, Robin C. Anderson<sup>3</sup>, Marcia O. Franco<sup>4</sup>, and Tryon A. Wickersham<sup>1</sup>, <sup>1</sup>Texas A&M University, College Station, TX, <sup>2</sup>Texas A&M University-Kingsville, Kingsville, TX, <sup>3</sup>Southern Plains Agricultural Research Center, Agricultural Research Service, USDA, College Station, TX, <sup>4</sup>Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.

Effects of monensin inclusion and the subspecies of cattle on utilization of bermudagrass hay (13.7% CP) were evaluated using ruminally cannulated steers (5 *Bos indicus*, BI and 5 *Bos taurus*, BT; 398 kg BW). Subspecies were concurrently subjected to a 2 period, 2 treatment cross-over design. Treatments consisted of 0 (CON) or 200 (MON) mg·hd<sup>-1</sup> monensin (Rumensin 90; Elanco Animal Health, Greenfield, IN) fed daily in 0.91 kg DDGS. Steers were group housed during adaptation periods and moved to individual covered pens to facilitate sampling. Periods were 70 d in length: 20 d adaptation, 5 d sample collection, 17 d continuation of treatment application (for ruminal sampling), and 28 d withdrawal between periods. Hay, ort, and fecal grab samples were collected d 21–25 for determination of intake and digestion. Data were analyzed using the MIXED procedure of SAS 9.3 (SAS Inst. Inc., Cary, NC) with terms in the model including treatment, subspecies, subspecies × treatment and period, with animal as a random effect. No subspecies × treatment interactions were observed ( $P \geq 0.21$ ). Monensin tended to increase forage OM intake (FOMI;  $P = 0.08$ ) from 19.5 to 20.3 g/kg BW and total OM intake (TOMI;  $P = 0.07$ ) from 21.3 to 22.2 g/kg BW. No effect of subspecies ( $P \geq 0.16$ ) was observed for FOMI or TOMI. Organic matter digestibility was not affected by monensin ( $P = 0.97$ ; 60.6 and 60.7 for CON and MON, respectively) and was not different ( $P = 0.28$ ) between subspecies (58.0 and 63.3% for BI and BT, respectively). When the combined effects of TOMI and OMD were evaluated as total digestible OM intake (TDOMI), no response ( $P = 0.71$ ) was observed from monensin supplementation. *Bos taurus* steers had greater ( $P = 0.04$ ) TDOMI than BI steers (14.3 vs 12.2 g/kg BW). Monensin tended to increase forage NDF intake (FNDFI;  $P = 0.07$ ) from 16.5 to 17.2 g/kg BW and total NDF intake (TNDFI;  $P = 0.08$ ) from 17.4 to 18.1 g/kg BW, but had no effect ( $P \geq 0.73$ ) on NDF digestibility (NDFD; 66.4 and 66.0% for CON and MON, respectively) or total digestible NDF intake (TDNDFI; 11.7 and 11.9 g/kg BW for CON and MON, respectively). Although FNDFI, TNDFI, and NDFD were not different ( $P \geq 0.16$ ) between subspecies, total digestible NDF intake was greater ( $P = 0.02$ ) for BT than BI steers (12.7 vs 10.9 g/kg BW). Overall, monensin increased FOMI and FNDFI and had no effect on OMD or NDFD. *Bos taurus* had greater TDOMI and TDNDFI compared with BI steers.

**Key Words:** cattle subspecies, ionophore, digestibility

**T352 Effect of rumen protected B vitamins supplementation during the receiving period on the productive performance of beef cattle.** Helene Leclerc\*<sup>1</sup>, Diana A. Espinosa<sup>2</sup>, Essi Evans<sup>3</sup>, Roberto Zambrano Gaytan<sup>2</sup>, and Juan de Dios Garza Flores<sup>2</sup>, <sup>1</sup>Jefo Nutrition, St-Hyacinthe, QC, Canada, <sup>2</sup>Rancho El 17, Hermosillo, Sonora, Mexico, <sup>3</sup>Technical Advisory Services, Bowmanville, ON, Canada.

A study was conducted to evaluate the performance of beef cattle given a rumen protected B vitamins blend during a 21-d receiving period.

Twelve loads of cattle (899 head) were equally split based on weight and shrink and were assigned to 15 pens/treatment. The test treatment consisted of 2g/head/day of rumen protected B vitamins (folic acid, pyridoxine, pantothenic acid and biotin; Jefo Nutrition, St-Hyacinthe, Quebec, Canada). No added B vitamins were given to control cattle, and a common diet was provided to both groups. Receiving ration consisted of 38% alfalfa hay, 30% steam flaked corn, 10% corn distillers grain, 2% soybean meal, 11% sugar cane molasses, 3% wheat straw and 6% minerals and vitamins. Animals were weighted individually on d 1 and d 21 of the trial to calculate total and daily gain weights. Animals were fed twice daily. Daily pen feed intakes were measured. All data were analyzed as a randomized complete block, using pens as experimental units. The general linear model included treatment as fixed and block as random effects. Days to recover shrink were reduced (15.36 vs. 16.63 d;  $P < 0.05$ ), for the group of cattle fed the protected B vitamins blend. No significant difference was observed between the control and treatment group, for morbidity status (2.16 vs. 1.52%) and mortality rate (0.43 vs. 0.87%). The inclusion of the protected B vitamins blend in the diet significantly improved total gain weight (46.26 vs. 42.40 kg;  $P < 0.05$ ), average daily gain (2.20 vs. 2.01 kg;  $P < 0.05$ ), and feed/gain (3.90 vs. 4.33;  $P < 0.05$ ); without affecting average daily feed intake. It is concluded that addition of the protected B vitamins blend in the diet improves the productivity of beef cattle during the receiving period, without affecting feed intake and the morbidity rate.

**Key Words:** protected B vitamins, beef cattle, receiving

**T353 Effects of including virginiamycin in feedlot diets containing monensin under commercial conditions in Mexico.** Jorge R. Kawas<sup>2</sup>, Rene Alvarado<sup>2</sup>, Milton A. Gorocica-Buenfil\*<sup>1</sup>, and Francis L. Fluharty<sup>3</sup>, <sup>1</sup>Phibro Animal Health de Mexico, Queretaro, Qro. Mexico, <sup>2</sup>MNA de Mexico, San Nicolas de los Garza, NL, Mexico, <sup>3</sup>The Ohio State University, Wooster, OH.

A trial was conducted in a commercial feedlot in northeastern Mexico to determine the effects of supplemental virginiamycin (Vm) in combination with monensin (Mn) on finishing cattle performance. Upon arrival, 2,256 crossbred heifers (BW = 276.0 ± 3.33 kg) were dewormed, vaccinated against respiratory and clostridial pathogens and implanted with a trenbolone acetate (TBA) implant (200 mg TBA + 20 mg estradiol benzoate). At processing, animals were allotted to 22 pens (approx. 100/pen) using a randomized complete block design. Two treatment groups were evaluated, both receiving a high-concentrate (+90%) diet. One treatment group (MN) had the basal diet supplemented with 400 mg/hd/d of Mn; the other treatment group (VM) diet was supplemented with 250 mg/hd/d of Vm and 300 mg/hd/d of Mn. Cattle were gradually adapted to their final diet (15.5% protein, 1.51 Mcal NEg/kg) over a 21–28 d period. When cattle had 44 d on feed (DOF), they were reimplanted with the same TBA implant. Zilpaterol chlorhydrate was provided to all cattle at 0.15 mg/kg BW for 30 d and was withdrawn 4 d before harvest. Cattle were harvested after 100 DOF. At harvest, HCW was recorded. Data were analyzed using the PROC MIXED analysis of SAS for a randomized complete block design. Pen was used as the experimental unit and block was included in the model as a random factor. After 44 DOF when cattle were reimplanted, the VM group had greater ADG (1.51 vs. 1.38;  $P < 0.05$ ). At harvest, total ADG was 6% greater in the VM group (1.40 vs. 1.32;  $P < 0.05$ ), and G:F was improved 7% in the VM group (0.179 vs. 0.167;  $P < 0.05$ ). Hot carcass weight was 4.9 kg greater ( $P < 0.01$ ) in the VM group than in the MN group (251.7 kg vs.

246.8 kg). These results agree with previous research in commercial feedlots in Mexico and Brazil. Under the parameters of this study, virginiamycin inclusion in a feedlot diet containing monensin improved feedlot performance and carcass weight.

**Key Words:** feedlot, virginiamycin, performance

**T354 Growth performance of yellow cattle in southern China weaned at different ages.** C. Wang\*<sup>1</sup>, Y. F. Xia<sup>1</sup>, H. L. Mao<sup>1</sup>, Y. Tu<sup>2</sup>, C. G. Jiang<sup>2</sup>, H. F. Wang<sup>1</sup>, Q. Y. Diao<sup>2</sup>, and D. X. Ren<sup>3</sup>, <sup>1</sup>*College of Animal Science and Technology, Zhejiang A & F University, Hangzhou-Lin'an, Zhejiang Province, China*, <sup>2</sup>*Feed Research Institute, Chinese Academy of Agricultural Sciences, Beijing, China*, <sup>3</sup>*Institute of Dairy Science, Zhejiang University, Hangzhou, Zhejiang Province, China*.

This study was conducted to examine the effects of weaning ages on performance of a small-type yellow cattle in southern China. Thirty female calves were selected according to birth weight, and stratified into 10 blocks of 3 and allocated within block to 3 treatments that the calf was considered weaned when it consumed 1000 (W1000), 750 (W750), or 500 (W500) g of starter for 2 consecutive days. All calves had colostrum within 4 h after born, and then were fed milk up to 3 L/d until d 7. Liquid feed composed of half whole milk and half milk replacer (MR, 93.2% DM, 23.9% CP, and 13.1% EE) were fed to calves at 3L/d until d 13 and 3 L/d MR for the remainder of the study. Starter (17.9% CP, 10.9 MJ/kg ME) was fed ad libitum from d 10, and grass hay and water were available ad libitum. Milk, MR, starter, and hay intakes were recorded daily. All Calves were weighted and body height, body length, circumference of cannon bone were measured after birth and every 30 d until d 150. Data except for weaning age and BW change were analyzed using the Proc MIXED of SAS while weaning age and BW change were analyzed using the Proc GLM of SAS. The results showed that average weaning age was  $49 \pm 4.8$ ,  $58 \pm 4.4$ , and  $63 \pm 4.6$  for W500, W750, and W1000, respectively. Calves in W500 and W750 had higher starter intake than that of W1000 ( $P < 0.05$ ). The body measurements were not significant different between groups ( $P > 0.05$ ), however, calves in W1000 tend to have a low body height and body length ( $0.05 < P < 0.15$ ). There was no significant difference in BW and ADG on d 30 when any of the calves was weaned ( $P > 0.05$ ). On d 60, BW of calves in W1000 was higher than that of W500 ( $P < 0.05$ ) and tend to be higher than that of W750 ( $P = 0.10$ ). However, calves in W1000 lost their BW advantage on d 90. Calves in W750 and W500 had higher ADG from 1 to 150 d compared with that of W1000. It is concluded that starter-dependent weaning can help to enhance the adaptation to the solid feed with advantages on weight gain and feed intake.

**Key Words:** weaning, yellow cattle, growth performance

**T355 Decreasing the frequency of energy supplementation to beef steers impairs growth and antibody production against bovine viral diarrhoea virus 1b.** Luis F. Artioli\*<sup>1</sup>, Philippe Moriel<sup>1</sup>, Rodrigo S. Marques<sup>2</sup>, and Reinaldo Cooke<sup>2</sup>, <sup>1</sup>*North Carolina State University, Waynesville, NC*, <sup>2</sup>*Oregon State University, Burns, OR*.

We evaluated the effects of decreasing the frequency of energy supplementation on growth performance and measurements of innate and humoral immune response of preconditioning beef steers. At weaning (d -7), Angus steers (n = 24; BW =  $221 \pm 31$  kg; age =  $177 \pm 19$  d) were allocated to a single drylot pen with free-choice access to fescue hay. On d 0, steers were stratified by BW and age, and randomly assigned to 1 of 8 drylot pens (3 steers/pen). Steers were provided daily ad libitum access

to ground fescue hay (17% CP and 58% TDN; DM basis) and similar weekly concentrate offer (12 kg/steer) from d 0 to 42. Treatments were randomly assigned to pens (4 pens/treatment) and consisted of dividing and providing the weekly concentrate offer either 3 (S3; 3.8 kg on Mondays, Wednesdays and Fridays) or 7 times (S7; 1.7 kg daily) per week. Supplements included 50% soyhulls and 50% corn gluten feed (17% CP and 72% TDN; DM basis). Steers were vaccinated using the SelectVAC protocol (Zoetis) on d 7 and 21. Shrunken BW was obtained on d 0 and 42, and blood samples from jugular vein collected on d -7, 0, 7, 8, 9, 10, 14, 21, 22, 23, 24, 28, 35 and 42. Although BW did not differ from d 0 to 42 ( $P \geq 0.33$ ), S7 steers had greater mean ADG ( $1.3$  vs.  $1.0 \pm 0.07$  kg/d;  $P = 0.01$ ), mean hay DMI ( $2.8$  vs.  $2.2 \pm 0.08$  kg/d;  $P < 0.01$ ) and total DMI from d 0 to 42 ( $190$  vs.  $165 \pm 4$  kg;  $P = 0.02$ ), but similar G:F compared with S3 steers ( $P = 0.14$ ;  $0.29$  vs.  $0.26$ ). Plasma concentrations of glucose, insulin, cortisol and ceruloplasmin did not differ ( $P \geq 0.33$ ) between treatments. However, S3 steers had greater plasma haptoglobin concentrations on d 8 ( $1.95$  vs.  $1.20 \pm 0.13$  mg/mL;  $P < 0.01$ ) and 10 ( $1.39$  vs.  $0.84 \pm 0.13$  mg/mL;  $P < 0.01$ ), and less mean serum bovine viral diarrhoea virus-1b (BVDV1b) titers than S7 steers ( $1.5$  vs.  $2.5 \pm 0.31$  log<sub>2</sub> base;  $P = 0.03$ ). Therefore, decreasing the frequency of energy supplementation from 7 to 3 times weekly during a 42-d preconditioning period decreased hay and total DMI, growth and antibody production against serum BVDV-1b, and increased plasma haptoglobin concentrations of beef steers.

**Key Words:** supplementation frequency, immune, steer

**T356 Pre- and postpartum herbage allowances of grasslands affected productive and reproductive performances of primiparous beef cows.** Mariana Carriquiry\*<sup>1</sup>, Martín Claramunt<sup>2</sup>, Ana L. Astessiano<sup>1</sup>, and Pablo M. Soca<sup>3</sup>, <sup>1</sup>*Facultad de Agronomía, Montevideo, Uruguay*, <sup>2</sup>*Facultad de Veterinaria, Paysandú, Uruguay*, <sup>3</sup>*Facultad de Agronomía, Paysandú, Uruguay*.

The aim of this work was to evaluate the effect of herbage allowance of grasslands (Campo biome) during the prepartum and postpartum on productive and reproductive responses of primiparous beef cows. Fifty-four Hereford cows ( $5.9 \pm 0.5$  BCS in a 1–8 scale and  $472 \pm 35$  kg BW) were used in a randomized block design with 2 spatial replications and a factorial arrangement of prepartum (fall; high vs. low, PREH vs. PREL) and postpartum (spring-summer; high vs. low, POSTH vs. POSTL) herbage allowance (4 vs. 2.5 kgDM/kgBW of annual mean for high vs. low). Cows were in a continuous grazing system. Cow BW, BCS, and calf BW were determined monthly and at calving and milk production and composition were determined using a milking machine at 130 d postpartum. Calving to conception interval (CCI) and early (during first month of mating period) and total pregnancy were registered using ultrasound. Means from a mixed model were considered to differ when  $P \leq 0.05$ . Cow BW and BCS were greater from mid-winter to the end of summer (last third of gestation to weaning) for PREH than PREL cows and during the postpartum for POSTH than POSTL cows. Milk energy output was between 1.2 and  $1.6 \pm 0.8$  Mcal NEL/d greater for PREH than PREL cows and for POSTH than POSTL cows. Calf BW did not differ at birth but at weaning was  $30 \pm 5$  kg greater for POSTH than POSTL cows. The CCI tended ( $P = 0.10$ ) to be shorter for PREH than PREL cows ( $129$  vs.  $139 \pm 6$  d) and was shorter for POSTH than POSTL cows ( $126$  vs.  $141 \pm 6$  d). Early pregnancy was greater for POSTH than POSTL cows ( $54$  vs.  $39\%$ ) while total pregnancy did not differ among treatments (averaged 77%). Cow BCS at calving affected CCI, early and total pregnancy (1 unit of BCS at calving decreased CCI by 21 d and increased early and total pregnancy by 50%). Effects of PRE and

POST herbage allowances had an additive effect on productive and/or reproductive responses of primiparous beef cows grazing native pastures.

**Key Words:** beef cattle, rangeland, forage allowance

**T357 The effects of adding 3-nitrooxypropanol and monensin to a finishing diet on methane production using the rumen simulation technique (Rusitec).** A. Romero-Perez<sup>1,2</sup>, E. K. Okine<sup>2</sup>, L. L. Guan<sup>2</sup>, S. M. Duval<sup>3</sup>, M. Kindermann<sup>4</sup>, and K. A. Beauchemin<sup>1</sup>, <sup>1</sup>Agriculture and Agri-Food Canada, Lethbridge Research Centre, Lethbridge, AB, Canada, <sup>2</sup>Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB, Canada, <sup>3</sup>DSM Nutritional Products France, Research Centre for Animal Nutrition and Health, Saint Louis Cedex, France, <sup>4</sup>DSM Nutritional Products, Basel, Switzerland.

3-Nitrooxypropanol (NOP), an enzymatic inhibitor that has consistently reduced methane (CH<sub>4</sub>) emissions in sheep, dairy and beef cattle (up to 59% reduction on a long-term basis), and monensin (MON), an ionophore that has a moderate, and sometimes transitory, effect on CH<sub>4</sub> reduction were used in this study. The objective was to evaluate the effect of NOP, MON and the combination of both on CH<sub>4</sub> production when added to a finishing diet (85% barley grain, 10% barley silage, and 5% vitamin-mineral supplement; DM basis) using Rusitec fermenters. Two Rusitec apparatuses each equipped with 8 fermenters were used in a completely randomized block design with 2 blocks (apparatus) and 4 treatments: Control, NOP (2 mg), MON (2 mg) and NOP+MON (2 mg+2 mg). Within each apparatus, 2 fermenters were randomly assigned to a treatment. Treatments were supplied daily with 10 g of diet. The experiment included an adaptation period without treatment supplementation (8 d), a treatment period (7 d), and a recovery period where treatments were discontinued (3 d). During the treatment period, DM digestibility was not affected. Total VFA and molar proportion of propionate and butyrate were not affected ( $P > 0.05$ ) but acetate was reduced ( $P < 0.01$ ) with addition of NOP (8.3%) and NOP+MON (14.9%). Methane production was reduced ( $P < 0.01$ ) by 69.4 and 68.2% with NOP and NOP+MON respectively, while H<sub>2</sub> production was increased ( $P < 0.01$ ) by 76.7 and 75.2% respectively for the same treatments compared with Control. Treatments had no effect on the copy number of the 16S rRNA gene for total bacteria ( $P > 0.21$ ); however, that for methanogens was reduced ( $P < 0.01$ ) with NOP and NOP+MON treatments. During the recovery period on d 18, no effect ( $P > 0.20$ ) was observed for CH<sub>4</sub> and H<sub>2</sub> production; however, a trend for lower acetate concentration with NOP and NOP+MON was still observed ( $P = 0.08$ ). Treatments including NOP were effective in reducing CH<sub>4</sub> production in vitro using a finishing diet with a concomitant increase in H<sub>2</sub> production; however, the combination of NOP and MON did not lead to greater CH<sub>4</sub> reduction than NOP alone.

**Key Words:** 3-nitrooxypropanol, monensin, methane

**T358 Nutritional performance and metabolic characteristics of cattle fed tropical forage with nitrogen and starch supplementation.** Marcia de Oliveira Franco\*, Edenio Detmann, Alexandre Ribeiro Lopes, Luana Marta de Almeida Rufino, and Erick Darlison Batista, *Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.*

Effects of nitrogen supplementation with and without starch supplementation on nutritional performance and metabolic characteristics of cattle fed low and medium-quality tropical forages were evaluated using ruminal and abomasal cannulated steers. Four European × Zebu

bulls (381 kg BW) were distributed according to a 4 × 4 Latin square. Each experimental period of 28 d was divided into 2 sub-periods: in the first sub-period 2 animals received low-quality hay and 2 animals received medium-quality hay (*Brachiaria decumbens*); the following supplementation schemes were evaluated in the second sub-period: low-quality hay with nitrogen supplementation (300 g CP/d); low-quality hay with nitrogen (300 g CP/d) and starch (225 g/d) supplementation; medium-quality hay with nitrogen supplementation (300 g CP/d); and medium-quality hay with nitrogen (300 g CP/d) and starch (225 g/d) supplementation. In the absence of supplementation, the medium-quality forage provided higher ( $P < 0.05$ ) intake, digestibility, nitrogen balance (NB), and efficiency of nitrogen utilization (EFNU). The greater performance of animals fed medium-quality forage was attributed to anabolic stimuli, supported by higher ( $P < 0.05$ ) serum concentration of IGF1. Comparing sub-periods, the supply of supplements depressed ( $P < 0.05$ ) medium-quality forage intake, but did not affect ( $P > 0.05$ ) low-quality forage intake. There was no effect ( $P > 0.05$ ) of supplementation on NDF digestibility as compared with the sub-period without supplementation. However, comparison between supplements indicated depression ( $P < 0.05$ ) in NDF digestibility when starch was included. Supplementation increased ( $P < 0.05$ ) NB, EFNU, and serum concentration of IGF1 in animals fed low-quality forage. Nitrogen supplementation increases nitrogen retention in animals, an effect attributed mainly to anabolic stimuli. However, this effect is more prominent when animals are fed low-quality forages. No positive impact on animal metabolism was obtained with the combination of supplemental nitrogen and starch.

**Key Words:** *Brachiaria decumbens*, digestibility, nitrogen balance

**T359 Nutritional performance and metabolic characteristics of cattle fed low-quality tropical forage and supplemented with nitrogen associated with different starch proportions.** Marcia de Oliveira Franco\*, Edenio Detmann, Marcilia Medrado Barbosa, Gabriel Cipriano Rocha, and Claudia Batista Sampaio, *Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.*

Effects of nitrogen supplementation associated with different starch proportions on nutritional performance and metabolic characteristics of cattle fed low-quality tropical forage (*Brachiaria decumbens* hay, 7.4% CP) were evaluated using ruminal and abomasal cannulated steers. Five European × Zebu young bulls (186 kg BW) were distributed according to a 5 × 5 Latin square. The following treatments were evaluated: control (only forage), supplementation with 300 g CP/d (0:1), supplementation with 300 g starch/d and 300 g CP/d (1:1), supplementation with 600 g starch/d and 300 g CP/d (2:1), and supplementation with 900 g starch/d and 300 g CP/d (3:1). Supplements increased ( $P < 0.05$ ) DM intake, but did not affect ( $P > 0.05$ ) forage intake. There was a cubic effect ( $P < 0.05$ ) of starch, which was attributed to the highest forage intake (g/kg BW) when using the 2:1 starch:CP ratio. Supplements increased ( $P < 0.05$ ) OM digestibility, but did not affect ( $P > 0.05$ ) NDF digestibility. There was a positive linear effect ( $P < 0.05$ ) of the amount of starch supplemented on OM digestibility. Total NDF digestibility was not affected ( $P > 0.05$ ) by the amount of starch supplemented. Ruminal ammonia nitrogen concentrations were higher ( $P < 0.05$ ) in supplemented animals, however, a negative linear effect ( $P < 0.05$ ) of amount of starch was observed. Supplements increased ( $P < 0.05$ ) the nitrogen balance (NB) and efficiency of nitrogen utilization. These effects were attributed to increased body anabolism, supported by higher ( $P < 0.05$ ) serum concentration of IGF1. Increasing the amount of starch tended to linearly increase the NB ( $P < 0.06$ ) and serum concentration of IGF1 ( $P < 0.05$ ). However, comparison of NB means between treatments with supplementation showed a higher value for the 2:1 starch:CP ratio.



Nitrogen supplementation in cattle fed low-quality tropical forage increases nitrogen retention in the animal body. Additional supply of starch increases nitrogen retention by increasing energy availability for both rumen and animal metabolism.

**Key Words:** fiber, nitrogen balance, Zebu

**T360 Effect of rumen protected carbohydrate supplementation on performance in feedlot finishing steers during heat stress.** Juan P. Russi<sup>1,3</sup>, Patricio Davies<sup>4</sup>, Nicolas DiLorenzo<sup>2</sup>, and Alejandro E. Relling<sup>1</sup>, <sup>1</sup>Facultad de Cs Veterinarias, UNLP, La Plata, Buenos Aires, Argentina, <sup>2</sup>University of Florida, Mariana, FL, <sup>3</sup>RUPCA LLC, Merced, CA, <sup>4</sup>INTA, Gral Villegas, Buenos Aires, Argentina.

Finishing steers during the summer can be challenging due to the effects of high temperatures and humidity on DMI. The objective of this study was to evaluate the inclusion of a rumen-protected carbohydrate (RUPCA; US Patent # 8,507,025) on performance of finishing steers during heat stress. Temperature-humidity index average measured every day during the experiment was  $72 \pm 4.9$ . Crossbred steers ( $n = 135$ ;  $355 \pm 20$ kg) were used in a 62-d experiment. Steers were blocked by initial BW and placed into 15 pens. Steers within blocks were randomly assigned to 3 treatments: T0 = fed 91.4% of a basal diet (% DM), 22.3% corn silage, 65.9% dry corn, 0.6% sunflower meal, 0.5% urea, 2% minerals and vitamins and 8.6% of a supplement containing (% DM) 58.1% soybean meal, 38.9% soluble carbohydrates, 2% urea and 1% minerals salts; T1 = fed the basal diet plus 4.3% supplement and 4.3% RUPCA; and T2 = fed basal diet plus 8.6% RUPCA. The supplement and RUPCA consisted of the same ingredients, differing on the processing of the carbohydrate (i.e., protected or not from ruminal degradation). Body weight was measured on d 0, 15, 39 and 62 relative to the beginning of treatments feeding (d 0). Pen DMI was measured on d 10, 18, 25, 31, 35, 46, 51, 56, 60 and 62. Back-fat on the 12th rib (BF) and LM area were measured on d 1 and 62. Data were analyzed as a randomized complete block design with repeated measures using a mixed model of SAS. Initial BW was used as a covariate. There were no differences between treatments on final BW, BF or LM area on d 62 ( $P > 0.10$ ). Treatment  $\times$  day interactions were observed for G:F, ADG and DMI ( $P < 0.05$ ) suggesting a different response to treatments during periods of heat stress (Table 1). Feeding RUPCA may be beneficial for finishing steers under heat stress.

**Table 1 (Abstr. T360).**

Item	Treatment			SEM	P-value		
	T0	T1	T2		Trt	Day	Trt $\times$ Day
DMI, kg/d	9.9 <sup>ab</sup>	9.8 <sup>a</sup>	10.0 <sup>b</sup>	0.07	0.04	<0.001	0.001
Initial BW, kg	287	285	285	0.6	0.23	—	—
Final BW, kg	352	357	353	3.1	0.51	—	—
ADG, kg	1.00	1.11	1.07	0.065	0.38	<0.001	0.039
G:F	0.105	0.116	0.109	0.0051	0.32	<0.001	0.004
Backfat 12th rib (62 d), mm	0.58	0.61	0.60	0.017	0.68	—	—
LM area (62 d), cm <sup>2</sup>	57.6	54.9	56.1	0.97	0.30	—	—

<sup>ab</sup>Means without common superscript differ ( $P < 0.05$ ).

**Key Words:** carbohydrate, rumen, bypass energy

**T361 Partially replacing corn with glycerin increases total VFA, propionate, and ruminal NH<sub>3</sub>-N concentrations in finish-ing beef diets evaluated in a dual-flow continuous culture system.** Pedro Del Bianco Benedeti<sup>1,2</sup>, Lorryny Galoro da Silva<sup>1</sup>, Eduardo Marostegan de Paula<sup>1</sup>, Teshome Shenkoru<sup>1</sup>, Hugo Monteiro<sup>1</sup>, Brad Amorati<sup>1</sup>, Yehling Yeah<sup>1</sup>, Marcos Marcondes<sup>2</sup>, and Antonio Faciola<sup>1</sup>, <sup>1</sup>University of Nevada, Reno, NV, <sup>2</sup>Federal University of Viçosa, Viçosa, MG, Brazil.

Glycerin, the main biodiesel production by-product, has the potential to partially replace corn as an energy source for cattle. The objective of this study was to evaluate the effects of partially replacing dry ground corn (DGC) with glycerin on ruminal fermentation using a dual-flow continuous culture system. Six fermenters were used in a replicated 3x3 latin square arrangement with 3 periods of 10 d each, with 7 d for diet adaptation and 3-d for sample collections. Three dietary inclusion levels of glycerin were tested (0, 15, and 30% on DM basis). All diets contained 75% concentrate and were formulated to meet NRC (2000) recommendations. Fermenters were fed 72 g of DM/d equally divided in 2 meals per day. Liquid and solid dilution rates were adjusted to 11 and 5.5%/h, respectively. On d 8, 9, and 10, samples of digesta effluent were collected for ruminal NH<sub>3</sub>-N and VFA analyses. Nutrient flow and digestibility, N balance, and microbial growth were also measured. Data were analyzed using the MIXED procedure in SAS. Ruminal traits are presented in Table 1. Concentrations of total VFA, propionate, and NH<sub>3</sub>-N increased linearly ( $P < 0.05$ ) and concentrations of acetate, butyrate, *iso*-valerate, and BCVFA, as well as the acetate: propionate ratio decreased linearly ( $P < 0.05$ ) as glycerin replaced DGC. These results suggest that partially replacing DGC with glycerin may change ruminal fermentation, increasing total VFA, propionate, and NH<sub>3</sub>-N concentrations, which may increase energy status and microbial yield in beef cattle.

**Table 1 (Abstr. T361).** Ruminal traits

Item	Glycerin (%)			SEM	P-value	
	0	15	30		L	Q
NH <sub>3</sub> -N, mg/100 mL	8.7	10.6	17.6	1.22	<0.01	NS
Total VFA, mM	113.0	122.9	124.8	1.76	<0.01	NS
Acetate, % of total VFA	59.0	49.0	40.1	2.13	<0.01	NS
Propionate, % of total VFA	22.3	35.4	47.8	2.54	<0.01	NS
Butyrate, % of total VFA	15.7	13.4	10.1	0.68	<0.01	NS
Valerate, % of total VFA	1.18	0.96	1.02	0.05	NS	NS
Isovalerate, % of total VFA	1.43	0.97	0.58	0.10	<0.01	NS
Acetate:propionate	2.65	1.41	0.84	0.19	<0.01	NS
BCVFA, % of total VFA	2.98	2.62	2.03	0.31	<0.01	NS

**Key Words:** ammonia, glycerin, volatile fatty acids

**T362 Herbage allowance of grasslands during calf fetal and early life: Effects on body weight and composition.** Mariana Carriquiry<sup>1</sup>, Martín Claramunt<sup>2</sup>, Alberto Casal<sup>1</sup>, Ana L. Astessiano<sup>1</sup>, and Pablo M. Soca<sup>3</sup>, <sup>1</sup>Facultad de Agronomía, UdelaR, Montevideo, Uruguay, <sup>2</sup>Facultad de Veterinaria, UdelaR, Paysandú, Uruguay, <sup>3</sup>Facultad de Agronomía, Paysandú, Uruguay.

Fifty-four Hereford calves (31 males, 23 females) were used in a randomized block design (2 spatial replications) to evaluate the effect of herbage allowance of grasslands during their fetal and early life (130 d of gestation to weaning) on calf BW and body composition during the first year of age. Their dams (primiparous,  $5.9 \pm 0.5$  BCS 1–8 scale,  $472 \pm 35$  kg BW) were allocated in a factorial arrangement of parturum

(fall,  $130 \pm 12$  d of gestation to calving; high vs. low, PREH vs. PREL) and postpartum (spring-summer, birth to weaning; high vs. low, POSTH vs. POSTL) herbage allowance (4 vs. 2.5 kgDM/kgBW of annual mean). Males were castrated at birth. Calf BW was determined at birth and monthly thereafter until the first year of age ( $390 \pm 12$  d). Body composition was determined at 200 (weaning) and 390 d using the urea dilution technique. Means from a mixed model repeated analysis were considered to differ when  $P \leq 0.05$ . Calf birth weight was not affected by PRE and was greater in male than female calves ( $33.7$  vs.  $32.5 \pm 0.2$  kg). Calf BW at 200 and 390 d or pre and postweaning average daily gains (ADG) were not affected by PRE but calf BW at 200 and 390 d was  $30 \pm 3$  kg greater in POSTH than POSTL calves due to a greater preweaning ADG ( $+152 \pm 0.02$  g/d). Carcass fat increased ( $10.7$  vs.  $14.2 \pm 1.2\%$ ) while water ( $62.4$  vs.  $60.1 \pm 0.7\%$ ) and protein ( $19.1$  vs.  $18.3 \pm 0.3\%$ ) decreased from 200 to 390 d. Body composition was affected by the interaction between PRE and POST as carcass fat was greater while water and protein percentages were less in PREL-POSTH than PREL-POSTL calves, being intermediate in PREH-POSTL and PREH-POSTH calves ( $10.3, 15.2, 12.2$  and  $12.6 \pm 1.2\%$  for fat,  $62.7, 59.6, 61.6$  and  $61.0 \pm 0.7\%$  for water and  $19.2, 18.2, 18.8$  and  $18.8 \pm 0.3\%$  for protein for PREL-POSTL, PREL-POSTH, POSTH-PREL, and POSTH-POSTH, respectively). Improve forage allowance during calf lactation period modified body composition of calves born from cows grazing low herbage allowance during gestation, as the increase in calf BW was associated with increased fat in detriment of protein deposition.

**Key Words:** beef cattle, rangeland, developmental programming

**T363 Effects of a standardized blend of phytochemicals on performance of beef cattle in two distinct dietary contexts using multiple trial analysis method.** Clementine Oguey<sup>1</sup> and Christian Bruneau<sup>\*2</sup>, <sup>1</sup>Pancosma SA, Le Grand Saconnex, GE, Switzerland, <sup>2</sup>Pancosma, St Hyacinthe, QC, Canada.

The optimization of performance of naturally fed beef cattle and the replacement of monensin (MON) are concerns constantly looked after by producers. Many phytochemicals have been reported to influence production efficiency of dairy and beef animals. Objective was to assess the effect of a standardized protected blend of cinnamaldehyde, eugenol and capsaicin oleoresin (XT, XTRACT Ruminant, code X60-7065, Pancosma) on performance of beef cattle in 2 distinct commercial contexts: on top of a blank diet or as a substitute to MON, by 2 multiple trial analyses. A first set of data regrouped 10 trials organized in 7 studies (1290 growing cattle; mean initial BW of 276 kg; mean duration of 88 d, mean XT dose of 1.1 g/hd/d). All trials reported side by side comparisons of an unsupplemented control void of ionophore to the inclusion of XT in beef cattle. A second set of data regrouped 4 trials organized in 4 studies, all reporting side-by-side comparisons of the use of MON vs. XT (1004 growing cattle; mean initial BW of 215 kg; mean duration of 85 d, mean XT dose of 950 mg/hd/d; mean MON dose of 265 mg/hd/d). Outcomes selected were DMI, ADG and feed to gain ration (F:G). For each set of data, data were analyzed using a mixed model with the TRIAL variable as a random effect and the TRT variable as a fixed effect. Mean values were calculated using the LSMEANS procedure of XLstat, weighting the data for the variance among trials. Results showed that when added on top to a blank diet, XT did not affect DMI of beef cattle (mean: 7.18 kg/hd/d,  $P = 1.0$ ), but increased ADG by 108 g/hd/d ( $P < 0.05$ ) and reduced F:G by 7.5% ( $P < 0.05$ ). The second analysis showed that XT numerically increased ADG and DMI compared with MON by respectively 2.4 and 4.7% ( $P = 0.2$ ) without altering F:G (mean 4.82 g/g,  $P = 0.7$ ). These findings are in line with available data on MON, and suggest that the addition

of XT on top of a basal diet improves performance of beef cattle and can be safely used to replace MON.

**Key Words:** phytochemical, beef cattle performance, multiple analysis

**T364 Nutrient intake and productive performance of beef cattle fed diets containing soybean, corn, or sorghum silages.**

Lilian Oliveira Rosa<sup>\*1</sup>, Odilon Gomes Pereira<sup>1</sup>, Karina Guimarães Ribeiro<sup>1</sup>, Sebastião de Campos Valadares Filho<sup>1</sup>, Stefanie Alvarenga Santos<sup>2</sup>, Rilene Ferreira Diniz Valadares<sup>2</sup>, and Andressa Fernanda Campos<sup>3</sup>, <sup>1</sup>Viçosa Federal University, Viçosa, Minas Gerais, Brazil, <sup>2</sup>Bahia Federal University, Salvador, Bahia, Brazil, <sup>3</sup>Sao Paulo State University, Jaboticabal, Sao Paulo, Brazil.

The aim of this work was to evaluate the nutrient intake and productive performance of Nelore cattle fed with diets containing soybean, corn, or sorghum silages as forage sources. The experimental diets was composed for: 1 - soybean silage (SS); 2 - corn silage (CS); 3 - sorghum silage (SOS); 4 - 50% SS:50% CS; and 5 - 50% SS:50% SOS, on dry matter basis. Diets consisted of 60% silage and 40% concentrate (corn, soybean meal, urea, and mineral mixture), formulated to be isonitrogenous (12% CP, DM basis). Forty-five crossbred Holstein-Zebu bulls none castrated, with initial live weight of 360 kg were allotted in a randomized blocks design with 5 treatments and 9 replicates. The animals were kept in individual pens of approximately 10 m<sup>2</sup>, with protected feeders and waterier. The experiment lasted 99 d, divided in 15 d of initial adaptation plus 3 periods of 28 d each. All data were analyzed using MIXED procedure of SAS and differences between means were determined using the DIFF, which differentiates means based on Tukey's test. Significance was declared at  $P < 0.05$ . The dry matter (DM) intake was affected by the silages ( $P < 0.01$ ), being SS silage that got lowest value among silages. The soybean silage (SS silage) provided lowest values of organic matter (OM), crude protein (CP), neutral detergent fiber (NDF) and non-fibrous carbohydrate (NFC) intakes, kg/d. The ether extract (EE) intake was highest in soybean silage, as only forage source or associated with CS or SOS. The maximum NDF intake, in % BW, was found to animals fed diets with sorghum silage as only forage source (0.88% BW) or associated with soybean silage (0.74% BW). The average daily gain (ADG) was lower in animals fed diets with soybean silage (0.433 kg/d) compared the others diets, that were not different statistically, with average to 1.163 kg/d. However, no effect of different silages ( $P > 0.05$ ) was observed on dressing percentage, on average 54.5%. Our results suggest that soybean silage, as only forage source, results in lower animal performance. However, when this silage was associated with corn and sorghum silages, it has similar animal performance that of animals fed corn and sorghum silages as only forage sources.

**Key Words:** average daily gain, carcass dressing, forage source

**T365 Nutrient intake, total digestibility, ruminal pH, and ammonia concentration of beef cattle fed diets containing soybean, corn, or sorghum silages.**

Lilian Oliveira Rosa<sup>\*1</sup>, Odilon Gomes Pereira<sup>1</sup>, Karina Guimarães Ribeiro<sup>1</sup>, Sebastião de Campos Valadares Filho<sup>1</sup>, Stefanie Alvarenga Santos<sup>2</sup>, Rilene Ferreira Diniz Valadares<sup>2</sup>, and Andressa Fernanda Campos<sup>3</sup>, <sup>1</sup>Viçosa Federal University, Viçosa, Minas Gerais, Brazil, <sup>2</sup>Bahia Federal University, Salvador, Bahia, Brazil, <sup>3</sup>Sao Paulo State University, Jaboticabal, Sao Paulo, Brazil.

The aim of this work was to evaluate the nutrient intake, total digestibility and ruminal pH and ammonia concentration of beef cattle fed

diets containing soybean, corn, or sorghum silages as forage sources. The experimental diets was composed for: 1 - soybean silage (SS); 2 - corn silage (CS); 3 - sorghum silage (SOS); 4 - 50% SS:50% CS; and 5 - 50% SS:50% SOS, on dry matter basis. Diets consisted of 60% silage and 40% concentrate (corn, soybean meal, urea, and mineral mixture), formulated to be isonitrogenous (12% CP, DM basis). Five adult crossbred Holstein-Zebu bulls, male no castrated, rumen-cannulated with an initial weight of 442 kg were distributed in a 5 × 5 Latin square design. The animals were kept in individual pens of approximately 10 m<sup>2</sup>, with protected feeders and waterier. Effects of diets ( $P < 0.01$ ) were observed on the intakes of all nutrients, with the lowest intakes ( $P < 0.05$ ) of dry matter (DM), organic matter (OM), crude protein (CP), neutral detergent fiber (NDF) and neutral detergent fiber corrected to ash and protein (NDFap) being observed in the animals fed with soybean silage as only forage source. For TDN intake, the highest values were found for diets containing corn silage and it associated with soybean silage. For most nutrients, the highest digestibilities values were found with the diets containing corn silage as only forage source or when it was associated with soybean silage. There was effect to collection time ( $P < 0.05$ ) for ruminal pH (T, hours), adjusting quadratic model ( $Y = 6.0452 + 0.2648 \times T - 0.09955 \times T^2$ ,  $r^2 = 83.4\%$ ), but no effects were observed to diets (D) and collection times and diets interaction ( $T \times D$ ) for the same parameter. However, no effects were detected to diets (D), collection time (T) and collection time and diets interaction ( $T \times D$ ) for ruminal ammonia concentration, with average of 6.25 mg/dL. Our results suggest that soybean silage, as only forage source, results in lower nutrients intake. However, animals fed with this silage had similar ruminal parameters compared the others diets.

**Key Words:** dry matter intake, forage source, ruminal parameter

**T366 Enteric methane emissions in cattle fed diets containing sugar cane or corn silage.** Lays Mariz<sup>1,4</sup>, Stefanie Alvarenga Santos<sup>2</sup>, Laura Franco Prados<sup>1</sup>, Paloma de Melo Amaral<sup>\*1,4</sup>, Diego Zanetti<sup>1</sup>, Gustavo Chamon de Castro Menezes<sup>1</sup>, Sebastiao Valadares Filho<sup>1</sup>, Antonio Faciola<sup>4</sup>, and Luiz Gustavo Pereira<sup>3</sup>, <sup>1</sup>Federal University of Vicosa, Vicosa, MG, Brazil, <sup>2</sup>School of Veterinary Medicine and Animal Science of the Federal University of Bahia (UFBA), Salvador, BA, Brazil, <sup>3</sup>Embrapa Dairy Cattle, Juiz de Fora, MG, Brazil, <sup>4</sup>University of Nevada, Reno, NV.

The objective of this study was to evaluate enteric methane emissions in cattle fed diets containing sugar cane or corn silage. Five rumen-cannulated steers ( $336 \pm 16.6$  kg of initial BW) were used in a 5 × 5 Latin square arrangement. The study lasted 105 d and consisted of 5 periods of 21-d each. Animals were housed and fed individually. The 5 experimental diets contained 60% forage and 40% concentrate. Treatments consisted of 5 different forage sources: corn silage (CS), fresh sugar cane (FSC), regular sugar cane silage (SCS0%), sugar cane silage treated with 0.4% calcium oxide (SCS0.4%), and sugar cane silage treated with 0.8% calcium oxide (SCS0.8%). Forage CP levels were adjusted to 11% using a mixture of urea/ammonium sulfate (9:1). Sulfur hexafluoride tracer gas technique was used to measure methane emissions. Feed intake and refusals were measured daily and methane emissions were measured for 5 consecutive days. Data were analyzed using the MIXED procedure in SAS. Data is presented in Table 1. Animals fed CS had higher DMI ( $P < 0.01$ ) than animals fed sugar cane. Methane emissions when expressed in g/d or in g/kg of DMI did not change among treatments. However, when expressed in g/kg of digestible NDF (DNDF) was lower for CS diet ( $P = 0.05$ ). Methane energy loss did not change among diets; however, it was numerically lower for the CS diet. Treated SCS did not improve intake and did not reduce

methane emissions. The results from this study suggest that compared with sugar cane diets, corn silage diets may reduce methane emissions per unit of DNDF, which may lead to higher energy efficiency.

**Table 1 (Abstr. T366).** Mean values for intake and enteric methane emissions in steers

Item	Treatment					SEM	P-value
	CS	FSC	SCS 0%	SCS 0.4%	SCS 0.8%		
DMI (kg/d)	7.11 <sup>a</sup>	5.23 <sup>b</sup>	3.87 <sup>b</sup>	4.29 <sup>b</sup>	4.08 <sup>b</sup>	0.26	0.01
Methane emissions							
g/d	204.01	138.27	147.9	141.21	139.57	16.13	0.47
g/kg DMI	23.77	26.73	37.33	30.01	30.23	432.58	0.19
g/kg DNDF	164.37	298.47	201.43	169.17	220.82	14.69	0.05
Methane energy loss							
Gross energy intake (%)	7.08	7.78	10.93	8.79	9.12	0.67	0.22

**Key Words:** corn silage, sugar cane, methane emissions

**T367 Effects of oscillating dietary crude protein on nutrient intake, digestibility, performance, and carcass traits of finishing crossbred bulls in feedlot.** Paloma de Melo Amaral<sup>\*1,3</sup>, Stefanie Alvarenga Santos<sup>2</sup>, Laura Franco Prados<sup>1</sup>, Lays Mariz<sup>1,3</sup>, Lyvian Cardoso Alves<sup>1</sup>, Ana Clara Baiao Menezes<sup>1</sup>, Faider Alberto Castano Villadiego<sup>1</sup>, Flavia Adriane de Sales Silva<sup>1</sup>, Sebastiao Valadares Filho<sup>1</sup>, and Antonio Faciola<sup>3</sup>, <sup>1</sup>Federal University of Vicosa, Vicosa, MG, Brazil, <sup>2</sup>School of Veterinary Medicine and Animal Science of the Federal University of Bahia (UFBA), Salvador, BA, Brazil, <sup>3</sup>University of Nevada, Reno, NV.

Protein is a costly nutrient and excessive dietary N is an important environmental concern. The finishing period may offer the possibility of reducing dietary CP without negatively affecting beef-cattle production. This reduction may be achieved by oscillating dietary CP during the finishing period. Therefore, the goal of this study was to evaluate the effects of fixed or oscillating dietary CP on nutrient intake, digestibility, performance, and carcass traits of crossbred bulls in the finishing period. Twenty-four bulls ( $417 \pm 54$  kg of initial BW) were used in a complete randomized block design in a 2 × 2 factorial arrangement with 6 replications per treatment. The treatments were: Constant 11% CP (11–11), constant 13% CP (13–13), oscillating 11–13% CP (11–13), and oscillating 13–11% CP (13–11). The experiment lasted 72 d and oscillating treatments switched diets at d 37. Animals were housed and fed individually. Data were analyzed using the MIXED procedure in SAS and significance was declared at  $\alpha = 0.05$ . Partial data are presented in Table 1. There were no interactions ( $P > 0.05$ ) between fixed and oscillating CP level. There were no effects ( $P > 0.05$ ) of CP levels on intakes of DM, OM, NFC, and TDN. There were no effects ( $P > 0.05$ ) of CP levels on ADG or in carcass traits. We concluded that there were no benefits of oscillating dietary CP levels for crossbred bulls with ADG of approximately 2 kg/d in feedlots. There were no performance or carcass traits benefits of feeding more than 11% CP in the diets of crossbred bulls in feedlots.



**Table 1 (Abstr. T367).** Effects of constant or oscillating CP levels on intake, performance, and carcass traits

Item	Dietary CP				SEM	P-value		
	11-11%	11-13%	13-11%	13-13%		Pi	Pf	Pi × Pf
Intake, kg/d								
DM	12.1	11.8	10.6	11.3	1.5	0.14	0.80	0.46
OM	11.5	11.2	10.1	10.8	1.4	0.14	0.80	0.46
NFC	6.0	5.9	5.4	5.7	0.8	0.16	0.81	0.46
TDN	8.6	8.3	7.7	8.4	1.0	0.36	0.63	0.52
Performance, kg/d								
ADG	2.1	2.0	1.9	2.0	0.3	0.85	0.89	0.59
Carcass traits <sup>1</sup>								
HCDP, %	58.4	59.0	58.6	59.9	0.02	0.70	0.52	0.30
SFT, mm	2.9	3.1	2.8	3.1	1.12	0.86	0.86	0.66

<sup>1</sup>HCDP = hot carcass dressing percentage, SFT = subcutaneous fat thickness.

**Key Words:** oscillating protein, beef cattle, feedlot

**T368 Identification and removal of outliers in feed databases for beef cattle.** Huyen Tran<sup>\*1</sup>, William Weiss<sup>2</sup>, Galen Erickson<sup>3</sup>, and Phillip S. Miller<sup>3</sup>, <sup>1</sup>National Animal Nutrition Program, University of Kentucky, Lexington, KY, <sup>2</sup>The Ohio State University, Wooster, OH, <sup>3</sup>University of Nebraska, Lincoln, NE.

Accurate feed composition data are critical for diet formulation and determination of nutrient requirements of animals. Large feed databases are available; however, they often contain misidentified feed names and can have biased nutritive values. The first 2 objectives of this project were to identify and characterize outliers in feed databases and to develop feed composition tables for beef cattle. Approximately 1.5 million feed composition records provided by 3 commercial laboratories were sorted, screened, and reclassified. Histograms were used to visualize sample distribution. For most forages, feeds were classified as haylage or hay when DM <70 or ≥70, respectively. Grains were classified as high moisture (DM <80%) and dry grains (DM ≥80%). Any nutrient with a value outside mean ± 3.5 SD was removed (method A). Data were analyzed by laboratory before individual means and variance were weighted for sample size for calculation of the overall mean and SD. The third objective was to compare performance of method A to a combination of univariate and multivariate approaches for identifying outliers. Fifteen feeds were randomly selected representing grains, forages, byproducts, and oilseeds and screened for outliers. Feeds with missing key nutrients were removed. Principal component and clustering analyses of SAS were used in the multivariate approach. Among 1.5 million data classified as 352 feeds, 45.7% of the data seemed to be misidentified, leaving 196 feeds for analysis. Outliers were characterized as inaccurate DM classification, transformation of data, decimal point issues, erroneous data, or terminology inconsistency. Method A removed 1.4% of samples and decreased means by 0.9% and SD by 15.2% for CP. The multivariate analysis removed a larger percentage of samples (33.3%) and decreased means by 1.6% and SD by 38.8% for CP. Clustering analysis defined 8 of 15 feeds with >1 cluster. The multivariate method was powerful in decreasing the SD and clustering feeds. Removing outliers based on 3.5 SD (Method A) was simple to use; but this method was inefficient in clustering feeds classified by economic values or maturity. A National Research Support project supported by USDA-NIFA and the State Agricultural Experiment Stations.

**Key Words:** feed composition, data processing, outlier mining

**T369 Effects of energy and nitrogen supplementation of cheatgrass on ruminal fermentation using a dual-flow continuous culture system.** Lorryny Galoro da Silva<sup>\*1</sup>, Farnaz Malekjahani<sup>1,4</sup>, Pedro Del Bianco Benedetti<sup>1,2</sup>, Eduardo Marostegan de Paula<sup>1</sup>, Teshome Shenkoru<sup>1</sup>, Paloma de Melo Amaral<sup>1,2</sup>, Lays Mariz<sup>1,2</sup>, Hugo Monteiro<sup>1,3</sup>, and Antonio Faciola<sup>1</sup>, <sup>1</sup>University of Nevada, Reno, NV, <sup>2</sup>Federal University of Viçosa, Viçosa, MG, Brazil, <sup>3</sup>Maringa State University, Maringa, PR, Brazil, <sup>4</sup>Ferdowsi University, Mashhad, Iran.

Cheatgrass (CG; *Bromus tectorum*), an annual grass that is one of the main components of sagebrush community in Western US contributes to the fuel-load for wild fires. One fuel-reduction strategy is livestock grazing. The objective of this study was to determine the effects of molasses and urea supplementation on a CG-based diet on ruminal digestibility, rumen microbial fermentation and bacterial N synthesis. Diets were randomly assigned to 8 dual-flow continuous culture fermenters in a 2 × 2 factorial arrangement of treatments (urea and molasses) in a 4 × 4 Latin square design with four 10-d experimental periods consisted of 7 d for diet adaptation and 3 d for sample collection. Fermenters were fed 72 g/d of DM divided in 4 portions of 1 of 4 diets: CG, CG plus urea (CGU), CG plus molasses (CGM), and CG plus urea and molasses (CGUM). Liquid and solid flow rates were adjusted to 10 and 5%/h, respectively. A 500-mL sample was taken on d 8, 9, and 10 and analyzed for rumen traits, nutrient digestibility, and microbial growth. Data were analyzed in SAS. Partial data are presented in the table. The NH<sub>3</sub>-N concentration was higher in the diets containing urea, indicating a higher N availability for microbial growth. Total VFA concentration was not increased by molasses; however, it was lower when urea alone was added. Acetate molar proportion decreased when molasses alone was added. However, propionate molar proportion was increased when molasses was added. Results from this experiment indicate that CG utilization is improved by a combination of N and energy supplementation, which may reduce CG fuel-load in areas where CG is widely spread.

**Table 1 (Abstr. T369).**

Item	Treatment				SEM	P-value
	CG	CGU	CGM	CGUM		
NH <sub>3</sub> -N, mg/dL	3.19 <sup>c</sup>	21.49 <sup>a</sup>	0.82 <sup>c</sup>	13.75 <sup>b</sup>	0.94	<0.01
Total VFA, mmol	60.34 <sup>a</sup>	38.85 <sup>b</sup>	67.05 <sup>a</sup>	68.59 <sup>a</sup>	3.31	<0.01
Acetate, %	72.36 <sup>a</sup>	73.73 <sup>a</sup>	66.16 <sup>b</sup>	65.95 <sup>b</sup>	1.56	<0.01
Propionate, %	19.17 <sup>b</sup>	19.66 <sup>b</sup>	24.40 <sup>a</sup>	25.38 <sup>a</sup>	1.49	<0.01
Butyrate, %	8.12 <sup>a</sup>	6.03 <sup>b</sup>	9.12 <sup>a</sup>	8.10 <sup>a</sup>	0.64	0.02
Isobutyrate, %	0.05 <sup>b</sup>	0.38 <sup>a</sup>	0.06 <sup>b</sup>	0.03 <sup>b</sup>	0.07	<0.01
Valerate, %	0.04	0.02	0.16	0.04	0.05	0.23
Isovalerate, %	0.04 <sup>ab</sup>	0.02 <sup>b</sup>	0.21 <sup>a</sup>	0.04 <sup>b</sup>	0.05	0.07
Acetate:Propionate	3.26 <sup>ab</sup>	3.98 <sup>a</sup>	2.83 <sup>b</sup>	2.66 <sup>b</sup>	0.28	0.02
Total BCVFA, %	0.05	0.14	0.17	0.04	0.04	0.13

**Key Words:** cheatgrass, continuous culture, molasses

**T370 Interactions between physical form of the feed and previous experience on concentrate spillage in Holstein calves.** Maria Devant<sup>\*1</sup>, Alex Bach<sup>2,1</sup>, Josep Ribó<sup>3</sup>, and Anna Solé<sup>1</sup>, <sup>1</sup>IRTA-Ruminant Production, Animal Nutrition, Management, and Welfare Research Group, Caldes Montbui, Spain, <sup>2</sup>ICREA, Barcelona, Spain, <sup>3</sup>Grup Alimentari Guissona, Guissona, Spain.

Twenty-four Holstein bulls (137 ± 2.9 kg of BW and 117 ± 3.02 d old) were housed individually in pens (1.2 m × 1.45 m), and fed concentrate and straw in separate feeders. During the first 2 wk, half of the calves

were fed the same concentrate either in meal (MF) or pellet (PF) form. After these first 2 wk, half of the MF calves continued to be fed MF (MFMF) and half received pellet (MFPF), whereas half of the PF calves were MF (PFMF) and the other half pellet (PFPF) for 3 additional weeks. During the entire 5 wk concentrate and straw intake were recorded daily, attendance (number of visits and duration) at the feeder (concentrate and straw) were recorded through a sensor placed at the access door to the feeder, and concentrate spillage around the feeder was collected weekly. Data were analyzed using a mixed-effects model with repeated measures split in 2 analyses; the first model for the first 2 wk contrasted MF vs PF, and a second model for the last 3 wk analyzed the data as a 2 × 2 factorial design (MFMF, MFPF, PFMF, PFPF). During the first 2 wk of study, apparent mean concentrate ( $4.90 \pm 0.128$  kg/d) and straw ( $0.30 \pm 0.30$  kg/d) intakes were not affected by treatments, however concentrate spillage was doubled ( $P < 0.05$ ) in MF ( $5.9 \pm 0.70\%$ ) than in PF ( $2.8 \pm 0.70\%$ ) calves. Number of daily visits did not differ between treatments ( $14.3$  and  $15.4 \pm 0.48$ , for MF and PF, respectively), but time devoted to each visit tended ( $P = 0.10$ ) to be lesser in PF ( $9.2 \pm 0.83$  min) than in MF ( $11.1 \pm 0.83$  min). In the last 3 wk of study, previous experience did not affect apparent concentrate or straw intake. Previous experience had a positive impact on feed spillage when previous presentation form was PF; thus, concentrate spillage was similar between PFMF ( $2.5 \pm 0.87\%$ ) and PFPF ( $2.6 \pm 0.87\%$ ). In contrast, previous experience did not affect concentrate spillage when calves were initially exposed to MF, and MFMF calves spilled more concentrate ( $6.8 \pm 0.87\%$ ) than MFPF calves ( $2.3 \pm 0.87\%$ ). In conclusion, feeding calves a pellet upon arrival to a grower facility could have long-lasting positive effects due to a reduction on concentrate spillage.

**Key Words:** beef, feed spillage, feed presentation form

**T371 Effect of zinc amino acid complex on growth performance and carcass characteristics of finishing beef steers fed ractopamine hydrochloride.** C. K. Larson and M. E. Branine\*, *Zinpro Corporation, Eden Prairie, MN.*

Ractopamine hydrochloride (RAC) is fed to cattle before slaughter to improve growth performance and carcass lean meat yield. Zinc (Zn) is required for RAC function; however, Zn level and source required to optimize RAC response is unknown. The objective of this research was to evaluate the response of Zn provided as an amino acid complex (Zn-AAC) on growth, feed efficiency, and carcass merit in growing-finishing beef steers. Two well-controlled studies were conducted at commercial feedlot research facilities in TX and OK. At the TX and OK sites, 3,849 (initial BW =  $344 \pm 2.3$  kg) and 2,105 (initial BW =  $311 \pm 3.5$  kg) steers, respectively, were randomized across 8 pens/treatment. Common treatments at each site were: (1) Control (CON) = basal diet with no Zn-AAC or no RAC; (2) RAC = 320 mg RAC·hd<sup>-1</sup>·d<sup>-1</sup> (TX) and 200 mg RAC·hd<sup>-1</sup>·d<sup>-1</sup> (OK); (3) Zn-AAC + RAC = RAC plus 360 mg Zn-AAC·hd<sup>-1</sup>·d<sup>-1</sup>. At both sites, RAC was fed for the final 28-d before slaughter. Basal diets provided steam-flaked corn as the primary grain source. Total dietary Zn concentration in CON diets was 62 and 76 mg Zn·kg<sup>-1</sup>DM for OK and TX, respectively. Individual and combined study analyses were conducted using SAS on growth performance and carcass data. Pen was the experimental unit. Combined study analyses indicated RAC increased carcass-adjusted final BW ( $P \leq 0.02$ ), ADG ( $P \leq 0.01$ , feed efficiency ( $P \leq 0.01$ ), HCW ( $P \leq 0.03$ ) and ribeye area ( $P \leq 0.01$ ) compared with CON. Adding Zn-AAC to RAC produced additional improvements in carcass-adjusted final BW ( $P \leq 0.09$ ); ADG ( $P \leq 0.01$  and HCW ( $P \leq 0.11$ ). Marbling score, back fat thickness and calculated yield grade were not affected by treatment. Incidence and severity of liver abscesses were reduced ( $P \leq 0.02$ ) with Zn-AAC +

RAC compared with CON and RAC. Feeding Zn-AAC to cattle fed RAC produced additional improvements in growth and carcass yield and reduced prevalence of total and severe (A<sup>+</sup>) liver abscesses.

**Key Words:** feedlot, ractopamine, zinc

**T372 Sources of nonfiber carbohydrate in sugarcane silage based diets.** Viviane B. Ferrari\*, Nara R. B. Consolo, Rafael T. Sousa, Frederich D. Rodriguez, and Luis Felipe P. Silva, *University of São Paulo, São Paulo, Brazil.*

The aim of this study was to evaluate sources of nonfiber carbohydrates (NFC) and levels of concentrate, in sugarcane silage based diets, on intake, ADG, final body weight (FBW) and G:F ratio. The trial was repeated in 2 years: 2012 and 2013, to increase the power of the experiment. In each year, 54 Nellore young bulls, with initial body weight of 365 kg, were randomly distributed in 18 pens, with 3 animals per pen. The experimental design was a randomized block design, with 6 replicates, in a 2 × 3 factorial arrangement of treatments. Treatments were 2 levels of concentrate (diets 60% or 80% concentrate on DM basis) and 3 sources of NFC: steam-flaked corn (SFC), pelleted citrus pulp (PCP), or ground corn (GC). Steam-flaked corn and PCP replaced 70% of the ground corn in the diet, and all diets had sugarcane silage as the roughage source. Animals were weighted at the beginning of the experiment and at the end of each period (4 periods of 21 d). Intake was regulated for pen by daily weighting of diet and orts, allowing for 5 to 10% of orts. Samples of feedstuffs and orts were weekly collected for chemical analyses and determination of nutrients intake. There was a NFC × Diet interaction on DMI ( $P = 0.01$ ), as substituting GC for SFC and PCP decreased DMI, but only at the 60% concentrate diet ( $P < 0.01$ ). There was also a NFC × Diet interaction for NDFI ( $P = 0.02$ ). At the 60% concentrate diet, SFC decreased NDFI when compared with GC and PCP ( $P < 0.05$ ). At the 80% concentrate diet, NDFI was higher for PCP than for GC and SFC ( $P < 0.01$ ). Substituting PCP for GC decreased FBW ( $P < 0.01$ ), with no difference between GC and SFC ( $P = 0.26$ ). Similarly, PCP decreased ADG compared with GC ( $1.27$  vs.  $1.40$ ;  $P = 0.04$ ), independently of diet. Increasing the concentrate level in the diet improved G:F ratio ( $0.149$  vs.  $0.137$ ;  $P < 0.01$ ), but there was no effect of NFC sources on G:F ratio ( $P = 0.74$ ), nor there was a NFC × Diet interaction ( $P = 0.21$ ). Pelleted citrus pulp as main carbohydrate source decreased performance of young bulls compared with ground and steam-flaked corn.

**Key Words:** average daily gain, steam-flaking, feed intake

**T373 Effects of starch content on in vitro ruminal fermentation of ground and dry-rolled barley grain.** Uchenna Y. Anele<sup>1</sup>, Basim Refat<sup>1,4</sup>, Mary-Lou Swift<sup>2</sup>, Yanli Zhao<sup>1,3</sup>, Tim McAllister<sup>1</sup>, and Wenzhu Yang\*<sup>1</sup>, <sup>1</sup>Agriculture and Agri-Food Canada, Research Centre, Lethbridge, AB, Canada, <sup>2</sup>Alberta Agriculture & Rural Development, Lethbridge, AB, Canada, <sup>3</sup>Inner Mongolia Agricultural University, Hohhot, Inner Mongolia, China, <sup>4</sup>Zagazig University, Zagazig, Egypt.

Rapid digestion of barley starch in the rumen is hypothesized that starch content of barley grain can influence both rates of gas production (GP) and dry matter disappearance (DMD). Rumen fermentation of ground and dry-rolled barley grain differing in starch content was evaluated using a batch culture technique. The study was arranged in a 2 starch contents (low vs. high) × 2 processing (ground vs. dry-rolled) factorial design. Barley samples were collected monthly from 10 different feedlots in southern Alberta during one year. Samples were ranked according to

the starch content into low (<60%) and high (>60% of DM). Ten barley samples with 5 low (57.1 ± 1.7%) and 5 high (66.0 ± 0.7% of DM) starch were either ground 2 mm or dry-rolled with processing index (PI) of 75%. Gas production and DMD were estimated at 3, 6, 12 and 24 h of incubation using rumen fluid from 3 fistulated beef heifers fed diet containing 70% barley silage and 30% barley grain (DM basis). Cumulative GP (mL/g DM) was fitted to a model  $GP = B(1 - e^{-c(t-lag)})$ . Starch content × processing interaction was not significant for in vitro GP kinetics. Rate of GP (%/h) was greater ( $P < 0.01$ ) in both high starch (18.3) and ground (20.6) versus, respectively, low starch (16.1) and rolled (13.9) barley samples. Similarly, both high starch (40.0) and ground (44.9) samples had greater ( $P < 0.01$ ) absolute initial GP during the first hour of incubation compared with, respectively, low starch (34.6) and rolled (29.7) barley. Data from DMD at different times of incubation were fitted to a model  $DMD = a + b(1 - e^{-c(t-L)})$ . Starch content × processing interactions were noted for the b fraction ( $P < 0.03$ ) and rate of DMD ( $P < 0.01$ ). Consistently, both high starch and ground barley had greater ( $P < 0.05$ ) a, b and c of DM versus low starch and dry-rolled barley. However, the a, b, and c of starch did not differ between low and high starch barley. Ground barley had greater ( $P < 0.01$ ) c (ground vs. rolled; 8.9 vs 6.1%) compared with rolled barley. The results indicated that starch content of barley had significant effect on in vitro rate of GP and the rate of DMD; processing (ground vs. dry-rolled) increased the extent and rate of DMD and starch.

**Key Words:** barley starch content, in vitro fermentation, grain processing

**T374 Effect of sainfoin hay and pomegranate peel extracts on in vitro fermentation and protein degradation using the Rusitec technique.** Basim Refat<sup>1,2</sup>, Uchenna Y. Anele<sup>1</sup>, Zhixiong He<sup>1,3</sup>, S. M. Bassiony<sup>2</sup>, G. A. Abdel-Rahman<sup>2</sup>, and Wenzhu Yang<sup>1</sup>, <sup>1</sup>Agri-culture and Agri-Food Canada, Research Centre, Lethbridge, AB, Canada, <sup>2</sup>Faculty of Agriculture, University of Zagazig, Zagazig, Egypt, <sup>3</sup>Institute of Subtropical Agriculture, The Chinese Academy of Science, Changsha, Hunan, China.

Pomegranate peel extracts (PPE) have been found to exert beneficial effects on animal health due to their antioxidant, antibacterial and immunological effects. Sainfoin hay that contains moderate to high content of condensed tannins, was reported the reduced rumen proteolysis and decreased urine N excretion in beef cattle. The objective of this study was to determine the effect of sainfoin hay extract (SHE) and PPE supplementation on in vitro rumen fermentation and protein metabolism of high-protein finishing diet using rumen simulation technique (Rusitec). The experiment was a completely randomized design with 3 treatments: Control (10% barley silage, 60% barley grain and 30% wheat distillers grain), and control supplemented with SHE (4.1 g catechin equivalent/kg DM) or with PPE (2.8 g tannic acid equivalents/kg DM). The experiment consisted of 10 d of adaptation and 7 d of data collection. Concentration of volatile fatty acid (VFA) was lower (44.1 or 41.6 vs. 48.2 mM;  $P < 0.02$ ) but molar proportion of acetate was higher (37.2 or 37.4 vs. 34.9%;  $P < 0.01$ ) with SHE or PPE vs. control. Branched-chain VFA decreased ( $P < 0.01$ ) with PPE (0.9%) compared with control. Concentrations (mg/100 mL) of large (10.1) and small peptides (6.1) were not affected but that of NH<sub>3</sub>-N was lower ( $P < 0.01$ ) with PPE (6.3) vs. control (8.1). Digestibility of DM, starch and crude protein decreased ( $P < 0.02$ ) with SHE (58.7, 74.7 and 43.0%, respectively) and PPE (55.5, 68.9 and 43.4%, respectively) supplementation compared with control (63.3, 83.2 and 51.0%, respectively). Bacterial N production (averaged 52 mg/d) and bacterial efficiency (averaged 10.3 g bacterial N/kg digested OM) were not affected by SHE or PPE

supplementation. Overall, there were no differences in VFA concentrations, N fractions and nutrient digestibility between SHE and PPE except for the concentrations of branched-chain VFA and NH<sub>3</sub>-N which were higher ( $P < 0.01$ ) with SHE versus PPE supplementation. These results indicated that supplementation of high-grain diet with SHE or PPE increased rumen by-pass protein; however, suppression of feed fermentation may decrease the feeding value of high-grain diet.

**Key Words:** tannins extracts, fermentation, Rusitec

**T375 Effects of starch content and processing method on in situ rumen digestibility of barley grain in beef heifers.** Yanli Zhao<sup>1,2</sup>, Sumei Yan<sup>2</sup>, Uchenna Y. Anele<sup>1</sup>, Mary-Lou Swift<sup>3</sup>, Tim A. McAllister<sup>1</sup>, and Wenzhu Yang<sup>\*1</sup>, <sup>1</sup>Lethbridge Research Centre, Agriculture and Agri-Food Canada, Lethbridge, AB, Canada, <sup>2</sup>College of Animal Science, Inner Mongolia Agricultural University, Hohhot, Inner Mongolia, China, <sup>3</sup>Alberta Agriculture and Rural Development, Lethbridge, AB, Canada.

The inherent variability in barley chemical composition leads to differences in animal performance. The objective of this study was to investigate the effects of starch content of barley grain and processing method on in situ rumen digestion of DM and starch. Barley samples (n = 120) were collected monthly from 10 feedlots in Southern Alberta for one year. Samples were ranked according to their starch content into low (<60%) and high (>60% of DM). Ten barley samples with 5 low (57.1 ± 1.7%) and 5 high (66.0 ± 0.7% of DM) starch were either ground 2 mm or dry-rolled with processing index (PI) of 75% (PI = 100 × bulk density processed/bulk density whole). Three beef heifers (650 BW) fitted with rumen cannulas and fed diet consisting of 70% barley silage and 30% barley grain were used for in situ incubation. The study was arranged in a 2 (low and high starch) × 2 (ground and rolled) factorial design. Digestion kinetics of DM after 0, 3, 6, 12, 24, and 48 h of incubation was estimated using the model:  $y = a + b(1 - e^{-ct})$ . Effective rumen digestibility (ED) was estimated using  $ED = a + bc/(c + k)$  with  $k = 6\%/h$ . Particle size distribution of rolled barley differed between low and high starch barley; the particles retained on the 3.35-mm sieve was less (17.8 vs. 25.3%;  $P < 0.01$ ) but the particles on 2.36-mm sieve was greater (40.5 vs. 32.1%;  $P < 0.01$ ) for low vs. high starch barley. Starch content × processing interactions were not noted. Low starch barley had less ( $P < 0.01$ ) a (12.2 vs. 14.8%) and ED of DM (60.4 vs. 64.8%) versus high starch barley. The low starch barley also had less ( $P < 0.01$ ) a (14.2 vs. 23.1%) and ED of starch (68.4 vs. 74.6%) but had greater ( $P < 0.03$ ) b (75.5 vs. 70.5%) compared with high starch barley. As expected, rumen digestion kinetics of DM and starch differed between ground and dry rolled (PI = 75%) samples. Ground samples had greater ( $P < 0.01$ ) a (21.6 vs. 5.4%), c (33.1 vs. 10.1%/h) and ED (71.6 vs. 53.6%) of DM but smaller b (59.8 vs. 77.8%;  $P < 0.01$ ) compared with rolled barley. These results indicated that starch content of barley grain and manipulating processing method could effectively alter rumen digestion of barley grain.

**Key Words:** barley starch content, in situ rumen digestion, beef heifer

**T376 Effect of physical form of concentrate on performance, eating pattern, and behavior in Holstein bulls fed finishing high-concentrate diets.** Marçal Verdú<sup>\*1</sup>, Alex Bach<sup>2,1</sup>, and Maria Devant<sup>1</sup>, <sup>1</sup>IRTA-Ruminant Production, Animal Nutrition, Management, and Welfare Research Group, Caldes Montbui, Spain, <sup>2</sup>ICREA, Barcelona, Spain.



Pelleting concentrate is one of the predominant grain processing methods in Mediterranean beef feeding systems. A good pellet quality is related to improvements in performance and feed efficiency, but it involves an extra cost at manufacturing. One hundred twelve bulls ( $272 \pm 4.4$  kg of BW and  $216 \pm 1.0$  d of age) were randomly allocated in 6 pens provided with a concentrate single feeder with lateral protections, a straw feeder, and a water bowl. Pens were assigned to 1 of 2 dietary treatments according to the physical form of concentrate: pellet (PF), and crumble (CF) to simulate a worse pellet quality. The experimental design was 3 Latin Squares with periods of 28 d replicated twice. Concentrate intake and eating pattern were recorded daily, and concentrate wastage and animal BW every 14 d. Behavior was registered on d 20 of each period by scan sampling to analyze the general activity and social behavior. Data were analyzed using a mixed-effects model. The PF bulls consumed more concentrate ( $P < 0.01$ ), but had lesser ( $P < 0.01$ ) waste and day-to-day CV of concentrate intake compared with CF ( $7.0$  and  $6.7 \pm 0.08$  kg of DM/d,  $0.06$  and  $0.11 \pm 0.004$  kg of DM/d,  $16.7$  and  $21.3 \pm 0.99\%$ , respectively). However, ADG ( $P = 0.11$ ) and feed efficiency were not affected by physical form. The eating pattern of PF bulls was characterized ( $P < 0.01$ ) by a lesser meal size and meal duration, but a greater meal frequency and eating rate than CF bulls ( $630.9$  and  $668.4 \pm 17.32$  g/meal,  $3.6$  and  $4.2 \pm 0.18$  min/meal,  $11.8$  and  $10.9 \pm 0.32$  meals/d,  $193.0$  and  $174.4 \pm 11.79$  g/min, respectively). Social behavior was not influenced by physical form. In conclusion, the decrease of pellet quality modified eating behavior increasing meal duration and decreasing concentrate intake, may be bulls fed crumbles were sorting avoiding fines. A long-term study is necessary, as it could be hypothesized that the effects of decreased pellet quality on decreased concentrate intake could impair performance.

**Key Words:** bull, eating pattern, physical form of concentrate

### T377 Carcass and sensory traits and free amino acid contents among quality grades in loin and rump of Korean cattle steer.

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This study was performed to compare carcass traits, sensory characteristics, physiochemical composition, and contents of nucleotides, collagen, and free amino acids among quality grades (QG) and to understand the association between QG and above parameters in loin and rump of Korean cattle steer. Loin and rump samples were obtained from 48 Korean cattle steers with each of 4 quality grades (QG 1++, 1+, 1, and 2; average 32 mo of age). Carcass weight and marbling score (MS) were highest in QG 1++, whereas texture score measured by a meat grader was highest in QG 2. A correlation analysis revealed that MS ( $r = 0.98$ ;  $P < 0.01$ ) and fat content ( $r = 0.73$ ;  $P < 0.01$ ) had strong positive correlations with QG and that texture had a strong negative correlation ( $r = -0.78$ ) with QG. Fat content in loin was highest but protein and moisture contents were lowest in QG 1++. Our results confirmed that a major determinant of QG is the MS; thus, intramuscular fat (IMF) content. The CIE L\*, a\*, and b\* values in loin were highest in QG 1++. Numeric values of shear force in loin were lowest in QG 1++, whereas those of tenderness, juiciness, and overall acceptability tended to be highest in QG 1++ without statistical significance. QG was strongly correlated with juiciness ( $r = 0.81$ ;  $P < 0.01$ ) and overall acceptability ( $r = 0.87$ ;  $P < 0.001$ ). All sensory characteristics were higher ( $P < 0.05$ ) in loin than those in rump. Adenosine-5'-monophosphate (AMP) and inosine-5'-monophosphate (IMP) contents in both loin and rump did not differ among QGs. No nucleotide (AMP, IMP, inosine, hypoxanthine) was

correlated with any of the sensory traits. Total, soluble, and insoluble collagen contents in loin were higher in QG 1++ than those in QG 1. All 3 collagens had lower content in loin than that in rump. All 3 collagens were positively correlated with tenderness, juiciness, and overall acceptability. Glutamic acid content did not significantly differ among the 4 QGs in either loin or rump. In conclusion, it is confirmed that QG is associated with sensory traits but nucleotide contents in beef may not be a major factor determining meat palatability in the present study.

**Key Words:** Korean cattle steer, carcass trait, quality grade

**T378 Plasma creatinine concentration of beef heifers fed with different lipid sources and frequency supplementation.** Marcia Cristina A. Santana\*<sup>1</sup>, Ricardo A. Reis<sup>2</sup>, Gabriel M. P. Melo<sup>2</sup>, Viviane C. Modesto<sup>3</sup>, Telma T. Berchielli<sup>2</sup>, Jucilene Cavali<sup>4</sup>, and Juliana F. H. Rodrigues<sup>4</sup>, <sup>1</sup>EMATER, Goiania, Goias, Brazil, <sup>2</sup>Unesp, Jaboticabal, Sao Paulo, Brazil, <sup>3</sup>Unesp, Ilha Solteira, Sao Paulo, Brazil, <sup>4</sup>UNIR, Presidente Médici, Rondônia, Brazil.

This research aims to evaluate urea plasma responses under different lipid sources and supplementation frequencies. The experiment was conducted throughout a 4-mo period during the dry season. The experiment was completely random, using a  $3 \times 2$  factorial arrangement (3 supplements and 2 supplementation frequencies). The supplements were derived from 3 different sources, soybean grains, soybean oil and protected fat (Megalac-E), the 2 supplementation frequencies were (D) daily or 3 d of week (Monday, Wednesday and Friday) called "alternate" (A). In the 4-mo experimental period, August–November, blood samples were taken from the jugular vein 4 h after the morning feeding. In all treatments, no creatinine levels differences were observed during August, October and November ( $P > 0.05$ ). However, on September were observed lower values of creatinine for Megalac-E daily and soybean oil alternately supplied. Therefore, the highest creatinine plasma levels observed for all treatments were on September and the lower showed on November. Overall, these data indicated that the creatinine plasma level can be influenced according to the feeding strategy during the dry season.

**Table 1.** Plasma creatinine concentration of heifers supplemented with different lipid sources at two different frequencies (mg/dL)<sup>1</sup>

	D-SG	A-SG	D-SO	A-SO	D-ML	A-ML
August	1.2 <sup>A,b</sup>	1.2 <sup>A,b</sup>	1.0 <sup>A,b</sup>	1.1 <sup>A,bc</sup>	1.2 <sup>A,b</sup>	1.3 <sup>A,b</sup>
September	2.8 <sup>A,a</sup>	2.8 <sup>A,a</sup>	2.6 <sup>A,a</sup>	2.2 <sup>B,a</sup>	2.4 <sup>AB,a</sup>	2.6 <sup>A,a</sup>
October	1.1 <sup>A,b</sup>	1.4 <sup>A,b</sup>	1.2 <sup>A,b</sup>	1.4 <sup>A,b</sup>	1.1 <sup>A,b</sup>	1.3 <sup>A,b</sup>
November	0.7 <sup>A,c</sup>	0.8 <sup>A,c</sup>	1.0 <sup>A,b</sup>	0.8 <sup>A,c</sup>	0.8 <sup>A,b</sup>	0.8 <sup>A,b</sup>

a-c,A,B Means with lowercase superscripts in columns and uppercase superscripts in rows differ ( $P < 0.05$ ).

<sup>1</sup>D = daily; A = alternately; SG = soybean; SO = soy oil, and ML = Megalac-E.

**Key Words:** soybean, lipid supplement, dry season

**T379 Efficacy of supplying lasalocid sodium via a self-fed trace mineralized salt block supplement to growing beef calves grazing warm season grass.** Brandon Stewart\*<sup>1</sup>, Paul Beck<sup>1</sup>, John Tucker<sup>2</sup>, Tom Hess<sup>2</sup>, and Don Hubbell<sup>2</sup>, <sup>1</sup>University of Arkansas SWREC, Hope, AR, <sup>2</sup>University of Arkansas LFRS, Batesville, AR.

Two experiments were conducted at the University of Arkansas Southwest Research and Experiment Station (Hope, AR) and Livestock and Forestry Branch Station (Batesville, AR) in northern Arkansas. Growing beef calves ( $n = 96$  steers at the Hope site and 48 steers and 48 heifers at the Batesville site, BW =  $215 \pm 14.5$ ) grazed 0.8 ha warm-season grass

based pastures (n = 24/site, predominantly bermudagrass) during a 56-d summer grazing study to determine the effects of supplementation of growing steers grazing warm-season perennial grass pastures with Lasalocid via self-limited block (Bovatec 2.2 Block, Zoetis Animal Health, Inc.) on growth performance. Pastures were stocked with 4 calves/pasture with 24 total pasture replicates/treatment. Because of drought conditions this study was initiated on June 12 and terminated on August 8 at the Batesville site and was initiated on June 27 and terminated on August 22 at the Hope site. At the Hope site, lack of grazable forage made it necessary to offer grass hay (10% CP and 55% TDN) and soybean hulls at 2 lb/head/d to all pastures for the duration of the study. Beginning and ending weights were collected full on 2 consecutive days, and interim weights were collected full on 28-d intervals. Calves in each pasture were offered free-choice access to either a non-medicated control trace mineralized salt block (Control) or Bovatec 2.2 block (Bovatec). Daily

block intakes at the Hope site averaged  $12 \pm 8.1$  g/calf with an average Bovatec dose of  $59 \pm 39$  mg/head/day. At Batesville, daily intake of control averaged  $28 \pm 12$  g/calf and intake of Bovatec averaged  $20 \pm 5.3$  g/head/day (supplying  $100 \pm 25$  mg Bovatec). There was no treatment  $\times$  site interaction ( $P = 0.44$ ) for performance in this study. At the end of the study there was no difference ( $P \geq 0.16$ ) between Control and Bovatec treatments for BW ( $267$  vs  $269 \pm 3.9$  kg, respectively) or ADG ( $0.75$  vs.  $0.81 \pm 0.0435$  kg/d, respectively). The results of this study indicate that Bovatec offered in a self-fed trace mineralized salt block supplement did not supply a large enough dose of lasalocid to increase growth rate of beef calves grazing warm-season grass pastures.

**Key Words:** growing calves, lasalocid, bermudagrass pasture

## Ruminant Nutrition: Dairy II

**T380 Dietary grain source and oil supplement: Milk fat synthesis and milk fatty acid profile of Holstein cows.** Shahryar Kargar\*<sup>1</sup>, Gholam Reza Ghorbani<sup>2</sup>, Veerle Fievez<sup>3</sup>, and David J. Schingoethe<sup>4</sup>, <sup>1</sup>Shiraz University, Shiraz, Iran, <sup>2</sup>Isfahan University of Technology, Isfahan, Iran, <sup>3</sup>Ghent University, Melle, Belgium, <sup>4</sup>South Dakota State University, Brookings.

Effects of grain type and dietary oil supplement on milk fat depression, milk fatty acid (FA) profile, and lactational performance of dairy cows were evaluated using 8 multiparous Holstein cows in a duplicated 4 × 4 Latin square design with a 2 × 2 factorial arrangement of treatments. Experimental diets contained either ground barley or ground corn supplemented with either fish oil or soybean oil at 2% of dietary dry matter (DM). Experimental diets contained 28.5 and 31.2% of cereal grain in corn- and barley-based diets, respectively, as the sole source of grain. The forage component of the experimental diet was a mixture of corn silage (19.0% of DM) and alfalfa hay (21.0% of DM). Treatment periods were 25 d, with the final 7 d used for sample and data collection. Data were composited within period and subjected to MIXED MODEL procedure of SAS (SAS Institute, 2003) to account for effects of square, period within square, cow within square, treatments (grain type and oil supplement), and the interaction between grain type and oil supplement. Total FA intake was greater in corn-based diets and also in soybean oil supplemented diets ( $P \leq 0.05$ ). Fish oil decreased intake of all 18-carbon FA but increased intakes of other FA including C16:0, C20:5, and C22:6 ( $P \leq 0.05$ ). No significant differences existed in the DM intake and yield of milk or milk components between barley- and corn-based diets ( $P > 0.05$ ). Fish oil negatively affected feed intake and yields of milk and milk components as compared with soybean oil ( $P \leq 0.05$ ). Although milk fat yield was not affected ( $P > 0.05$ ), the barley-based diets increased the concentration and yield of medium-chain FA but decreased the concentration of long-chain FA as compared with corn-based diets ( $P \leq 0.05$ ). Corn-based diets increased concentration and yield of both *trans*-11 C18:1 and *cis*-9, *trans*-11 C18:2 which was a reflection of greater intake of *cis*-9, *cis*-12 C18:2 (25.1 g/d) as substrate for rumen biohydrogenation ( $P \leq 0.05$ ). Severity of MFD was greater for fish oil- vs. soybean oil which evidenced by the increased concentration and yield of biohydrogenation intermediates associated with MFD (especially *trans*-10 C18:1) in milk fat ( $P \leq 0.05$ ). However, fish oil increased concentration and yield of both *trans*-11 C18:1 and *cis*-9, *trans*-11 C18:2 as compared with soybean oil ( $P \leq 0.05$ ). Results indicated that there was no interaction between the type of grain and oil supplement on induction of MFD and milk fat yield. Although milk fat yield was not affected, milk fat composition was differently modified in barley- vs. corn-based diets. Observed lower milk fat yield for fish oil supplemented diets was coincided with the increased concentration and yield of biohydrogenation intermediates associated with MFD in milk fat.

**Key Words:** grain and oil, milk fat depression, dairy cow

**T381 Effect of biotin and pantothenic acid supplementation on performance and concentration of avidin-binding substances (ABS) in lactating dairy cows.** Gonzalo Ferreira, Alston N. Brown\*, and Christy Teets, *Department of Dairy Science, Virginia Polytechnic Institute and State University, Blacksburg, VA.*

We hypothesized that pantothenic acid reduces the absorption of biotin in lactating dairy cows. Twenty-four Holstein cows (8 primiparous and

16 multiparous) were assigned to a 4 × 4 replicated Latin square design with 18-d periods. Cows were 109 ± 30 d in milk and weighed 591 ± 70 kg at the beginning of the experiment. Treatments consisted of a control diet (CON) containing 53% concentrate and 47% forage, CON supplemented with 20 mg/d biotin (BIO), CON supplemented with 475 mg/d pantothenic acid (PAN), and CON supplemented with 20 mg/d biotin + 475 mg/d pantothenic acid (BIOPAN). Data were analyzed with a Proc Mixed model that included the effects of square (fixed, 5 df), treatment (fixed, 3 df), treatment × square interaction (fixed, 15 df), period (random, 3 df), cow within square (random, 18 df), and the residual error (51 df). No differences among treatments were observed for dry matter intake ( $P < 0.46$ ), milk yield ( $P < 0.68$ ), fat concentration ( $P < 0.85$ ) and yield ( $P < 0.88$ ), protein concentration ( $P < 0.28$ ) and yield ( $P < 0.07$ ), lactose concentration ( $P < 0.90$ ) and yield ( $P < 0.25$ ), and milk urea nitrogen ( $P < 0.49$ ). Biotin supplementation increased the concentration of avidin-binding substances (ABS) in milk ( $P < 0.03$ ). This increase was similar for BIOPAN ( $P < 0.01$ ). Relative to CON, pantothenic acid did not reduce the concentration of ABS in milk ( $P < 0.99$ ). In conclusion, under the conditions of this experiment, pantothenic acid did not affect the absorption of biotin in lactating dairy cows.

**Table 1 (Abstr. T381).** Effect of biotin and pantothenic acid supplementation on performance and concentration of avidin-binding substances (ABS) in milk of dairy cows

Item	CON	BIO	PAN	BIOPAN	SEM	$P <$
DMI, kg/d	21.6	21.5	21.2	21.1	0.49	0.46
Milk yield, kg/d	41.9	41.9	42.1	41.1	1.41	0.68
Fat, %	2.83	2.79	2.88	2.90	0.19	0.85
Fat, kg/d	1.18	1.18	1.22	1.18	0.08	0.88
Protein, %	3.01	3.07	3.03	3.04	0.06	0.28
Protein, kg/d	1.26	1.31	1.30	1.25	0.04	0.07
Lactose, %	4.88	4.90	4.89	4.89	0.04	0.90
Lactose, kg/d	2.06	2.09	2.11	2.02	0.07	0.25
MUN, mg/dL	15.39	15.83	15.42	15.73	0.58	0.49
ABS in milk, ng/mL	28.9 <sup>b</sup>	39.6 <sup>a</sup>	28.9 <sup>b</sup>	43.0 <sup>a</sup>	3.36	0.01

**Key Words:** biotin, pantothenic acid, vitamin

**T382 Assessment of in vitro ruminal fermentation characteristics of lactation dairy diets supplemented with slow-release urea using continuous cultures.** F. Mason<sup>1</sup>, K. Neal<sup>2</sup>, S. Y. Yang<sup>2</sup>, J.-S. Eun<sup>\*2</sup>, and M. Spanghero<sup>1</sup>, <sup>1</sup>Department of Agricultural and Environmental Science, University of Udine, Udine, Italy, <sup>2</sup>Department of Animal, Dairy, and Veterinary Sciences, Utah State University, Logan, UT.

The present study investigated the effects of supplementing slow-release urea (SRU; Optigen, Alltech, Nicholasville, KY) in 3 lactation dairy diets with different forage-to-concentrate ratios (F:C) on in vitro ruminal fermentation characteristics. The experiment was performed in a 3 (F:C) × 2 (without vs. with SRU supplementation) factorial design with 4 independent runs of continuous cultures (n = 4). The 3 F:C included high-forage (HF; 64:36), medium-forage (MF; 51:49), and low-forage diet (LF; 34:66) on a DM basis, and alfalfa hay and cone silage were forage sources at equal proportions. Supplementing SRU considerably reduced dietary concentrations of mixture of soybean meal and canola meal (50:50 on a DM basis) in the SRU supplemented diets. Continuous culture apparatus consisted of 700-mL working volume fermentation



vessels to measure major fermentation end products. Each culture was offered a diet of 20 g DM/d in 2 equal portions at 0800 and 2000 h. Decreasing forage proportion reduced ruminal pH ( $P < 0.01$ ), but supplementing SRU did not influence ruminal pH. Manipulating F:C did not affect total VFA concentration; however, under HF supplementing SRU increased total VFA concentration, while SRU supplementation in MF decreased total VFA concentration, leading to an interaction between F:C and SRU ( $P = 0.02$ ). Decreasing forage proportion decreased acetate proportion, but increased propionate proportion, resulting in a tendency to decrease acetate-to-propionate ratio ( $P = 0.09$ ). In contrast, SRU supplementation did not affect individual VFA proportions. Decreasing F:C decreased ammonia-N concentration, whereas supplementation of SRU tended to increase concentration of ammonia-N ( $P = 0.08$ ). While methane production decreased with decreasing F:C ( $P < 0.01$ ), supplementing SRU tended to increase methane production ( $P = 0.07$ ) mainly due to its sizable increase in MF, leading to a tendency of F:C  $\times$  SRU interaction ( $P = 0.06$ ). Overall results in the current study indicate that supplementing SRU would be more beneficial in HF with enhanced ruminal fermentation evidenced by increased total VFA concentration.

**Key Words:** forage-to-concentrate ratio, ruminal fermentation, slow-release urea

**T383 Ruminal fermentation characteristics of lactation dairy diets with different forage-to-concentrate ratios without or with lipid extract algae in continuous cultures.** S. Y. Yang\*<sup>1</sup>, K. Neal<sup>1</sup>, J.-S. Eun<sup>1</sup>, A. J. Young<sup>1</sup>, and R. C. Sims<sup>2</sup>, <sup>1</sup>Department of Animal, Dairy, and Veterinary Sciences, Utah State University, Logan, UT, <sup>2</sup>Department of Biological Engineering, Utah State University, Logan, UT.

The current in vitro experiment was performed to test the effects of supplementing lipid extract algae (LEA) in lactation dairy diets on ruminal fermentation in a 2 (level of forage in diets)  $\times$  2 (without vs. with LEA) factorial design with 4 independent runs of continuous cultures ( $n = 4$ ). Diets with LEA completely replaced mixture of soybean meal and canola meal (50:50 in a DM basis). The data in this experiment were analyzed using the Proc Mixed procedure of SAS using a model that included fixed effects of level of forage, LEA supplementation, and their interaction and a random effect of fermentor within independent run. Feeding LEA decreased culture pH, regardless of level of forage, but the decrease of culture pH was greater under high-forage diet compared with low-forage diet, resulting in an interaction between level of forage and LEA. Under high-forage diet, total VFA concentration increased with feeding LEA, but it was not affected in low-forage diet, leading to a tendency ( $P = 0.08$ ) of level of forage and LEA interaction. Adding LEA decreased ammonia-N concentration both in high- and low-forage diet. Overall results in this experiment indicate that feeding LEA in lactation dairy diets did not interfere with in vitro ruminal fermentation. The decreased ammonia-N concentration due to feeding LEA may have resulted from less degradation of N fraction in LEA compared with mixture of soybean meal and canola meal.

*Contd.*

**Table 1 (Abstr. T383).** In vitro effects of lipid extract algae supplemented in dairy diets

Item	Diet <sup>1</sup>				SEM	<i>P</i> <sup>2</sup>		
	HF		LF			FC	LEA	INT
	-LEA	+LEA	-LEA	+LEA				
Mean culture pH	6.25 <sup>a</sup>	6.04 <sup>b</sup>	6.07	6.00	0.047	<0.01	0.05	0.05
Total VFA, mM	29.1 <sup>b</sup>	34.0 <sup>a</sup>	34.4	32.5	2.77	0.30	0.60	0.08
Individual VFA <sup>3</sup>								
Acetate (A)	46.1	47.3	47.3	47.7	3.92	0.21	0.33	0.53
Propionate (P)	36.5	36.8	40.6	38.7	2.64	0.01	0.45	0.30
A:P	1.26	1.29	1.19	1.24	0.168	0.06	0.54	0.47
Ammonia-N, mg/dL	8.04	5.36	6.54	3.10	0.403	<0.01	<0.01	0.31

<sup>a-b</sup>Means within a row with different superscripts differ ( $P < 0.05$ ).

<sup>1</sup>HF-LEA = high-forage diet (HF; 60% forage:40% concentrate) without lipid extract algae (LEA); HF+LEA = HF with LEA; LF-LEA = low-forage diet (LF; 40% forage:60% concentrate) without LEA; and LF+LEA = LF with LEA.

<sup>2</sup>FC = forage-to-concentrate ratio in the diet; LEA = supplementation of LEA; and INT = interaction between FC and LEA.

<sup>3</sup>Expressed as mol/100 mol.

**Key Words:** continuous culture, lactation dairy diet, lipid extract algae

**T384 In vitro ruminal metabolism of a lactation dairy diet supplemented with virgin coconut oil and pine bark extract in continuous cultures.** S. Y. Yang\*<sup>1</sup>, R. W. S. Ningrat<sup>2</sup>, K. Neal<sup>1</sup>, B. R. Min<sup>3</sup>, and J.-S. Eun<sup>1</sup>, <sup>1</sup>Department of Animal, Dairy, and Veterinary Sciences, Utah State University, Logan, UT, <sup>2</sup>Faculty of Animal Sciences, Andalas University, Padang, Indonesia, <sup>3</sup>Department of Agricultural and Environmental Sciences, Tuskegee University, Tuskegee, AL.

The present study investigated effects of virgin coconut oil (VCO; 7.0% C8:0, 5.4% C10:0, 48.9% C12:0, and 20.2% C14:0) and pine bark extract (PBE; 75% condensed tannins), either separately or in combination as supplements to a lactation dairy diet on in vitro ruminal fermentation profiles. The experiment was performed in a completely randomized design with 4 independent runs of continuous cultures. Four dietary treatments included: 1) control (CONT; TMR without supplement), 2) TMR with 3% VCO (VCOT), 3) TMR with 3% PBE (PBET), and 4) TMR with 3% VCO and 3% PBE (VPT). Each culture of 700-mL working volume fermentation content was offered a diet of 20 g DM/d in 2 equal portions at 0800 and 2000 h. The data in this study were analyzed using the Proc Mixed procedure of SAS using a model that included fixed effect of dietary treatments (CONT, VCOT, PBET, and VPT) and a random effect of fermentor within independent run. Culture pH was maintained at least at 6.13 across dietary treatments, and supplementing VCO and/or PBE did not influence culture pH. Total VFA concentration was similar in response to the supplements. Supplementation of VCO decreased acetate proportion, but did not affect propionate proportion, resulting in a tendency to decrease acetate-to-propionate ratio ( $P = 0.10$ ). In contrast, PBE supplementation increased acetate proportion, while it did not influence propionate proportion, leading to a tendency to increase acetate-to-propionate ratio ( $P = 0.10$ ). Cultures offered VCOT and VPT increased butyrate proportion ( $P = 0.01$ ). Supplementing PBE decreased ( $P < 0.01$ ) ammonia-N concentration both in PBET and VPT, whereas VCO supplementation resulted in no effect on ammonia-N concentration. Cultures offered VCO and PBE supplementation, either separately or in combination, showed no response on methane production, although feeding VCOT numerically decreased methane production by 13.5% compared with CONT. The decrease in ammonia-N concentration when PBE-containing diets (PBET and VPT) were offered is likely attributed

to condensed tannins in PBE; however, their concentration would not be enough to lessen ruminal methanogenesis. In addition, the concentration of VCO used in this study may have not been enough to manipulate ruminal fermentation.

**Key Words:** continuous culture, pine bark extract, virgin coconut oil

**T385 Methane production from dairy cows fed regular or brown midrib corn silage.** Fadi Hassanat\*<sup>1</sup>, Rachel Gervais<sup>2</sup>, and Chaouki Benchaar<sup>1</sup>, <sup>1</sup>*Agriculture and Agri-Food Canada, Dairy and Swine Research and Development Centre, Sherbrooke, QC, Canada*, <sup>2</sup>*Département des Sciences Animales, Université Laval, Québec, QC, Canada*.

This study investigated the effects of feeding dairy cows regular (RCS) or brown midrib corn silage (BMCS) based diets on enteric CH<sub>4</sub> emissions. In a crossover design (35-d periods), 16 lactating multiparous Holstein cows (BW = 691 ± 77 kg; DIM = 119 ± 27; milk yield = 43.5 ± 6.4 kg/d) were fed (ad libitum; 5% orts on as-fed basis) a TMR (65:35 forage:concentrate ratio) containing 59% (dry matter basis) RCS or BMCS. Production of CH<sub>4</sub> was measured (3 consecutive days) using respiration chambers. Rumen fermentation characteristics were assessed over 2 consecutive days, and milk performance and nutrient apparent-total-tract digestibility were determined over 6 consecutive days. Effects of dietary corn silage cultivar were determined using the MIXED Procedure of SAS and significance was declared at  $P \leq 0.05$ . Compared with feeding RCS, feeding BMCS increased ( $P \leq 0.01$ ) dry matter intake (+ 1.6 kg/d), milk yield (+ 3.2 kg/d) without affecting milk fat (3.87% ± 0.11) and milk protein (3.50% ± 0.05) contents. Dietary treatment did not affect ruminal pH, protozoa numbers, volatile fatty acid profile and apparent total-tract digestibility of organic matter, including its fiber fraction. Daily enteric methane emission averaged 476 g/d (±21) and was not affected by dietary treatment. Methane energy losses (proportional to gross energy intake) were 7% lower ( $P = 0.05$ ) in cows fed BMCS (5.16%) compared with cows fed RCS (5.52%). Similarly, CH<sub>4</sub> emitted per kg of milk was lower ( $P = 0.02$ ) in cows fed BMCS (12.7 g/kg) than in cows fed RCS (14.3 g/kg). It is concluded that replacing RCS with BMCS in dairy cow diets has the potential to lower CH<sub>4</sub> production per unit of energy consumed or unit of milk produced. Possible influence of feeding RCS or BMCS-based diets on gaseous emissions from manure needs to be evaluated.

**Key Words:** CH<sub>4</sub> production, dairy cow, brown midrib corn silage

**T386 Determination of in vivo and in situ bioavailability of a rumen-protected lysine product, AjiPro-L.** Makoto Miura\*<sup>1</sup>, Atsushi Haruno<sup>1</sup>, Hiroyuki Sato<sup>1</sup>, Yuki Miyazawa<sup>1</sup>, Eri Ikegami<sup>1</sup>, Takeshi Fujieda<sup>1</sup>, and Izuru Shinzato<sup>2</sup>, <sup>1</sup>*Research Institute for Bioscience Products & Fine Chemicals, Ajinomoto Co. Inc., Kawasaki, Kanagawa, Japan*, <sup>2</sup>*Ajinomoto Heartland Inc., Chicago, IL*.

Past studies indicated that the 2nd generation AjiPro-L (A2G, Ajinomoto Co., Inc.) has higher bioavailability than the 1st generation product (A1G) based on in vivo plasma lysine technique (Whitehouse et al., 2014; Tucker et al., 2014). The objective of this study was to compare the other characteristics between A2G and A1G. Three ruminally fistulated lactating cows fed a diet composed of corn silage, grass silage and concentrates were used to determine in situ rumen stability. A1G and A2G were placed in Nylon bags (1 g per bag, 5 × 7 cm, pore size 53 ± 10 μm) and incubated in the rumen for 6, 12 and 24 h, respectively. After washing and drying bags, lysine (Lys) content in the residual products was analyzed. Three dry cows, fistulated both ruminally and

duodenally and fed a corn silage based diet, were used to determine rumen-bypassed and fecal excreted Lys from each product. A1G or A2G was ruminally administrated along with the highly protected L-arginine (HP-Arg; Robinson et al., 2011) as a control marker. Duodenal digesta was collected every 2 or 3 h for 48 – 54 h and was homogenized to extract free Lys and Arg. Changes of Lys and Arg concentrations in digesta were plotted to calculate the area under the curve (AUC). The proportion of AUC of Lys to Arg was defined as the bypass rate of AjiPro-L. To determine the fecal excretion, feces from each cow were collected for 72 h after ingestion of AjiPro-L. Feces were homogenized with hot water to extract free Lys from the products. Amounts of Lys excreted in feces were calculated by analyzing free Lys concentration in the extract. Statistical differences were tested by an ANOVA. In situ ruminal protection of A2G was lower ( $P < 0.01$ ) than A1G (97 vs. 99% at 6 h, 94 vs 97% at 12 h and 84 vs 90% at 24 h, respectively), but still reasonably high. The bypass rate of Lys from A2G (75 ± 11%) was not different ( $P = 0.45$ ) from A1G (82 ± 14%), but fecal excreted Lys from A2G (33 ± 6%) was significantly less ( $P < 0.01$ ) than A1G (51 ± 1%). Our results indicate that higher bioavailability of A2G than A1G was attributed to improved intestinal digestibility, supporting the results obtained by the in vivo plasma Lys response method.

**Key Words:** rumen-protected lysine, dairy cow, bioavailability

**T387 Handling characteristics of AjiPro-L in the practical use.** Makoto Miura\*<sup>1</sup>, Yuki Miyazawa<sup>1</sup>, Eri Ikegami<sup>1</sup>, Mizuki Tanida<sup>1</sup>, Takeshi Fujieda<sup>1</sup>, and Izuru Shinzato<sup>2</sup>, <sup>1</sup>*Research Institute for Bioscience Products & Fine Chemicals, Ajinomoto Co. Inc., Kawasaki, Kanagawa, Japan*, <sup>2</sup>*Ajinomoto Heartland Inc., Chicago, IL*.

When choosing a rumen-protected lysine product (RPL), not only the product efficacy but also handling stability in the practical use should be paid attention to. The 2nd generation AjiPro-L (A2G; Ajinomoto Co., Inc.) has been proven to possess higher bioavailability than the 1st generation product (A1G) based on in vivo plasma lysine technique (Whitehouse et al., 2014; Tucker et al., 2014). The objective of this study was to compare handling characteristics between A1G and A2G. A1G or A2G was mixed in a concentrate mix, and the mixing homogeneity was examined. The mix was loaded into a feed truck, transported for 160 km, unloaded into a feed bin, and was discharged. Samples were collected at the time of loading into the truck and discharging from the feed bin. The samples were sieved to separate the particles of A1G or A2G to calculate the inclusion rate. Results showed the inclusion rate of A2G remained unchanged between before and after transportation ( $P > 0.05$ ) while that of A1G decreased after transportation (4.0 vs. 3.6%,  $P < 0.05$ ), suggesting superior mixing homogeneity of A2G over A1G probably thanks to a smaller particle size of A2G (1 – 2 mm in diameter) than A1G (6 – 7 mm). Next, TMR mixing stability was evaluated with the same procedure as Ji et al. (2012). Dacron bags containing 1 g of RPL were mixed with TMR for 6 min in a Super Data Ranger. Then, bags were incubated in the rumen of 3 fistulated cows for 6, 12, and 24 h. After incubation, bags were washed and dried. Amounts of Lys in the residual product were analyzed for calculation of in situ protection. In situ protection of A2G mixed with TMR was slightly lower than A1G (97 vs. 99% at 6 h, 93 vs. 97% at 12 h and 85 vs. 91% at 24 h, respectively,  $P < 0.01$ ). The protection of both RPLs was not affected by mechanical mixing ( $P > 0.05$ ). Finally, storage stability of A2G was examined under –22 to 104°F by using the in vitro procedure (Miyazawa et al., 2014). In vitro ruminal protection or intestinal dissolution was not deteriorated at least for 6 mo under the ambient temperature tested.

Results demonstrate that A2G possesses sufficient handling stability for a use under practical conditions.

**Key Words:** rumen-protected lysine, handling, stability

**T388 Effects of corn silage hybrids on metabolic parameters and lactational performance of transition dairy cows.** A. W.

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Developing solutions to lessen metabolic stress experienced by transition dairy cows is very critical to improve lactational performance. The objective of this study was to determine metabolic parameters [concentrations of nonesterified fatty acids (NEFA) and  $\beta$ -hydroxybutyric acid (BHBA)] and lactational performance of dairy cows fed brown midrib corn silage (BMRCs)-based diets during the transition period when compared with conventional corn silage (CCS)-based diets. At 4 wk before parturition, 40 dry multiparous Holstein cows were randomly assigned treatments. The treatment groups consisted of 2 close-up transition diets (CCS-based and BMRCs-based diets) offered to 2 groups of 20 cows each. After calving, 10 cows from each prepartum group were individually fed either a CCS-based lactation diet or a BMRCs-based lactation diet. Four dietary treatments tested postpartum included: 1) CC = CCS-based close-up diet + CCS-based lactation diet; 2) CB = CCS-based close-up diet + BMRCs-based lactation diet; 3) BB = BMRCs-based close-up diet + BMRCs-based lactation diet; and 4) BC = BMRCs-based close-up diet + CCS-based lactation diet. Statistical analysis during the pre- and postpartum period parameters were performed by a split-plot-in-time design with repeated measures procedure using the Proc Mixed procedure of SAS with a model that included effects of treatment and week and their interaction and a random effect of cow within treatment. During the dry period, there was a tendency ( $P = 0.09$ ) for a diet  $\times$  test week interaction for DMI; initially, CCS-based diet had greater DMI, but as parturition neared, BMRCs-based diet increased DMI, resulting in greater DMI than CCS-based diet at parturition. Postpartum concentrations of BHBA and NEFA were lower for BB and BC diets, with CC and CB diets having the greatest concentrations of both BHBA and NEFA ( $P < 0.05$ ). In contrast, cows fed BB and BC diets increased milk yield compared with those fed CC and CB diets. There was a tendency for BB to have greater milk yield than CB on wk 1 (32.7 vs. 27.9 kg/d,  $P = 0.09$ ) and greater milk yield on wk 4 (45.1 vs. 38.5 kg/d). Overall data in the present study indicate that cows fed BMRCs-based diet in the dry period but CCS-based diet in lactation had positive carry-over effects on metabolic parameters and lactational performance possibly due to a consistent pattern of feed intake during dry period.

**Key Words:** brown-midrib corn silage, dairy cow, transition period

**T389 Effects of thermal processed clay in nonmedicated**

**rations in Japanese dairy farms.** Fang Chi\*<sup>1</sup>, Fumiaki Atarashi<sup>2</sup>, Kenji Wada<sup>2</sup>, Hiroshi Endo<sup>2</sup>, San Ching<sup>1</sup>, and LeAnn Johnston<sup>1</sup>, <sup>1</sup>Amlan International, Chicago, IL, <sup>2</sup>Okitama Food Animal Clinic, Federation of Agricultural Mutual Aid Association, Yamagata Prefecture, Japan.

Calibrin-Z (CZ), a thermally processed calcium montmorillonite, has been shown to enhance dairy productivity due to its barrier function in the digestive tract. Two classic dairy farms from different locations in Yamagata Prefecture, Japan, were used in this on-farm feeding trial. The objective was to investigate the effects of CZ feeding on lactating cow performance under a common dairy feeding regimen in Japan. Concentrate feed was purchased from local feed companies, corn and

grass silage was produced on-farm, and CZ was added to the Total Mixed Ration (TMR) with 20 g/cow/d for 3 mo. The TMR was formulated to meet Japanese nutrients requirements standard, and collected for mycotoxin analyses. Milk yield and quality and fecal firmness were recorded daily. Blood was drawn before and 30 d after CZ was fed, and albumin, BUN, GGT, hematocrit, and white blood cell counts were determined. Disease incidence and general health were recorded. Analysis of variance was used to determine the effects of CZ. Mycotoxin concentrations, such as aflatoxin, DON, fumonisin, and zearalenone were detectable in the TMR, but were below Japanese regulations. Manure firmness was numerically increased on one farm, but no changes were found on the other farm over the 3 mo feeding period. There were no differences in general blood chemistry; however, CD4+ and CD14+ were higher ( $P < 0.05$ ) after CZ was fed for 30 d. In addition, CD3+ and CD335+ were numerically higher ( $P > 0.05$ ) when CZ was included in the ration. Interestingly, a 47% (Farm A) or 40% (Farm B) reduction of disease incidences was observed as compared with the same period of the prior year. Among them, mastitis decreased 60% on Farm A, and enteritis decreased 100% on Farm B. Milk production was increased ( $P < 0.05$ ) on both farms (Farm A, 25.0 vs. 26.4 kg; Farm B, 27.0 vs. 28.4 kg/cow/d) after CZ was fed; while milk fat, protein, lactose, bacteria count, urea nitrogen, and somatic cell count remained unchanged. In conclusion, addition of Calibrin-Z to Japanese non-medicated rations may enhance general health in lactating cows; consequently, increased milk production and a reduction of disease occurrence were observed on the studied farms.

**Key Words:** dairy milk production, white blood cells, calcium montmorillonite

**T390 Associations of behavior and production in lactating**

**dairy cows.** Carleigh Johnston and Trevor J. DeVries\*, *Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada.*

Identification of the associations of cow behavior with productivity is important for supporting recommendation of strategies that optimize milk quality, efficiency, and yield. Thus, the objective of this study was to collate data from studies of the behavior of lactating dairy cows and identify associations between measures of behavior of dairy cattle and milk production and efficiency. A database containing behavior and production data for 132 dairy cow-week observations (mean of 7 d of consecutive data per cow) was assembled from 5 studies. Cows averaged (mean  $\pm$  SD) 1.8  $\pm$  0.9 lactations, 108.4  $\pm$  42.7 DIM, and 654.6  $\pm$  71.4 kg BW during each observation week. Production data included: DMI (27.0  $\pm$  3.1 kg/d), milk yield (43.0  $\pm$  7.0 kg/d), milk fat content (3.60  $\pm$  0.49%), milk fat yield (1.53  $\pm$  0.30 kg/d), milk protein content (3.05  $\pm$  0.25%), milk protein yield (1.29  $\pm$  0.17 kg/d), and production efficiency (1.60  $\pm$  0.22 kg milk/kg DMI). Behavioral data included: feeding time (230.4  $\pm$  35.5 min/d), feeding rate (0.13  $\pm$  0.03 kg/min), meal frequency (9.0  $\pm$  2.0 meals/d), meal size (3.2  $\pm$  0.9 kg/meal), daily mealtime (279.6  $\pm$  51.7 min/d), rumination time (516.0  $\pm$  90.7 min/d), and lying time (621.5  $\pm$  142.8 min/d). Data were analyzed in multi-variable mixed-effect regression models to identify which behavioral variables, when accounting for other cow-level factors (DIM, parity, BW), were associated with measures of production. DMI was associated with feeding time (+0.02 kg/min;  $P = 0.002$ ) and tended to be associated with rumination time (+0.003 kg/min;  $P = 0.1$ ) and meal frequency (+0.2 kg/meal;  $P = 0.06$ ). Similarly, milk yield was associated with feeding time (+0.03 kg/min;  $P = 0.005$ ) and rumination time (+0.02 kg/min;  $P < 0.001$ ), and tended to be associated with meal frequency (+0.3 kg/meal;  $P = 0.1$ ). Milk fat yield was associated with meal frequency



(+0.02%/meal;  $P = 0.05$ ). Efficiency of production was associated with rumination time (+0.0006/min;  $P < 0.001$ ) and tended to be associated with meal time per day (+0.0005/min;  $P = 0.09$ ). Overall, our results indicate that nutrition, management, and housing factors that improve time spent feeding, in more frequent meals, and time spent ruminating may have a positive impact on milk yield and component production, as well as production efficiency.

**Key Words:** behavior, production, rumination

**T391 Growth performance and sorting behavior of heifers offered diets with forage dilution.** Wayne Coblenz<sup>\*1</sup>, Nancy Esser<sup>2</sup>, Patrick Hoffman<sup>3</sup>, and Matt Akins<sup>3</sup>, <sup>1</sup>US Dairy Forage Research Center, Marshfield, WI, <sup>2</sup>University of Wisconsin, Marshfield, WI, <sup>3</sup>University of Wisconsin, Madison, WI.

Dairy heifers consuming high-quality forage diets are susceptible to excessive weight gains and over-conditioning, which often has been controlled by dilution with straw that is sortable by dairy heifers. Our objectives were: i) to compare the growth performance of dairy heifers offered a high-quality forage diet (CONTROL) with diets containing 1 of 3 diluting agents that included eastern gamagrass haylage (EGG), chopped wheat straw (STRAW), or chopped corn fodder (FODDER); and ii) evaluate sorting behaviors of heifers offered these forage diets. Holstein heifers ( $n = 128$ ) were stratified (32 heifers/block) on the basis of initial BW (heavy,  $560 \pm 27.7$  kg; medium-heavy,  $481 \pm 17.7$  kg; medium-light,  $441 \pm 22.0$  kg; and light,  $399 \pm 14.4$  kg), and then assigned to 1 of 16 identical research pens (4 pens/block; 8 heifers/pen) in a randomized complete block design with the 4 research diets as treatments. Diets were offered in a 118-d feeding trial with heifers crowded to 133% of capacity at the feed bunk. Compared with CONTROL, inclusion of low-energy forages was effective in reducing DMI (11.06 vs. 10.04 kg/d;  $P < 0.01$ ) and energy intake (7.39 vs. 5.95 kg TDN/d;  $P < 0.01$ ). Concentrations of physically effective fiber (pef) particles did not change during the 24-h period following feeding for either the CONTROL ( $P \geq 0.56$ ) or EGG ( $P \geq 0.75$ ) diets; however, this response for pef particles masked the competing (and cancelling) responses for individual large and medium particles comprising pef, which heifers sorted with discrimination and preference, respectively. Sorting against pef particles was detected for STRAW as a linear ( $P < 0.01$ ) function of time from feeding, and much more severely for the FODDER diet, which exhibited linear, quadratic and cubic effects ( $P < 0.01$ ). Sorting of forage particles by heifers could not be related directly to heifer performance. Compared with CONTROL, ADG was reduced by dilution in all cases (1.16 vs. 0.91 kg/d;  $P < 0.01$ ), but ADG for STRAW was approximately 0.2 kg/d less than EGG (0.98 kg/d) or FODDER (0.97 kg/d), despite exhibiting sorting characteristics intermediate between those diets.

**Key Words:** heifer, sorting, dilution

**T392 Response of lactating cows to a blend of essential oils and pepper extract.** Rayana B. Silva<sup>1</sup>, Renata A. N. Pereira<sup>3,2</sup>, Rafael C. Araújo<sup>4</sup>, and Marcos N. Pereira<sup>\*1,2</sup>, <sup>1</sup>Universidade Federal de Lavras, Lavras, MG, Brazil, <sup>2</sup>Better Nature Research Center, Ijaci, MG, Brazil, <sup>3</sup>Empresa de Pesquisa Agropecuária de Minas Gerais, Lavras, MG, Brazil, <sup>4</sup>GRASP Ind. e Com. LTDA, Curitiba, PR, Brazil.

Secondary plant compounds can modify rumen fermentation, cutaneous blood flow, sweat secretion, diet digestibility, and feeding behavior of dairy cows. Twenty-eight, individually fed Holsteins ( $181 \pm 102$  DIM) received a standard diet for 2 wk and 2 dietary treatments for 8 wk, in a

covariate adjusted randomized block design. Treatments were: Control (CTL) or a microencapsulated blend of essential oils and pepper extract (BEO: Carvacrol, cinamaldehyde, eugenol, and capsaicin. Activo Premium. 150 mg/kg DM). BEO decreased DMI (20.1 vs. 19.5 kg/d;  $P = 0.05$ ) and increased milk yield (30.1 vs. 30.8 kg;  $P = 0.04$ ), improving the milk to feed ratio (1.53 vs. 1.63;  $P < 0.01$ ). Milk fat content tended to increase with BEO ( $P = 0.10$ ), but protein, lactose, total solids, MUN, and SCC did not differ ( $P > 0.21$ ). BCS and BW were similar ( $P > 0.39$ ). BEO reduced ruminal acetate (57.8 vs. 51.4% of VFA;  $P = 0.01$ ) and increased propionate (26.1 vs. 31.3% of VFA;  $P = 0.02$ ), decreasing the A/P ratio (2.3 vs. 1.7;  $P = 0.03$ ). Total-tract digestibility of DM, OM, and NDF were similar ( $P > 0.20$ ), but there were trends for increased non-NDF OM digestibility ( $P = 0.07$ ) and digestible OM intake ( $P = 0.06$ ) on BEO. Cows on BEO ingested a greater proportion of the daily feed intake from 6AM to 12PM ( $P = 0.02$ ) and tended to have longer duration of the first meal ( $P = 0.08$ ), whereas CTL cows tended to ingest more from 7PM to 6AM ( $P = 0.06$ ). BEO induced selective sorting in favor of small feed particles ( $P < 0.01$ ) and refusal of large particles ( $P < 0.03$ ) in the morning. Ingestion and rumination times were similar ( $P > 0.19$ ). Viscosity of manure was reduced by BEO ( $P = 0.04$ ). Treatments had no effect on PUN along the day, plasma glucose content, and the urinary secretion of allantoin ( $P > 0.42$ ). BEO increased the concentration of  $O_2$  in jugular blood ( $P < 0.01$ ), but had no effect on pH,  $pCO_2$ ,  $HCO_3^-$ , total  $CO_2$ , and base excess ( $P > 0.46$ ). Sweating rate at d 14 was increased by BEO ( $P = 0.05$ ). No difference was detected in rectal and skin temperatures and in respiratory frequency ( $P > 0.21$ ). The supplementation of dairy cows with BEO affected feeding behavior, reduced the A/P ratio in rumen fluid, and improved feed efficiency.

**Key Words:** capsaicin, essential oil, feeding behavior

**T393 Supplementation of herbage-based diets with corn meal or liquid molasses changes the milk fatty acids profile in grazing dairy cows.** S. F. Reis<sup>1</sup>, A. F. Brito<sup>\*1</sup>, P. Y. Chouinard<sup>3</sup>, K. J. Soder<sup>2</sup>, and S. Ross<sup>1</sup>, <sup>1</sup>University of New Hampshire, Durham, NH, <sup>2</sup>USDA-Agricultural Research Service, University Park, PA, <sup>3</sup>Université Laval, Québec City, Québec, Canada.

Previous studies showed that feeding carbohydrate sources with different NSC profiles (e.g., starch vs. sucrose) and rates of ruminal degradation altered the milk fatty acids (FA) profile in dairy cows. This study evaluated the impact of corn meal (CM) or liquid molasses (MOL) on the milk FA profile of organically certified Jersey cows fed herbage-based diets. Ten multiparous (107 DIM) and 10 primiparous (131 DIM) lactating cows were blocked by parity, DIM, and milk yield and randomly assigned to 1 of 2 dietary treatments: CM or MOL. Diets were formulated to contain (DM basis): 74% herbage, 12% grass-legume mix hay, 12% CM or MOL, and 2% minerals and vitamins premix. The study lasted 110 d (June to September). Cows had access to pasture with mixed botanical composition (41.2% grass and 20% legume) for approximately 16 h daily in a strip grazing management system and were milked twice daily. Milk samples were collected monthly for 4 consecutive milkings, pooled by cow/period, and analyzed for milk FA using GLC. Herbage averaged (DM basis) 18.5% CP and 56.3% NDF, and 20% linoleic acid and 41.5%  $\alpha$ -linolenic acid (% total FA). Milk FA was expressed as % total milk FA. Milk proportions of most saturated and unsaturated FA were affected by month, which are explained by variation in herbage botanical and FA composition, and milk yield. The milk proportion of total saturated FA was highest in cows fed MOL (71.8 vs. 71%). Milk proportions of  $\alpha$ -linolenic acid (0.88 vs. 0.77%), *trans*-10,*cis*-12 CLA (0.090 vs. 0.078%), and PUFA (4.57 vs. 4.43%) were highest in cows fed MOL. Conversely, milk linoleic acid (1.12 vs. 1.72%), *trans*-10:18:1

(0.16 vs. 0.19%), *trans*-11 18:1 (2.62 vs. 2.76%), *cis*-9,*trans*-11 CLA (0.83 vs. 0.87%), and MUFA (22.8 vs. 23.7%) were lowest with MOL. The proportion of n-3 FA was highest (1.19 vs. 1.03%), whereas that of n-6 FA (1.30 vs. 1.35%) was lowest in cows fed MOL. Supplementing herbage-based diets with energy sources containing different NSC profiles resulted in marked changes in milk FA composition.

**Key Words:** milk fatty acids, molasses, herbage

**T394 Health, milk, milk components, milk quality and reproduction evaluated in Holstein cows fed OmniGen-AF from dry-off through 120 days in milk.** Amanda E. Holland<sup>\*1</sup>, Frank E. Rivera<sup>1</sup>, James D. Chapman<sup>1</sup>, and Lane O. Ely<sup>2</sup>, <sup>1</sup>Phibro Animal Health Corporation, Quincy, IL, <sup>2</sup>University of Georgia, Athens, GA.

Three hundred sixty-one multiparous late lactation cows were selected using DC305 and randomly assigned to 1 or 2 diet additives before dry-off; basal diet + placebo (PL, n = 181) or basal diet + OmniGen-AF (OG, n = 180). Diet groups were balanced by parity, DIM, calving date, 305ME, milk and health history. Both additives were fed at 56g/h/d, added directly to the dry and lactation TMR's. Cows were fed the assigned diet additives from dry-off through 120 d in milk (DIM) and health, milk, milk quality and reproduction evaluated. Data were analyzed by PROC GLM with significance tested at  $P < 0.05$  and only cows completing 120 DIM were included. Average parity and days dry for cows fed the PL and OG were 3.7, 59.3 d and 3.6, 58.3 d, respectively. Cows fed OG had fewer retained placenta (-6.6%,  $P = 0.023$ ) and metritis (-5%,  $P = 0.044$ ) cases than PL fed cows. Mastitis cases were similar, however cows fed OG tended to have fewer repeat cases ( $P = 0.107$ ). No differences were detected in the other recorded health metrics. Individual milk and milk components were recorded and were used to calculate week-4 milk (W4), energy and fat corrected milks (ECM, FCM). No differences were detected between PL and OG fed cows for test-day (TD) milk 1-4 (44.6kg vs. 45.5kg), W4 (43.5kg vs. 44.7kg) or peak milk (49.9kg vs. 50.4kg). OG fed cows produced more ECM (+1.5kg,  $P = 0.008$ ) and FCM (+1.7kg,  $P = 0.007$ ) through TD 1-4 than PL cows. OG cows had higher % milk fat (MF) at TD 1 (+0.3) and kg MF at TD 1 (+0.13), 2 (+0.09) and 3 (+0.09) and 0.05 kg more milk protein (MP) at TD 2 than PL cows. Compared with PL, the OG cows made milk with more kg MF (1.5 vs. 1.6,  $P = 0.002$ ), kg MP (1.3 vs. 1.5,  $P = 0.053$ ) and % MF (3.45 vs. 3.56,  $P = 0.032$ ). TD 1-4 SCC's were not different. All reproductive measures were not different, however OG cows tended to have fewer days to first breeding ( $P = 0.12$ ) and greater days carried calf ( $P = 0.088$ ) than PL cows. In conclusion, feeding OG to multiparous cows from dry-off through 120 DIM resulted in differences in peripartum disease incidence, milk and milk components.

**Key Words:** OmniGen-AF, health, components

**T395 Supplementing lactating cow diets with long chain fats has minimal effects on total-tract NDF digestibility: A quantitative review.** Kristina A. Weld<sup>\*</sup> and Louis E. Armentano, University of Wisconsin-Madison, Madison, WI.

Supplementing fat to lactating cows may reduce total-tract NDF digestibility (ttNDFd). The objective was to analyze the effects of different types of fat across studies. The numerical difference between ttNDFd and dry matter intake (DMI) for the supplemented minus control diet were calculated within study and these differences were analyzed in SAS 9.4 using Proc Mixed (Table 1). Studies were weighted based on the inverse of the standard error of ttNDFd squared. The model contained the

fixed effect of fat type and study as a random effect. Fatty acid content of treatment diets never exceeded 10% of DM (mean = 6.2, SD = 1.4). Supplementation of fats containing 12 and 14 carbon chain fatty acids (C12, C14) decreased both DMI and ttNDFd. Oil decreased ttNDFd by 2.1 percentage units, but the DMI decrease was not significant. No other type of fat consistently decreased ttNDFd, though calcium salts (n = 13 of palm oil, n = 6 of unsaturated C18) increased ttNDFd by 2.2 percentage units and decreased DMI by 1.45 kg/day. A regression analysis across all types of fat, using Proc Glim in SAS 9.4, of the difference in ttNDFd versus the difference in DMI within study shows a positive relationship. This suggests that the increase in ttNDFd for calcium salts is not explained through the decrease in DMI. Overall, these results indicate that the addition of a fat supplement, in which the fatty acids are C16 or greater in length, has minimal effects on ttNDFd. Feeding calcium salts in place of free oils would likely increase ttNDFd, but decrease DMI.

**Table 1 (Abstr. T395).** Effects of fat supplemented diets compared with control diets by fat type

Fat type	No. of studies	ΔttNDFd		ΔDMI,		ΔFA		content	
		percentage units	SE	P-value	kg/d	SE	P-value	%DM	SE
C12/C14	6	-11.32	1.67	0.0001	-4.04	0.66	0.0001	3.8	0.72
Oil	11	-2.13	0.97	0.04	-0.59	0.39	0.13	2.9	0.41
Animal vegetable blend	8	-1.15	1.36	0.41	-0.64	0.54	0.18	3.8	0.58
High C16	5	-0.31	1.43	0.83	-0.31	0.57	0.34	2.9	0.72
Tallow	22	-0.27	1.17	0.82	-0.88	0.47	0.07	2.5	0.62
Hydrogenated fat	14	0.42	1.36	0.76	0.42	0.54	0.37	3.4	0.48
Calcium salts	19	2.2	0.83	0.01	-1.48	0.33	0.0001	3.2	0.37

**Key Words:** NDF digestibility, fat, DMI

**T396 Sodium and potassium carbonates added to continuous cultures of ruminal microorganisms had similar effects on reducing biohydrogenation intermediates linked to milk fat depression.** Kaylin Young<sup>1</sup>, Elliot Block<sup>2</sup>, Joseph Harrison<sup>3</sup>, and Thomas Jenkins<sup>\*1</sup>, <sup>1</sup>Clemson University, Clemson, SC, <sup>2</sup>Arm and Hammer Animal Nutrition, Princeton, NJ, <sup>3</sup>Washington State University, Puyallup, WA.

In previous studies, the addition of potassium carbonate to continuous cultures of ruminal microorganisms decreased the production of *trans*-10 18:1 and *trans*-10,*cis*-12 conjugated linoleic acid (CLA), consistent with in vivo reports of higher milk fat percentages in lactating cows fed supplemental potassium carbonate. This experiment compared sodium carbonate to potassium carbonate to determine if they had similar effects on biohydrogenation intermediates. A single basal diet was fed (60 g/d) to 4 dual-flow continuous fermenters for four 10-d periods divided into 7 d for adaptation and 3 d for collection of samples. The diet consisted of alfalfa pellets and concentrate mix (47/53, DM basis), and contained 3.3% (DM basis) soybean oil. At each feeding (0800 and 1600 h), 16 mL were injected into each fermenter containing either only distilled water (CON), 1.4 g carbonate as  $K_2CO_3$ , 1.4 g carbonate as  $Na_2CO_3$ , or 1.4 g carbonate as a 50:50 mixture of  $K_2CO_3$  and  $Na_2CO_3$  (MIX). The experiment was analyzed as a randomized block design with differences among treatments determined by 3 orthogonal contrasts: (1) control vs. carbonate; (2) MIX vs. both single carbonates; and (3)  $K_2CO_3$  vs.  $Na_2CO_3$ . Carbonate sources did not differ ( $P > 0.05$ ) so MIX,  $K_2CO_3$ , and  $Na_2CO_3$  means were averaged to give a single combined carbonate value. CON and carbonate values differed ( $P < 0.05$ ) respectively for

culture pH (6.05 and 6.36), acetate (58.9 vs. 65.5 mol/100 mol), propionate (27.2 vs. 22.3 mol/100 mol), acetate/propionate (2.12 and 3.19), and production of *trans*-10 C18:1 (504.1 and 251.7 mg/d), *trans*-11 C18:1 (253.6 and 355.5 mg/d), and *cis*-9, *trans*-11 CLA (8.4 and 11.7 mg/d). CON and carbonate also differed ( $P < 0.10$ ) in production of *trans*-10, *cis*-12 CLA (19.7 and 12.8 mg/d). The results showed that sodium carbonate was equally effective as potassium carbonate in reducing the production of biohydrogenation intermediates that have been linked to the cause of milk fat depression.

**Key Words:** lipid, biohydrogenation, carbonates

**T397 Effects of starch content and fermentability, and culture pH on biohydrogenation of unsaturated fatty acids and NDF digestibility in batch culture.** Yan Sun\*, Michael S. Allen, and Adam L. Lock, *Michigan State University, East Lansing, MI.*

Effects of starch content (SC) and fermentability (SF), and culture pH on biohydrogenation (BH) of unsaturated fatty acids (FA) and NDF digestibility (NDFD) in batch culture were evaluated. Cultures (4 replicates/treatment/time point) were incubated at pH 5.8 or 6.2 for 0, 12, and 24 h and included alfalfa hay (55 or 70% of DM), and either dry ground corn (DC) or high moisture corn (HMC) as starch sources (30 or 45% of DM) for a SC of 22 or 33% DM. The alfalfa hay was treated with corn oil (2% of DM) to increase the total unsaturated FA content of cultures. Effects of SC, SF, culture pH, and their interactions were determined. BH extent for *cis*-9,*cis*-12 18:2 and NDFD increased over time across all treatments (both  $P < 0.001$ ). For main effects of treatments, 33% SC compared with 22% SC and pH 5.8 compared with pH 6.2 reduced BH extent of *cis*-9,*cis*-12 18:2 (35 vs. 39% and 23 vs. 51%, respectively; both  $P < 0.001$ ) and NDFD (13 vs. 16% and 8 vs. 21%, respectively; both  $P < 0.001$ ). Compared with DC, HMC decreased NDFD (13 vs. 16%;  $P < 0.001$ ). HMC increased BH extent of *cis*-9,*cis*-12 18:2 at pH 5.8, and decreased it at pH 6.2 compared with DC (interaction  $P < 0.001$ ) with no overall effect of SF ( $P = 0.90$ ). Contents of *trans*-10,*cis*-12 18:2 (CLA) and *trans*-10 18:1 increased over time for all treatments (both  $P < 0.001$ ). Overall, 33% SC compared with 22% SC and pH 5.8 compared with pH 6.2 increased content of CLA by 20 and 67% respectively (both  $P < 0.001$ ). Although SF did not affect CLA content overall ( $P = 0.13$ ), it interacted with time (interaction  $P < 0.001$ ); HMC increased CLA at 12 h, and decreased it at 24 h compared with DC. The 33% SC compared with 22% SC, HMC compared with DC, and pH 5.8 compared with pH 6.2, increased content of *trans*-10 18:1 by 17, 28, and 43% respectively (all  $P < 0.001$ ). In conclusion, higher SC and lower culture pH decreased BH of *cis*-9,*cis*-12 18:2 and NDFD, and increased formation of CLA and *trans*-10 18:1. Compared with DC, HMC increased formation of *trans*-10 18:1, decreased NDFD, and interacted with culture pH and time for BH of *cis*-9, *cis*-12 18:2 and the formation of CLA.

**Key Words:** biohydrogenation, starch, conjugated linoleic acid

**T398 The effect of replacing corn silage with sugarcane on milk yield and intake of lactating dairy cows: An analysis using CNCPS v6.5.** Edgar A. Collao-Saenz\*<sup>1</sup>, Andreas Foskolos<sup>2</sup>, Ryan J. Higgs<sup>2</sup>, Vera L. Banys<sup>1</sup>, Marcos N. Pereira<sup>3</sup>, and Michael E. Van Amburgh<sup>2</sup>, <sup>1</sup>*Universidade Federal de Goiás, Jataí, GO, Brazil*, <sup>2</sup>*Cornell University, Ithaca, NY*, <sup>3</sup>*Universidade Federal de Lavras, Lavras, MG, Brazil.*

Sugarcane is an option for feeding dairy cattle in the tropics. Reductions in DMI and milk yield (MY) are frequently reported when sugarcane replaces corn silage in dairy rations, due to the low NDF digestibility.

This meta-analysis was conducted to evaluate DMI and MY response of dairy cows to the replacement of corn silage with sugarcane. A database was compiled from 13 Brazilian publications involving 50 treatments from the last 12 years. The nutrient composition of the diets was calculated using CNCPS v. 6.5 feed library. Treatments were stratified in 3 categories: no replacement (SC0), 25–75% replacement (SCA) and 100% replacement (SCB) and the outcomes and predictions were analyzed with a mixed effects model (random: study; fixed: replacement level). Then, a mixed model regression analysis was used to investigate the potential relationships among reported nutrient composition of the diets (CP, NPN, NDF, NFC, Lignin and CNCPS estimated fractions) and MY and DMI as dependent variables. The best models were selected based on the highest BLUP ( $R^2_{BLUP}$ ) correlation coefficient and the lowest root mean square error (RMSE). Total replacement reduced MY (21.9 vs. 18.6 kg/d) and DMI (19.1 vs. 16 kg/d). DMI was the single most important factor explaining MY ( $R^2_{BLUP} = 0.973$ ; RMSE = 0.865). When CP and RUP (%CP) were included into the DMI model, accuracy and precision were increased ( $R^2_{BLUP} = 0.979$ ; RMSE = 0.75). Total NDF of the diet did not affect MY, but sugarcane NDF content affected it negatively. Sugar (CHOA4;  $R^2_{BLUP} = 0.948$ ; RMSE = 0.66) and sugarcane peNDF ( $R^2_{BLUP} = 0.941$ ; RMSE = 0.68) were the 2 primary factors negatively related to DMI. The replacement of corn silage with sugarcane reduced MY and DMI of dairy cows, diet sugarcane NDF content was the nutrient most related to the response.

**Key Words:** DMI, corn silage, NDF

**T399 Effect of dietary energy source and protein supply on dairy cow performance.** Helio Rezende Lima Neto<sup>1</sup>, Helene Lapierre<sup>2</sup>, and Lorraine Doepel\*<sup>1</sup>, <sup>1</sup>*University of Calgary, Calgary, Alberta, Canada*, <sup>2</sup>*Agriculture and Agri-Food Canada, Sherbrooke, Quebec, Canada.*

This study examined the interaction of dietary energy source and metabolizable protein (MP) supply on cow performance. Eight Holstein cows were used in a double 4 × 4 Latin square design with 21-d periods, with dietary energy source and MP supply tested in a factorial arrangement: high fiber, high protein (HFHP); high fiber, low protein (HFLP); high starch, high protein (HSHP); and high starch, low protein (HSLP). Diets were formulated to supply 100% of the NE<sub>L</sub> requirement, and 100% (HP) or 70% (LP) of MP requirement. There were no interactions between energy source and MP supply on the parameters measured. Intakes of NE<sub>L</sub> (Mcal/d) and MP (g/d) were: HFHP: 31.5, 2148; HFLP: 29.5, 1492; HSHP: 37.8, 2699; HSLP: 36.2, 2007. Milk yield (34.4 vs. 29.9 kg/d) and DMI (22.6 vs. 18.4 kg/d) were higher in cows fed HS compared with those fed HF, whereas DMI tended to be lower in cows fed LP vs. HP (19.9 vs. 21.1). Milk protein (3.04 vs. 2.85%) and lactose (4.62 vs. 4.56%) concentrations were higher in cows fed HS than those fed HF, but milk fat concentration was higher with HF compared with HS (4.22 vs. 3.49%). Milk protein (1.04 vs. 0.85 kg/d) and lactose (1.59 vs. 1.36 kg/d) yields were higher in cows fed HS than those fed HF. Milk protein yield tended to be higher with HP than with LP (962 vs. 925 g/d), and milk urea-N was higher with HP than LP (14.5 vs. 7.9 mg/dL). Dietary energy affected milk yield and composition more than protein supply did. Although diets were formulated to be iso-energetic, the large decrease in DMI in cows fed HF mainly drove the effect of HS vs. HF. The smaller effects of protein supply were not affected by energy source.

*Contd.*



**Table 1 (Abstr. T399).**

Parameter	Treatments				SEM	P-value	
	HFHP	HFLP	HSHP	HSLP		HF vs. HS	HP vs. LP
DMI, kg/d	19.0	17.8	23.1	22.0	0.85	<0.01	0.06
Yield, kg/d							
Milk	30.4	29.4	34.9	33.9	1.28	<0.01	0.20
Protein	0.87	0.83	1.06	1.02	0.04	<0.01	0.09
Fat	1.31	1.22	1.19	1.16	0.07	0.16	0.34
Lactose	1.38	1.34	1.62	1.57	0.06	<0.01	0.24
Milk composition, %							
Protein	2.87	2.85	3.04	3.04	0.06	<0.01	0.84
Fat	4.22	4.21	3.52	3.46	0.16	<0.01	0.80
Lactose	4.56	4.55	4.62	4.63	0.04	0.04	0.91
MUN, mg/dL	15.7	10.4	13.2	5.4	0.80	<0.01	<0.01

**Key Words:** energy source, metabolizable protein, milk yield

**T400 Effect of dietary energy source and protein supply on mammary amino acid metabolism of dairy cows.** Helio Rezende Lima Neto<sup>1</sup>, Helene Lapierre<sup>2</sup>, and Lorraine Doepel<sup>1\*</sup>, <sup>1</sup>University of Calgary, Calgary, AB, Canada, <sup>2</sup>Agriculture and Agri-Food Canada, Sherbrooke, QC, Canada.

The objective of this study was to determine the interaction of dietary energy source and metabolizable protein (MP) supply on AA arterial concentration and mammary arterial-venous difference (AVdiff). Eight Holstein cows were used in a double 4 × 4 Latin square design with 21-d periods. Energy source and MP supply were tested in a factorial arrangement with the following dietary treatments: high fiber, high protein (HFHP); high fiber, low protein (HFLP); high starch, high protein (HSHP); and high starch, low protein (HSLP). Diets were formulated to supply 100% of the NE<sub>L</sub> requirement, and 100% (HP) or 70% (LP) of MP requirement. Arterial concentrations of Ile, Leu, Phe, and the branched-chain AA (BCAA) were higher with HF compared with HS, regardless of MP supply, whereas concentrations of His, Leu, and BCAA were higher, or tended to be higher, with HP compared with LP. Group 1 AA-N (His, Met, Trp, Phe+Tyr) concentration tended to be higher with HP than LP but Group 2 AA-N (Ile, Leu, Ly, Val) concentration was not different between HP and LP. Metabolizable protein supply did not affect AVdiff except for Ile, Leu and BCAA, in which it was greater for HP than LP ( $P < 0.10$ ). Energy source did not affect AVdiff. The concentration of total essential AA-N (TEAAN) was higher with HF and HP relative to HS and LP, but AVdiff was not affected by treatment. Protein supply had a greater influence on AVdiff than did energy source, whereas energy source and MP supply influenced essential AA concentrations. Dietary energy source and MP affected several production variables; those data are presented in a companion abstract.

*Contd.*

**Table 1 (Abstr. T400).**

Arterial concentration, $\mu\text{M}$	Treatments				SEM	P-value	
	HFHP	HFLP	HSHP	HSLP		HF vs. HS	HP vs. LP
G1AAN	327.9	287.4	306.9	291.9	19.24	0.56	0.07
G2AAN	540.8	481.5	448.4	432.4	24.89	<0.01	0.12
BCAA	438.5	366.0	349.6	329.0	19.78	<0.01	0.02
TEAAN	881.3	799.5	792.7	748.3	40.19	0.06	0.09
Ile	91.4	92.0	75.8	75.9	4.95	<0.01	0.94
Lys	51.2	57.7	49.3	51.9	3.06	0.16	0.11
Met	22.9	21.9	22.8	21.1	1.07	0.62	0.13
Phe	59.2	51.6	47.2	49.7	3.08	0.02	0.36

**Key Words:** metabolizable protein, energy source, amino acid

**T401 Effect of supplemental level of Optigen on the milk performance and plasma biochemical indices of dairy goat.** Wang Hui<sup>1</sup>, Xue Neil<sup>2</sup>, and Luo Jun<sup>\*1</sup>, <sup>1</sup>Alltech-NWAFU Animal Science Research Alliance, Northwest A&F University, Yangling, Shaanxi, China, <sup>2</sup>Alltech China, Chaoyang District, Beijing, China.

The experiment was conducted to investigate the effects of different levels of Optigen, a new slow-release non-protein product, on milk performance and plasma biochemical indices of Xinong Saanen dairy goat, thus to select the optimum supplemental level in the diet. Forty-eight healthy lactating Xinong Saanen dairy goats with average BW of  $60 \pm 8.98$  kg were assigned to 4 groups with 12 goats in each group. Goats in the 4 groups were fed experimental diets containing 0, 0.25, 0.5, or 0.75% Optigen, respectively. There was a pretrial period of 1 wk followed by an experimental period of 16 wk. The results showed that (1) the dietary Optigen level did not significantly affect average daily feed intake ( $P > 0.05$ ); (2) goats in the group with dietary Optigen level of 0.5% have a significantly high average daily milk yield ( $P < 0.05$ ); (3) the dietary Optigen level had significant influences on milk fat ( $P < 0.05$ ), but there was no significant difference among 3 experimental group containing 0.25, 0.5, and 0.75% Optigen in the diet ( $P > 0.05$ ); milk protein percentage, milk lactose percentage and SNF percentage were all unaffected ( $P > 0.05$ ) by the supplementation of Optigen; (4) the dietary Optigen level had significant influences on plasma urea nitrogen ( $P < 0.05$ ), goats with 0.5% Optigen supplement in the diet demonstrated the lowest plasma urea nitrogen content, and followed by that with 0.75% and 0.25% Optigen supplement in the diet. Plasma triglyceride, total cholesterol and total protein were unaffected ( $P > 0.05$ ) by the supplementation of Optigen. In conclusion, dietary supplementation of Optigen can increase milk fat content and reduce plasma urea nitrogen content. 0.5% supplementation of Optigen in the diet can obtain a higher fat content and milk production.

**Key Words:** Optigen, milk performance, dairy goat

**T402 A sensory additive increases milk and protein responses to concentrate supplementation in grazing dairy cows.** R. Pulido<sup>1</sup>, M. Ruiz<sup>1</sup>, F. Bargo<sup>\*2</sup>, G. Tedó<sup>2</sup>, R. Cussen<sup>3</sup>, J. Acuña<sup>3</sup>, J. R. Roche<sup>4</sup>, and I. R. Ipharraguerre<sup>2</sup>, <sup>1</sup>UACH, Valdivia, Chile, <sup>2</sup>Luacta SA, Barcelona, Spain, <sup>3</sup>BestFed, Osorno, Chile, <sup>4</sup>Down to Earth Advice Ltd., Hamilton, New Zealand.

Forty 5 Holstein dairy cows (62 DIM, 501 kg BW) were assigned to a 3 × 2 incomplete Latin square (LS) design replicated 15 times to evaluate the effect of a sensory additive (ProEfficient, PE) on milk response to concentrate supplementation on pasture. Cows were blocked by parity

in 15 blocks and within blocks randomly assigned to 3 concentrate treatments: 1 kg/d mineral concentrate (MC); 7 kg/d starch-based concentrate (CC); and 7 kg/d of CC with 30 g/d of PE (PEC). Cows grazed a perennial ryegrass pasture during 56 d divided into 2 28-d periods. Targeted herbage allowance and pre grazing herbage mass were 25 kg DM/cow and 2400 kg DM/ha, respectively. Concentrates were fed twice daily at milking. The MC was composed of 60% wheat/barley/oat mix, 10% rapeseed meal, and 30% of a mineral premix, and averaged 10.4% CP, 17.2% NDF, and 64.3% in vitro DM digestibility (IVDMD). The CC and PEC were composed of 65% corn, 13.5% triticale/oat/wheat mix, 10% soybean meal, 5% rapeseed meal, 2% sugar beet molasses, and 4.5% mineral premix, and averaged 14.1% CP; 14.7% NDF, 88.4% IVDMD. Ryegrass pasture averaged 22.3% CP, 41.9% NDF, and 77.7% IVDMD. Data were analyzed using a mixed model that included the fixed effects of treatment, block, period, their 2-way interactions, and the random effect of cow within block. On average, supplementation with the starch-based concentrate increased ( $P < 0.05$ ) milk yield 4.4 kg/d compared with MC (24.3 vs. 28.7 kg/d, SEM 0.284). However, cows supplemented with PEC produced more ( $P < 0.05$ ) milk than CC cows (29.0 vs. 28.4 kg/d, respectively; SEM 0.284) because the milk production response to concentrate supplementation was improved 16% ( $P < 0.05$ ) by the addition of PE into the concentrate (0.79 kg vs. 0.68 kg milk/kg concentrate for PEC and CC, respectively). Supplementation with PEC also increased ( $P < 0.05$ ) milk protein percentage (3.74 vs. 3.43%, SEM 0.11) and yield (1.084 vs. 0.978 kg/d, SEM 0.039) compared with CC. The milk and protein responses triggered by PE might be associated with a reduction in the substitution rate of CC for pasture or changes in the concentration of the orexigenic hormone ghrelin.

**Key Words:** sensory additive, milk response to supplementation, grazing dairy cow

**T403 Effect of intensified milk feeding on immune status and hepatic energy metabolism of calves.** Christine T. Schäff<sup>1</sup>, Tadeusz Stefaniak<sup>2</sup>, Paulina Jawor<sup>2</sup>, and Harald M. Hammon<sup>1</sup>, <sup>1</sup>Leibniz Institute for Farm Animal Biology (FBN), Dummerstorf, Germany, <sup>2</sup>Faculty of Veterinary Medicine, Wrocław University of Environmental and Life Science, Wrocław, Poland.

Ad libitum milk feeding of neonatal calves improves growth, which is reflected by increased plasma concentrations of glucose, insulin, and IGF-I. Thus, we hypothesized that ad libitum milk replacer feeding during the first 5 wk of life affects hepatic energy metabolism and immune status before weaning in neonatal calves. Holstein calves were fed colostrum for 3 d followed by milk replacer (MR) feeding (125 g/l water) until wk 9 either restricted to 6 l/d all the time (RES;  $n = 14$ ) or ad libitum until d 35 and then stepwise reduced in wk 6 to 6 l/d and fed like RES thereafter (ADLIB;  $n = 14$ ). Calves had free access to concentrate and hay and were slaughtered at d  $60 \pm 2$ . Blood samples were taken on d 1, 2, and 7, then weekly until wk 8 for determination of plasma concentrations of immunoglobulin (Ig) G1, G2, M, haptoglobin, and fibrinogen. Liver samples were taken at slaughter to determine glycogen concentration and mRNA abundance of IGF-I, IGF and insulin receptor, IGF binding proteins 2, 3, and 4, glucose-6-phosphatase, pyruvate carboxylase, cytosolic phosphoenolpyruvate carboxykinase (*PCK1*), and glucose transporter GLUT 2 by quantitative real-time PCR. Data were analyzed by Mixed Model (plasma data) and General Linear Model (mRNA and glycogen data) of SAS. Plasma concentrations of all Ig equally increased ( $P < 0.05$ ) after first colostrum intake in both groups. Plasma concentrations of fibrinogen ( $P < 0.05$ ) increased during 1st wk in both group and tended to be greater ( $P < 0.1$ ) in RES than in ADLIB, whereas plasma haptoglobin concentrations were not affected

by feeding. Hepatic glycogen concentration and mRNA abundance were unaffected by feeding except abundance of *PCK1* was greater ( $P < 0.05$ ) in ADLIB than in RES indicating possible influence of intensified milk feeding on gluconeogenesis. The hepatic somatotrophic axis was not influenced at slaughter by different early feeding. Therefore, intensive milk feeding for 5 wk had no long-lasting effects on hepatic energy metabolism and Ig status in blood plasma, but may partly affect hepatic acute phase protein response in milk-fed calves.

**Key Words:** calf, ad libitum feeding, energy metabolism

**T404 Effect of lactation stage and rate of increase of concentrate allowance on rumen adaptation in dairy cows.** Kasper Dieho<sup>1</sup>, André Bannink<sup>2</sup>, Thomas J. Schonewille<sup>3</sup>, and Jan Dijkstra<sup>1</sup>, <sup>1</sup>Animal Nutrition Group, Wageningen University, Wageningen, the Netherlands, <sup>2</sup>Animal Nutrition, Wageningen UR Livestock Research, Wageningen, the Netherlands, <sup>3</sup>Department of Farm Animal Health, Faculty of Veterinary Medicine, Utrecht University, Utrecht, the Netherlands.

We studied the effects of lactation stage and postpartum rate of increase of concentrate allowance on daily fermentable organic matter (FOM) intake, rumen papilla surface area, and fractional VFA absorption rate ( $k_a$ VFA). The concentrate treatment aimed to create a transient difference in daily FOM intake. Twelve rumen-cannulated, Holstein-Friesian cows had free access to either a dry period ration or a basal lactation ration. Starting at 4 DIM, concentrate allowance increased at a high (1.0 kg DM/d; HIGH,  $n = 6$ ) or a low rate (0.25 kg DM/d; LOW,  $n = 6$ ) up to a maximum of 10.9 kg DM/d. On sampling days (-50, -30, -10, 3, 9, 16, 30, 44, 60, and 80 DIM), the rumen contents were evacuated and papilla biopsies taken. Thereafter,  $k_a$ VFA was measured using the empty washed rumen technique with 46 L McDougall buffer (pH 6.0, 39°C), containing 120 mM VFA (60% acetic, 25% propionic, and 15% butyric acid) and a marker (Co-EDTA). From -50 to 3 DIM, FOM intake was  $5.7 \pm 0.3$  kg/d ( $P = 0.29$ ), papilla surface area decreased from  $34.4$  to  $28.0 \pm 2.0$  mm<sup>2</sup> ( $P = 0.02$ ) and  $k_a$ VFA decreased from 0.51 to  $0.33 \pm 0.03$  /h ( $P < 0.01$ ), with no differences between future treatment groups HIGH and LOW ( $P \geq 0.33$ ). From 3 to 80 DIM, FOM intake, papilla surface area, and  $k_a$ VFA increased to  $15.0 \pm 0.3$  kg/d,  $63.4 \pm 2.0$  mm<sup>2</sup>, and  $0.75 \pm 0.03$  /h respectively (all  $P < 0.01$ ). A treatment by DIM interaction occurred for daily FOM intake ( $P < 0.01$ ), which was greater in group HIGH at 16 DIM ( $12.3$  vs.  $10.1 \pm 0.3$  kg/d,  $P < 0.01$ ), and for papilla surface area ( $P = 0.01$ ), which was greater in group HIGH from 16 DIM ( $46.0$  vs.  $33.2 \pm 2.0$  mm<sup>2</sup>,  $P < 0.01$ ) through to 44 DIM ( $60.5$  vs.  $49.7 \pm 2.0$  mm<sup>2</sup>,  $P = 0.01$ ). However, a treatment by DIM interaction did not occur for  $k_a$ VFA ( $P = 0.28$ ), which was similar for group HIGH and LOW at 16 DIM ( $0.60$  vs.  $0.56 \pm 0.03$  /h,  $P = 0.53$ ) and 44 DIM ( $0.69$  vs.  $0.69 \pm 0.03$  /h,  $P = 0.95$ ). In conclusion, lactation stage but not concentrate treatment affected  $k_a$ VFA. This suggests  $k_a$ VFA increases with, but is not limited by, papilla surface area during the first weeks of lactation, which was affected by the rate of increase of concentrate allowance.

**Key Words:** VFA absorption, rumen papillae

**T405 Breed and stage of lactation affect the content of bioactive fatty acids in milk.** Melissa L. Bainbridge<sup>1</sup>, Laura M. Cersosimo<sup>1</sup>, André-Denis G. Wright<sup>2</sup>, and Jana Kraft<sup>1</sup>, <sup>1</sup>University of Vermont, Burlington, VT, <sup>2</sup>University of Arizona, Tucson, AZ.

Dairy products are a significant contributor to the fat intake of the American diet and contain several bioactive fatty acids (FA) that possess health benefits. The objective of this study was to compare the

content and profile of bioactive FA in milk between Holstein (HO), Jersey (JE), and F<sub>1</sub> HO × JE crossbreds (CB). Twenty-two primiparous cows (n = 7 HO, n = 7 CB, n = 8 JE) were followed across a lactation. All cows were fed a consistent TMR at a 70:30 forage to concentrate ratio. Sample periods (SP) were defined as 3 d in milk (DIM), 93 DIM, 183 DIM, and 273 DIM. Milk sample and data collection occurred on days -2 to 2 relative to the SP. Data were analyzed using a repeated measures general linear mixed model in SAS. HO and CB, had a higher content of n-3 FA in milk fat than JE (0.60 and 0.63 vs. 0.47 g/100g FA; *P* < 0.01) and a lower n-6/n-3 ratio (3.2 and 3.1 vs. 3.8; *P* < 0.01). SP had an effect on the n-6/n-3 ratio of milk fat, with the lowest value observed at 3 DIM (2.6; *P* < 0.01) and the highest at 183 DIM (3.9; *P* < 0.01). The content of conjugated linoleic acids (CLA) in milk fat was affected by breed and SP. HO and CB had a higher proportion of CLA than JE (0.50 and 0.48 vs. 0.37 g/100g FA; *P* < 0.01). Overall, CLA content decreased over lactation (0.54, 0.45, 0.40, and 0.40 g/100g FA for 3, 93, 183 and 273 DIM, respectively; *P* < 0.01). Breed had no effect on content of total branched-chain fatty acids (BCFA) in milk fat, however, breed affected individual BCFA. Content of *iso* 14:0 in milk fat was lower in HO than in JE (0.10 vs. 0.13 g/100g FA; *P* < 0.05) and content of *iso* 17:0 was higher in HO and CB than in JE (0.33 and 0.31 vs. 0.27g/100g FA; *P* < 0.01). Total BCFA were affected by SP, with 93 and 183 DIM (1.51 and 1.59 g/100g FA) having a lower proportion of BCFA than 3 and 273 DIM (1.71 and 1.74 g/100g FA; *P* < 0.01). In conclusion, HO and CB exhibited a higher content of bioactive FA in milk than JE. Across a lactation, the greatest content of bioactive FA in milk occurred postpartum.

**Key Words:** branched-chain fatty acids, n-3 fatty acids, CLA

**T406 Rumen microbial protein outflow, and plasma amino acid levels, in early lactation multiparity Holstein cows on commercial California dairy farms.** Nadia Swanepoel<sup>\*1,2</sup>, Peter H. Robinson<sup>1</sup>, and Lourens J. Erasmus<sup>2</sup>, <sup>1</sup>*Department of Animal Science, University of California, Davis, CA*, <sup>2</sup>*Department of Animal and Wildlife Sciences, University of Pretoria, Pretoria, South Africa*.

Metabolic models are unlikely to accurately predict microbial crude protein outflow (MCP) from the rumen and amino acid (AA) limitations since they are generally based on static values of unknown accuracy. Practical and reliable methods to identify MCP and AA limitations are required to evaluate nutritional limitations in commercial groups of dairy cows in 'real time'. Our objective was to determine normal ranges of plasma AA, and MCP, in early lactation multiparity Holstein dairy cows fed a range of rations as a benchmark to determine high, low and normal levels. Multiparity high cow groups on 19 commercial dairies in California were selected. One load of total mixed ration (TMR) was collected at feeding, and milk yields were obtained from monthly milk tests on each dairy. Spot urine samples were collected from voluntarily urinating cows between 38 and 151 d in milk (DIM) to focus on early lactation. Specific gravity of each urine sample was determined to estimate urine flow. Urine samples from each dairy were analyzed for allantoin, which was used, with estimated urine volume, to calculate MCP. Blood was collected from the tail vein of 12 cows/dairy (65 to 76 DIM) and assayed for AA. Group averages among the dairies were 33.6 to 52.5 kg/d milk yield, 1309 to 2149 g/d MCP, 2.8 to 4.4, 8.4 to 13.9, 5.9 to 9.7, 22.9 to 38.5, 16.9 to 27.5 and 11.3 to 18.6 µg/mL for Met, Lys, His, Val, Leu, and Ile respectively. Correlation analysis was performed using the STEPWISE Procedure of SAS with a backward elimination of parameters. Milk and DIM were poor MCP predictors (*r*<sup>2</sup> < 0.01 and 0.05 respectively) and neutral detergent fiber (NDF; *P* = 0.02, *r*<sup>2</sup> = 0.23) was the only TMR nutrient that was predictive of MCP,

with higher diet NDF levels leading to increased MCP flow. Multiple correlation with all TMR nutrients (% DM) created the best fit model: MCP (g/d) = -5793.1 + (118.8 × NDF) + (150.1 × Fat) + (87.0 × Starch) + (74.0 × nonfiber carbohydrate) [*r*<sup>2</sup> = 0.50]. Results show that plasma AA (from 6 cows) and MCP (from 8 cows) are sufficient to determine whether plasma AA and/or MCP values in a group of cows are low, high, or normal thereby suggesting changes in rations to address the identified issue(s).

**Key Words:** allantoin, spot urine samples

**T407 Evaluating different physical concentrate forms in preweaned calves.** Marta Terré<sup>1</sup>, Maria Devant<sup>\*1</sup>, and Alex Bach<sup>2,1</sup>, <sup>1</sup>*IRTA, Caldes de Montbui, Spain*, <sup>2</sup>*ICREA, Barcelona, Spain*.

In Experiment 1, 37 crossbreed calves were used in a cafeteria study to evaluate calves preferences for different physical form presentation of concentrates. The same concentrate with 4 different physical presentations: meal (ML), pellet (PL), pellet and whole cereal grains (WHL), and pellet and flaked cereal grains (FLK) were simultaneously available from 4 to 45 d age (weaning). A preference diet ratio was calculated: daily concentrate DM intake for each concentrate presentation divided by daily total DM intake divided by the total number of concentrate offered, and an ANOVA with repeated measures was used to analyze the preference ratio. In Experiment 2, 63 Holstein calves were randomly distributed in 2x2 factorial design to evaluate performance of calves fed 2 different protein sources in the milk replacer (MR) and 2 concentrates with different presentation form: calves received either a MR containing dried whey as a protein source, and a pellet concentrate with whole grains of corn and barley or corn and barley flakes; or calves received a MR with dried skim milk as protein ingredient, and a pellet concentrate with whole grains of corn and barley or corn and barley flakes. Animals were weighed weekly, and intakes were recorded daily. Data were analyzed with an ANOVA with repeated measures. In Experiment 1, during the first week of study, calves had a lower (*P* < 0.01) preference for FLK than for WHL; however, thereafter calves showed the greatest (*P* < 0.01) preference for FLK at 2, 4, and 5 week of study. In Experiment 2, there were no differences between milk replacer types, and starter concentrate intake was greater (*P* < 0.05) in whole-grain than in flaked-grain concentrates from the 5 wk of study until weaning. However, no differences in growth or gain to feed ratio were observed. It is concluded that preweaned calves prefer concentrates based on pellets and flaked-grains, and when flaked-grains are compared with whole-grains, lower concentrate intake is observed, but their performance was not impaired.

**Key Words:** calves, physical form, preference

**T408 Fetal programming on dairy cows: Effect of dam's parity and days in milk at conception on first-lactation milk yield in dairy cows.** Ayelen Chiarle<sup>1</sup>, Ramiro Rearte<sup>1</sup>, Santiago Corva<sup>1</sup>, R. Luzbel de la Sota<sup>1,2</sup>, Mauricio Giuliodori<sup>1</sup>, and Alejandro Relling<sup>\*1,3</sup>, <sup>1</sup>*Fac. Cs Veterinarias, UNLP, La Plata, Argentina*, <sup>2</sup>*CCT La Plata, CONICET, Argentina*, <sup>3</sup>*IGEVET, CCT La Plata, CONICET, Argentina*.

Fetal programming is defined as nutritional and/or endocrine changes that happen in the cow during gestation that may change expression of some genes on the adult life of the progeny. These changes may have an impact on the progeny's performance. Therefore, the objective of the current study was to evaluate the effect of dam's parity (1 through 8) and days in milk (DIM) at conception on the daughter's first lactation milk yield accumulated up to 150 d (MY150, kg). Data from



dairy cows (n = 159.886) and their daughters (n = 201.547) from 1131 Argentinean dairy herds from ARPECOL (La Plata, BA, Argentina) were included in the study. Data were analyzed as completely randomized block design with a mixed model (SAS 9.3). The model included the fixed effects of parity (1 through 8) and DIM at conception, divided into quartiles (1st: >77 d, 2nd: 77–115 d; 3rd: 115–190 d; 4th: <190); and the random effects of dam and dairy herd (block). Mean DIM for each quartile were 57, 94, 148 and 296 d. Linear, quadratic and cubic polynomial contrasts were used for mean separation. The daughters' MY150 decreases (*P*-value Linear < 0.01, Quadratic < 0.01, Cubic > 0.10) when parity number increases. Mean MY150 were 2915, 2879, 2862, 2853, 2844, 2846, 2837 and 2835 kg for 1st, 2nd, 3rd, 4th, 5th, 6th, 7th and 8th parity, respectively. In addition, daughters' MY150 increases (*P*-value Linear < 0.01, Quadratic < 0.05, Cubic > 0.10) when DIM at conception increases. Mean MY150 were 2851, 2858, 2863, and 2863 kg for those conceived on average at 57, 94, 148 and 296 DIM, respectively. In summary, physiological changes related to parity and DIM at conception have an impact on progeny's MY150.

**Key Words:** dairy cattle, fetal programming, progeny milk yield

**T409 Transcriptome profile in cows resistant to milk fat depression.** Adriana Siurana<sup>1</sup>, Sergio Calsamiglia<sup>\*1</sup>, David Gallardo<sup>3</sup>, and Angela Canovas<sup>2</sup>, <sup>1</sup>*Animal Nutrition and Welfare Service, Universitat Autònoma de Barcelona, Bellaterra, Spain*, <sup>2</sup>*Servei Veterinari de Genètica Molecular, Universitat Autònoma de Barcelona, Bellaterra, Spain*, <sup>3</sup>*Departament de Genètica Animal, Centre de Recerca en Agrigenòmica, Universitat Autònoma de Barcelona, Bellaterra, Spain*.

Feeding linseed to dairy cows results in milk fat depression (MFD), but there is a wide range of sensitivity among cows. The objectives of this study were (1) To compare the mRNA expression of transcripts expressed in milk somatic cells in cows resistant or sensitive to MFD; (2) to identify metabolic pathways and transcription factors affected by MFD in resistant cows under different dietary conditions (no fat supplemented or linseed rich-diet). Four cows were selected from a dairy farm after a switch from a control diet to a linseed-rich diet. Among them, 2 cows (R-MFD) were resistant to MFD showing high milk fat content in both control (CTR) (4.06%) and linseed-rich diet (LIN) (4.36%); and 2 cows (S-MFD) were sensitive to MFD decreasing milk fat content after the change into the LIN diet (3.56 to 2.54%). Fresh milk samples were collected from each cow the week before and 2 weeks after the diet change. Transcriptome analysis was performed using RNA-sequencing technology with a HiSeq2000 platform. Differential expression analysis between cows S-MFD vs. R-MFD allowed to detect a large number of genes differentially expressed (DE) in both diets, CTR (n = 1,111) and LIN (n = 1,669). In addition, 726 genes were differentially expressed between CTR and LIN diets in cows R-MFD whereas only 38 genes were identified in cows S-MFD. Results showed an overexpression in genes and pathways related to fatty acid synthesis and lipid metabolism such as TR/RXR and PPAR $\alpha$ /RXR $\alpha$  Activation pathways in cows R-MFD (CTR vs. LIN). Several genes and transcription factors such as *FOXO3*, *MTOR*, *PPARA*, *PPARG*, *SREBF1* and *MYC* were identified acting as key regulators in R-MFD cows with linseed-rich diet. These results suggest that R-MFD cows could be activating a compensatory mechanism to increase the fatty acid synthesis in linseed-rich diets. It would be interesting to combine these results with the study of the structural variation in the whole transcriptome of S-MFD and R-MFD cows to contribute to the better understanding of molecular mechanisms affecting the MFD in cows.

**Key Words:** milk fat depression, fatty acid synthesis, RNA-sequencing

**T410 Apparent synthesis of vitamin B<sub>6</sub> and folates in the rumen of lactating dairy cows fed alfalfa or orchardgrass silages.** D. S. Castagnino<sup>\*1,2</sup>, M. Seck<sup>1,2</sup>, K. L. Kammes<sup>3</sup>, J. A. Voelker Linton<sup>3</sup>, M. S. Allen<sup>3</sup>, R. Gervais<sup>2</sup>, P. Y. Chouinard<sup>2</sup>, and C. L. Girard<sup>1</sup>, <sup>1</sup>*Agriculture and Agri-Food Canada, Sherbrooke, QC, Canada*, <sup>2</sup>*Département des Sciences Animales, Université Laval, Québec, QC, Canada*, <sup>3</sup>*Department of Animal Science, Michigan State University, East Lansing, MI*.

Effects of forage family (legume vs. grass) on apparent ruminal synthesis and post-ruminal supply of vitamin B<sub>6</sub> and folates were evaluated in 2 experiments. Diets containing either alfalfa (AL) or orchardgrass (OG) silages as the sole forage were offered to ruminally and duodenally cannulated lactating Holstein cows in crossover design experiments. Experiment 1 (E1) compared diets containing AL and OG (~27% total NDF) offered to 8 cows in 2 15-d treatment periods. Experiment 2 (E2) compared diets containing AL and OG (~30% total NDF) offered to 13 cows in 2 18-d treatment periods. Intake, duodenal flow and apparent ruminal synthesis (ARS) for each vitamin were analyzed using SAS mixed procedure. The experiments were analyzed separately. Differences were declared significant at *P* < 0.05. Vitamin B<sub>6</sub> intakes were greater for AL than OG in E1 (368 vs. 236 ± 18.2 mg/d; *P* < 0.01) and E2 (180 vs. 77 ± 5.1 mg/d; *P* < 0.01). In contrast, folates intakes were lower for AL than OG in E1 (7.6 vs. 9.2 ± 0.51 mg/d; *P* = 0.02) and E2 (8.7 vs. 10.2 ± 0.29 mg/d; *P* < 0.01). In E1, duodenal flows of vitamins were not affected by treatment (vitamin B<sub>6</sub>, 72 ± 6.2 mg/d, *P* = 0.63; folates, 33 ± 4.2 mg/d, *P* = 0.15). In E2, duodenal flows of vitamin B<sub>6</sub> were greater for AL than OG (59 vs. 31 ± 5.2 mg/d; *P* < 0.01) but the duodenal flows of folates were not affected by treatment (39 ± 2.7 mg/d; *P* = 0.78). Alfalfa increased the apparent ruminal degradation of vitamin B<sub>6</sub> compared with OG in E1 (-294 vs. -167 ± 14.5 mg/d; *P* < 0.01) and E2 (-121 vs. -46 ± 4.9 mg/d; *P* < 0.01). The ARS of folates were not affected by treatment in either experiment (E1: 25 ± 3.9 mg/d, *P* = 0.19; E2: 30 ± 2.5 mg/d, *P* = 0.81). Combining data from both experiments, vitamin B<sub>6</sub> intake was correlated positively with its duodenal flow (*r* = 0.68, *P* < 0.01) but negatively with ARS synthesis of the vitamin (*r* = -0.98, *P* < 0.01). Folates intake was correlated positively with its duodenal flow (*r* = 0.68, *P* < 0.01) and its ARS (*r* = 0.59, *P* < 0.01). Forage family affected the ruminal balance of vitamin B<sub>6</sub> but had little or no effect on the post-ruminal supply of vitamin B<sub>6</sub> and folates.

**Key Words:** dairy cow, vitamin B<sub>6</sub>, folates

**T411 Noninvasive indicators to identify lactating dairy cows with the greater risk of subacute rumen acidosis.** Xiaosheng Gao<sup>\*</sup> and Masahito Oba, *University of Alberta, Edmonton, Alberta, Canada*.

The objective of the study was to evaluate if milk urea nitrogen (MUN) and milk fat content could be used as the noninvasive indicator to identify cows with a greater or lower risk of subacute ruminal acidosis (SARA). Our hypothesis was that cows greatly vary in their risk to develop SARA even if they are fed a common diet, and that cows with lower MUN and milk fat content would have a greater risk of SARA while cows with higher MUN and milk fat content would have a lower risk of SARA. In a screening study, 35 late-lactating Holstein cows (DIM = 250 ± 71.1; BW = 601 ± 45.4 kg) were fed a high grain diet containing 35% forage and 65% concentrate mix ad libitum for 21 d. Concentration of MUN ranged from 5.7 to 13.9 mg/dL among the 35 cows, and the average milk fat content was 3.5%. Then, 5 cows with highest MUN concentrations with milk fat higher than 3.5% were selected as animals that presumably have a low risk of SARA (LOW), and 5 cows with lowest MUN concentrations with milk fat less than 3.5% were selected as animals

that presumably have a high risk of SARA (HIGH). Then these 10 animals were ruminally cannulated during the subsequent dry period. As one LOW cow was culled due to fatty liver, 9 animals (DIM = 122 ± 33.2; BW = 615 ± 49.1 kg) were used for the subsequent study in the following lactation. All cows were fed a high grain diet consisting of 35% forage and 65% concentrate mix ad libitum for 21 d. Ruminal pH was measured every 30 s for 72 h. All response variables were evaluated for the group effect using the PROC TTEST procedure of SAS (version 9.2, SAS Institute Inc., Cary, NC). Minimum (5.75 vs. 5.30;  $P = 0.02$ ) and mean ruminal pH (6.35 vs. 6.04;  $P = 0.02$ ) was higher for LOW compared with HIGH animals. In addition, duration of rumen pH below 5.8 was shorter in LOW animals (52.5 vs. 395 min/d;  $P = 0.04$ ). These results suggested that MUN and milk fat content in late-lactating cows fed a high grain diet may be used to identify cows that have a higher or lower risk of SARA.

**Key Words:** subacute ruminal acidosis, milk urea nitrogen, milk fat

**T412 Effects of different dosages of *Saccharomyces cerevisiae* fermentation product on lactation performance of dairy cows under heat stress.** Wen Zhu\*<sup>1</sup>, B. X. Zhang<sup>1</sup>, K. Y. Yao<sup>1</sup>, I. Yoon<sup>2</sup>, Ruby Chung<sup>2</sup>, J. K. Wang<sup>1</sup>, J. A. Ye<sup>1</sup>, and J. X. Liu<sup>1</sup>, <sup>1</sup>*Institute of Dairy Science, College of Animal Science, Zhejiang University, Hangzhou, China*, <sup>2</sup>*Diamond V, Cedar Rapids, IA*.

The objective of this study was to evaluate the effects of different dosages of *Saccharomyces cerevisiae* fermentation product (SCFP; Original XP; Diamond V, Cedar Rapids, IA) on lactation performance of mid-lactation dairy cows under heat stress. Eighty-one multiparous Holstein lactating dairy cows were divided into 27 blocks of 3 cows each based on milk yield (23.6 ± 0.20 kg/d), parity (2.88 ± 0.91) and days in milk (204 ± 46 d). The cows were randomly assigned within blocks to one of 3 dietary treatments: 0 (control), 120, or 240 g/d of SCFP mixed with 240, 120, or 0 g of corn meal, respectively. The experiment lasted for 9 weeks, using the first week as adaption period. All data were analyzed through the PROC MIXED program of SAS with the covariance type AR (1) for repeated measures. Means were separated using the PDIF option in the LSMEANS statement. Linear and quadratic effects of treatment were tested using orthogonal polynomial contrasts. During the experimental period, average daily Temperature-Humidity Index (measured at 08:00, 14:00 and 20:00) was above 72 for 47 out of 56 d (84%). Rectal temperatures tended to decrease linearly ( $P = 0.07$ ) for cows supplemented with SCFP compared with control cows at 14:30, but were not different at 06:30 ( $P > 0.05$ ). Dry matter intake was not affected ( $P > 0.05$ ) by SCFP supplementation. Milk yield increased linearly ( $P = 0.02$ ) with increasing dosage of SCFP. Feed efficiency (milk yield/dry matter intake) was highest ( $P = 0.04$ ) for cows fed 240 g/d SCFP. Net energy balance also increased linearly ( $P < 0.01$ ) with increasing dosage of SCFP. Cows supplemented with SCFP gained ( $P < 0.01$ ) body weight, while control cows lost body weight. Concentrations of milk urea nitrogen decreased linearly ( $P < 0.01$ ) with increasing dosage of SCFP, while no difference was observed among the treatments in conversion of dietary N to milk N. In summary, supplementation of SCFP alleviated the negative effect of heat stress in lactating Holstein dairy cows and allowed cows to maintain higher milk production, feed efficiency and net energy balance. Effects of SCFP were dose-dependent and greater effects were observed from higher doses.

**Key Words:** heat stress, yeast culture, lactation performance

**T413 Effects of *Saccharomyces cerevisiae* fermentation products on lactation performance, rumen fermentation and microbial communities in dairy cows fed a diet containing low quality forages.** Wen Zhu\*<sup>1</sup>, Z. H. Wei<sup>1</sup>, N. N. Xu<sup>1</sup>, Fan Yang<sup>1</sup>, I. Yoon<sup>2</sup>, Ruby Chung<sup>2</sup>, J. K. Wang<sup>1</sup>, J. A. Ye<sup>1</sup>, and J. X. Liu<sup>1</sup>, <sup>1</sup>*Institute of Dairy Science, College of Animal Science, Zhejiang University, Hangzhou, China*, <sup>2</sup>*Diamond V, Cedar Rapids, IA*.

The objective of this study was to evaluate the effects of various dosages of *Saccharomyces cerevisiae* fermentation product (SCFP; Original XP; Diamond V) on lactation performance, rumen microbial communities and rumen fermentation in mid-lactation dairy cows fed a total mixed ration containing low quality forages. Eighty multiparous Holstein dairy cows were blocked based on days in milk (180 ± 45 d), parity (3.24 ± 1.06) and milk yield (26.6 ± 0.79 kg/d), and were randomly assigned to one of 4 treatments. Treatments consisted of 0, 60, 120, or 180 g/d of SCFP per head mixed with 180, 120, 60, or 0 g of corn meal, respectively. The forage in the basal diet was comprised (DM basis) of 15% corn stover, 7% Chinese wild ryegrass, and 17.3% corn silage. The experiment lasted for 10 weeks, with the first 2 weeks for adaption. The data were analyzed using the PROC MIXED program of SAS with the covariance type AR (1) for repeated measures. Means were separated by using the PDIF option in the LSMEANS statement. Orthogonal polynomial contrasts were used for linear and quadratic effects testing. Dry matter intake decreased linearly ( $P = 0.02$ ), while milk yield increased linearly ( $P = 0.05$ ) with the increasing dosage of SCFP, resulting in improved ( $P < 0.01$ ) feed efficiency compared with the control. Contents of milk components were similar ( $P > 0.05$ ) among the treatments. Neither rumen pH ( $P = 0.70$ ) nor ammonia-nitrogen concentration ( $P = 0.82$ ) was affected by SCFP supplementation. Increasing dosage of SCFP linearly increased ( $P < 0.05$ ) concentrations of ruminal total volatile fatty acids, acetate, propionate and butyrate, with no difference ( $P > 0.05$ ) in molar proportion of individual acids. The population of fungi and cellulolytic bacteria increased linearly ( $P < 0.01$ ), but those of lactate-utilizing and lactate-producing bacteria decreased linearly ( $P < 0.01$ ) with increasing dosage of SCFP. The estimated microbial protein yield in the rumen increased linearly ( $P < 0.01$ ) in response to SCFP supplementation. In summary, supplementation of SCFP improved the lactation performance of dairy cows consuming diets containing low quality forages by optimizing rumen fermentation through manipulating rumen populations. Effects of SCFP were dose-dependent and greater effects were observed from higher doses.

**Key Words:** low-quality forage, yeast culture, lactation performance

**T414 Response of dairy cows to monensin on diets differing in starch content and source.** Eugenio F. Barbosa<sup>1</sup>, Julia D. L. Dias<sup>1</sup>, Fabiana F. Cardoso<sup>1</sup>, Túlio H. R. Souza<sup>1</sup>, Lucas C. Resende<sup>1</sup>, Ozana F. Zacaroni<sup>1</sup>, Renata A. N. Pereira<sup>3,2</sup>, and Marcos N. Pereira\*<sup>1,2</sup>, <sup>1</sup>*Universidade Federal de Lavras, Lavras, MG, Brazil*, <sup>2</sup>*Better Nature Research Center, Ijaci, MG, Brazil*, <sup>3</sup>*Empresa de Pesquisa Agropecuária de Minas Gerais, Lavras, MG, Brazil*.

The response of dairy cows to monensin can interact with diet composition. We evaluated the response of lactating cows to monensin supplementation of diets differing in rumen fermentable starch. Twenty-eight Holsteins (157 ± 76 DIM) were individually fed a standard TMR for 3 wks and monensin (MON. 300 mg/d) or control (CTL) for 9 wks, in a covariate adjusted randomized block design. A low-starch diet (LS. 25.8% starch in DM) was offered from d 1 to 35 and a high-starch diet (HS. 30% starch in DM) from d 36 to 63. HS was formulated by replacing whole cottonseed and finely ground mature corn by an isonitrogenous mixture of soybean meal and high moisture corn. Data were collected

daily or after adaptation to LS and HS, and were analyzed as repeated measures with PROC MIXED of SAS. The mean square for cow nested within treatment (MON vs. CTL) was the error term for the treatment effect. Digestible OM intake (DOMI) was increased by HS (15.3 vs. 14.4 kg/d,  $P < 0.01$ ), but was similar for MON (14.6 kg/d) and CTL (15.1 kg/d,  $P = 0.41$ ). Milk yield did not respond to MON (32.3 kg/d,  $P = 0.32$ ). MON reduced DMI and increased Milk/DMI, ECM/DMI, and ECM/DOMI only when HS was fed ( $P < 0.01$  for the interaction of treatment and wk/diet). Plasma glucose content was increased by MON and the response interacted similarly with wk/diet ( $P < 0.05$ ). MON reduced the A/P ratio in rumen fluid in both diets (2.43 vs. 1.81,  $P < 0.01$ ) and HS reduced ruminal pH ( $P < 0.01$ ) and increased protozoa content ( $P < 0.01$ ). MON reduced total-tract NDF digestibility more in LS (52.1 vs. 41.3% of intake) than in HS (57.0 vs. 52.0,  $P < 0.05$  for the interaction). The daily urinary allantoin excretion did not respond to MON ( $P = 0.40$ ), neither PUN at 0, 1.5, and 3 h post feeding ( $P > 0.27$ ). MON induced lower degree of refusal of long feed particles and of preferential intake of short particles in the afternoon than CTL ( $P < 0.05$ ). Ingestion time (min/d and min/kg of DMI) and the number of daily meals were reduced by HS ( $P < 0.05$ ). HS increased pCO<sub>2</sub>, HCO<sub>3</sub><sup>-</sup>, total CO<sub>2</sub>, and base excess of jugular blood ( $P < 0.04$ ) and tended to reduce blood pH ( $P = 0.10$ ). The positive effect of MON on feed efficiency occurred only when ruminally available starch was increased in the diet.

**Key Words:** monensin, starch, feed efficiency

#### T415 Growth performance of dairy heifers fed reduced-fat distillers grains in replacement of forage in limit-fed rations.

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Our objective was to determine the maximum inclusion rate of reduced-fat distillers dried grains (RFDDGS) in dairy heifer rations. A 16-wk randomized complete block design study was conducted using 48 Holstein heifers (199 ± 2 d of age) to evaluate effects of diet on growth performance. Treatments were 30% RFDDGS (30DG), 40% RFDDGS (40DG), and 50% RFDDGS (50DG) with the rest of the diets consisting of grass hay and 1.5% mineral mix. Using Calan gates rations were limit-fed at 2.65, 2.50, and 2.35% of BW (DM basis) for 30DG, 40DG, and 50DG, respectively to have similar intakes of CP and energy among treatments. Frame sizes, BW, and BCS were measured on 2 consecutive days during wk 0, 2, 4, 6, 8, 10, 12, 14, and 16 of the feeding period. Data were analyzed using the MIXED procedures with repeated measures in SAS 9.4. Changes per day were analyzed by regression analysis. There were no interactions of treatment by wk. Heifer DMI linearly decreased ( $P < 0.01$ ) with increased dietary concentrations of RFDDGS (6.49, 6.21, and 5.84 kg/d; SEM = 0.117 for 30DG, 40DG, and 50DG, respectively). Body weights (264.1, 266.2, and 266.4 kg; SEM = 4.98) and ADG (0.89, 0.94, and 0.97 ± 0.083 kg/d) were similar ( $P > 0.05$ ) among treatments. Gain to feed (0.14, 0.16, and 0.18 kg; SEM = 0.0034) linearly increased ( $P < 0.01$ ). Hip height (124.8, 124.7, and 124.8 cm; SEM = 0.36) and hip width (35.6, 35.8, and 35.8 cm; SEM = 0.57) were similar ( $P > 0.05$ ) among treatments. Body length (112.5, 112.9, and 113.1 cm; SEM = 0.18) tended ( $P = 0.06$ ) to linearly increase across treatments. There was a quadratic effect ( $P < 0.05$ ) for withers height (120.9, 121.7, and 121.6 cm; SEM = 0.29), paunch girth (172.5, 173.9, and 172.5 cm; SEM = 1.29), and BCS (3.11, 3.12, and 3.07; SEM = 0.018), and a quadratic tendency ( $P = 0.08$ ) for heart girth (140.9, 140.6, and 140.9 cm; SEM = 0.41). Limit-feeding diets with greater inclusion rates of RFDDGS resulted in improved gain to feed ratios

and maintained frame growth without increasing BCS, demonstrating that replacing forage with RFDDGS does not negatively affect heifer growth performance.

**Key Words:** distillers grains, dairy heifer, growth performance

#### T416 Ruminal degradation and intestinal digestibility of camelina and carinata meal compared with other protein sources.

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Our objective was to determine DM and CP ruminal degradability and intestinal digestibility of camelina meal (CM) and carinata meal (CR) compared with other oilseed meals, soybean meal (SBM) and reduced-fat distillers dried grains (DG). In situ measurements were done using 3 multiparous, ruminally-cannulated Holstein cows (BW 848.6 ± 94.7 kg). Six feeds were evaluated: CM, CR, canola meal (CN), linseed meal (LN), DG, and SBM. Duplicate 5 g samples were weighed into 10 × 20 cm nylon bags and ruminally incubated for 0, 2, 4, 8, 12, 24 and 48 h. Six additional bags of each feed were incubated at 12 h for use in determination of in vitro intestinal digestibility of CP. Residues were incubated with pepsin and pancreatin solutions for 1 h and 24 h, respectively. Rumen degradation constants for DM and CP were estimated using the NLIN procedures in SAS 9.3. Intestinally digestible protein (IDP), intestinally absorbable dietary protein (IADP = ruminally undegradable protein (RUP) × IDP), and total digestible protein (TDP = ruminally degradable protein (RDP) + IADP) were evaluated using MIXED procedures in SAS. Ruminally degradable DM (RDDM) was greatest in CM, CR, and SBM. The CM and CR had the greatest RDP and least RUP of the feeds. The IDP was less in CM and CR compared with SBM and LN, but greater than CN and DG. The CR and CM had less IADP compared with the other feeds. The TDP was similar for CM and CR compared with SBM and LN, but greater than CN and DG. Overall, results indicate that CM and CR are highly degradable in the rumen and are comparable protein sources to SBM and LN for total digestibility.

**Table 1 (Abstr. T416).**

Item	CM	CR	CN	LN	DG	SBM	SEM
CP, % DM	40.9	43.9	45.3	40.4	35.0	53.9	—
RDDM, % of DM	65.0 <sup>a</sup>	63.0 <sup>a</sup>	50.9 <sup>c</sup>	55.8 <sup>b</sup>	50.8 <sup>c</sup>	65.0 <sup>a</sup>	0.64
RDP, % of CP	76.4 <sup>a</sup>	70.5 <sup>b</sup>	52.0 <sup>d</sup>	61.2 <sup>c</sup>	44.1 <sup>e</sup>	58.4 <sup>c</sup>	0.95
RUP, % of CP	23.6 <sup>e</sup>	29.4 <sup>d</sup>	48.0 <sup>b</sup>	38.8 <sup>c</sup>	55.9 <sup>a</sup>	41.6 <sup>c</sup>	0.95
IDP, % of RUP	80.9 <sup>b</sup>	80.9 <sup>b</sup>	70.9 <sup>c</sup>	81.6 <sup>b</sup>	63.2 <sup>c</sup>	90.5 <sup>a</sup>	2.61
IADP, % of CP	19.1 <sup>c</sup>	23.8 <sup>c</sup>	34.0 <sup>ab</sup>	31.7 <sup>b</sup>	35.4 <sup>ab</sup>	37.6 <sup>a</sup>	1.42
TDP, % of CP	95.5 <sup>a</sup>	94.4 <sup>a</sup>	86.0 <sup>b</sup>	92.9 <sup>a</sup>	79.5 <sup>c</sup>	96.0 <sup>a</sup>	1.22

<sup>a-c</sup>Values with unlike superscripts differ by  $P < 0.05$ .

**Key Words:** camelina meal, carinata meal, digestibility

#### T417 The decline in digestive efficiency of US dairy cows during the last 44 years. Sarah B. Potts\*, Melissa Shaughnessy, and Richard A. Erdman, University of Maryland, College Park, MD.

Since 1970, US milk production per cow has more than doubled resulting in large increases in feed intake. It is well established that increasing feed intake reduces diet digestibility. Our objective was to determine if the digestive efficiency of US dairy cows has also changed in the last 44 years. We assembled a data set consisting of diet digestibility measured via total collection or iNDF procedures with lactating dairy cows from studies published in the *Journal of Dairy Science* from 1970



to 2014. The data set contained 121 treatment means from 38 studies conducted at 16 US institutions. Based on regression analysis, milk yield and DMI increased by 20 kg/d and 8 kg/d between 1970 and 2014, respectively. Temporal effects on digestibility (DM, CP, or NDF) were determined using the regression model: Digestibility = YEAR<sub>1970</sub> + CP + NDF; where YEAR<sub>1970</sub> is the publication year minus 1970 and CP and NDF are diet constituents (% of diet DM) included to account for their effects on digestibility. DM digestibility decreased 0.13 percentage units per year ( $P < 0.01$ ) for a total reduction of 5.7 percentage units since 1970. However, there were no temporal effects on CP and NDF digestibilities ( $P > 0.3$ ). To account for the potential effect of feed intake on digestibility, DMI as a percentage of BW (DMI<sub>BW</sub>) was included in the regression model. With DMI<sub>BW</sub> in the model, DM digestibility decreased 0.08 percentage units per year ( $P = 0.03$ ) for a total reduction of 3.5 percentage units. This suggests that only a portion (39%) of the decline in digestibility was due to level of feed intake. In contrast, CP digestibility was unchanged over time ( $P = 0.94$ ), and NDF digestibility increased 0.14 percentage units per year ( $P = 0.05$ ) when the model included DMI<sub>BW</sub>. Increased NDF digestibility was likely due to the use of more highly digestible NDF sources in more recent studies. These results suggest that digestive efficiency has decreased by nearly 6 percentage units over the last 44 years. Perhaps genetic selection of cows for high milk production has inadvertently resulted in cows with high feed intake capacity irrespective of effects on digestive efficiency.

**Key Words:** digestibility, digestive efficiency, dairy cow

**T418 Ruminal degradation and intestinal digestibility of microbially treated soybean meal and dried distillers grains compared with the original feeds.** Jennifer L. Casperson<sup>1</sup>, Jill L. Anderson\*<sup>1</sup>, Jason R. Croat<sup>2</sup>, and William R. Gibbons<sup>2</sup>, <sup>1</sup>Dairy Science Department, South Dakota State University, Brookings, SD, <sup>2</sup>Department of Biology and Microbiology, South Dakota State University, Brookings, SD.

Our objective was to determine if microbial (fungal) treatment of soybean meal (SBM) and dried distillers grain with solubles (DG) affected ruminal degradation and intestinal digestibility of DM and CP. In situ measurements were done using 3 mid-lactation, multiparous, ruminally-cannulated Holstein cows (BW 847 ± 96). Four feeds were evaluated: SBM, treated SBM (TSBM), DG, and treated DG (TDG). Duplicate 5 g samples in nylon bags (10 × 20 cm) were incubated in the rumen for 0, 1, 2, 4, 8, 12, 24 and 48 h. Ruminal degradation constants for DM and CP were estimated using the NLIN procedures in SAS 9.3. Six additional bags of each feed were ruminally incubated for 12 h. Residues were used to determine in vitro intestinal digestibility of CP via incubation with pepsin and pancreatin solutions for 1 h and 24 h, respectively. Intestinally digestible protein (IDP), intestinally absorbable dietary protein (IADP = Ruminal undegradable protein (RUP) × IDP), and total digestible protein (TDP = Ruminal degradable protein (RDP) + IADP) were compared using MIXED procedures in SAS. Total CP was more in TSBM and TDG compared with original feeds. Ruminally degradable DM (RDDM) was less in TSBM and DG than in SBM, TDG. The RDP was increased in TDG versus DG and decreased in TSBM versus SBM. Effects were opposite for RUP. The IDP, IADP were less in DG and TDG compared with SBM and TSBM. The TDP was greatest in SBM and least in DG. Results indicate that microbial-treatment increased protein content of SBM and DG. Digestibility of CP was increased for TDG versus DG, but was not increased in TSBM versus SBM.

Contd.

**Table 1 (Abstr. T418).**

Item	SBM	TSBM	DG	TDG	SEM
CP, % DM	49.3	59.0	29.5	42.5	—
RDDM, % of DM	67.3 <sup>a</sup>	54.1 <sup>b</sup>	54.3 <sup>b</sup>	67.1 <sup>a</sup>	1.27
RDP, % of CP	57.8 <sup>b</sup>	47.2 <sup>c</sup>	40.5 <sup>c</sup>	69.6 <sup>a</sup>	1.52
RUP, % of CP	42.2 <sup>b</sup>	52.8 <sup>a</sup>	59.5 <sup>a</sup>	30.4 <sup>c</sup>	1.52
IDP, % of RUP	89.3 <sup>a</sup>	84.2 <sup>b</sup>	61.4 <sup>c</sup>	57.7 <sup>c</sup>	0.98
IADP, % of CP	37.8 <sup>ab</sup>	44.5 <sup>a</sup>	36.5 <sup>b</sup>	17.5 <sup>c</sup>	1.52
TDP, % of CP	95.5 <sup>a</sup>	91.7 <sup>b</sup>	77.0 <sup>d</sup>	87.2 <sup>c</sup>	0.50

<sup>a-d</sup>Values with unlike superscripts differ by  $P < 0.05$ .

**Key Words:** microbially treated, rumen degradability

**T419 Meta-analysis of feeding trials to estimate energy requirements of dairy cows under tropical conditions.** André S. Oliveira\*, Universidade Federal de Mato Grosso, Campus Sinop, Sinop, Mato Grosso, Brazil.

A meta-analysis of feeding trials was conducted to determine the metabolizable energy requirements for the maintenance (ME<sub>m</sub>) and efficiency of metabolizable energy intake (MEI) for the milk production (k<sub>L</sub>) of *Bos taurus* and *Bos taurus* × *Bos indicus* crossbreds dairy cows in the tropics. The database contained 236 means treatments from 60 feeding trial studies. The data set was classified by genotype: *Bos taurus* (37 studies; n = 150 treatments means; MEI = 45.9 ± 6.6 Mcal/d; BW = 570 ± 45 kg; BW variation = 0.30 ± 0.37 kg/d; BCS = 2.7 ± 0.2; milk production = 22.3 ± 5.0 kg/d; milk fat = 3.64 ± 0.37%; milk protein = 3.23 ± 0.25%; milk lactose = 4.43 ± 0.21%; and milk energy = 0.70 ± 0.04 Mcal/kg) and *Bos taurus* × *Bos indicus* crossbred (23 studies; n = 86 treatments means; MEI = 31.6 ± 7.2.6 Mcal/d; BW = 487 ± 48 kg; BW variation = 0.07 ± 0.52 kg/d; BCS = 3.2 ± 0.5; milk production = 13.7 ± 2.0 kg/d; milk fat = 3.63 ± 0.55%; milk protein = 3.11 ± 0.28%; milk lactose = 4.49 ± 0.20%; and milk energy = 0.70 ± 0.06 Mcal/kg). A linear regression of MEI (Mcal/kg BW<sup>0.75</sup>/d) with the milk energy output adjusted to a zero energy balance (E<sub>L(0)</sub>, Mcal/kg BW<sup>0.75</sup>/d) was fitted using a mixed model with the study as the random effect and the genotype as the fixed effect. The ME<sub>m</sub> was calculated as the intercept and k<sub>L</sub> as the reciprocal of the slope of the regression. The dairy cow genotype affected the ME<sub>m</sub> ( $P = 0.013$ ) and k<sub>L</sub> ( $P = 0.033$ ). *Bos taurus* MEI (Mcal/kg BW<sup>0.75</sup>/d) = 0.180 (±0.022,  $P < 0.001$ ) + 1.546 (±0.153,  $P < 0.001$ ) × E<sub>L(0)</sub>,  $R^2 = 0.675$ , MSPE = 0.0007; and *Bos taurus* × *Bos indicus* MEI (Mcal/kg BW<sup>0.75</sup>/d) = 0.147 (±0.024,  $P < 0.001$ ) + 1.771 (±0.288,  $P < 0.001$ ) × E<sub>L(0)</sub>,  $R^2 = 0.859$ , MSPE = 0.0004. *Bos taurus* × *Bos indicus* crossbred dairy cows have ( $P < 0.05$ ) an ME<sub>m</sub> requirement that is 18% lower (0.147 ± 0.024 vs. 0.180 ± 0.022 Mcal/kg BW<sup>0.75</sup>/d), but a k<sub>L</sub> that is 12% lower (0.57 ± 0.09 vs. 0.65 ± 0.06) than that of *Bos taurus* cows. These results support the hypothesis that *Bos taurus* × *Bos indicus* crossbred dairy cows have a lower ME<sub>m</sub> requirement but lower energetic efficiency for milk production than do *Bos taurus* dairy cows under tropical conditions.

**Key Words:** *Bos indicus*, maintenance, metabolizable energy

**T420 Effect of a ration change from a TMR to a pasture-based ration on rumen bacteria in dairy cows.** Melanie Schären\*<sup>1</sup>, Ulrich Meyer<sup>1</sup>, Sven Dänicke<sup>1</sup>, and Gerhard Breves<sup>2</sup>, <sup>1</sup>Friedrich Loeffler Institut (FLI)-Federal Research Institute for Animal Health - Institute for Animal Nutrition, Braunschweig, Germany, <sup>2</sup>University of Veterinary Medicine Hannover, Foundation - Institute for Physiology, Hannover, Germany.

In spring, dairy cows are often gradually transitioned from a TMR to a pasture-based ration. This study aims to investigate how the populations of the liquid (LAB), particle (PAB) and epithelium (EAB) associated bacteria in the rumen are influenced by this nutritional change. A 10-wk trial (w1–10) was performed in spring 2014, including 10 rumen-fistulated dairy cows of the German Holstein breed (182 ± 24 d in milk, 23.5 ± 3.5 kg milk/day, means ± SD). The cows were divided into a pasture- and a confinement group (PG and CG, n = 5). The CG stayed on a TMR-based ration (35% corn silage, 35% grass silage, 30% concentrate; DM basis), while the PG was gradually transitioned from a TMR- to a pasture-based ration (w1: TMR-only, w2: 3 h/day on pasture, w3 and 4: 12 h/day on pasture, w5–10: pasture-only).

In w1, w5 and w10 of the trial samples of solid and liquid rumen contents were taken and after emptying the rumen, papillae biopsies were taken from the ventral rumen sac. The bacterial DNA was isolated from each sample and a SSCP-analysis was performed. Similarity and cluster analysis was done using GelComparII. Data were analyzed as repeated measures using PROC MIXED of SAS Enterprise Guide 6.1. Cluster analysis displayed a clustering of LAB, PAB and EAB in both ration types and all three points in time. LAB and PAB had a similarity of 82 ± 5% (means ± SD) and differ strongly from the EAB with a similarity of 39 ± 8% EAB compared with LAB, and 37 ± 8% EAB compared with PAB (p < 0.001). When comparing the samples of the different bacteria populations in w5 and w10 to their reference sample in w1, a significant greater decrease in similarity over time in all three bacteria populations was observed for the PG compared with the CG (p < 0.01; illustrated in Table 1). To further identify the different bacterial populations 16S DNA sequencing of all samples is currently being performed.

**Table 1 (Abstr. T420).** Change in bacterial populations of the LAB, PAB, and EAB over time expressed in average similarity (in %) of samples compared with their reference sample in week 1 (means ± SD)

	LAB CG	PAB CG	EAB CG	LAB PG	PAB PG	EAB PG
Week 5	93 ± 6%	97 ± 2%	91 ± 4%	76 ± 10%	86 ± 8%	82 ± 8%
Week 10	91 ± 3%	94 ± 2%	88 ± 4%	61 ± 13%	85 ± 8%	67 ± 8%

**Key Words:** dairy cow, pasture, rumen microbiota

**T421 Morphology change and expression of genes related to tight junctions, cytokines, proliferation and apoptosis in the rumen of lactating dairy cows fed corn stover or rice straw replacing alfalfa as forage source.** Bing Wang<sup>\*1,3</sup>, Mei Liu<sup>1,3</sup>, X. B. Huang<sup>1,3</sup>, J. S. Wu<sup>2</sup>, and J. X. Liu<sup>1,3</sup>, <sup>1</sup>Institute of Dairy Science, College of Animal Sciences, Zhejiang University, Hangzhou, China, <sup>2</sup>Department of Veterinary Science, College of Animal Sciences, Zhejiang University, Hangzhou, China, <sup>3</sup>MOE Key Laboratory of Molecular Animal Nutrition, Zhejiang University, Hangzhou, China.

The objective of this study was to characterize the morphology and expression of the genes related to tight junction protein, cytokines, proliferation, and apoptosis in the gastrointestinal tracts (GIT) of lactating cows. Eighteen multiparous Holstein dairy cows were individually fed, and randomly assigned into 1 of 3 treatments. Isonitrogenous diets with forage-to-concentrate ratio at 45:55 contained similar concentrate and 15% corn silage, with 3 forage sources (DM basis): 23% alfalfa hay and 7% Chinese wild rye hay (AH); 30% corn stover (CS); and 30% rice straw (RS). The portion with length above 19 mm was the largest in RS, followed by AH, least in CS. After 14-wk feeding, all the cows were slaughtered. The digesta, tissue, and relevant epithelium of the GIT were collected immediately after dissection. The morphology of the GIT

tissues was analyzed according to the hematoxylin-eosin stained method and the histomorphometric analysis was performed using Image J Plus software (Media Cybernetics, Bethesda, MD). The qRT-PCR reaction of the related genes in rumen epithelium was performed using the 2 × SYBR Premix Ex Taq (Tli RNaseH Plus) kit (Takara, Otsu, Japan). The variance of the data was analyzed as a completely randomized design using PROC MIXED of SAS. The DM of rumen digesta was heavier in diet RS than in AH (P < 0.01). The thickness of ruminal mucosa was greater in RS than in AH (P < 0.05) or CS (P < 0.05), but the thickness of mucous epithelium was greater in AH (P < 0.05) or RS (P < 0.05) than in CS. The rumen butyrate concentration was greater in diet CS than RS (P < 0.01) or AH (P < 0.01), and had a negative correlation with the mRNA abundance of BCD (P = 0.01). Abundance of mRNA of IGF-1R was upregulated in AH compared with CS (P = 0.047). Abundance of mRNA of claudin-1 (P = 0.09), claudin-4 (P = 0.10), and ZO-1 (P = 0.07) tended to increase in AH compared with CS. The expression of gene related to cytokines showed no difference among the treatments. The expression of IGF-1 tended to increase in AH (P = 0.066) compared with CS. Our finding suggested that feeding of rice straw to dairy cow caused the change in ruminal mucosa mostly related to cell proliferation and apoptosis, which may be not only attributed to the physically effective fiber effects but also the butyrate factor.

**Key Words:** cow, gastrointestinal tract, morphology

**T422 Effect of a calcium oral bolus administered after calving on the metabolic parameters of dairy cows.** J. M. Béguin<sup>\*1</sup>, R. P. Dagorne<sup>1</sup>, and P. Courty<sup>2</sup>, <sup>1</sup>Néolait, Yffiniac, France, <sup>2</sup>ESITPA, Mont-Saint-Aignan, France.

The objective of this study was to evaluate the effect of postpartum administration of a calcium bolus (Dietevit Flash, Néolait, Yffiniac, France), on the metabolic parameters of multiparous dairy cows. Holstein cows (n = 38) from 3 commercial dairy farms in Western France, were randomly assigned to a control group (group A) and a trial group (group B). Cows in group B received at calving 2 boluses and 2 boluses at 12–18 h after calving. The 4 boluses supplied calcium (65 g as calcium formiate and calcium propionate), magnesium (5 g), vitamin E (4,000 IU), vitamin D3 (50,000 IU) and niacin (12 g). Blood samples were taken 12 d before calving, 12 and 28 h after calving and 10 d after calving. Blood serum was then analyzed for calcium, magnesium and vitamin E at the Frank Duncombe's Lab, France. The metabolic profiles of the 2 groups were compared using repeated measures ANOVA (SPSS v18). The incidence of cows with postpartum hypocalcemia (calcium < 80mg/L) in both groups was compared using the Pearson Chi-squared test. Compared with group A, cows in group B had higher calcemia at 12 h postpartum (87.3 vs. 79.1 mg/L, P = 0.02) and 28 h postpartum (86.3 vs. 76.4 mg/L, P = 0.01), lower magnesemia at 28 h postpartum (19.0 vs. 22.1 mg/L, P = 0.03) and higher blood levels of vitamin E at 28 h postpartum (2.2 vs. 1.4 µg/L, P = 0.04). Prevalence of hypocalcaemia was significantly decreased in group B (7.1% vs. 37.5 and 25% vs. 56.2%, P = 0.01) at respectively 12 and 28 h postpartum. In conclusion, the Dietevit Flash bolus significantly improves calcemia in postpartum multiparous dairy cows. The Dietevit Flash helps secure the difficult peripartum phase by decreasing the prevalence of subclinical hypocalcaemia in a significant way during the first 2 d of lactation.

**Key Words:** dairy cow, bolus, subclinical hypocalcemia

**T423 Urea kinetics in dairy cows fed soybean meal (SBM), canola meal (CM), corn high protein dried distillers grains (HPDDG) or wheat dried distillers grains with solubles (WDDGS).** D. R. Ouellet<sup>\*1</sup>, G. Maxin<sup>2</sup>, and H. Lapierre<sup>1</sup>, <sup>1</sup>Dairy and Swine R&D Centre, Agriculture and Agri-Food Canada, Sherbrooke, QC, Canada, <sup>2</sup>INRA UMR 1213 Herbivores, Saint-Genès-Champagnelle, France.

Inclusion of CM in dairy rations decreases plasma urea concentration (Martineau et al., J. Dairy Sci. 97:1603). The objective of this study was to compare the effects of feeding diets containing SBM, CM, HPDDG or WDDGS as single protein supplement on urea kinetics. Eight rumen-fistulated Holstein cows were used in a replicated 4 × 4 Latin square with 14-d periods. Dietary treatments consisted of inclusion of these protein in diet: SBM (13.7%), CM (20.8%), HPDDG (20.4%) or WDDGS (22.8%). The 4 diets were isonitrogenous (17.2%CP) and isoenergetic (1.56 Mcal NE<sub>L</sub>/kg DM; NRC 2001), with a fixed forage:concentrate ratio of 62:38. From d 10–13, total collection of urine was performed. On d 12, cows were infused in a jugular vein <sup>15</sup>N<sub>2</sub>-urea (0.5 mmol/h) and urine spot samples were collected after 24 h of infusion; enrichment of <sup>15</sup>N<sub>2</sub>-urea was determined to estimate urea kinetics. Differences among treatments were analyzed using adjusted Tukey-Kramer multiple comparisons test. Results are given as LSM ± SEM for SBM, CM, HPDDG and WDDGS, respectively. The N intake and milk N were similar between treatments (659, 660, 657, and 665 ± 23 g/d and 1.13, 1.15, 1.15, and 1.13 ± 0.01 kg/d; Maxin et al., JDS 96:7806). The RDP supply (NRC 2001) averaged 2622, 2672, 2110 and 2959 g/d. Urea production (UER) tended (*P* = 0.08) to be lower for WDDGS vs. SBM (353 vs. 409 ± 18 g urea-N/d) whereas CM (391) and HPDDG (405) were intermediate. Urea gut entry rate (GER) averaged 265, 272, 267 and 210 ± 18 g urea-N/d, with only WDDGS being lower (*P* < 0.05) than the other diets. The proportion of GER/UER was also lower (*P* < 0.05) for WDDGS: 0.65, 0.69, 0.65, and 0.59 ± 0.02. Urinary N excretion was lower (*P* < 0.06) for CM (118 ± 6 g urea-N/d) than HPDDG (139), WDDGS (144) and SBM (144). Cows fed WDDGS had a lower urea production and GER, but urinary excretion was similar to HPDDG and SBM, in disagreement with estimated higher RDP supply. The intermediate urea production for the CM supplement coupled with the numerically higher proportion returned to the gut lead to a lower urinary urea-N excretion which can explain the decreased plasma urea concentration also observed in this study: 8.5, 7.8, 8.8, and 8.9 ± 0.4 mM (Maxin et al., J. Dairy Sci. 96:7806).

**Key Words:** urea kinetics, protein supplement, dairy cow

**T424 Determining bioavailability of Lys in rumen-protected Lys products using the plasma free AA dose-response technique.** Nancy L. Whitehouse<sup>\*1</sup>, Andre B. D. Pereira<sup>1</sup>, Adam C. Crowther<sup>1</sup>, Devan L. Chirgwin<sup>1</sup>, Andre F. Brito<sup>1</sup>, Charles G. Schwab<sup>2</sup>, and Jack E. Garrett<sup>3</sup>, <sup>1</sup>University of New Hampshire, Durham, NH, <sup>2</sup>Schwab Consulting, LLC, Boscobel, WI, <sup>3</sup>QualiTech Inc, Chaska, MN.

Two experiments were conducted using the technique to determine the bioavailability of Lys in rumen-protected (RP)Lys supplements manufactured by QualiTech Inc. Bioavailability estimates are calculated by dividing the slope of the regression line relating changes in plasma Lys concentrations from feeding an RPLys supplement by the slope of the regression line obtained by continuous abomasal infusion of Lys. Experiment 1 examined the efficacy of the company's first generation product. Experiment 2 was to determine the effect of changes in core/matrix composition and amount of encapsulation on Lys availability. Six multiparous ruminally-cannulated Holstein cows were used in Exp. 1 in a replicated 3 × 3 Latin square (193 DIM). Treatments were 1) 0 g/d Lys, 2) 60 g/d infused Lys from Lys-HCl (80% Lys), and 3) 60 g/d fed Lys

from RPLys (56% Lys). Five multiparous ruminally-cannulated cows were used in Exp. 2 in a 5 × 5 Latin square (108 DIM). Treatments were 1) 0 g/d Lys, 2) 60 g/d infused Lys from Lys-HCl (80% Lys), 3) 60 g/d fed Lys from product A (30.4% Lys), 4) 60 g/d fed Lys from product B (47.4% Lys), and 5) 60 g/d fed Lys from product C (37.3% Lys). Before feeding, the products were mixed with 2 kg of the Lys-adequate basal TMR, placed in ziploc bags, and stored for 8 h at 4°C. Fed treatments were offered to cows 30 min before each of 3 daily feedings. Fed treatments not consumed after 15 min were placed in the rumen. Tail vein samples were collected 2, 4, 6 and 8 h after the morning feeding the last 3 d of each period. Deproteinized plasma samples were composited by cow/d and stored (–80°C) until AA analysis. Lysine concentrations (μM) were expressed as a proportion of total AA before regression analysis. Data were analyzed using the PROC MIXED and REG procedures of SAS. Lysine was the only plasma AA that increased linearly in response to treatments (*P* < 0.02). Calculated Lys bioavailability estimates were 27.1% in Exp. 1, and 17.0, 24.5, and 28.5%, respectively, for Products A, B, and C in Exp. 2. The technique was sensitive enough to confirm differences between Product A vs. products B and C in Exp. 2.

**Key Words:** amino acid, lysine, bioavailability

**T425 Blood ketone bodies incidence and concentration from intensively housed early-lactation dairy cows in Brazil.** Rafahel C. Souza<sup>1</sup>, Rogério C. Souza<sup>1</sup>, Breno M. Sousa<sup>2</sup>, and Andre B. D. Pereira<sup>\*3</sup>, <sup>1</sup>Pontifícia Universidade Católica de Minas Gerais, Betim, MG, Brazil, <sup>2</sup>Centro Universitário UniBH, Belo Horizonte, MG, Brazil, <sup>3</sup>University of New Hampshire, Durham, NH.

Ketosis is a metabolic syndrome that can increase farm costs and result in milk production losses during a lactation. This syndrome mostly affects high production cows and causes are usually associated with negative energy balance during early to mid-lactation. The objective of this study was to evaluate the concentration of ketone bodies in the blood of 732 early-lactation [25.2 ± 15.8 d in milk (DIM)] high producing Holstein cows (32.98 kg/d of milk yield) from 10 intensively managed farms in Brazil. A portable electronic diagnosis equipment (Ketovet, TaiDoc technology, Taiwan, China) was used for measurement of blood ketone bodies and results are expressed in mM. Blood was sampled from the tail's artery or vein, and the volume of 1 drop was added to a reagent strip set for β-hydroxybutyrate (BHBA) analysis and 5 s was the average time needed for the equipment to yield results. Animals with values of blood BHBA of less than 1.1 mM were considered normal and free of metabolic syndrome. Animals with values between 1.2 and 3.5 mM were diagnosed with subclinical ketosis, and animals with values above 3.5 were diagnosed with clinical ketosis. Animals were blocked by days in milk (0–15, 16–30, 31–45, >46) and farm was used as a random effect. Data were analyzed using a t student test of least square difference and significance was declared as *P* < 0.05. From the 732 animals evaluated, 448 (47.85%) were considered free from the syndrome, 279 (38.11%) had subclinical ketosis and 5 (0.68%) had clinical levels of BHBA. Similar BHBA levels were found for cows between 0 and 15 DIM (1.07 mM), between 16 and 30 DIM (1.15 mM) and between 31 and 45 DIM (1.06 mM), but were significantly higher for cows with more than 46 DIM (1.34 mM, SEM = 1.34, *P* < 0.01). Results of this experiment suggest that cows with more than 46 DIM had subclinical ketosis and values of BHBA that were higher than for all cows with less than 45 DIM.

**Key Words:** transition period, metabolic syndrome, ketone bodies



**T426 Evaluation of camelina meal as a feedstuff for growing dairy heifers.** Rhea D. Lawrence\* and Jill L. Anderson, *Dairy Science Department, South Dakota State University, Brookings, SD.*

Our objective was to evaluate growth performance of dairy heifers fed camelina meal (CM) compared with linseed meal (LN) or reduced-fat distillers dried grains with solubles (DG). A 12-wk randomized complete block design study was conducted using 33 Holstein and 9 Brown Swiss heifers (144.8 ± 22 d of age; body weight (BW) 171.8 ± 24.3 kg) with 3 treatments. Treatments were 10% of the diet as CM, LN, or DG (DM basis). Concentrate mixes also included corn and soybean meal, at slightly different inclusion rates to make diets isocaloric and isonitrogenous. All diets contained 60% grass hay and 40% concentrate mix. Diets were limit-fed to 2.65% of BW (DM basis) using a Calan gate feeding system. Frame sizes, BW, and body condition scores (BCS) were measured on 2 d during wk 0, 2, 4, 6, 8, 10, and 12. Data were analyzed using MIXED procedures with repeated measures and ADG was found using REGRESSION procedures of SAS 9.4. Heifer DMI was similar between CM and LN, but greater ( $P = 0.03$ ) for DG (4.83, 4.82, and 5.03 kg/d; SEM = 0.17 for CM, LN, and DG, respectively). Body weights (199.5, 210.9, and 205.1 kg; SEM = 2.86) were found to be less ( $P < 0.01$ ) for heifers fed CM and greatest for LN, which could be due to unequal initial BW. Treatments had similar ( $P > 0.05$ ) ADG (0.67, 0.75, 0.71 kg/d; SE = 0.15). Gain to feed was similar for CM and DG, but greater ( $P < 0.05$ ) for LN (0.14, 0.17, 0.15 kg/d; SEM = 0.006). Hip height (116.3, 115.8, and 115.4 cm; SEM = 0.67), withers height (112.3, 112.3, and 112.4 cm; SEM = 0.47), and heart girth (128.6, 128.3, and 128.9 cm; SEM = 0.81) were similar ( $P > 0.05$ ) among treatments. Hip width (31.7, 31.3, and 31.4 cm; SEM = 0.27) and body length (105.2, 102.0, and 103.3 cm; SEM = 0.77) were greater ( $P < 0.05$ ) for CM compared with LN and DG. Body condition score (3.16, 3.10, and 3.17; SEM = 0.02) was greater ( $P < 0.01$ ) for CM and DG compared with LN. Feeding CM maintained frame growth and ADG but decreased gain to feed compared with DG and LN. The decreased feed efficiency may be related to glucosinolates in the CM. Further research should be conducted before implementing CM as a primary protein source for growing heifers.

**Key Words:** camelina meal, dairy heifer

**T427 Evaluation of the incidence of subclinical ketosis for F<sub>1</sub> Gir x Holstein lactating dairy cows supplemented with medium-chain fatty acids.** Rafahel C. Souza<sup>1</sup>, Rogério C. Souza<sup>1</sup>, Vanessa A. Teixeira<sup>1</sup>, Joaquim H. C. M. Souza Junior<sup>1</sup>, Igor C. Leal<sup>1</sup>, Andre B. D. Pereira<sup>\*2</sup>, and Maria I. V. Melo<sup>1</sup>, <sup>1</sup>*Pontifícia Universidade Católica de Minas Gerais, Betim, MG, Brazil*, <sup>2</sup>*University of New Hampshire, Durham, NH.*

The use of medium-chain fatty acids (MCFA) during the periparturient period can alleviate the effects of negative energy balance and improve animal health in early lactation dairy cows. The objective of this study was to evaluate blood β-hydroxybutyrate (BHBA) of 30 early-lactation F<sub>1</sub> Gir × Holstein cows supplemented with MCFA. Diets were formulated according to the NRC (2001) model and contained 50% forage as corn silage and chopped sugar cane. Concentrate was based on ground corn, soybean meal, citrus pulp and minerals in 2 treatments: (1) no addition of MCFA (0FA); and (2) addition of 40 g of MCFA [40FA, 0.186% of dry matter as toplac rumacel (Nutrifarma, Taio, PR, Brazil)]. The experiment was a completely randomized design and cows were supplemented with MCFA during the periparturient period (from 21 d before parturition until 21 d after). A portable electronic diagnosis equipment (Ketovet, TaiDoc technology, Taiwan, China) was used for measurement of BHBA and results are expressed in mM. Blood was

sampled from the tail artery or vein, and the volume of 1 drop was added to a reagent strip set for BHBA analysis. Samples were collected and analyzed at the day of parturition and then again after 42 and 70 d in milk. Animals with values above 1.2 mM were diagnosed with subclinical ketosis. Data were analyzed using the MIXED procedure of SAS and significance was declared at  $P < 0.05$ . At the day of parturition, 73.4% of cows in 0FA (11 out of 15) had subclinical ketosis while only 6.6% (1 out of 15 cows) of cows on 40FA had subclinical ketosis ( $P < 0.05$ ). The proportion of cows with subclinical ketosis was reduced to 40% for 0FA and slightly increased to 13.4% for 40FA after 42 d in milk and this proportion remained for after 70 d in milk, being significantly different between treatments ( $P < 0.05$ ). Results of this study suggests that F<sub>1</sub> Gir × Holstein cows receiving diets based on corn silage and chopped sugar cane supplemented with MCFA may have lower incidence of subclinical ketosis when compared with non-supplemented cows.

**Key Words:** dairy cow, medium-chain fatty acids, subclinical ketosis.

**T428 Milk production and composition of F<sub>1</sub> Gir x Holstein lactating cows supplemented with medium-chain fatty acids during the periparturient period.** Rafahel C. Souza<sup>1</sup>, Rogério C. Souza<sup>1</sup>, Vanessa A. Teixeira<sup>1</sup>, Igor G. Leal<sup>1</sup>, Joaquim H. C. M. Souza Junior<sup>1</sup>, Andre B. D. Pereira<sup>\*2</sup>, and Maria I. V. Melo<sup>1</sup>, <sup>1</sup>*Pontifícia Universidade Católica de Minas Gerais, Betim, MG, Brazil*, <sup>2</sup>*University of New Hampshire, Durham, NH.*

Subclinical ketosis is usually associated with negative energy balance in early lactation. The use of medium-chain fatty acids (MCFA) during the periparturient period can alleviate the negative energy balance and improve animal health. The objective of this study was to evaluate 30 F<sub>1</sub> Gir × Holstein lactating dairy cows for milk production and composition when supplemented with MCFA during the periparturient period. Diets were formulated according to the NRC (2001) model and contained 50% forage as corn silage and chopped sugar cane. Concentrate was based on ground corn, soybean meal, citrus pulp and minerals in 2 treatments: (1) No addition of MCFA (0FA) and (2) Addition of 40 g of MCFA (40FA, 0.186% of DM) as Toplac Rumacel (Nutrifarma, Taio, PR, Brazil). The experiment was a completely randomized design and cows were supplemented with MCFA during the periparturient period (from 21 d before parturition until 21 d after). Analyses of milk yield and composition were done weekly and collections were made on d 7, 14, 21, 28, 35, 42, 49, 56, 63 and 70 after parturition for each cow. Data were analyzed using the MIXED procedure of SAS and means were compared using the Tukey adjustment with significance declared as  $P < 0.05$ . Milk yield was higher for cows on 40FA (26.97 kg/d) when compared with cows on 0FA (25.50 kg/d,  $P < 0.05$ ), but this trend only happened from 35 d after parturition until 70 d. Milk fat and protein yields were not different between treatments and days sampled. Somatic cell count was lower (64,260 cells/mL) for cows on treatment 40FA compared with 0FA (98,800 cells/mL,  $P < 0.05$ ) suggesting that MCFA can improve mammary health. Results of this study suggest that supplementation of MCFA improved milk production and composition of F<sub>1</sub> Gir x Holstein cows when compared with non-supplemented cows.

**Key Words:** dairy cow, milk production and composition, medium-chain fatty acids

**T429 Effects of casein, glucose, and acetate infused into the abomasum of feed-restricted cows on milk and milk components yield.** Marina A. C. Danes<sup>\*1</sup>, Michel A. Wattiaux<sup>1</sup>, and Glen A. Broderick<sup>2</sup>, <sup>1</sup>*University of Wisconsin-Madison, Madison, WI*, <sup>2</sup>*Broderick Nutrition & Research LLC.*

Amino acids and glucose have been shown to regulate protein synthesis in the mammary gland through cell signaling. Acetate might also have an effect through AKT pathway, since it is the main energy source for the mammary secretory cell. Six Holstein multiparous cows, averaging 55 kg milk/d, were used in a 6x6 Latin square with 14-d periods. In each period, cows were fed the same diet for 10 d, after which they were feed-restricted for 4 d to 85% of ad libitum intake and abomasally infused for 1 of 6 treatments: acetate (A), glucose (G), each at 5% of ad libitum metabolizable energy intake (Mcal/d), casein (C) at 15% of ad libitum metabolizable protein intake (g/d), A+C (AC), G+C (GC), or a saline solution (S, negative control). Data from d 13 and 14 of each period were analyzed with Proc Mixed of SAS, using data from the d 9 and 10 as covariate. LS means are reported in the table. Casein by itself had no effect on any of the variables, compared with the negative control. This could be a consequence of lack of energy due to feed restriction, since glucose infused with casein numerically increased milk protein yield by 121 g/d. Glucose increased milk yield and nitrogen use efficiency, as indicated by the lowest MUN. Acetate had no effect on any of the variables, rejecting our hypothesis that it would enhance milk protein synthesis by improving energy status of the cell. Instead, acetate was used for fat synthesis as shown by the numerically greater fat yield for the acetate treatments. Interestingly, the infusion of casein along with acetate increased milk fat secretion more than acetate alone, which might be related to the effect of specific AA on fat synthesizing enzymes recently reported in the literature.

**Table 1 (Abstr. T429).**

Item	A	AC	C	G	GC	S	SEM	P-value
Milk, kg/d	37.2 <sup>B</sup>	39.2 <sup>AB</sup>	38.4 <sup>AB</sup>	42.2 <sup>A</sup>	41.3 <sup>AB</sup>	37.1 <sup>B</sup>	1.8	<0.10
Lactose, kg/d	1.67 <sup>AB</sup>	1.79 <sup>AB</sup>	1.71 <sup>AB</sup>	1.89 <sup>A</sup>	1.84 <sup>AB</sup>	1.65 <sup>B</sup>	0.08	<0.10
Fat, kg/d	1.59	1.7	1.43	1.54	1.44	1.44	0.12	NS
Protein, kg/d	1.13 <sup>B</sup>	1.19 <sup>AB</sup>	1.19 <sup>AB</sup>	1.27 <sup>AB</sup>	1.31 <sup>A</sup>	1.15 <sup>AB</sup>	0.07	0.11
MUN, mg/dL	11.4 <sup>bc</sup>	15.1 <sup>a</sup>	12.2 <sup>bc</sup>	10.4 <sup>c</sup>	11.2 <sup>bc</sup>	12.4 <sup>b</sup>	0.8	<0.01

Means followed by different lowercase (uppercase) letters differ at  $P < 0.10$  ( $P < 0.15$ ).

**Key Words:** milk protein synthesis, mammary metabolism

**T430 The effect of pelletized corn stover replacing alfalfa hay on lactation performance, blood parameters and rumen fermentation in mid-lactation dairy cows.** H. Z. Sun<sup>\*1,2</sup>, Z. H. Wei<sup>1,2</sup>, Wen Zhu<sup>1,2</sup>, X. Xie<sup>1,2</sup>, J. K. Wang<sup>1,2</sup>, L. L. Guan<sup>3</sup>, and J. X. Liu<sup>1,2</sup>, <sup>1</sup>*Institute of Dairy Science, College of Animal Sciences, Zhejiang University, Hangzhou, China*, <sup>2</sup>*MoE Key Laboratory of Molecular Animal Nutrition, Zhejiang University, Hangzhou, China*, <sup>3</sup>*Department of Agricultural, Food & Nutritional Science, University of Alberta, Edmonton, Canada*.

This study was conducted to investigate the effect of pelletized corn stover replacing alfalfa hay on lactation performance, blood parameters and rumen fermentation dairy cows. Twenty multiparous Holstein dairy cows were blocked based on days in milk ( $219 \pm 49.5$  d, mean  $\pm$  SD) and milk yield ( $25.4 \pm 4.03$  kg, mean  $\pm$  SD), and were randomly assigned into one of 2 dietary treatments with similar concentrate mixture, but different forage ingredients (as DM basis): 15% of pelletized corn stover pellets (CS) or 12.2% of alfalfa hay (AH). Two diets had similar content of net energy for lactation (1.57 vs. 1.58 Mcal/kg DM), but differed in crude protein content (15.8 vs. 16.7 for CS and AH). After feeding for 3 weeks, the rumen fluid and blood samples were collected. The data were analyzed using MIXED procedure of SAS software. The DM intake of diet did not differ ( $P = 0.72$ ) between 2 diets, with an average of 16.5

kg/d. Milk yield in AH-fed cows was 0.87 kg/d higher than the CS-fed, though the difference was not significant ( $P = 0.53$ ). No difference was observed between 2 diets in milk protein (3.37 vs. 3.38%,  $P = 0.97$ ), milk fat (4.15 vs. 4.25%,  $P = 0.76$ ), lactose (4.78 vs. 4.76%,  $P = 0.83$ ), total solid (13.7 vs. 12.9%,  $P = 0.16$ ), milk urea nitrogen (16.0 vs. 13.9%,  $P = 0.19$ ), and somatic cell counts ( $206$  vs.  $325 \times 10^3$ /mL,  $P = 0.39$ ). The concentration of plasma glucose was significantly lower in the CS-fed cows than in AH-fed animals (1.82 vs. 2.23 mM,  $P = 0.02$ ), whereas the other blood parameters (NEFA, BHBA, Ca and P) were not different ( $P > 0.05$ ). The concentrations of ruminal total and individual volatile fatty acids showed no difference between 2 groups. From the above results, it is indicated that the pelletized corn stover can be used to replace alfalfa hay in the diet for mid-lactation dairy cows without adverse effects on lactation performance and rumen fermentation.

**Key Words:** pelletized corn stover, alfalfa hay, dairy cow

**T431 Supplementation with artificial sweetener improves milk yield and composition and alters nutrient partitioning in lactating dairy cows.** Emma H. Wall<sup>\*1,2</sup> and David M. Bravo<sup>2</sup>, <sup>1</sup>*Pancosma, Geneva, Switzerland*, <sup>2</sup>*InVivo Animal Nutrition & Health, Saint-Nolff, France*.

Supplementation with Sucram (SUC, Pancosma, Geneva, Switzerland) increases intestinal expression of sodium-glucose co-transporter 1, glucose uptake, and mucosal growth in ruminants. This trial aimed to determine the productive implications of this response in dairy cows. Primi- and multiparous lactating Holstein dairy cows were housed together in a freestall pen and were milked using an automated milking system (AMS). For 8 weeks, SUC was blended with a carrier and was dispensed at the AMS for SUC cows ( $n = 91$ ) at a rate of 2.72 kg/d (SUC dose: 2g/cow/d); control cows received no additive ( $n = 89$ ). Individual cow milk production and composition, milking frequency, and BW were recorded daily. Average DMI of the pen was monitored daily and did not change throughout the study. A treatment by stage of lactation interaction was observed: in cows that were greater than 100 DIM, there was no effect of SUC on any variables measured ( $P > 0.20$ ). In contrast in cows less than 100 DIM, SUC enhanced milk production (44.5 vs. 45.6 kg/d;  $P \leq 0.10$ ). Milk fat (1.62 vs. 1.71 kg/d) and protein (1.41 vs. 1.46 kg/d) yields were also improved with SUC ( $P < 0.02$ ); therefore, energy-corrected milk production was greater for SUC cows (46.0 vs. 48.0 kg/d;  $P < 0.01$ ). There was a parity by treatment interaction for BW such that primiparous cows maintained condition regardless of treatment ( $P > 0.50$ ) whereas multiparous cows supplemented with SUC lost BW during the trial ( $P < 0.001$ ). Interestingly, there was also a parity by treatment interaction for number of daily milkings, which was increased by SUC in primiparous animals only ( $P < 0.001$ ). This indicates that the increase in milk yield in those animals may have been due to greater frequency of milk removal whereas in multiparous cows it was due to mobilization of body reserves. These findings reveal that in early to mid-lactation, supplementation with SUC improves milk production performance and optimizes nutrient partitioning.

**Key Words:** automated milking system, feed additive, Sucram

**T432 Effect of fish oil on transportation of fatty acids in plasma lipoproteins of lactating and nonlactating cows.** Einar Vargas-Bello-Pérez<sup>\*1</sup>, Gonzalo Íñiguez-González<sup>1</sup>, Philip C. Garnsworthy<sup>2</sup>, and Juan J. Lóor<sup>3</sup>, <sup>1</sup>*Pontificia Universidad Católica de Chile, Santiago, Chile*, <sup>2</sup>*The University of Nottingham, Loughborough, UK*, <sup>3</sup>*University of Illinois, Urbana, IL*.

The aim of this study was to elucidate the effect of dietary fish oil (FO) and a blend of FO and hydrogenated palm oil (PO) on transportation of fatty acids (FA) in plasma lipoproteins of lactating and nonlactating cows. Three lactating and 3 nonlactating Holstein cows (fitted with rumen cannulas) were used in 2 different 3 × 3 Latin square designs that included 3 periods of 21 d. Dietary treatments for lactating cows consisted of a basal diet (Control; no fat supplement), and fat-supplemented diets containing FO (500 g/d/cow) and FOPO (250 FO + 250 PO g/d/cow hydrogenated palm oil). For nonlactating cows dietary treatments consisted of a basal diet (Control; no fat supplement), and fat-supplemented diets containing FO (170 g/d/cow) and FOPO (85 FO + 85 PO g/d/cow). Compared with control and FOPO, FO increased plasma contents of C16:0, C16:1, C17:1, C18:2n6t, C18:2 c9t11 and C20:5n3. Nonlactating cows had higher plasma contents of C18:1t11, C18:1n9c, C20:3n6, C18:2n6c, C23:0 and C22:6n3. Only C18:2n6c was higher in lactating than nonlactating cows (37 vs. 26 g/100g). Compared with control and FOPO, FO increased HDL contents of C18:1t11, C18:2n6t, C18:2 c9t11 and C20:5n3, and increased LDL contents of C18:1t9, C18:1t11 and C20:5n3. Nonlactating cows had higher LDL contents of C18:1t9 and C18:1t11 (1.98 vs. 0.89 g/100g and 2.7 vs. 1.14 g/100g) than lactating cows. In plasma, nonlactating cows had higher contents of total SFA (46 vs. 43 g/100g), MUFA (16 vs. 11 g/100g) and C18:1 tFA (5 vs. 3 g/100g) and lower content of total PUFA (39 vs. 46 g/100g) than lactating cows. Total content of C18:1 tFA was higher in nonlactating cows than in lactating cows (7 vs. 3 g/100g). Overall, results demonstrate clear differences in plasma transport of FA, which depend on dietary FA source and lactation state.

**Key Words:** fish oil, plasma, lipoprotein

**T433 Development and evaluation of predictive models of intake for crossbred Holstein-Zebu dairy cows.** V. L. Souza<sup>\*1</sup>, T. Z. Albertini<sup>1</sup>, R. Almeida<sup>2</sup>, G. B. Mourão<sup>1</sup>, J. K. Drackley<sup>3</sup>, and D. P. D. Lanna<sup>1</sup>, <sup>1</sup>Esalq/USP, Piracicaba, SP, Brazil, <sup>2</sup>Universidade Federal do Paraná, Curitiba, PR, Brazil, <sup>3</sup>University of Illinois, Urbana, IL.

Equations to predict dry matter intake (DMI) of crossbred Holstein-Zebu dairy cows were developed and compared by using data of 161 treatment means from 38 Brazilian studies (n = 446 cows, milk production average = 16.60 ± 5.70 kg/d). A data set was developed of Holstein × Zebu lactating dairy cows of different degrees of breeding in confinement or pasture systems. The data were evaluated using mixed nonlinear models including study as a random effect. Body weight (BW), 4% fat corrected milk (4% FCM) and weeks of lactation (WOL) were used as independent variables in the model. The proposed model to estimate DMI (kg/d) of crossbred cows was:  $[0.5552 (\pm 0.06636) \times 4\% \text{ FCM} + 0.06332 (\pm 0.009455) \times \text{BW}^{0.75}] \times [1 - e^{(-0.7732 (\pm 0.7019) \times (\text{WOL} - 1.629 (\pm 1.913))}]$ . The accuracy of the model was compared with 4 previously published equations by use of an independent data set. The mean square error of prediction (MSEP), mean bias, concordance correlation coefficient (CCC), and analysis of linear regression were used for evaluating models. The new model showed the lowest values of MSEP and highest CCC and r<sup>2</sup> compared with the other 4 equations (Table 1). The Brazilian model from Freitas et al. (2006), despite being developed in tropical conditions, showed the highest value of the MSEP. The new equations to predict DMI can be used in the formulation of diets for crossbred dairy cows under tropical conditions.

**Table 1 (Abstr. T433).**

Variable	DMI (kg/d)	New model (2015)	NRC (2001)	Fox et al. (2004)	Traxler (1997)	Freitas et al. (2006)
Mean, kg	16.10	16.88	16.11	15.05	16.95	15.85
Mean bias, Y - X, kg	—	-0.78	-0.01	1.05	-0.85	0.25
MSEP, kg × kg	—	1.64	2.45	2.73	2.76	6.98
MSEP Decomposition, %						
Mean bias, %	—	37.001	0.003	40.458	26.338	0.863
Systematic bias, %	—	3.469	0.052	5.327	0.781	63.86
Random errors, %	—	59.531	99.945	54.215	72.882	35.227
CCC	—	0.90	0.82	0.80	0.80	0.75
r <sup>2</sup>	—	0.88	0.69	0.81	0.74	0.69
P-value (a = 0)	—	0.495	0.889	0.229	0.244	<0.001
P-value (b = 1)	—	0.125	0.884	0.049	0.506	<0.001

**Key Words:** crossbred dairy cow, intake, meta-analysis

**T434 Soil contamination in forages: Estimation and geographical distribution.** J. R. Knapp<sup>\*1</sup>, W. P. Weiss<sup>2</sup>, R. T. Ward<sup>3</sup>, and K. R. Perryman<sup>4</sup>, <sup>1</sup>Fox Hollow Consulting LLC, Columbus, OH, <sup>2</sup>Dept. of Animal Sciences, The Ohio State University, Wooster, OH, <sup>3</sup>Cumberland Valley Analytical Services, Hagerstown, MD, <sup>4</sup>Micronutrients Inc., Indianapolis, IN.

Titanium (Ti) concentration in forages is considered by agronomists to be the gold standard in determining soil contamination of herbaceous materials. However, Ti is not measured in routine nutrient analysis of feed ingredients. Soil contamination of forages reduces the concentration of all nutrients, and soil Fe can decrease the absorption and utilization of dietary copper and perhaps other minerals. The objectives of this study were to determine the relationship between Fe and ash concentrations and to estimate soil contamination from available analytes in commonly used forages in US dairy operations. Data from Cumberland Valley Analytical Services covering the period from 2009 to 2014 for corn silage, legume hay, mixed mostly legume (MML) silage, and mixed mostly grass (MMG) silage were used. Soil contamination was estimated using 2 independent measures, residual ash and Fe enrichment. Residual ash (RA) was calculated according to Cary et al. (1986). Fe enrichment was calculated using the measured forage Fe concentrations and soil Fe concentrations obtained from a US Geological Survey database for the matching location. Linear regression analysis was used to evaluate relationships between total ash, RA, and forage Fe concentrations. As expected, RA was highly correlated with total ash with R<sup>2</sup> > 0.90 (P < 0.001). Fe was also highly correlated with RA and total ash with R<sup>2</sup> ranging from 0.47 to 0.63 (P < 0.001). The relationships between Fe and ash concentrations is good across forages, and 1% or greater soil contamination occurs in a significant portion of forages (Table). Level of soil contamination was associated with geographic location. In particular, corn silages grown in the western US had higher (P < 0.001) levels of total ash, RA, and Fe than corn silages from the eastern and midwestern US.

*Contd.*



**Table 1 (Abstr. T434).** Level of soil contamination (% DM), percentage of samples (Prop), total ash (% DM), and Fe concentrations (mg/kg) of commonly used dairy forages, given as mean  $\pm$  SD

Soil contamination		Corn silage	Legume hay	MML silage	MMG silage
<1%	Prop	63.5	18.3	8.3	19.3
	Total Ash	3.28 $\pm$ 0.47	9.20 $\pm$ 1.24	9.21 $\pm$ 1.19	6.08 $\pm$ 1.56
	Fe	133 $\pm$ 83	212 $\pm$ 128	265 $\pm$ 151	219 $\pm$ 170
1 to 4%	Prop	36.0	71.9	78.1	57.6
	Total Ash	5.39 $\pm$ 0.94	10.68 $\pm$ 1.25	10.55 $\pm$ 1.24	8.41 $\pm$ 1.77
	Fe	234 $\pm$ 139	353 $\pm$ 238	423 $\pm$ 261	355 $\pm$ 252
>4%	Prop	0.5	9.8	13.6	23.1
	Total Ash	8.02 $\pm$ 0.60	13.35 $\pm$ 1.35	13.51 $\pm$ 1.56	12.07 $\pm$ 2.03
	Fe	555 $\pm$ 313	872 $\pm$ 553	1155 $\pm$ 576	850 $\pm$ 605

**Key Words:** trace mineral, soil contamination, copper availability

**T435 Comparison of two methods to quantify free amino acids in cow plasma.** Nelson Lobos<sup>\*1</sup>, Glen Broderick<sup>2</sup>, Nancy Whitehouse<sup>5</sup>, Daniel Luchini<sup>3</sup>, Michel Wattiaux<sup>1</sup>, and Peter Crump<sup>4</sup>, <sup>1</sup>Department of Dairy Science, University of Wisconsin-Madison, Madison, WI, <sup>2</sup>Broderick Nutrition & Research LLC, Madison, WI, <sup>3</sup>Adisseo S.A.S., Alpharetta, GA, <sup>4</sup>Department of Computing and Biometry, University of Wisconsin-Madison, Madison, WI, <sup>5</sup>Department of Biological Sciences, University of New Hampshire, Durham, NH.

The objective was to determine how measurements of plasma free AA by gas chromatography (GC) after chloroformate derivatization compared with values obtained by ion-exchange chromatography (HPLC) followed by ninhydrin derivatization. Plasma was obtained from lactating dairy cows (mean DIM = 148) part of a trial were Met was abomasally infused (12 or 24 g Met/d) or fed in rumen-protected form (15 or 30 g Met/d). Blood samples (n = 89) were collected from coccygeal vessels into evacuated tubes. After centrifugation, plasma was split into 2 aliquots and stored frozen at  $-80^{\circ}\text{C}$ . Each set was analyzed either by HPLC in a commercial laboratory, or in-house by GC using a kit (EZ:faast, Phenomenex). This method involved deproteinization with TCA, AA collection using solid phase extraction, derivatization with chloroformate, GC separation in a capillary column and flame ionization detection. Quantitation was based on area under the curve, using the internal standard ratio method (Norvaline). Data were analyzed by Pearson correlation ( $r$ ; Table 1). However Bland-Altman posited that correlation is misleading to compare methods and instead proposed the inspection of difference plots and limits of agreement. Calculation of concordance correlation coefficients provided a quantitative alternative (CCC; Table 1). Overall correlation between methods was 0.95 ( $P < 0.001$ ); nevertheless, correlations varied greatly for individual AA from 0.97 for Met to 0.15 for Glu. For Met, most data were within the limits of agreement established by Bland-Altman approach. Furthermore among all AA, CCC for Met was the closest to 1 (indicating almost total agreement between methods; Table 1). For research focusing on Met, GC offers a cheaper and faster alternative to HPLC.

*Contd.*

**Table 1 (Abstr. T435).** Free AA (except Arg and Trp) in plasma measured by two methods

Amino acid	GC ( $\mu\text{M}$ )	HPLC ( $\mu\text{M}$ )	$r$	CCC
His	50.2	62.8	0.42	0.18
Ile	107.6	101.6	0.96	0.94
Leu	121.2	145.2	0.92	0.36
Lys	72.4	80.8	0.35	0.67
Met	32.8	33.9	0.97	0.99
Phe	40.0	46.1	0.65	0.37
Thr	186.9	117.9	0.96	0.25
Val	348.0	266.1	0.89	0.25
Ala	351.7	267.9	0.88	0.21
Asn	44.0	57.1	0.84	0.26
Asp	9.4	3.9	0.51	0.07
Cys	18.9	21.0	0.35	0.56
Glu	56.1	39.7	0.15	0.08
Gln	110.6	200.6	0.78	0.10
Gly	463.3	348.8	0.92	0.36
Pro	102.2	81.4	0.84	0.25
Ser	127.1	95.3	0.89	0.32
Tyr	30.5	42.1	0.60	0.31

**Key Words:** amino acid, methionine, chromatography

**T436 Fecal starch and starch digestibility: An indirect inter-relationship.** C. E. Owens<sup>1</sup>, R. A. Zinn<sup>2</sup>, and F. N. Owens<sup>\*3</sup>, <sup>1</sup>Duke University, Durham, NC, <sup>2</sup>University of California, El Centro, CA, <sup>3</sup>DuPont Pioneer, Johnston, IA.

Published equations to calculate total-tract starch digestibility (SD) from the concentration of starch in fecal dry matter (FS) have regression slopes that differ by more than 5 fold. Why these slopes differed and methods to improve the accuracy and precision of prediction were explored. Mathematically, SD and FS are correlated. However, 2 additional factors, starch content of dietary DM (DS) and indigestibility of diet DM (IDM =  $100 - \text{DMD}$ , %) alter this relationship. Published literature data (204 diets fed to lactating dairy cows; 191 diets fed to feedlot cattle) that provided or allowed SD, FS, DS, and IDM to be calculated were compiled to examine this relationship. When DS and IDM were ignored, the relationship was imprecise (SD, % =  $99.16 - 1.07 \text{ FS} \%$ ;  $R^2 = 0.81$  for lactating cows; SD, % =  $99.98 - 0.44 \times \text{FS} \%$ ;  $R^2 = 0.88$  for feedlot cattle). Including DS improved precision (SD, % =  $100.13 - 0.341 \times \text{FS} \% / \text{DS} \%$ ,  $R^2 = 0.92$  for lactating cows; SD, % =  $100.29 - 0.257 \times \text{FS} \% / \text{DS} \%$ ,  $R^2 = 0.91$  for feedlot cattle); adding IDM increased precision further (SD, % =  $100 - 1 \times \text{FS} \% / \text{DS} \% \times \text{IDM} \%$ ;  $R^2 = 1.00$ ;  $P = 1/\infty$  for all cattle). Because starch is a component of dietary DM, as SD increases IDM will decrease; hence the relationship of SD to FS is curvilinear, not linear unless the digestibility of non-starch components increases when SD increases. Knowledge of IDM and the dietary and fecal concentrations of any component of the diet including NDF allows apparent digestibility to be computed directly and avoids the need to predict digestibility from in vitro assays. Direct measurement often proves more economical analytically than in vitro procedures and avoids dependence on tabular feedstuff values in publications or computerized diet formulation programs. Combined with intake of DM, direct digestibility measurement can quantify the amount of digested energy available for production by ruminants. As with all analytical procedures, accuracy of digestibility estimates relies on representative sampling and appropriate analyses of diets and feces.

**Key Words:** starch digestibility, fecal starch, equation

**T437 Starch and NDF digestibility by high-producing lactating cows: A field study.** B. Powel-Smith, L. J. Nuzback, W. C. Mahanna, and F. N. Owens\*, *DuPont Pioneer, Johnston, IA.*

In vivo measurements are the gold standard against which in vitro procedures digestibility estimates must be judged. To quantify the relationships of in vitro ruminal disappearance to total-tract digestibility of starch and NDF, samples were obtained from the top string of lactating cows from 32 commercial herds in the Upper Midwest. Silage, TMR, grain, and fecal samples were assayed at a commercial laboratory for some of the following: starch, 7h in vitro starch digestion, NDF digestibility at 24 and 240 h, and lignin at. For calculating in vivo digestibility, lignin and UNDF served as indigestible markers. All cows were fed kernel processed corn silage (25 to 40% DM) plus dry ground or high moisture ear corn or grain; diets averaged 26% starch and 31% NDF. Except for 2 herds, starch digestibility exceeded 95% being related closely to fecal starch concentration ( $R^2 = 0.98$ ). However, in vitro starch disappearance at 7 h was not related to total-tract starch digestibility ( $R^2 = 0.00$ ). Total-tract digestion averaged 55% for NDF or 60% for available NDF (NDF minus UNDF240). In vivo NDF digestibility of the TMR tended to decline as its UNDF content increased ( $R^2 = 0.21$ ), but 24 h in vitro NDF disappearance of the TMR proved to be poorly related to NDF digestibility in vivo ( $R^2 = 0.13$ ). Kernel processing score was not related to starch digestibility, but all silage had fermented more than 6 mo before feeding. Processing score tended to be greater and presence of whole and half kernels in silage was less with shredlage than conventional kernel processing rolls but total-tract starch digestibility was not altered by processing roll type. Dry matter digestibility calculated from UNDF differed by as much as 10% from digestibility calculated from lignin. Total-tract starch digestibility was not correlated with surface area of particles or mean particle size of the dry ground corn fed. To obtain reliable and accurate estimates of energy availability for ruminants, nutritionists should rely on total-tract digestion measurements.

**Key Words:** in vivo digestibility, NDF, starch

**T438 Effects of diets with different energy levels on growth performance and rumen environment of heifer.** Yan Tu\*<sup>1,2</sup>, Xiang Cui<sup>1</sup>, Tao Ma<sup>1,2</sup>, Bing-wen Si<sup>1,2</sup>, Nai-feng Zhang<sup>1,2</sup>, and Qi-yu Diao<sup>1,2</sup>, <sup>1</sup>*Feed Research Institute of Chinese Academy of Agricultural Sciences, Key Laboratory of Feed Biotechnology of the Ministry of Agriculture, Beijing, China,* <sup>2</sup>*Beijing Key Laboratory for Dairy Cow Nutrition, Beijing, China.*

Thirty-two Chinese Holstein heifers (98 d old) were randomly divided into 4 groups with 8 heifers each (8 replications). Each group was subjected to one of the following 4 total mixed rations (TMR) with 2.52 (A), 2.82 (B), 2.99 (C), and 3.21 (D) Mcal of metabolic energy (ME) per kg (dry matter basis) diet for 82 d. The energy gradient was adjusted by changing the composition of concentrate and adding rumen bypass fat. The concentrate to roughage ratio of the TMR was fixed at 59:52:40:48. Growth performance was determined at 98, 120, 150, and 180 d of age, and rumen fermentation parameters were determined at 98, 120, 150, and 180 d of age, and microbial populations in rumen fluid from 181-d-old heifers were determined by real time polymerase chain reaction analysis. During 98 to 180 d of age, the average daily gain (ADG) did not differ among the groups ( $P > 0.05$ ). However, during 151 to 180 d of age, ADG of the heifers of group D (1.11 kg/d) were greater ( $P < 0.05$ ) than those of group A (0.77 kg/d). Feed utilization efficiency was improved in groups C and D ( $P < 0.05$ ) when compared with that in group A. Dietary ME did not influence ruminal pH value and  $\text{NH}_3\text{-N}$  concentration ( $P > 0.05$ ). Increasing level of ME significantly increased

ruminal proportion of propionate ( $P < 0.01$ ) and reduced that of acetate ( $P < 0.05$ ) and the ratio of acetate to propionate ( $P < 0.01$ ). Dietary treatments had no significant effect on the concentration of total volatile fatty acid ( $P > 0.05$ ). *Lachospira multipara* of group C tended to be higher than that of group D ( $P < 0.10$ ), and the numerical quantity of protozoan, *R. flaeafaciens*, *R. albus*, *B. fibrisolvens* and *P. ruminicola* of group C were greater than the other groups ( $P < 0.05$ ). Current study suggested that heifers supplemented with 2.99 Mcal of ME per kg of diet (group C) had better feed utilization efficiency and more abundant microbial flora compared with those fed other levels of ME.

**Key Words:** heifer, rumen fermentation parameter, ruminal microorganism

**T439 Effect of tallow and soybean oil addition to calf starters fed to dairy calves from birth to four months of age on calf performance and digestion.** T. Mark Hill\*, H. Gale Bateman II, James M. Aldrich, James D. Quigley, and Rick L. Schlotterbeck, *Nurture Research Center, ProVimi North America, Brookville, OH.*

Energy demands for calves can increase during periods of heat and cold stress. One way to potentially increase energy intake is to increase the energy density of the feed with fat. Trial 1a compared a control starter with no added fat or oil (CON) to starters with 2% tallow (TAL) and 2% soybean oil (SOY). Starters were 20% CP and 45 to 47% starch. Male Holstein calves (n = 48) that were initially 3 to 5 d of age were fed a 27% CP, 17% fat milk replacer at 0.66 kg DM daily and fully weaned by 42 d of a 56-d trial. Trial 1b estimated the digestion of the diets (employed chromic oxide as an indigestible digesta flow marker) using a subset of 5 weaned calves per treatment between d 52 and 56. Trial 2 used 48 Holstein calves initially 59 to 61 d of age fed starters CON and SOY blended with 5% chopped grass hay over a 56-d trial. Trial 3 used 48 Holstein calves initially 59 to 61 d of age fed starters CON and TAL blended with 5% chopped grass hay over a 56-d trial. Treatments were compared in a randomized ANOVA using repeated measures. In Trials 1a and 1b, pre-planned contrasts compared CON vs. TAL and CON vs. SOY ( $P < 0.05$ ) Compared with CON, calves fed SOY had reduced starter intake, ADG, and digestion of DM, OM, and CP vs. CON before 8 weeks of age ( $P < 0.05$ ). Compared with CON, calves fed SOY had reduced ( $P < 0.05$ ) ADG and change in hip width from 2 to 4 mo of age. Compared with CON, calves fed TAL had reduced ( $P < 0.05$ ) ADG and tended ( $P < 0.1$ ) to have reduced change in hip width from 2 to 4 mo of age. Calculated ME intake was not increased in any trial by added fat or oil. Tallow and soybean oil inclusion at 2% of the starter feed was not advantageous for calf growth before 4 mo of age.

**Key Words:** fat, oil, digestion

**T440 Relationships between udder resistance and dietary levels of copper and zinc.** Alfredo J. Escribano<sup>1</sup>, Juan Jose Mallo\*<sup>1</sup>, Luis Miguel Jiménez<sup>2</sup>, and Nuria Roger<sup>2</sup>, <sup>1</sup>*NOREL Animal Nutrition, Madrid, Spain,* <sup>2</sup>*Servet Talavera S.L., Talavera de la Reina, Toledo, Spain.*

Mastitis constitutes the main health issue in dairy cows farms, because it is widespread and leads to high economic losses. Copper and especially zinc have shown to increase cows' resistance to mammary infections. However, the levels of these minerals in rations varies greatly, and then, there is a gap of knowledge with regard to the relationship between the levels of trace minerals in the diets used to feed dairy cows, and the incidence of mammary diseases. To contribute in this sense, we carried out this study, with the objective to evaluate the relationship between

the level of zinc and copper and udder health parameters. For this purpose, 55 farms representative of the Spanish production systems were selected. The statistical procedure followed was based on bivariate correlation analysis and ANOVA. Results showed that negative and significant correlations existed between the level of dietary copper and the following indicators: Somatic Cell Counts ( $R^2 = 0.138$ ,  $P = 0.008$ ) and monthly rate of clinical mastitis ( $R^2 = 0.093$ ,  $P = 0.031$ ). However, no significant correlations were found with the rest of indicators: mastitis prevalence, new infections rate, curation rate in lactation, curation rate in the dry period, and new infections rate at calving. Regarding zinc levels, a significant correlation was found with the curation rate in the dry period ( $R^2 = 0.136$ ,  $P = 0.008$ ). As it can be observed, weak and few correlations were found. Surprisingly and contrary to most of the scientific knowledge, the effect of zinc had a low impact on udder health and resistance. These results could be showing interactions between minerals levels and other dietary compounds. Also, as levels of zinc were above the recommendations, a low response to increasing zinc levels was found. On the contrary, as not all diets were satisfying animals' needs for copper, higher responses to the level of this mineral were found. Further research is needed to have a deeper knowledge about both minerals interactions and the levels of minerals above which host's resistance do not increase significantly.

**Key Words:** dairy, mastitis, mineral

**T441 Use of a fermented ammoniated condensed whey product in lactating dairy cattle.** John P. McNamara<sup>\*1</sup>, Drina Huisman<sup>1</sup>, Heather M. Hastings<sup>1</sup>, and Gerald Poppy<sup>2</sup>, <sup>1</sup>Washington State University, Pullman, WA, <sup>2</sup>Packerland Whey Product Inc., Luxemburg, WI.

An existing fermented ammoniated condensed whey product (LactoWhey) has been in use in dairy rations for more than 30 years; however the last published studies were almost 30 years ago. It was of interest to test the efficacy of using this product in more modern dairy rations typical of the northwestern US dairy region which contain a wide variety of protein sources and by-product feeds. Therefore, a field trial was conducted at 3 commercial dairies and one university dairy. Lactowhey is liquid fermented ammoniated condensed whey containing 44% CP equivalent and 38% lactic acid equivalent from ammonium lactate. Lactowhey was added to the ration to replace other protein sources (primarily SBM and Canola meal) on a CP equivalent basis. Rations were balanced and fed to meet requirements to multiple pens on each commercial dairy (or through individual feeding gates on the university dairy) starting from an average of 66 DIM for 90 d. On the commercial dairies, milk tank weights and composition were recorded as well as DHIA test day milk and composition. A subset of 18 to 20 cows in each pen was followed for individual milk and composition, and rumen fluid and blood was sampled at 0, 3 and 13 wk of treatment (at the university dairy all cows ( $n = 20$  per treatment group) were also sampled at 6 wk of treatment. Milk production ranged from 35.9 to 45.5 kg/d at start of trial and 41.7 to 35.2 kg/d during treatment period, with no effect of treatment. Milk composition averaged 3.80% and 3.21% and 12.3 mg/dl for fat and protein and MUN across the period and farms, with no effect of treatment. There were no differences due to treatment in BW or BCS change, or rumen pH or in blood NEFA, glucose or BUN. The proportion of rumen bacterial DNA as measured by microbiome analysis was primarily *Prevotella* species, with a variety of other species represented. Species proportions varied among the 4 dairies and among individual cows, with a generally consistent pattern within dairies and individual cows. There was no consistent effect of the fermented whey on bacterial proportions. The work confirms the utility of this fermented

whey product to support high milk production in commercial herd when used to replace other protein sources.

**Key Words:** ammoniated whey, protein substitution, lactation

**T442 Evaluating varying dietary protein and energy levels for economical productive performance of Nili-Ravi buffalo heifer calves.** Zeeshan Muhammad Iqbal<sup>\*1</sup>, Muhammad Abdullah<sup>1</sup>, Khalid Javed<sup>1</sup>, Makhdoom Abdul Jabbar<sup>1</sup>, and Juan J. Loo<sup>2</sup>, <sup>1</sup>University of Veterinary and Animal Sciences, Lahore, Pakistan, <sup>2</sup>University of Illinois at Urbana-Champaign, Urbana, IL.

The optimal dairy management programs are based on rearing of heifers at a low cost without compromising growth rate. The aim of this study was to investigate the effect of dietary protein and energy level on growth and metabolic parameters of Nili-Ravi buffalo heifers. The experiment was conducted with 30 female buffalo calves (6–8 mo age) divided randomly into 3 treatment groups ( $n = 10$ /group). The animals in treatment A, B and C were offered 3 different levels of concentrate (CP 17% and 2.6 Mcal ME/kg) at 0.5%, 1% and 1.5% of body weight, respectively. In addition to this, all animals were given chopped green fodder (sorghum and maize) and fresh water ad libitum. All data were collected on individual animals for a total of 8 mo. As designed, the average daily protein and energy ( $585.94 \pm 15.36$  g,  $13.08 \pm 0.34$  MJ/kg) intake was greater in treatment C and lower ( $374.70 \pm 10.09$  g,  $9.56 \pm 0.26$  MJ/kg) in treatment A. The differences in intake did not affect ( $P > 0.05$ ) average daily gain of heifers ( $497.32.69 \pm 17.92$ ,  $503.63 \pm 19.09$  and  $532.77 \pm 20.67$  g/d). Thus, feed efficiency estimates were greater for treatment A ( $0.135 \pm 0.004$ ) as compared with treatments B and C ( $0.113 \pm 0.003$ ,  $0.108 \pm 0.004$ ). Dietary treatments did not affect ( $P > 0.05$ ) blood profile but serum urea concentration was greater in treatment B and C ( $50.08 \pm 2.05$ ,  $51.41 \pm 2.29$ ) compared with treatment A ( $42.34 \pm 1.59$ ). Increasing nutrient intake had no effect ( $P > 0.05$ ) on the digestibility of organic matter, fat, ash, ADF. However, the digestibility of diet DM, crude protein and ADF was greater in treatment C ( $71.14 \pm 0.96$ ,  $65.85 \pm 0.88$ ,  $56.09 \pm 1.02\%$ ) as compared with treatment A ( $66.06 \pm 2.32$ ,  $61.21 \pm 1.72$ ,  $51.43 \pm 2.13\%$ ) and B ( $67.22 \pm 1.09$ ,  $62.01 \pm 1.20$ ,  $52.91 \pm 1.32\%$ ). The cost for production (USD) of per kg gain was higher in treatment C (\$2.42) and lower in lower in treatment A (\$1.34) and B (\$1.96), respectively. As increasing dietary concentrate intake had no effect on growth rate, the estimated efficiency of utilization of nutrients was better and cost per kg gain was lower with the 0.5% concentrate inclusion. Thus, this level of concentrate in the diet at a rate of 0.5% of body weight appears most economical for rearing Nili-Ravi buffalo heifers from 6 to 14 mo of age.

**Key Words:** protein, energy, heifer

**T443 Trace mineral variation in dairy forages: Where are the hot spots?** J. R. Knapp<sup>\*1</sup>, W. P. Weiss<sup>2</sup>, R. T. Ward<sup>3</sup>, and K. R. Perryman<sup>4</sup>, <sup>1</sup>Fox Hollow Consulting LLC, Columbus, OH, <sup>2</sup>Dept. of Animal Sciences, The Ohio State University, Wooster, OH, <sup>3</sup>Cumberland Valley Analytical Services, Hagerstown, MD, <sup>4</sup>Micronutrients Inc., Indianapolis, IN.

The objectives of this study were to quantify the variation in trace mineral (TM) concentrations in forages, evaluate the contribution of US geographical location and harvest season to the TM variation, and identify areas where high variation in TM concentrations would preclude using standard reference concentrations. Trace minerals of interest were Cu, Mn, Zn, and Fe. Data from Cumberland Valley Analytical Services covering the period from 2009 to 2014 with concentrations of major



nutrients as well as mineral concentrations were used. Data were statistically filtered to remove outliers based on macronutrient concentrations and misidentified feeds before analyses of TM variation. As expected, TM concentrations for corn silage, legume hay, mixed mostly legume (MML) silage, and mixed mostly grass (MMG) silage displayed skewed distributions. TM values were log normalized before ANOVA with location, season, and their interaction as independent effects and total ash concentration as a covariate. Variation in TM concentrations due to location and total ash were significant ( $P < 0.0001$ ) and the largest sources. Season was often non-significant ( $P > 0.20$ ), while the interaction between location and season was significant ( $P < 0.05$ ). Whereas forages in some US locations have consistently low TM concentrations, forages in other areas have both high concentrations and high variation. These results support sampling and analysis of forages and formulation of dairy rations for TM based on analytical results rather than reference values. Recommendations for sampling frequency will vary by location as a function of the observed variation in TM concentrations.

**Table 1 (Abstr. T443).** Lowest and highest variation in observed Cu and Zn concentrations among U.S. geographical locations, given as median and 5th (p5) to 95th (p95) percentiles in mg/kg

Item	Lowest variation			Highest variation		
	Median	p5	p95	Median	p5	p95
<b>Cu</b>						
Corn silage	6.0	4.9	7.3	7.4	3.2	16.4
Legume hay	9.6	7.2	11.4	8.2	6.0	34.0
MML silage	7.8	5.7	9.6	10.9	5.7	30.1
MMG silage	8.8	6.1	9.4	7.8	4.7	37.9
<b>Zn</b>						
Corn silage	23.2	21.4	28.5	26.6	15.3	60.7
Legume hay	24.0	21.1	29.5	27.8	20.9	49.2
MML silage	28.3	25.8	35.0	35.7	21.8	69.7
MMG silage	23.8	20.0	27.6	29.1	18.4	97.5

**Key Words:** copper, zinc, mineral variation

**T444 Evaluating carbon and oxygen flux variability and heat production in dairy cows using a portable automated gas quantification system.** Andre B. D. Pereira<sup>\*1</sup>, Andre F. Brito<sup>1</sup>, Santiago A. Utsumi<sup>2</sup>, Brianna J. Isenberg<sup>1</sup>, and Kelly S. O'Connor<sup>1</sup>, <sup>1</sup>University of New Hampshire, Durham, NH, <sup>2</sup>Michigan State University, W. K. Kellogg Biological Station, Hickory Corners, MI.

The objectives of this study were (1) to evaluate the variability in spot short-term measurements of CO<sub>2</sub> ( $Q_{CO_2}$ ) CH<sub>4</sub> ( $Q_{CH_4}$ ) emissions and O<sub>2</sub> consumption ( $Q_{O_2}$ ), and (2) estimate heat production (HP) from dairy cows fed diets with different RDP and NSC profiles. Eight multiparous and 4 primiparous lactating Holstein cows were blocked by days in milk and milk yield into replicated 4 × 4 Latin squares. Cows received corn silage, grass-legume haylage, roasted soybean, and (1) corn meal and urea (negative control), (2) corn meal, soybean meal, and rumen-protected Lys-Met (positive control), (3) field peas, corn meal, and soybean meal, or (4) field peas, corn meal, soybean meal, and rumen-protected Lys-Met. Gas measurements were performed using a portable automated gas quantification system (Greenfeed, C-Lock Inc., Rapid City, SD) fitted with sensors for CO<sub>2</sub>, CH<sub>4</sub> and O<sub>2</sub>. Cows were sampled 3 times daily (about 8 h apart) for 7 d, with sampling points advanced 2 h in the following day to account for diurnal variation of gas flux. Heart rate was monitored for 4 d to calculate  $Q_{O_2}$ /heartbeat ( $n = 4$  cows). Heat production was calculated according to Kaufmann et al. (2011): [(4.96 + 16.07/respiratory quotient) ×  $Q_{CO_2}$ ]. Filtering

of gas sampling by variable head position or low visitation to the gas quantification system ( $n = 2$ ) resulted in lower number of gas flux points than anticipated (316 valid measurements of 1,008 possible). Each gas sampling lasted, on average, 3.7 min. For all treatments, the within animal CV averaged 25.5% ( $Q_{CH_4}$ ), 15.4% ( $Q_{CO_2}$ ), and 22.3% ( $Q_{O_2}$ ) and the between animal CV averaged 7.0% ( $Q_{CH_4}$ ), 4.3% ( $Q_{CO_2}$ ), and 5.8% ( $Q_{O_2}$ ). There was no difference between treatments for  $Q_{CH_4}$  (mean = 373.5 g/d),  $Q_{CO_2}$  (mean = 6,293 L/d),  $Q_{O_2}$  (mean = 7,307 L/d), HP (mean = 149 MJ/d),  $Q_{O_2}$ /heartbeat (mean = 66.1 L/heartbeat), and HP/heartbeat (mean = 20.7 MJ/heartbeat). Short-term spot measurements of gas flux can be used to estimate HP in dairy cows. However, sampling rate needs to be optimized, when using the automated gas quantification system, to reduce variability.

**Key Words:** carbon emission, oxygen consumption, heat production

**T445 Influence of a grape seed and grape marc meal extract (GSGME) on performance parameters, hepatic expression of pro-inflammatory and ER stress-related genes and antioxidant status in dairy cows.** Denise K. Gessner<sup>1</sup>, Christian Koch<sup>2</sup>, Franz-Josef Romberg<sup>2</sup>, Anne Winkler<sup>3</sup>, Georg Dusel<sup>3</sup>, Eva Herzog<sup>1</sup>, Erika Most<sup>1</sup>, Anne-Kathrin Blässe<sup>\*4</sup>, Bernhard Eckel<sup>4</sup>, and Klaus Eder<sup>1</sup>, <sup>1</sup>Justus Liebig University Giessen, Giessen, Germany, <sup>2</sup>Lehr- und Versuchsanstalt Hofgut Neumühle, Neumühle, Germany, <sup>3</sup>University of Applied Sciences Bingen, Bingen, Germany, <sup>4</sup>Dr. Eckel GmbH, Niederzissen, Germany.

At the onset of lactation, inflammation and stress of the endoplasmic reticulum (ER) occurs in the liver resulting in impaired hepatic function with undesirable effects on the health status of high yielding dairy cows. The present study investigated the hypothesis that feeding a polyphenol-rich grape seed and grape marc extract (GSGME) has the potential to suppress the inflammatory process and ER stress in the liver during early lactation. Twenty-eight German Holstein cows received either a total mixed ration (TMR, control group) or the same TMR supplemented with 1% GSGME (polyphenol content: 43 mg/g; Anta Ox, Dr. Eckel GmbH) from wk 3 ap to wk 9 pp (experimental unit: single cow). Milk, blood, and liver samples were taken. Data were statistically evaluated by the linear mixed-effects model of R. Dry matter intake of the cows did not differ between the 2 groups. However, the GSGME group had a greater milk performance ( $\Delta$  milk yield: + 3.7 kg/d;  $P < 0.05$ ;  $\Delta$  energy corrected milk: + 2.9 kg,  $P < 0.05$ ) from wk 2 to 9 of lactation. Cows of the GSGME group had lower hepatic mRNA concentrations of FGF21, a key marker of ER stress ( $P < 0.05$ ) and a tendency toward lower hepatic levels of triglycerides and cholesterol ( $P < 0.15$ ). A reduction of mRNA concentrations of various other ER stress response-related and pro-inflammatory genes could be observed in the GSGME group. Feeding GSGME had no effect on the antioxidative status ( $\alpha$ -tocopherol,  $\beta$ -carotene, TBARS and total antioxidant capacity in plasma). In conclusion, the study suggests that feeding polyphenol-rich plant extracts during early lactation could be a useful strategy to prevent inflammatory processes and ER stress in the liver and thus improve milk yield and animal health in dairy cows.

**Key Words:** grape seed extract, grape seed meal extract, dairy cow

**T446 Effects of *Saccharomyces cerevisiae* fermentation products on dairy calf: I. Pre-weaning performance and post-weaning stress.** G. M. Alugongo<sup>\*1</sup>, J. X. Xiao<sup>1</sup>, R. Chung<sup>2</sup>, S. Z. Dong<sup>1</sup>, S. L. Li<sup>1</sup>, I. Yoon<sup>2</sup>, and Z. J. Cao<sup>1</sup>, <sup>1</sup>State Key Laboratory of Animal Nutrition, Department of Animal Nutrition and Feed Sciences, China Agricultural University, Beijing, China, <sup>2</sup>Diamond V, Cedar Rapids, IA.

The aim of the study was to evaluate the effects of *Saccharomyces cerevisiae* fermentation products (SCFP) on performance and health of calves during the first 63 d of age. New Zealand Holstein calves (30 male and 30 female) at 2 d of age were blocked by sex and date of birth then randomly assigned within blocks to one of 3 treatments. A texturized grain starter was fed ad libitum containing 0 (Control), 0.5 or 1% SCFP (Original XPC, Diamond V) of DM. The basal diet contained steam-flaked corn (33.1%), wheat bran (7.6%), Soybean meal (14.3%), Extruded soybean (4.2%), canola meal (8.0%), and DDGS (16.0%) and was without antibiotics. In addition, the supplemented calves were fed 1 g/d SCFP (SmartCare, Diamond V) in milk from d 2 to 30. All calves were fed 4 L of colostrum within 1 h of birth and were subsequently fed milk twice daily until weaned at 56 d of age. Performance and health of all weaned female calves were monitored until 63 d of age to determine the effect of pre-weaning treatment of SCFP on weaning stress. Starter intake, fecal score and medical treatment were recorded daily. Body weight measures and blood samples were collected on d 2, 28, 56, and 63. Serum was analyzed for BUN, NEFA, IGF-1, glucose, albumin, total protein and oxidative biomarkers. Data were analyzed by MIXED procedure in SAS with contrast statement to declare Control vs. all SCFP, and 0.5% SCFP vs. 1% SCFP in starter grains. Calf was the experimental unit. Body weight, DMI, blood parameters and oxidative biomarkers did not differ. Supplementation of SCFP lowered ( $P < 0.05$ ) fecal scores, and diarrhea frequency. Number of medical treatments and medication costs were also lower ( $P < 0.05$ ) with SCFP. After weaning, SCFP reduced diarrhea in a dose-dependent manner with greater effect seen with 1% SCFP in starter grain. Overall, SCFP reduced diarrhea and pre-weaning medical costs and helped calves better manage weaning stress. The effects of SCFP were greater with the higher dosage of SCFP in starter grain.

**Table 1 (Abstr. T446).**

Item <sup>1</sup>	Treatment				Contrasts ( <i>P</i> -value)	
	CON	SCFP1	SCFP2	SEM	CON vs. SCFP1 and SCFP2	SCFP1 vs. SCFP2
ADG (kg/d)						
PrW	0.70	0.72	0.73	0.03	0.29	0.84
PoW	1.09	1.12	1.12	0.11	0.78	0.98
Fecal scores						
PrW	1.86 <sup>a</sup>	1.53 <sup>b</sup>	1.52 <sup>b</sup>	0.04	< 0.001	0.62
PoW	2.19 <sup>a</sup>	1.85 <sup>b</sup>	1.71 <sup>b</sup>	0.12	< 0.001	0.21
Diarrhea frequency						
PrW	20.81 <sup>a</sup>	10.80 <sup>b</sup>	9.20 <sup>b</sup>	1.83	<0.001	0.41
PoW	42.86 <sup>a</sup>	21.43 <sup>b</sup>	2.86 <sup>c</sup>	7.62	<0.001	0.03
Medications						
PrW	0.20 <sup>a</sup>	0.15 <sup>b</sup>	0.15 <sup>b</sup>	0.02	< 0.001	0.86
Price (\$)						
PrW	0.56 <sup>a</sup>	0.44 <sup>b</sup>	0.44 <sup>b</sup>	0.05	0.01	0.97

<sup>a,b,c</sup> $P < 0.05$ .

<sup>1</sup>PrW = pre-weaning; PoW = post-weaning; Medications = average no. of medical treatments/calf/day; Price = average cost of medication/calf/day.

**Key Words:** calf, *Saccharomyces cerevisiae* fermentation product (SCFP), health

**T447 Fibrolytic enzyme and corn silage differing in particle size for lactating dairy cows.** Gilson S. Dias Júnior\*<sup>1</sup>, Alan S. Pereira<sup>1</sup>, Fabiana F. Cardoso<sup>1</sup>, Renata A. N. Pereira<sup>3,2</sup>, and Marcos N. Pereira<sup>1,2</sup>, <sup>1</sup>Universidade Federal de Lavras, Lavras, MG, Brazil, <sup>2</sup>Better Nature Research Center, Ijaci, MG, Brazil, <sup>3</sup>Empresa de Pesquisa Agropecuária de Minas Gerais, Lavras. MG, Brazil.

The majority of Brazilian dairy farms use pull-type, one row forage harvesters for ensiling corn. For such machines, a kernel processor (KP) was developed (MU9000462-0 U2 INPI patent). We evaluated the response of cows to corn silage (CS) short particles-no KP (S, 3 mm theoretical cut) compared with long particles-KP (L, 8.5 mm). The supplementation of fibrolytic enzyme (F, Fibrozyme, Alltech) was evaluated in a 2 × 2 factorial. Sixteen individually fed Holsteins (110 ± 62 DIM) received treatments in 28-d periods, 4 × 4 LS. Corn was harvested at half milk line maturity and ensiled in 3 m diameter bag silos for 138 d. Silage S had 44.8% of particles below the 8 mm screen and L had 25.3%. Diets contained 31.6% of DM as the treatment CS, 17.1% of a standard CS, 1.9% Tifton hay, and 31.7% starch. Diet S had 1.9% of DM as NDF above the 19 mm screen, 12.3% from 8 to 19 mm, and 17.8% below 8 mm, and L had 3.0%, 15.6%, and 13.9%, respectively. The TMR was mixed manually twice per day immediately before feeding to avoid particle size reduction. Silage L increased 4% FCM (29.5 vs. 28.2 kg/d,  $P = 0.09$ ) and fat yield ( $P = 0.04$ ), and had no impact on DMI (22.0 kg/d,  $P = 0.50$ ). There were trends for reduced milk fat content and yield in response to F ( $P < 0.15$ ). Silage S reduced ruminal fluid pH and the proportion of acetate in VFA, but increased butyrate and protozoa content ( $P < 0.05$ ). Total-tract OM and NDF digestibility tended to be increased by F only when S reduced nutrient digestibility ( $P = 0.09$  for interaction of CS and F). The daily urinary allantoin secretion was increased only when F supplemented S ( $P = 0.03$  for interaction of CS and F). Fecal viscosity and DM content were also reduced when F was added to S ( $P < 0.02$  for interaction of CS and F). The loss of visible damaged corn kernels in feces was reduced by F ( $P = 0.05$ ), but the excretion of intact kernels was similar ( $P = 0.92$ ). Silage L tended to increase rumination bouts/bolus ( $P = 0.10$ ) and the duration of the first daily meal ( $P = 0.08$ ), and increased chewing activity per day and per unit of DMI ( $P < 0.03$ ). There was a trend for reduced PUN on F ( $P = 0.07$ ). Fibrolytic enzyme was beneficial to digestion only when fiber digestibility was reduced by short particles CS. Long particles CS increased lactation performance and chewing activity.

**Key Words:** fibrolytic enzyme, corn silage, particle size

**T448 Meta-analysis to examine the effect of supplemental sugar on dairy cow performance as influenced by diet nutrient components.** Stephen M. Emanuele\*<sup>1</sup>, Mary Beth de Ondarza<sup>2</sup>, and Charles J. Sniffen<sup>3</sup>, <sup>1</sup>Quality Liquid Feeds, Dodgeville, WI, <sup>2</sup>Paradox Nutrition LLC, West Chazy, NY, <sup>3</sup>Fencrest LLC, Holderness, NH.

A data set was compiled from published research (n = 85) that tested the effect of supplemental dietary sugar. Forage NDF ranged from 17 – 29% DM and calculated total sugar from 2.3 - 11% DM. Mixed model analysis was conducted using JMP statistical software (SAS Inst. Inc., Cary, NC) with nutrient parameters (CNCPS 6.1 with NDS platform, RUM&N Sas, Italy) as main effects. Model fit used treatment (Control vs. 1.5–3% added sugar vs. 3–5% added sugar vs. 5–7% added sugar), DIM (<150 or >150 DIM) within treatment, and Milk Yield (<33 or >33 kg/d) within treatment as independent factors with experiment as the random effect. Responses were analyzed with the following nutrients (%DM) in the model: starch, soluble fiber, forage NDF, ammonia, RDP, and Protein B2. Protein B2 is true protein that can contribute to dietary RUP. Number of cows per treatment was included as a weighting

factor. Nutrients with a positive effect on 3.5% FCM yield were sugar (+1.45 kg) ( $P < 0.04$ ), starch ( $P < 0.01$ ) and soluble fiber ( $P < 0.05$ ). Higher producing cows (>33 kg milk/d) had a greater response (2.14 kg FCM/d) to sugar than cows producing <33 kg of milk ( $P < 0.0001$ ). Nutrients with a positive effect on milk protein yield were sugar (+0.07 kg;  $P < 0.05$ ) and starch ( $P < 0.0001$ ). Cows producing > 33kg milk had a greater milk protein response to sugar (0.09 vs. 0.05 kg/d than cows producing <33 kg milk/d, ( $P < 0.001$ )). Sugar did not affect milk fat yield ( $P = 0.16$ ), although starch and soluble fiber had a positive effect ( $P < 0.05$ ). Sugar alone did not affect feed efficiency but starch ( $P = 0.04$ ), soluble fiber ( $P = 0.01$ ), and Protein B2 ( $P = 0.03$ ), all had a positive effect with a mean increase of 0.06 units with 3–5% supplemental sugar. Nonlinear statistical analysis using JMP Neural Net function determined optimal total calculated sugar to be 6.75% of diet DM. Optimal range for starch when feeding supplemental sugar was 20 – 25% of diet DM. Sugar did not affect milk fat or protein percent. In conclusion, 3 to 5% supplemental sugar (6 to 8% total sugar (%DM)), and starch (%DM) increased FCM (kg/d), and milk protein yield. To optimize response to supplemental sugar, the diet should contain >17% forage NDF, 10–11% RDP and less than 30% starch.

**Key Words:** sugar, meta-analysis, dairy

**T449 Pre- and post weaning performance and health of dairy calves fed milk replacers with plant and milk protein by-product sources balanced for selected amino acids.** Hugh Chester-Jones\*<sup>1</sup>, Dustin Dean<sup>2</sup>, David Ziegler<sup>1</sup>, and Kevin Halpin<sup>2</sup>, <sup>1</sup>University of Minnesota Southern Research and Outreach Center, Waseca, MN, <sup>2</sup>International Ingredients Corporation, St. Louis, MO.

One-hundred three (9 to 12 d old) individually fed Holstein heifer calves (41.8 ± 0.66 kg) were randomly assigned to 1 of 4 treatments to evaluate pre- (d 1–35) and post-weaning (d 36 to 49) calf performance and health when fed milk replacers (MR) with alternative protein sources. The study was conducted between April and August, 2014. All calves were fed a common all-milk non-medicated 22% CP:18% fat as-fed MR before the study was initiated. Calves were fed the non-medicated 22% CP:18% fat MR with either (1) All milk protein; (2) 33% of the protein replaced with dried partially hydrolyzed vegetable protein, partially autolyzed yeast and dairy solids; (3) 33% of the protein replaced with a milk extender made from dairy ingredients, edible lard, hydrolyzed vegetable proteins and yeast; or (4) 25% proteins from co-drying cheese and whey by-products. Milk replacers were formulated to be equivalent in lysine and sulfur amino acids. The MR were fed at 0.284 kg in 1.99 L water (12.5% solids) 2× daily from d 1 to 28 and 1× daily from d 29 to weaning at d 35. Calf starter (CS; 18% CP) and water were fed free choice d 1 to 49. During the first 7d of the study, calves were fed 1 part neomycin to 1 part oxytetracycline added to the MR solution to provide 22 mg/kg BW daily. There were no pre- or post weaning ADG differences ( $P > 0.05$ ). Calves averaged 0.74 kg/d gain for the 49 d study. There were no differences ( $P > 0.05$ ) in CS or total DMI intake which averaged 53.0 and 69.4 kg for the 49-d study, respectively. There were no differences in pre- and post weaning gain/feed. Gain/feed averaged 0.52 kg for the 49 d study. There were no differences in number of scouring d (d ≥ fecal score 3) pre- and post weaning. Days with fecal scores = 4 were higher ( $P < 0.05$ ) for Treatment 2 calves vs. those fed Treatments 3 and 4. Health treatment costs did not differ ( $P > 0.05$ ). Under the conditions of this study, replacing the total milk protein in MR with alternative sources resulted in calf performance and health similar to those fed an all milk protein MR.

**Key Words:** calf performance, milk replacer, alternative protein

**T450 The effect of biochemical fulvic acid (BFA) on heat stress and lactation performance in lactating cows.** Yifan Fan\*, Xiaoming Zhang, and Zhijun Cao, State Key Laboratory of Animal Nutrition, Department of Animal Nutrition and Feed Sciences, China Agricultural University, Beijing, China.

There was almost all cows suffered heat stress in hot and humid climate in most regions of China during summer. The objective of this study was to investigate the effect of biochemical fulvic acid (BFA) on heat stress and lactation performance in lactating cows. BFA is an aromatic nitrogen compound (Shenzhoulvyanmuye Co., Hebei Province, China). Its molecular construction determined it can be chelated trace elements. Thirty multiparous Holstein dairy cows were blocked into 2 groups by parity, days in milk, milk yield (MY) and somatic cell counts (SCC). This research was conducted from July through August 2014 (49 d). Two groups were fed same TMR. One was control group (CG) without BFA; the treatment group (TG) was added 25g BFA per cow per day. The quantity of different cows' parities is same in 2 groups (3 preparturient cows, 7 s lactation cows and 5 third lactation cows). The averaged days in milk of TG versus CG were 150 ± 35 d vs. 149 ± 38 d (Mean ± SD). The MY of TG and CG was 38.4 ± 4.8, 37.9 ± 6.6 kg, respectively. The SCC of TG was 4.17 × 10<sup>4</sup> compared with CG (5.06 × 10<sup>4</sup>). Cows were milked 3 times per day at 0430, 1230, and 2030 h. The MY, composition of milk, body temperature, blood samples, and respiratory rate were recorded. Data were statistically analyzed using the two-sample paired *t*-test for means procedure of SAS (version 9.2, SAS Institute Inc., Cary, NC). Although lactose, milk urea nitrogen (MUN), SCC, body temperature, and respiratory rate did not differ between TG and CG, milk yield (37.30 vs. 35.73 kg;  $P < 0.01$ ) and concentration of milk fat (3.54 vs. 3.13%;  $P < 0.05$ ) were significantly higher for TG compared with CG. However, the milk CP (3.29 vs. 3.18%;  $P < 0.01$ ) of CG was higher than TG. In the meanwhile, concentration of serum GSX-px (356 vs 207 μmol/L,  $P < 0.05$ ) was higher for TG compared with CG. These results suggested that adding BFA to lactating dairy cows during summer might increase milk yield, milk fat, relieve heat stress and not change the yield of milk CP.

**Key Words:** fulvic acid, heat stress, milk production

**T451 Pre- and post-weaning performance and health of dairy calves fed all-milk protein milk replacers or partially replacing milk protein with plasma and plant proteins in varying combinations.** Bruce Ziegler\*<sup>1</sup>, David Ziegler<sup>2</sup>, Hugh Chester-Jones<sup>2</sup>, Daniel Shimek<sup>1</sup>, Mary Raeth<sup>3</sup>, and David Cook<sup>4</sup>, <sup>1</sup>Hubbard Feeds, Inc., Mankato, MN, <sup>2</sup>University of Minnesota Southern Research and Outreach Center, Waseca, MN, <sup>3</sup>University of Minnesota Department of Animal Science, St. Paul, MN, <sup>4</sup>Milk Products, Chilton, WI.

One-hundred seven (2–5 d old) individually fed Holstein heifer calves (39.8 ± 0.70kg) were randomly assigned to 1 of 4 treatments to evaluate pre- (d 1–42) and post weaning (d 43–56) calf performance and health when fed milk replacers (MR) with alternative protein sources. The study was conducted between February and April, 2014. Calves were assigned to non-medicated 24% CP:20% fat MR with (1) All milk protein, (2) 25% of total protein from plasma, (3) 25% of total protein from plant peptide proteins, or (4) 12.5% plasma, 12.5% peptide proteins. All calves were fed a non-medicated 24% CP:20% fat MR at 0.34 kg in 2.39 L water (12.5% solids) 2× daily for the first 35 d and 1× daily d 36 to weaning at 42 d. Day 1 to 14, 1:1 neomycin:oxytetracycline was added to the MR solution to provide 22 mg/kg BW/d. Calf starter (CS; 18% CP as-fed) and water were fed free choice from d1. Milk replacer feeding rate was adjusted if ambient temperatures taken 0800 h were –23.3 to –28.3°C (+20% volume) or > –28.9°C (+40% volume). During the study, average



minimum temperatures were - 21.1, - 9.6 and 0.6°C for February (22 d < -17.8°C), March (3 d < -17.8°C) and April respectively. There were no pre- or post weaning ADG differences ( $P > 0.05$ ). Calves averaged 0.73 kg/d gain for the 56-d study. Milk replacer intake was similar for all treatments, averaging 25.1 kg. There were no differences ( $P > 0.05$ ) in CS or total DMI intake which averaged 51.5 and 76.7 kg for the 56-d study, respectively. There were no differences in pre- and post weaning gain/feed. Gain/feed averaged 0.58 kg for the 56 d study. Across treatments, calves doubled their initial BW and gained an average of 11.7 cm in frame growth. There were no differences in number of scouring d pre- and post weaning and health treatment costs. Under the conditions of this study, replacing 25% of the total milk protein in MR with alternative sources resulted in calf performance and health similar to those fed an all milk protein MR.

**Key Words:** calf performance, milk replacer, alternative protein

**T452 Flow of microbial crude protein out of the rumen when dairy cattle are supplemented with 2-hydroxy-4-methylthio-butanoic acid (HMTBa).** C. J. R. Jenkins<sup>1</sup>, S. C. Fernando<sup>1</sup>, C. L. Anderson<sup>3</sup>, N. D. Aluthge<sup>2</sup>, E. Castillo-Lopez<sup>4,1</sup>, H. A. Tucker<sup>5</sup>, G. I. Zanton<sup>5</sup>, D. Hostetler<sup>6</sup>, and P. J. Kononoff<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Nebraska-Lincoln, Lincoln, NE, <sup>2</sup>Food Science and Technology Department, University of Nebraska-Lincoln, Lincoln, NE, <sup>3</sup>School of Biological Sciences, University of Nebraska-Lincoln, Lincoln, NE, <sup>4</sup>Instituto de Investigaciones Oceanológicas, Universidad Autónoma de Baja California, Ensenada, México, <sup>5</sup>Novus International Inc., St. Charles, MO, <sup>6</sup>The School of Veterinary Medicine and Biomedical Sciences, University of Nebraska-Lincoln, Lincoln, NE.

Four multiparous, lactating Holstein cows (average DIM 169.5 ± 20.5 d), fitted with ruminal and duodenal cannulas, were used in a 4 × 4 Latin square with a 2 × 2 factorial arrangement of treatments to investigate the effects of 2-hydroxy-4-methylthio-butanoic acid (HMTBa; Alimet, Novus International, St. Charles, MO) when fed with diets deficient or in excess of metabolizable protein (MP) on milk production and composition, rumen microbial activity and protein flow out of the rumen, and rumen microbial community composition. Cows were housed in individual tiestalls and fed diets designated as “Low MP” or “High MP,” which were top dressed once daily with 250 g of a corn carrier or 250 g of a corn carrier containing 10% Alimet yielding 25 g of Alimet/hd/d. No interactions were observed between HMTBa and level of dietary MP, with the exception of ruminal acetate to propionate ratio ( $P = 0.04$ ). Milk yield was not affected by treatment and averaged 23.8 ± 2.06 kg. There was a tendency ( $P = 0.06$ ) for increased milk protein percent in cows receiving Low MP diets, averaging 3.30 ± 0.09% and 3.21 ± 0.09% for Low MP and High MP, respectively. The DM, OM, NDF, total-tract digestibilities were greater ( $P \leq 0.03$ ) in cows consuming the Low MP diet. Rumen pH was lower ( $P = 0.05$ ) in cows consuming High MP diets as well as in those consuming HMTBa ( $P < 0.01$ ). Rumen ammonia concentrations tended to be greater ( $P = 0.06$ ) in cows consuming HMTBa. Rumen VFA concentrations were greater ( $P = 0.02$ ) in cows consuming HMTBa. Duodenal DM flow, N flow, and bacterial N flow did not differ between treatments ( $P \geq 0.15$ ). The microbial community structure of cows receiving HMTBa was affected at the phylum level, as the relative abundance of *Fibrobacteres* was increased ( $P = 0.04$ ). Several correlations ( $P \leq 0.05$ ) between microorganisms and metadata were observed, including animal, HMTBa supplementation, level of dietary MP, DMI, digestibility, rumen ammonia, microbial N flow, and milk production. Results suggest that HMTBa affects rumen microbial

activity, irrespective of level of MP. Consequently, further investigation is warranted into the mechanism of these effects in the rumen.

**Key Words:** 2-hydroxy-4-methylthio-butanoic acid, microbial protein, rumen

**T453 Differences in microbial community structure associated with metabolizable protein and 2-hydroxy-4-methylthio-butanoic acid (HMTBa) supplementation when using in vitro and in vivo methods.** C. J. R. Jenkins<sup>1</sup>, N. D. Aluthge<sup>2</sup>, C. Anderson<sup>3</sup>, S. C. Fernando<sup>4,1</sup>, H. A. Tucker<sup>4</sup>, G. I. Zanton<sup>4</sup>, D. Hostetler<sup>5</sup>, and P. J. Kononoff<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Nebraska-Lincoln, Lincoln, NE, <sup>2</sup>Food Science and Technology Department, University of Nebraska-Lincoln, Lincoln, NE, <sup>3</sup>School of Biological Sciences, University of Nebraska-Lincoln, Lincoln, NE, <sup>4</sup>Novus International Inc., St. Charles, MO, <sup>5</sup>The School of Veterinary Medicine and Biomedical Sciences, University of Nebraska-Lincoln, Lincoln, NE.

The objective of the study was to investigate the effects of 2-hydroxy-4-methylthio-butanoic acid (HMTB; Alimet, Novus International, St. Charles, MO) and metabolizable protein, (Low MP or High MP) as observed in vitro and in vivo. Four multiparous, lactating Holstein cows (average DIM 169.5 ± 20.5 d), fitted with a ruminal and duodenal cannula were used in a 4 × 4 Latin square with a 2 × 2 factorial arrangement of treatments. Experimental periods were 28 d in length and cows received a top dress with either 0 or 25 g of HMTBa. On d 23 of each period and 4 h after feeding, 5 kg of whole ruminal contents were collected from all cows and rumen fluid was collected. Inoculum containing rumen fluid from each cow was added to tubes according to treatment and 0.5 g of each dietary treatment placed in a 100 mL fermentation tubes. After 24 h of fermentation, the fermentation was stop by snap freezing and a microbial pellet was isolated. DNA was isolated from the microbial pellet and was used for microbial community analysis. The Operational Taxonomic Unit(s) (OTU) based analysis of OTU in the Core Measureable Microbiome (CMM), revealed that the relative abundance of *Firmicutes* decreased ( $P = 0.01$ ) from the Low MP diet to the High MP diet (41.3 ± 4.9% and 30.2 ± 1.25%, respectively). In contrast, the proportion of *Tenericutes*, and *Spirochaetes* increased ( $P \leq 0.05$ ) from the Low MP to the High MP diet while an interaction between MP and HMTBa was observed ( $P \leq 0.05$ ) in *Bacteroidetes*, *Fibrobacteres*, and *Verrucomicrobia*. The relative abundance of phyla also differed ( $P \leq 0.01$ ) by method (in vitro vs in vivo) in *Firmicutes*, *Bacteroidetes*, *Proteobacteria*, TM7, *Tenericutes*, *Spirochaetes*, SR1, and *Verrucomicrobia*. The proportion of *Actinobacteria* tended to be higher ( $P = 0.02$ ) in vivo. Comparison of methods revealed differences in the average proportion of most phyla suggesting that in vitro fermentation after 24 h was not completely representative of that which was observed the in vivo.

**Key Words:** 2-hydroxy-4-methylthio-butanoic acid, rumen, bioinformatics

**T454 The effect of long-day photoperiod on behavior of lactating dairy cows.** Kira Macmillan\*, Santiago Espinoza, and Masahito Oba, University of Alberta, Edmonton, Alberta, Canada.

The objective of this study was to evaluate effects of photoperiod management on behavioral responses in lactating dairy cows. Thirty lactating cows (days in milk = 115 ± 33, body weight = 617 ± 70 kg) were subjected to a long-day photoperiod (LP; 16 h/d light) or a short day photoperiod (SP; 8 h/d light) in a tiestall barn (n = 15 per treatment). Data and samples were collected before animals were assigned

to treatments and after a 21-dadaptation period. Feeding behavior data were summarized for 4 time periods based on light management: Period 1 (7 p.m. to 3 a.m.; both treatments had no light), Period 2 (3 a.m. to 8 a.m.; only LP treatment had light), Period 3 (8 a.m. to 4 p.m.; both treatments had light), and Period 4 (4 p.m. to 7 p.m.; only LP treatment had light). All response variables were analyzed using a model including fixed effect of treatment, time, and treatment by time interaction with FIT Model Procedure of JMP. Period by treatment interaction was not observed for DMI, milk yield, lying time, or overall feeding behavior. However, in Period 2 (3 a.m. to 8 a.m.), there was a time by treatment interaction for lying time ( $P = 0.015$ ); LP treatment decreased lying time by 5.5 min/h while SP treatment did not. There was also tendency of an interaction for eating time ( $P = 0.064$ ); LP treatment increased eating time by 5.1 min/h while SP treatment did not. In addition, tendency for period by treatment interaction was observed for sorting behavior ( $P = 0.08$ ). The LP cows sorted, to a less extent compared with SP cows, against long particles (sorting index for particles on the screen with 19-mm pores: 91.4 vs. 78.0), and for small particles (sorting index for particles through the screen with 1.18-mm pores: 101.5 vs. 104.2). These results suggest that the provision of supplementary light may reduce sorting and modulate behavior in dairy cows, and that the behavior response to LP may differ depending on time of day in which supplementary light is given.

**Key Words:** long-day photoperiod, feeding behavior, sorting

**T455 Effects of *Saccharomyces cerevisiae* fermentation products on dairy calf: II) rumen fermentation and gastrointestinal development.** Jianxin Xiao<sup>\*1</sup>, Gibson Maswayi Alusingo<sup>1</sup>, Ruby Chung<sup>2</sup>, Dongshuang Zhao<sup>1</sup>, Shengli Li<sup>1</sup>, Ilkyu Yoon<sup>2</sup>, and Zhijun Cao<sup>1</sup>, <sup>1</sup>State Key Laboratory of Animal Nutrition, Department of Animal Nutrition and Feed Sciences, China Agricultural University, Beijing, China, <sup>2</sup>Diamond V, Cedar Rapids, IA.

The aim of the study was to evaluate the effects of *Saccharomyces cerevisiae* fermentation products (SCFP) in calf starter and milk on rumen fermentation and gastrointestinal tract development in the first 56 d of age. Fifteen Holstein bull calves were randomly assigned to one of 3 groups: a texturized calf starter containing 0 (Control), 0.5 or 1% SCFP (Original XPC, Diamond V) of DM from d 4 to 56. In addition, the supplemented calves were fed 1 g/d SCFP (SmartCare, Diamond V) in milk from d 2 to 30. All calves were fed 4 L of colostrum within 1 h of birth and were subsequently fed milk twice daily until weaned at d 56. Rumen fluid was collected by an esophageal tube 4 h after the morning feeding on d 28 and 56 to determine ruminal pH, ammonia N, and volatile fatty acids (VFA). At d 56, all 15 calves were harvested for slaughter weight, and rumen and intestinal morphological parameters. Papilla length (PL) and width (PW) were measured from 5 locations of rumen. Villus height (VH) and width (VW), crypt depth (CD) and villus height to crypt depth ratio (VCR) were measured in duodenum, jejunum and ileum. Data were analyzed by MIXED procedure in SAS with contrast statement to declare Control vs. all SCFP, and 0.5% SCFP vs. 1% SCFP in starter grains. Ruminal pH and ammonia N were not altered by SCFP. However, SCFP supplementation exhibited higher ( $P < 0.05$ ) ruminal butyrate concentration than Control at d 56. Supplementation of SCFP stimulated ( $P < 0.05$ ) PL in the rumen. Although SCFP had limited effect on VH, it reduced ( $P < 0.01$ ) the CD in jejunum, and increased ( $P < 0.05$ ) VCR in all segments of small intestine, especially when supplemented at a higher dosage in starter. In conclusion, SCFP improved gastrointestinal development, possibly

due to increased butyrate production, and greater effect was observed with higher dosage of SCFP.

**Key Words:** calf, *Saccharomyces cerevisiae* fermentation product (SCFP), gastrointestinal morphology

**T456 Effects of exogenous C16:0 and C18 fatty acids (FA) on milk lipid metabolism in bovine mammary epithelial cells.** N. Dan<sup>\*1</sup>, H. Zhang<sup>2</sup>, C. J. Ao<sup>1</sup>, and Khas-Erdene<sup>1</sup>, <sup>1</sup>College of Animal Science, Inner Mongolia Agricultural University, Hohhot, Inner Mongolia, China, <sup>2</sup>College of Animal Science, Inner Mongolia University for the Nationalities, Tongliao, Inner Mongolia, China.

The objective of this study was to examine the regulatory effects of C16:0, C18:0, C18:1c9, C18:2n6, and C18:3n3 on lipid metabolism of bovine mammary epithelial cells. After plating for 48h, bovine mammary epithelial cells cultured from Chinese Holstein cows were bathed in one of the 6 treatments: 100  $\mu$ M of C16:0 + 5  $\mu$ M of C18:0 + 100  $\mu$ M of C18:1c9 + 25  $\mu$ M of C18:2n6 + 1.2  $\mu$ M of C18:3n3 as Control; C18:0 + C18:1c9 + C18:2n6 + C18:3n3 as C16:0 absent treatment (A-C16:0); C16:0 + C18:1c9 + C18:2n6 + C18:3n3 as C18:0 absent treatment (A-C18:0); C16:0 + C18:0 + C18:2n6 + C18:3n3 as C18:1 absent treatment (A-C18:1); C16:0 + C18:0 + C18:1c9 + C18:3n3 as C18:2 absent treatment (A-C18:2); C16:0 + C18:0 + C18:1c9 + C18:2n6 as C18:3 absent treatment (A-C18:3). Key lipogenic genes were analyzed using quantitative PCR, and the FA composition in cells was quantified with gas chromatography. Data were analyzed by the ANOVA procedure of SAS. Cellular triglyceride contents in cells treated with A-C18:0, A-C18:2, A-C18:3 ( $P < 0.01$ ) and A-C18:1 ( $P < 0.05$ ) were decreased compared with the control. Absence of C16:0 induced greater level of LPL ( $P < 0.05$ ). The level of ACSL1 in A-C18:3 was higher ( $P < 0.05$ ) than that of A-C16:0 and A-C18:2. The mRNA abundance of FABP3 was enhanced by A-C18:3 ( $P < 0.05$ ). The expression of SCD was reduced by A-C18:1 ( $P < 0.05$ ). The abundance of CD36 showed reduction in A-C16:0, A-C18:1, and A-C18:2 ( $P < 0.05$ ). Expression of SREBP-1 in A-C18:0, A-C18:1 and A-C18:2 were lower than that of A-C18:3 ( $P < 0.05$ ). No effects of the treatments on ACACA and FASN were observed ( $P > 0.05$ ). Furthermore, C18:2n6 and C18:1c9 compositions in cells were increased by A-C16:0 and A-C18:0 ( $P < 0.05$ ), respectively. Percentage of C16:0 in A-C18:1, and that of C16:0 and C18:0 in A-C18:3 were enhanced ( $P < 0.05$ ), respectively. Cells in A-C18:3 had greater SFA and lower UFA compared with other treatments ( $P < 0.05$ ). In general, extracellular C18 FA availability has a strong effect on cellular triglyceride synthesis; different exogenous long-chain FAs showed no difference on inhibition of de novo FA synthesis, but greatly influence expression of lipogenic genes and milk FA composition.

**Key Words:** milk fat, lipid metabolism, gene expression

**T457 Does ruminal pH affect flow of N fractions in high-producing dairy cows?** Danilo Domingues Millen<sup>\*1</sup>, Charles Schwab<sup>2</sup>, and Sergio Calsamiglia<sup>3</sup>, <sup>1</sup>Sao Paulo State University, Dracena, Sao Paulo, Brazil, <sup>2</sup>Department of Animal and Nutritional Sciences, University of New Hampshire, Durham, NH, <sup>3</sup>Animal Nutrition and Welfare Service, Universidad Autonoma de Barcelona, Bellaterra, Spain.

There is continuous culture (Calsamiglia et al., 2008) and in situ (Devant et al., 2001) evidence that nonammonia nonmicrobial N (NANMN) flow from the rumen is increased under high concentrate-low pH conditions commonly found in high producing dairy cows. The objective of this study was to evaluate the effect of dietary conditions, including rumi-

nal pH, on the ruminal outflow of N fractions in lactating dairy cows. Peer-reviewed published studies ( $n = 45$  papers;  $n = 176$  data points) designed to investigate flow of N fractions to the small intestine in dairy cows were used. Rumen pH, organic matter truly digested in the rumen (OMTDR), efficiency of microbial protein synthesis (EMPS), and intakes of RUP (RUPI), N (NI), OM (OMI), DM (DMI), and concentrate (CONCI) were the independent variables. Microbial N (MN), NANMN, and nonammonia N (NAN) were the dependent variables in the analysis. Study was included as random effect. The relationship among variables was subjected to multivariate regression analysis by MIXED procedure of SAS. The contribution of each independent variable to the changes observed in each dependent variable was assessed by calculating semipartial correlations ( $pR^2$ ). The RUPI ( $P < 0.0001$ ;  $pR^2 = 0.47$ ), OMTDR ( $P < 0.0001$ ;  $pR^2 = 0.10$ ), EMPS ( $P = 0.003$ ;  $pR^2 = 0.02$ ) and CONCI ( $P = 0.05$ ;  $pR^2 = 0.01$ ) explained 60% of the

variation in NANMN. The contribution of rumen pH to the changes in NANMN was close to, but not significant ( $P = 0.13$ ). The average flow of NANMN at  $pH < 5.8$  ( $n = 14$ ) was 325 g N/d compared with 241 g N/d at  $pH > 5.8$  ( $n = 162$ ), with residuals showing some degree of lack of fit at low pH. However, the small number of data at  $pH < 5.8$  limits the power of the test. The EMPS ( $P < 0.0001$ ;  $pR^2 = 0.37$ ), OMTDR ( $P < 0.0001$ ;  $pR^2 = 0.31$ ), and DMI ( $P < 0.0001$ ;  $pR^2 = 0.19$ ) explained 87% of the variation in MN. The RUPI ( $P < 0.0001$ ;  $pR^2 = 0.27$ ), EMPS ( $P < 0.0001$ ;  $pR^2 = 0.21$ ), OMI ( $P < 0.0001$ ;  $pR^2 = 0.09$ ) and NI ( $P = 0.01$ ;  $pR^2 = 0.01$ ) explained 58% of the variation in NAN flow. The limited in vivo data does not allow to confirm the hypothesis that pH affects the flow of NANMN, but suggests that its flow is increased at low pH.

**Key Words:** pH, microbial, nonammonia



## Ruminant Nutrition: General II

### T205 Effects of prophylactic subcutaneous calcium treatment at calving on macro mineral status and health in Holstein cows.

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The objective of this study was to evaluate the effects of prophylactic subcutaneous (SC) Ca treatment at calving on DMI at 1 DIM, serum concentrations of Ca, P, Mg and subclinical hypocalcemia (SHC) and metritis incidence on a commercial HO herd. Cows were blocked based on parity [first (n = 143), second (n = 108) and third or greater (n = 124)], and randomly assigned to treatments after calving. Treatments were no treatment (Control; n = 190); 250 ml of 40% Ca borogluconate (CB) by SC injection at calving (1SC<sub>250</sub>; n = 72); 500 ml of 40% CB by SC injection at calving (1SC<sub>500</sub>; n = 63); 250 ml of 40% CB by SC injection at calving and at 12 to 18 h after the first injection (2SC<sub>250</sub>; n = 50). Blood samples were collected immediately after calving and at 1, 2, 4, and 7 DIM. Dry matter intake was measured for 24 h after calving in individual calving pens. Intake was analyzed with GLM procedure, including treatment, parity, and their interactions in the model. Serum Ca, P and Mg were analyzed as repeated measures with mixed procedure of SAS. Metritis (foul-smelling uterine discharge with fever) and SHC (Ca ≤8.5 mg/dl) were analyzed with PROC GENMOD with binomial distributions and logit link functions including treatment, parity, Ca status (≤8.5 vs >8.5 mg/dl) and their interactions. At 1 DIM, intake was greater (P = 0.004) for 1SC<sub>250</sub> (13.5 kg), 1SC<sub>500</sub> (15.0 kg) and 2SC<sub>250</sub> (15.6 kg) relative to control (12.4 kg). Compared with control (8.4 mg/dl), postpartum serum Ca was greater for 1SC<sub>250</sub>, 1SC<sub>500</sub> and 2SC<sub>250</sub> with 8.9, 9.2 and 9.0 mg/dl respectively. No treatment effect was found on serum P and Mg. The odds of SHC were 3.7, 3.0 and 14.0 times greater (P = 0.01) for control than 1SC<sub>250</sub>, 1SC<sub>500</sub> and 2SC<sub>250</sub> respectively. The odds to develop metritis tended to be 1.7 times higher for control than 2SC<sub>250</sub> (P = 0.07; 95% CI = 0.9- 3.3). These results suggest that prophylactic SC injections of Ca at calving can improve postpartum Ca status in HO cows and DMI at 1 DIM. Given the reduction on metritis and SHC with treatment, the evaluation of immune status warrant further investigation.

**Key Words:** calcium status, subcutaneous injection, metritis

### T458 Effect of time of gestation on fatty acid transporter and receptor mRNA concentration in bovine placenta.

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The aim of the study was to evaluate the effect of time of gestation on fatty acid transporter and receptor mRNA concentration in maternal and fetal bovine placenta. Placentas from 12 cows at different thirds of gestation (n = 4 per third) were sampled at slaughter to measure FATP-1, FATP-4, FABP-1 mRNA concentration in maternal (caruncles) and fetal (cotyledons) side. Once the placenta was removed, 1cm<sup>2</sup> was dissected and, divided into caruncles and cotyledons, stored in sterile tubes, dropped into liquid nitrogen and kept at -80°C until rtPCR analysis. Extraction of RNA was performed with TRIzol. Fetal and maternal placenta cDNA was subjected to qPCR assays using EvaGreen as intercalating dye (KAPA FAST, Biosystems, Woburn). Quantitative PCR was performed in a Rotor Gene Q thermocycler (Qiagen). Rela-

tive mRNA concentration was calculated by ddCt method using β actin as housekeeping gene. Data were analyzed as a complete randomized design with a 3 × 2 factorial arrangement, using the mixed procedure (SAS 9.3) with repeated measurements on space. Time of gestation, size of the placenta and their interaction were fixed factors, whereas animal was a random factor. There was a time by treatment interaction (P < 0.01) on FATP-1 mRNA expression of due to a greater mRNA expression in cotyledons on the first third of gestation as compared with the concentration in caruncles (Table 1). On the second and third thirds of gestation, the mRNA concentration in cotyledons decreased, reaching a similar concentration to that observed in caruncles (Table 1). FATP-4 and FABP-1 mRNA concentration were not different (P > 0.1, Table 1). We conclude that FATP-1 would play an important role in fatty acid transport during early fetal development.

**Table 1 (Abstr. T458).** Relative mRNA concentration of FATP-1, FATP-4, FABP-1 mRNA in different thirds of gestation on maternal (caruncles-M) and fetal (cotyledons-F) side on bovine placenta

Item	M			F			SEM	P-value		
	1	2	3	1	2	3		Side	Third	Third
FATP-1	0.63	0.30	0.94	2.14	1.26	0.92	0.29	0.01	0.15	0.07
FATP-4	1.85	0.90	1.94	1.94	0.99	0.78	0.78	0.55	0.42	0.53
FABP-1	3.07	2.01	2.62	3.29	4.09	3.77	1.42	0.35	0.99	0.80

**Key Words:** fatty acid transporter, fatty acid receptor, bovine placenta

### T459 Ensiling carinata meal with forages to decrease glucosinolate concentrations.

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Carinata meal (CM) has high quality protein, but it also has high concentration of sinigrin, a glucosinolate, which limits its use as a feedstuff. Our objective was to determine if ensiling CM with forages would decrease sinigrin concentration without compromising fermentation. Two trials were conducted, one on ensiling CM with alfalfa haylage (AH) and one with corn silage (CS). For both trials 3 blends of CM and forage were made of 0:100, 25:27, and 50:50 on a DM basis. In both approximately 637 g of DM for each the 3 blends were packed in 942 cm<sup>3</sup> microsilos in quadruplicate for 0, 7, 21 and 60 d of ensiling. Sinigrin was greatest (P < 0.01) in the 50:50 and decreased over time (P < 0.01) in the 25:75 and 50:50 in both trials. There was no treatment by d interaction for AH blends, but there was a treatment by d interaction for the CS blends for sinigrin. The pH decreased in all blends over time, but was greater in the 50:50 compared with the other blends. Acetic acid and lactic acid increased over time in all blends. Acetic acid was less in the AH blends with increased CM. There was no treatment effect on acetic acid for the CS blends. Lactic acid was less in both trials with increased inclusion of CM. In both trials, CP increased with inclusion of CM. The CP was similar over d of ensiling in AH blends, but tended (P = 0.05) to decrease over d in CS blends. In both trials, NDF was less with the addition of the CM and there was a treatment by d interaction (P < 0.01) in CS, and a tendency (P = 0.08) for interaction in the AH trial. Ensiling CM with forage decreases sinigrin concentration, without major detriment to silage fermentation.

**Table 1 (Abstr. T459).**

Item, %	Blend	0:100	25:75	50:50	SEM	Treatment	Day	Trt × Day
DM								
Sinigrin, mg/g	AH	—	2.24	5.78	0.13	<0.01	<0.01	0.65
	CS	—	3.67	7.12	0.17	<0.01	<0.01	<0.01
pH	AH	4.89	4.83	5.0	0.02	<0.01	<0.01	<0.01
	CS	3.96	4.23	4.47	0.04	<0.01	<0.01	0.90
Acetic acid	AH	1.32	1.05	0.75	0.04	<0.01	<0.01	<0.01
	CS	0.89	0.96	0.94	0.03	0.22	<0.01	0.61
Lactic acid	AH	3.71	2.57	1.77	0.07	<0.01	<0.01	<0.01
	CS	2.64	2.35	2.16	0.05	<0.01	<0.01	<0.01
CP	AH	24.6	29.6	32.6	0.96	<0.01	0.20	0.09
	CS	6.2	15.6	25.0	0.28	<0.01	0.05	0.19
NDF	AH	38.5	34.8	31.8	0.25	<0.01	<0.01	0.08
	CS	33.9	30.4	28.7	0.22	<0.01	0.05	<0.01

**Key Words:** carinata meal, glucosinolate, ensiling

**T460 Double-layered S/O/W emulsions as rumen delivery system for potential site-selective delivery of lysine in cows.** Yong-guang Guan and Qixin Zhong\*, *Department of Food Science and Technology, University of Tennessee, Knoxville, TN.*

Essential amino acids such as lysine are deficient in most ingredients fed to cows. Direct supplementation of lysine in cow diets however is questionable because the residence time of diets in the rumen is as long as 12 h and lysine can be hydrolyzed by some rumen microorganisms. Safe rumen delivery systems are therefore highly demanded to utilize essential amino acids. The objective of the present work was to study solid/oil/water (S/O/W) emulsions as a potential rumen delivery system that can release most lysine post-rumen. Spray-dried lysine solid particles were prepared and were suspended in soybean oil. The S/O suspension was emulsified in neutral aqueous suspensions with 2, 10, and 30% whey protein concentration (WPC), which was then acidified to pH 3.0 and mixed with a 0.5% sodium alginate solution at pH 2.0 to prepare double-layered emulsion droplets. The emulsion was then spray-dried. The dimension and morphology of droplets were studied using a laser diffraction particle size analyzer, optical microscopy, and scanning electron microscopy. The amount of unencapsulated lysine and the amount of lysine released from spray-dried powder after incubation in simulated rumen, abomasum, and intestinal juices at 37°C were determined using high performance liquid chromatography. All experiments were carried out at least in triplicate. ANOVA was carried out to determine significant differences between treatments at a significance level of 0.05. Results showed that increasing WPC concentration significantly ( $P < 0.05$ ) improved the encapsulation efficiency of lysine, up to 78.5% for the 30% WPC treatment. The particle dimension was around 10 µm based on particle size analysis and microscopy. About 10% lysine was released after 12-h incubation in the simulated rumen juice containing proteases. After an additional 3-h incubation in the simulated abomasum juice at pH 2 with pepsin, the cumulatively released lysine was less than 18%. After further in vitro digestion in the simulated intestinal juice at pH 7.0 with pancreatin, pepsin, and bile salt, the cumulatively released lysine was up to 56%. Therefore, the S/O/W emulsion can be developed as delivery systems to release most lysine post-rumen to supply important amino acids for cows.

**Key Words:** rumen delivery system, lysine, solid/lipid/water emulsions

**T461 Effects of dietary n-6 and n-3 fatty acid sources on intake, digestibility, rumen microbes and fatty acid profile in sheep.** Sardar M. Amanullah<sup>1,3</sup>, Sam Churl Kim<sup>\*1</sup>, Dong Hyeon Kim<sup>1</sup>, Hyuk Jun lee<sup>1</sup>, Young Ho Joo<sup>1</sup>, and Eun Tae Kim<sup>2</sup>, <sup>1</sup>*Division of Applied Life Science (BK21Plus, Insti. of Agric. & Life Sci.), Gyeongsang National University, Jinju, Gyeongsangnam-do, South Korea,* <sup>2</sup>*Dairy Science Division, National Institute of Animal Science, RDA, Cheonan, Chungcheongnam-do, South Korea,* <sup>3</sup>*Bangladesh Livestock Research Institute, Dhaka, Bangladesh.*

A study was conducted to know the effects of dietary oil sources rich in n-6 and n-3 FA on nutrient intake, digestibility, rumen microbial population and fatty acid (FA) profile in sheep. Four pre-pubertal female sheep (48.03 ± 3.69 kg) were housed individually in digestion crates in 4 × 4 Latin square design. Timothy hay and concentrate mixture (3:7 ratio) was the basal diet which was supplied at 2% of live weight on DM basis. Treatments were control (CON), corn oil (CO: n-6), linseed oil (LSO; n-3), and calcium salt of FA (Ca-Salt; protected n-6). Oil sources were pre-mixed with concentrate at 3% of fresh weight. Each period consisted with 10 d of adaptation and 5 d of sample collection. The orsts, feces and urine were collected daily before morning feed during the collection period. Blood sample was collected on collection d 4 after 1 h of morning feed, while rumen fluid sample was collected on d 5 at 3 h after morning feed by stomach tube. Data were analyzed using the GLM procedure of SAS. It was observed that intake and digestibility of nutrients were not affected by the supplementing oil sources ( $P > 0.05$ ). Real Time PCR revealed no differences in DNA concentration of methanogenic archaea ( $P = 0.047$ ), *Fibrobacter succinogens* ( $P = 0.307$ ) and *Ruminococcus flavefacience* ( $P = 0.327$ ) among treatments. On the other hand, DNA concentration of rumen ciliate (*Entodinium*) was reduced ( $P = 0.018$ ) by Ca-Salt, while *Ruminococcus albus* was reduced ( $P = 0.028$ ) by LSO compared with the others. The ruminal concentration of C18:0 was highest ( $P = 0.012$ ), but C16:0 ( $P = 0.001$ ) and C16:1n-9 ( $P = 0.018$ ) were lowest in LSO treatment. Other major 18-carbon FAs remained unaffected ( $P > 0.05$ ) in rumen contents. Plasma FA profile showed that, not only C18:3n-3 ( $P < 0.001$ ), but also C20:5n-3 ( $P = 0.033$ ) were increased by LSO supplementation. Consequently, n-6 to n-3 FA ratio ( $P = 0.007$ ) was decreased by LSO treatment. Results indicated that, increased concentration of n-3 FA and decreased n-6 to n-3 FA ratio in plasma can be achieved by supplementing linseed oil in ruminant's diet without affecting intake and digestibility of nutrients.

**Key Words:** fatty acid profile, oil source, rumen microbe

**T462 Approaches to confidence intervals for the energy requirements of beef cattle.** Hugo Colombarolli Bonfá\*, Edenio Detmann, Paulo Roberto Cecon, Sebastião de Campos Valadares Filho, and José Gilson Louzada Regadas Filho, *Universidade Federal de Minas Gerais, Viçosa, Minas Gerais, Brazil.*

The objective of this study was to propose approaches to the confidence intervals for the net and metabolizable energy requirements for maintenance and for the efficiency of utilization of metabolizable energy for maintenance and weight gain in beef cattle. A simulated population of 100,000 animals was used to demonstrate the distributional properties of the energy requirements. One hundred random samples (n = 100) were taken from a simulated population (n = 100,000). From those samples it was obtained through the Qui-Square and Kolmogorov-Smirnoff tests that net and metabolizable energy requirements for maintenance can be studied by using the properties of the normal distribution ( $P > 0.05$ ). This condition can be reinforced by the sigmoid pattern showed by the upper limits of the confidence intervals when plotted in a scatter graph. The confidence intervals approaches were proposed and demonstrated

using the properties of the normal distribution, and using approaches based on anamorphosis techniques and on utilization of a Taylor's series. A data set of 158 animals was used to demonstrate and validate the proposed approaches. The methods that were developed in this study allow obtaining the variance information and confidence intervals for the energy requirements of cattle more affordable by ruminant nutrition researchers, who can obtain confidence intervals for both energy requirements and efficiency of energy utilization based on information from one single experiment. The results demonstrated the feasibility of use of such approaches, which are relevant tools for the practice of inductive statistics and for the inter- and intra-experimental comparisons.

**Key Words:** efficiency of use of metabolizable energy, nutrient requirements of cattle, statistical inference

#### **T463 Predicting ruminal methane inhibition by condensed tannins using nonlinear exponential decay regression analysis.**

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Methane (CH<sub>4</sub>) is a potent greenhouse gas that is normally produced by microbial fermentation in the rumen and released to the environment during the eructation process. Prediction of ruminal CH<sub>4</sub> is important for ruminant nutrition, especially for determination of ME intake. Equations have been developed to predict ruminal CH<sub>4</sub> production based on dietary constituents, but none have considered condensed tannins (CT). Our objective was to develop an equation to predict ruminal CH<sub>4</sub> inhibition by CT. We gathered CH<sub>4</sub> production data from 24- to 48-h in vitro fermentation of diverse forages containing different concentrations of CT over the course of 3 years. Our analysis included 113 observations. The predictor variable CT was regressed on the response variable CH<sub>4</sub> using PROC NLIN of SAS and the Gauss-Newton method to converge the parameters of the nonlinear regression. We used the following exponential decay model to express the relationship between CT and CH<sub>4</sub>:  $Y = span \times e^{(-K \times X)} + plateau$ , where  $Y$  is CH<sub>4</sub>, g/kg FOM;  $span$  is the difference between  $Y$  when CT equals zero and the  $plateau$  ( $Y$  value at infinite), g/kg FOM;  $K$  is the fractional rate of decline, 1/% DM; and  $X$  is CT concentration, % DM. The following nonlinear exponential decay regression equation was developed:  $CH_4 = 113.6 \times e^{-0.1751 \times CT} - 2.18$  ( $r^2 = 0.52$ ;  $P < 0.0001$ ). This equation predicted that CH<sub>4</sub> production could be reduced by 50% when CT is about 3.85% DM. We used several statistics to evaluate the adequacy of this equation, including precision and accuracy. We determined that this equation is more accurate when screening CT-containing forages for their potential ability to mitigate CH<sub>4</sub> production by ruminants when the CT concentration is greater than 5% DM. We concluded that despite the large degree of variability in ruminal CH<sub>4</sub> production, this equation can be used as a tool for predicting potential ruminal CH<sub>4</sub> inhibition that occurs when feeding CT-containing forages to ruminants. Future research should focus on the development of predictive equations when other potential reducers of ruminal CH<sub>4</sub> are used in conjunction with CT.

**Key Words:** forage, greenhouse gas, modeling

#### **T464 Effect of lipid sources with different fatty acid profiles on intake and nutrient digestion of feedlot Nellore steers.**

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The present study was conducted to determine the effect of lipid sources with different fatty acid profiles on intake and nutrient digestion. Ten rumen and duodenal fistulated Nellore steers (268 body weight  $\pm$  27 kg) were distributed in a double 5  $\times$  5 Latin square (5 periods of 20 d, including 15 d for diet adjustment and 5 d for sample collection). Dietary treatments were: without fat (WF), palm oil (PO), linseed oil (LO), protected fat (PF; Lactoplus), and whole soybeans (WS). The roughage feed was corn silage (600 g/kg on a DM basis) plus concentrate (400 g/kg on a DM basis). Throughout the entire experimental period, the allowance was adjusted to allow refusals of approximately 100 g/kg in relation to the total amount consumed on the previous day. Feed refusals were collected and weighed before feeding for the first 5 d of each experimental period. Feces were collected for 5 d to estimate the digestibility of dietary constituents. Data were analyzed using the PROC MIXED of SAS. The higher intake ( $P < 0.001$ ) of DM, OM was found in animals on the diet with PF and WF. Animals fed with WS had intermediate intake, whereas the diet PO had the lowest intake. The intake of DM and OM of animals fed with LO did not differ from PO and WS diets. The higher EE intake ( $P < 0.001$ ) occurred in animals receiving PF. However, animals fed PO, showed intake similar to that of animals fed WF. There were no differences for the NDF intake ( $P > 0.05$ ). The treatments with PO and LO decreased total digestibility of DM ( $P = 0.021$ ), OM ( $P = 0.048$ ) and EE ( $P < 0.001$ ). Animals fed with the WF diet had lower ( $P < 0.001$ ) EE digestibility than other diets. The addition of LO resulted in the lowest NDF digestibility ( $P = 0.047$ ; 0.40 kg/kg), and the NDF digestibility was not different among the other diets. These observations may be linked to the number of double bonds in the fatty acid molecules and to the high availability of this source (LO), which tends to reduce cellulolytic bacteria proportion. Diets with lipid sources with different fatty acid profiles affected intake and nutrient digestion.

**Key Words:** beef cattle, lipid, metabolism ruminant

#### **T465 Experimental design and data-reporting needs to help support the advancement of nutrition research and nutrient requirement models.**

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The National Animal Nutrition Program, National Research Support Project 9, supports efforts in livestock nutrition, including the National Research Council's committees on the Nutrient Requirements of Animals. Our objective was to review the current status of experimentation and data reporting in animal nutrition literature and to provide suggestions for the advancement of animal nutrition research and the ongoing improvement of field-applied nutrient requirement models. Improved data reporting consistency and completeness represent a substantial opportunity to improve nutrition-related mathematical models. A body of nutrition research was reviewed and common phrases used to describe diets, animals, housing and environmental conditions were recorded and equivalent numerical data that could be reported were proposed. With the increasing availability of online supplementary material sections available in journals, a comprehensive checklist of data that should be included in publications was developed. To continue to improve our research effectiveness, studies utilizing multiple research methodologies to address complex systems and measure multiple variables will be necessary. From the current body of animal nutrition literature, a series of opportunities to integrate research focuses (nutrition, reproduction and



genetics) to advance the development of nutrient requirement models were identified. Examples of possibilities to integrate research methodologies include analysis of the energy cost of ionic gradients in cells and protein turnover in tissues. From our survey of experimentation and data reporting in animal nutrition, 4 key opportunities to advance animal nutrition knowledge were identified: 1) coordinated experiments must be designed to employ multiple research methodologies; 2) publication guidelines and restrictions should be updated to allow more complete data sets to be made available; 3) systems-oriented research approaches should be encouraged and supported; and 4) such new data should be more rapidly be integrated into our knowledge bases, research programs and practical applications

**Key Words:** experimental design, systems biology, NANP

**T466 Oxygen uptake by splanchnic tissues of sheep infused with different N compounds into the mesenteric vein.** Simone Stefanello, Gilberto V. Kozloski\*, Mariana P. Mezzomo, Alsiane S. Capelesso, Tiago Orlandi, Fernanda Hentz, and Diego Zeni, *Universidade Federal de Santa Maria, Santa Maria, RS, Brazil.*

Gluconeogenesis and ureagenesis might be integrated and energy expensive processes and, thus, a trial was conducted to measure the impact of the mesenteric infusion of different N compounds on oxygen uptake by splanchnic tissues of wethers. The trial was conducted with 4 multicatheterized wethers as a 4 × 4 Latin square with 210-min daily periods. The blood flow through portal-drained viscera (PDV) and liver was determined by downstream dilution of 15 g/L *p*-aminohippurate (PAH) infused continuously (1.5 mL/min) into the mesenteric vein. In parallel, wethers were continually infused into the mesenteric vein with a saline (0.15 M NaCl) solution during 90 min followed by the infusion, during more 120 min, of either: saline (control), 0.25 M NH<sub>4</sub>HCO<sub>3</sub>, 0.25 M L-alanine or 0.125 M L-arginine, all of them infused at a rate of 1.5 mL/min to provide 375 μmol N/min. Throughout infusion periods simultaneous arterial, portal and hepatic blood samples were taken at 30 min interval and analyzed for PAH and oxygen concentration. The PROC MIXED of SAS was used for variance analysis, which generated a residual error. The oxygen uptake during treatment infusion (i.e., 90 to 210 min) was compared with that of control period (i.e., first 90 min) within each treatment by F test. Oxygen uptake by PDV during treatment infusion was not different from that observed during saline infusion for any treatment, whereas only NH<sub>4</sub>HCO<sub>3</sub> infusion increased oxygen uptake by liver (Table 1). In conclusion, increased ammonia load increased energy expenditure by liver of wethers.

**Table 1 (Abstr. T466).** Oxygen uptake (mL/h) by portal-drained viscera (PDV) and liver of wethers infused with saline (NaCl) or with 375 μmol N/min of different N compounds into the mesenteric vein

Item	Time (min) <sup>1</sup>	Infusion treatments				SEM
		NaCl	Ammonia	Alanine	Arginine	
PDV	0-90	2871	2692	3210	3532	580.2
	90-210	2857	2579	3115	3583	280.5
<i>P</i> *		0.981	0.822	0.880	0.948	
Liver	0-90	1600	1617	2164	3154	316.6
	90-210	1917	2583	2460	2548	246.5
<i>P</i> *		0.453	0.045	0.385	0.157	

<sup>1</sup>Saline solution was used through the first 90 minutes in all treatments.  
\*Probability of the difference between 90-210 vs. 0-90 min means.

**Key Words:** ammonia, amino acids, ureagenesis

**T467 Net flux of metabolites by liver of sheep infused with different N compounds into the mesenteric vein.** Simone Stefanello, Gilberto V. Kozloski\*, Renato N. Libardoni, Gabriela P. Coradini, Sabrina Bäumer, Marta L. R. Leal, and André V. Soares, *Universidade Federal de Santa Maria, Santa Maria, RS, Brazil.*

Ureagenesis and gluconeogenesis might be integrated processes and, thus, a trial with 4 multicatheterized wethers was conducted to measure the impact of the mesenteric infusion of different N compounds on liver net flux of urea and glucose. The trial was conducted as a 4 × 4 Latin square with 210-min daily periods. The blood flow through portal-drained viscera (PDV) and total splanchnic tissues (ST) was determined by downstream dilution of 15 g/L *p*-aminohippurate (PAH) infused continuously (1.5 mL/min) into the mesenteric vein. In parallel, wethers were continually infused into the mesenteric vein with a saline (0.15 M NaCl) solution during 90 min followed by the infusion, during more 120 min, of either: saline (control), 0.25 M NH<sub>4</sub>HCO<sub>3</sub>, 0.25 M L-alanine or 0.125 M L-arginine, all of them infused at a rate of 1.5 mL/min to provide 375 μmol N/min. Throughout infusion periods simultaneous arterial, portal and hepatic blood samples were taken at 30 min interval and analyzed for PAH, urea and glucose. Liver net flux was the difference between TS and PDV values. The PROC MIXED of SAS was used for variance analysis, which generated a residual error. The hepatic net flux obtained during treatment infusion (i.e., 90 to 210 min) was compared with that of control period (i.e., first 90 min) within each treatment by F test. Hepatic net flux of urea was increased only for NH<sub>4</sub>HCO<sub>3</sub> whereas glucose net flux was increased only when alanine was infused into the mesenteric vein (Table 1). In conclusion, no clear relationship between ureagenesis and gluconeogenesis was observed in wethers.

**Table 1 (Abstr. T467).** Hepatic net flux (mg/h) of metabolites by wethers infused with saline (NaCl) or with 375 μmol N/min of different N compounds into the mesenteric vein

Item	Time (min) <sup>1</sup>	Infusion treatments				SEM
		NaCl	Ammonia	Alanine	Arginine	
Urea	0-90	431	437	585	538	68.9
	90-210	450	752	602	519	89.1
<i>P</i> *		0.796	0.044	0.888	0.889	
Glucose	0-90	2817	4220	3042	3799	540.5
	90-210	3825	4648	5502	4306	395.5
<i>P</i> *		0.194	0.757	0.018	0.556	

<sup>1</sup>Saline solution was used through the first 90 minutes in all treatments.  
\*Probability of the difference between 90-210 vs. 0-90 min means.

**Key Words:** amino acids, gluconeogenesis, ureagenesis

**T468 Evaluation of dairy and beef farm greenhouse gas emissions in different areas of Spain.** Ibdidhi Ridha and Sergio Calsamiglia\*, *Animal Nutrition and Welfare Service, Universitat Autònoma de Barcelona, Bellaterra, Spain.*

Ruminants are recognized sources of greenhouse gas (GHG) emissions. The objective of this paper was to estimate the carbon footprint of dairy and beef farms in different production systems in Spain. Component models for predicting all important sources of CH<sub>4</sub>, N<sub>2</sub>O, and CO<sub>2</sub> from primary and secondary sources in dairy and beef production were estimated with the Integrated Farm System Model (IFSM) and reported in CO<sub>2</sub> equivalent (eCO<sub>2</sub>/kg of energy-corrected milk (ECM) or kg body weight (BW)). The IFSM and Cornell Net Carbohydrate and Protein System (CNCPS) were used to evaluate dairy and beef farms in Spain for

GHG emissions and diet evaluation on methane production, respectively. Three dairy farms from each of 3 regions were selected: Mediterranean (Catalonia, Valencia and Murcia), Cantabric area (Galicia, Asturias and Cantabria) and Central zone (Castilla-La Mancha, Castilla-Leon and Madrid). The average carbon footprint (kg eCO<sub>2</sub>/kg of ECM) of all dairy farms was 0.83, with the Mediterranean farms being highest ( $P < 0.01$ ); 0.98) compared with the Central Zone (0.84) and the Cantabric area (0.67). Two extreme farms were selected for further simulations: the first one had the highest carbon footprint and non-enteric methane, while the second had the lowest carbon footprint and the highest enteric methane. These farms were simulated by the IFSM model using different management change scenarios (higher productivity, manure type collection, bedding type, anaerobic digester and storage type of manure) and dietary changes (modification of the ratio forage:concentrate, improved forages quality, inclusion of fat, use of ionophore). Management changes reduced methane emission up to 30% while dietary change reduced it up to 5%. Two beef farms fed 90:10 concentrate:straw and one fed with corn silage were used to simulate GHG emissions using the same models. The carbon footprint (eCO<sub>2</sub>/kg BW) was 6.98 in beef fed corn silage and 6.90 in beef fed without corn silage. Management strategies provided a greater potential to reduce methane emissions compared with dietary scenarios changes.

**Key Words:** carbon footprint, dairy and beef farm, Integrated Farm System Model (IFSM)

**T469 Detect the association of protein structures to protein nutrient utilization and availability of co-products from bio-fuel and bio-brewing processing.** Xuewei Zhang<sup>1</sup>, Limei Chen<sup>2,1</sup>, Yajing Ban<sup>2</sup>, and Peiqiang Yu<sup>\*2,1</sup>, <sup>1</sup>Department of Animal Science, Tianjin Agricultural University, Tianjin, China, <sup>2</sup>Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, SK, Canada.

The objective of this study was to identify the correlation between protein molecular structures and protein nutritional profiles of co-products from cereal grains after bio-fuel and bio-brewing processing in terms of (1) protein chemical profiles; (2) protein subfractions; (3) total digestible protein; (4) protein degradation and estimated intestinal CP digestibility. Five kinds of corn DDGS and 2 kinds of barley DDGS were collected from different manufactures in the north of China from 2012 to 2013. The protein molecular structure spectral feature were analyzed using advanced molecular spectroscopy technique at University of Saskatchewan. The protein subfractions were partitioned using CNCPS system. Total digestible protein, protein rumen degradation and intestinal digestion were determined using dry Holstein dry cows at University of Saskatchewan. Statistical analysis was performed using the PROC MIXED of SAS 9.3. The results showed that the co-products from corn and barley grains differed in both protein nutritional profiles and protein molecular structures in terms of  $\alpha$ -helix,  $\beta$ -sheet spectral intensity and their ratio and amide I, amide II spectral intensity and their ratio. Protein amide II height had a positive correlation with ( $P < 0.05$ ) PB2 fraction with  $R = 0.53$ , but that other protein amide parameters had no correlation with ( $P > 0.05$ ) PA, PB1, PB3 and PC fractions. Protein amide II height had a positive correlation with ( $P < 0.05$ ) TDN with  $R = 0.74$ . Protein amide II height has a negative correlation with ( $P < 0.05$ ) protein degradability (R\_DCP) with  $R = -0.67$ , and a positive correlation with ( $P < 0.05$ ) intestinal protein digestibility (I\_DCP) with  $R = 0.60$  and total-tract available protein (T\_ACP) with  $R = 0.58$ . For protein secondary structure, the  $\alpha$ -helix to  $\beta$ -sheet ratio was negatively correlated with ( $P < 0.05$ ) total protein digestibility (T\_DCP) with  $R = -0.56$  and positively correlated with ( $P < 0.05$ ) total digestible crude

protein (tdCP) with  $R = 0.55$ . In conclusion, protein molecular spectral parameters in the co-products are association with protein nutritional profiles and protein degradation and digestion.

**Key Words:** protein nutritive value, protein molecular structure, molecular spectroscopy

**T470 Effects of essential oils from wormwood hybrids on *in vitro* digestibility, microbial diversity and rumen fermentation of bermudagrass hay and soybean meal.** Seong Shin Lee<sup>\*1</sup>, Hee Yoon<sup>1</sup>, Hyuk Jun Lee<sup>1</sup>, Dong Hyeon Kim<sup>1,3</sup>, Sardar M. Amanullah<sup>1</sup>, Young Ho Joo<sup>1</sup>, Eun Tae Kim<sup>2</sup>, Adegbola T. Adesogan<sup>3</sup>, and Sam Churl Kim<sup>1</sup>, <sup>1</sup>Division of Applied Life Science (BK21Plus, Insti. of Agri. & Life Sci.), Gyeongsang National University, Jinju, Gyeongsangnam-do, South Korea, <sup>2</sup>Dairy Science Division, National Institute of Animal Science, RDA, Cheonan, Chungcheongnam-do, South Korea, <sup>3</sup>Department of Animal Science, Institute of Food and Agricultural Sciences, University of Florida, Gainesville, FL.

Effects of wormwood essential oil (EO) supplementations on *in vitro* digestibility and rumen fermentation characteristics of soybean meal (SBM) or bermudagrass hay (BH) were examined. Rumen fluid was collected from 2 cannulated Holstein cows, filtered through 2 layers of cheese cloth and mixed with Van Soest medium (1:2 ratio). Exactly 0.5 g of SBM or BH was treated with nothing or with 200 mL of wormwood EO from Ganghwa (GA), Injin (IN) or San (SA) wormwood hybrids grown at the Research Unit, Gyeongsang National University, South Korea. Three replicates of each treatment with 3 blanks were incubated at 39°C for 72 h in buffered-rumen fluid (40 mL). *In vitro* DM digestibility (DMD), pH, ammonia-N and volatile fatty acids (VFA) were measured. The population of certain rumen microbes was measured by Real-time PCR. Data were analyzed using the GLM procedure of SAS (SAS Inst., Cary, NC). Total VFA concentration (mM) of SBM was decreased ( $P < 0.05$ ) by GA and SA (157 vs. 143, and 146) but not IN (149). Other fermentation or digestibility measures and the methanogenic archaea population were unaffected by EO. However, the fold change of *Fibrobacter succinogens* was reduced ( $P < 0.05$ ) by GA, IN and SA (18.8 vs. 9.13, 2.36, 2.63), that of *Ruminococcus albus* was increased ( $P < 0.05$ ) by SA (10.8 vs. 24.5), that of *Streptococcus bovis* was increased ( $P < 0.05$ ) by GA and SA though greatest by SA ( $P < 0.05$ ) and that of *R. flavefaciens* was increased ( $P < 0.05$ ) by GA and IN. The DMD of BH was not increased by EO but GA decreased NDF digestibility. Ammonia-N concentration was increased ( $P < 0.05$ ) by SA (17.7 vs. 19.9 mg N/dl). Total VFA concentration was reduced ( $P < 0.05$ ) by GA (136.1 vs. 119.8 mM) but pH and VFA molar proportion did not differ among treatments ( $P > 0.05$ ). Adding the EO to SBM or BH had no beneficial effects on the fermentation or digestibility despite altering the ruminal bacteria population.

**Key Words:** essential oil, fermentation indices, rumen microbe

**T471 Dry matter intake and feeding behavior of cattle fed cottonseed and vitamin E.** Ricardo Galbiatti Sandoval Nogueira<sup>\*</sup>, Flavio Perna Jr, Eduardo Cuellar Orlandi Cassiano, Lizbeth Collazo Paucar, Mariane Cheschin Ernandes, Diana Carolina Zapata Vasquez, Adrielle Matias Ferrinho, Romulo Germano de Resende, Felipe Bispo Mendonça, Renata Gardenalli, Angélica Simone Cravo Pereira, and Paulo Henrique Mazza Rodrigues, University of São Paulo, Pirassununga, São Paulo, Brazil.

The objective of this study was to evaluate the dry matter intake and feeding behavior of cattle fed cottonseed and vitamin E. The experi-

ment was conducted University of São Paulo, Campus Pirassununga. Six cannulated non-pregnant, nonlactating cows were distributed in a replicated  $3 \times 3$  Latin square design. Feed was offered ad libitum twice daily. Feeding behavior was observed during 24 h 5 by 5 min. Treatments were: Control; Cottonseed (CS): 30.47% cottonseed included; vitamin E (VitE): 30.47% cottonseed plus 500 IU VitE included. Diets were isoenergetic and isonitrogenous. The ether extract content was 3.74, 8.32 and 8.32% for the Control, CS and VitE diets. Data were analyzed by SAS (v9.3) and significance declared at  $P < 0.05$ . Dry matter intake, dry matter intake in relation to body weight and per unit of metabolic size, total time and average time per event eating, ruminating, free and chewing (eating plus ruminating) were compared through orthogonal contrasts, where contrast 1 = CS and VitE vs. control, and contrast 2 = CS vs. VitE. Both diets provided ( $P > 0.05$ ) similar amounts of dry matter intake (15.44 and 15.40 vs. 14.64 kg animal day<sup>-1</sup>), dry matter intake in relation to body weight (1.74 and 1.74 vs. 1.66) and per unit of metabolic size (95.10 and 95.25 vs. 90.69). Treatments CS and VitE had greater time eating (219.1 and 215.0 vs. 190.8 min), ruminating (437.5 and 430.0 vs. 291.6 min), chewing (656.6 and 645.0 vs. 482.5 min) but less time free (775.0 and 786.9 vs. 947.5) compared with control. The average eating time per event was not different among treatments (33.1 and 36.9 vs. 32.4 min), but the average ruminating time per event (29.12 and 24.58 vs. 20.62 min) and chewing (30.0 and 27.3 vs. 24.16 min) was greater for CS and VitE compared with control. The average free time per event was less for CS and VitE than control (40.5 and 38.4 vs. 49.5 min). Including cottonseed in a diet at 30% did not decrease dry matter intake and the animals spent more time eating, ruminating, and chewing with less free time. Vitamin E did not affect dry matter intake parameters and feeding behavior.

**Key Words:** dry matter intake, feeding behavior, lipid

**T472 In situ degradability, rumen bacteria population, and in vitro gas production in cannulated steers fed diets with and without HMTBa.** Y. Liang, S. E. Bettis, M. Wehmeyer, G. I. Zanton, H. A. Tucker\*, and M. Vazquez-Anon, *Novus International Inc., St. Charles, MO.*

The objective of this study was to investigate changes in rumen environment when animals are fed a diet containing 2-hydroxy-4-(methylthio) butanoate (HMTBa). Six cannulated Holstein steers were utilized in a crossover design trial with two 42-d periods (28 d adaptation, 14 d sampling) and were fed a ration with hay, corn, and soyhulls with HMTBa (0.1%, DM basis) or without (CON). Whole rumen contents (WRC), rumen solids (RS) and rumen liquid (RL) samples were taken 3 h pre- and post-feeding on d 29 of each period. Samples were analyzed for bacteria sequencing. Additional rumen fluid, collected 3 h post-feeding was used to measure gas production using the ANKOM gas production system and alfalfa hay as substrate. On d 36 of each period, the effect of HMTBa supplementation on dry matter and NDF digestibility of alfalfa and grass hays were evaluated using in situ methods. In vitro gas production at 24 h was greater ( $P = 0.002$ ) in rumen fluid from steers fed HMTBa than CON. Supplementing HMTBa increased DM digestibility of alfalfa hay at 4 ( $P = 0.001$ ) and 12 h ( $P = 0.025$ ) of incubation and increased ( $P = 0.013$ ) that of grass hay at 24 h of incubation. In situ NDF digestibility was greater at 12 h of incubation for alfalfa hay ( $P = 0.005$ ) and 24 h of incubation for grass hay ( $P = 0.006$ ) with HMTBa supplementation. Feeding steers HMTBa tended ( $P = 0.074$ ) to increase Proteobacteria in RS and ( $P = 0.039$ ) Actinomycetales in RL. Supplementing HMTBa resulted in *Ruminococcaceae* accounting for a greater proportion of bacteria at the family level in RL ( $P = 0.048$ ), RS ( $P = 0.054$ ) and WRC ( $P = 0.037$ ). Furthermore,

feeding steers HMTBa increased ( $P = 0.028$ ) *Ruminococcus* in WRC, and tended to increase in RL ( $P = 0.054$ ) and RS ( $P = 0.052$ ). Greater population of *Ruminococcus* sp. was detected in WRC ( $P = 0.022$ ), RL ( $P = 0.031$ ), and RS ( $P = 0.025$ ) in HMTBa fed steers. In conclusion, supplementing HMTBa increases fibrolytic bacterial species, resulting in a greater gas production in vitro, and greater in situ DM and NDF digestibility of selected hays.

**Key Words:** 2-hydroxy-4-(methylthio) butanoate (HMTBa), rumen bacteria, NDF digestibility

**T473 Effect of sustained-release mineral dietetic feed bolus on plasma trace minerals status in grazing heifers.** J. M. Beguin<sup>\*1</sup>, R. P. Dagorne<sup>1</sup>, and R. Lecrubier<sup>2</sup>, <sup>1</sup>NEOLAIT, Yffiniac, France, <sup>2</sup>ESA, Angers, France.

The objective of this study was to evaluate, in grazing dairy heifers, the effect of an oral bolus on plasma trace mineral status. Heifers (n = 74, initial BW = 427 kg, age = 445 d) from 4 dairy farms in Western France, were randomly assigned to a control group (CONTROL) and a trial group (TEST). Heifers in TEST received 2 boluses (Dietevit Excell, Néolait, Yffiniac, France), at the start of grazing season. Boluses supplies zinc, manganese, copper, iodine, cobalt, selenium, vitamin A, vitamin D3 and vitamin E. Blood samples were taken before turn out in April (d 0), in July (d 106), and at the end of the grazing season in November (d 245). Plasma trace minerals analysis was carried out by ICP-MS (LEA Vendée, France). Glutathione peroxidase was also measured (LDH, Oniris, France). Metabolic profiles were compared using Repeated Measures ANOVA (SPSS v18). Samples of grass were taken from the grazing plots and were analyzed by ICP-MS (Sciencetec Analytical Services Ltd., United Kingdom). Grass trace mineral content per kg DM was zinc (25.4 mg), manganese (84 mg), iron (72 mg), copper (5.3 mg), cobalt (0.10 mg) and selenium (0.03 mg). Compared with CONTROL, heifers in TEST had the same trace minerals levels in d 0, higher selenium levels (42.6 vs. 25.7 µg/L in d 106 and 36.4 vs. 26.6 µg/L in d 245,  $P = 0.01$  and  $P = 0.03$ ), higher glutathione peroxidase levels (205 vs. 114 µg/L in d 106 and 205 vs. 129 µg/L in d 245,  $P = 0.01$  and  $P = 0.02$ ) and higher iodine levels (79 vs. 48 µg/L in d 106 and 62 vs. 56 µg/L in d 245,  $P = 0.01$  and  $P = 0.07$ ). There was no difference in plasma level for zinc (1064 vs. 999 µg/L in d 106 and 954 vs. 922 µg/L in d 245), manganese (2.8 vs. 2.8 µg/L in d 106 and 3.3 vs. 3.4 µg/L in d 245), copper (892 vs. 916 µg/L in d 106 and 995 vs. 1017 µg/L in d 245) and cobalt (1.4 vs. 1.2 µg/L in d 106 and 1.1 vs. 0.9 µg/L in d 245). Administration of 2 Dietevit Excell boluses to dairy heifers before turn out maintains healthy selenium, glutathione peroxidase and iodine plasma levels over a period of at least 245 d. Results show that Dietevit Excell gradually dissolves providing a sustained release of minerals over the grazing season.

**Key Words:** heifer, trace mineral, bolus

**T474 Crambe meal (*Crambe abyssinica*) inclusion in feed of Santa Inês crossbred lambs on blood serum urea.** Kariny Ferreira Moreira, Darcilene Maria Figueiredo\*, Adriano Oliveira Cruz, Ronald Matos dos Santos, Juscilene Aparecida Silva Pacheco, Cassiane Gomes dos Santos, Daniela Cordeiro Rocha, Marianne Schorer, and Aldrin Vieira Pires, *Universidade Federal dos Vales do Jequitinhonha e Mucuri, Diamantina, MG, Brazil.*

The objective of this study was to evaluate the effect of increasing levels of crude protein of crambe meal (*Crambe abyssinica*) (0, 25, 50, and 75%) on blood serum urea levels of lambs. Twenty-four Santa



Inês crossbred male lambs, with an average weight and age of 18 kg of body weight, and 4 mo, respectively. The experiment was conducted in a completely randomized design with 4 treatment and 6 replicates each. Animals remained 7 d for adaptation, and 3 periods of 28 d for data collection. Lambs were kept in individual pens (1.5 m x 1.0 m), equipped with trough and drinker. Animals received diets with 19% crude protein (% DM), and 65.4% total digestible nutrients (% DM), based on average daily gain of 200 g head<sup>-1</sup> with forage: concentrate ratio of 50:50. Lambs were fed ad libitum, twice a day, always at 0700 and 1500 h, allowing leftovers of approximately 20%. Sampling of urea serum in the lambs were through blood samples taken by puncture of the jugular vein at 13th day of the second experimental period immediately at 0, 3, 6 and 9 h after the morning feeding. Data were subjected to ANOVA and regression, at 5% probability. The blood serum urea level did not differ ( $P > 0.05$ ) among treatments and 25.96, 25.65, 24.23, and 21.70 g day<sup>-1</sup>, respectively, for diets with 0, 25, 50, and 75% crude protein inclusion of crambe meal. This result demonstrates the appropriateness of the use of nitrogen compounds in the rumen due to the availability of degradable organic matter. Therefore the use of crambe meal on diet has satisfactory effect with regard in lamb blood serum urea.

**Key Words:** alternative feed, protein feed, sheep breeding

**T475 Crambe meal (*Crambe abyssinica*) inclusion in food of Santa Inês crossbred lambs on urea nitrogen.** Kariny Ferreira Moreira, Darcilene Maria Figueiredo\*, Adriano Oliveira Cruz, Ronald Matos dos Santos, Juscelene Aparecida Silva Pacheco, Cassiane Gomes dos Santos, Daniela Cordeiro Barbosa, Marianne Schorer, and Aldrin Vieira Pires, *Universidade Federal dos Vales do Jequitinhonha e Mucuri, Diamantina, MG, Brazil.*

The objective of this study was to evaluate the increasing levels of crude protein of crambe meal (*Crambe abyssinica*) (0, 25, 50, and 75% DM) in lambs diets on the urinary excretion of urea nitrogen (UEUN). Twenty-four Santa Inês crossbred male sheep, with an average weight and age of 18 kg of body weight, and 4 mo, respectively. The experiment was conducted in a completely randomized design with 4 treatment and 6 replicates each. Animals remained 7 d for adaptation, and 3 periods of 28 d for data collection. Lambs were kept in individual pens (1.5 m x 1.0 m), equipped with trough and drinker. Animals received diets with 19% crude protein (% DM), and 65.4% total digestible nutrients (% DM), based on average daily gain of 200 g head<sup>-1</sup> with forage: concentrate ratio of 50:50. Lambs were fed ad libitum, twice a day, always at 0700 to 1500 h, allowing leftovers of approximately 20%. For determination of UEUN spot urine samples were collected by spontaneous urination on the 13th day of the second experimental period immediately at 0h, 3h, 6h and 9h after morning feeding. Data were subjected to ANOVA and regression, at 5% probability. There was no effect ( $P > 0.05$ ) between diets on the UEUN, and 2.49, 2.41, 2.47 and 2.66 g day<sup>-1</sup>, respectively for diets containing 0, 25, 50 and 75% of crude protein inclusion of crambe meal indicating low amount of nitrogen without being fully utilized by the animal. Therefore the inclusion of crude protein from the crambe meal for lambs feeding had a beneficial effect in relation to UEUN.

**Key Words:** alternative food, protein food, sheep breeding

**T476 Induction of subacute ruminal acidosis affects gene expression in rumen epithelial tissue.** J. C. McCann\*, S. Alqarni, S. Luan, F. C. Cardoso, and J. J. Loor, *University of Illinois at Urbana-Champaign, Urbana, IL.*

Subacute ruminal acidosis (SARA) negatively affects the dairy industry by decreasing dry matter intake, milk production, profitability, and increasing culling rate and death loss. These consequences are related to a loss of barrier function in the rumen epithelial tissue associated with a reduction in ruminal pH. Six lactating Holstein cows were used in a replicated 2 × 2 Latin square design to determine the effects of SARA induction on the rumen epithelial tissue. Experimental periods were 10 d with d 1 to 3 for ad libitum intake of control diet, followed by 50% feed restriction on d 4, and ad libitum access on d 5 of the control diet (control) or control diet + 4.6 kg of a 50:50 wheat/barley pellet (challenge). Rumen papillae biopsies were collected on d 1 and 6 of each period and stored at -80 C. Quantitative RT-PCR was used to determine expression genes related to barrier function with all reactions run in triplicate. Three reference genes (*CMTM6*, *ERC1*, and *MRPL39*) were selected due to stable expression across animals and times. Data were analyzed using the MIXED procedure of SAS with day, treatment, period, and square as fixed effects and cow as a random effect. Of the evaluated barrier function genes, the greatest relative abundance was observed for *CLDN1* followed by *CLDN4*. Expression of *CLDN1*, *CLDN4*, and *JAM2* was upregulated on d 6 ( $P < 0.1$ ). Greater expression of *TJPI/ZO-1* was observed for the control treatment ( $P = 0.086$ ). A treatment × day effect was observed for *OCLN* ( $P = 0.07$ ) as expression was upregulated on d 6 for the challenge treatment but downregulated for the control. No treatment effect ( $P > 0.1$ ) was observed for *CLDN1*, *CLDN4*, *JAM2*, *CAR*, *TLR2*, *TLR4*, and *IGFBP3*. Toll-like receptors recognize bacterial components and are capable of upregulating barrier function of epithelial tissue. Both *TLR2* and *TLR4* were downregulated on d 6 ( $P = 0.05$  and  $P = 0.1$ , respectively). Collectively, results suggest feed restriction and subsequent refeeding caused a greater effect on expression of barrier function genes than the additional starch in the challenge treatment. However the results may still be indicative of the rumen epithelium tissue response to SARA.

**Key Words:** acidosis, rumen, epithelium

**T477 Effects of monensin and essential oils from some Nigerian spices on methane production and ruminal fermentation in vitro.** Musibau A. Bamikole<sup>1,2</sup>, Ibukun M. Ogunade<sup>\*1</sup>, Felipe Amaro<sup>1</sup>, Yun Jiang<sup>1</sup>, Thiago F. Bernardes<sup>1</sup>, Darren D. Henry<sup>3</sup>, F. O. Ugiagbe<sup>2</sup>, U. J. Ikhatua<sup>2</sup>, Nicolas DiLorenzo<sup>3</sup>, and Adegbola T. Adesogan<sup>1</sup>, <sup>1</sup>University of Florida, Gainesville, FL, <sup>2</sup>University of Benin, Benin city, Nigeria, <sup>3</sup>North Florida Research and Education Center, University of Florida, Marianna, FL.

Plant extracts may be potential replacements for antibiotic feed additives used in livestock production and they are considered safer. Effects of essential oils (EO) extracted from rosemary (*Rosmarinus officinalis*; REO) leaves and clove (*Syzygium aromaticum*; COE) flower on in vitro rumen fermentation parameters including methane production were examined. A corn silage-based TMR (0.5 g; CP 16.6%; NDF 35.9%) was treated with CEO or REO at rates of 0 (Control), 10 (Low), 20 (Med) and 30 (High) µL/50 mL of rumen fluid-buffer inoculum (ratio 1:2) and with monensin (0.6 mg/50 mL). Each suspension was incubated in a 120-mL gas-tight culture bottle in triplicate at 39°C for 24 h in each of 2 runs. Fermentation parameters, gas and methane production, in vitro DM digestibility (DMD), and fermentation efficiency (FE; DMD g kg<sup>-1</sup>/gas volume) were measured. Data for each EO were separately analyzed with the Glimmix procedure of SAS. The DMD was reduced ( $P < 0.05$ ) by monensin, Med REO and High CEO (526, 531 and 513 vs. 555 g/kg). Gas volume (mL/g DM) was increased ( $P < 0.05$ ) by Low REO or CEO (84.5 vs. 92.4 and 96.2) and decreased ( $P < 0.05$ ) by their High rates or monensin (76.7, 63.6 and 66.8), respectively. The

FE was increased ( $P < 0.05$ ) by High REO or CEO or monensin (7.27, 8.69 and 7.92 vs. 6.58) and reduced ( $P < 0.05$ ) by Low REO (5.85), respectively. Methane production (mg/g DM digested) was reduced ( $P < 0.05$ ) by High REO and CEO and monensin (8.65, 8.02, and 7.10 vs. 11.44). The pH was increased ( $P < 0.05$ ) by monensin (5.75 vs. 5.66) but not by EO. Ammonia-N and VFA concentrations were unaffected by treatment except that monensin reduced ( $P < 0.05$ ) acetate concentration and increased valerate concentration. Also, monensin and Med and high CEO increased ( $P < 0.05$ ) butyrate concentration. High doses of essential oils from clove and rosemary decreased methane production and increased fermentation efficiency in a manner comparable to monensin

**Key Words:** essential oil, spice, in vitro fermentation

**T478 Effect of heating method on alteration of protein molecular structure in flaxseed: Relationship with changes in protein subfraction profile and digestion in dairy cows.** Nazir A. Khan<sup>1</sup>, Helen Booker<sup>2</sup>, Yajing Ban<sup>1</sup>, and Peiqiang Yu\*<sup>1</sup>, <sup>1</sup>Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, SK, Canada, <sup>2</sup>Department of Plant Sciences, University of Saskatchewan, Department of Plant Sciences, University of Saskatchewan, Saskatoon, SK, Canada.

This study evaluated the effect of heating methods on alteration of protein structure in flaxseed (*Linum usitatissimum* L.) in relation to changes in protein subfraction profile and digestion in dairy cows. Seeds from 2 flaxseed varieties, sampled from 2 replicate plots at 2 locations, were evaluated. The plots ( $n = 4$ ) were used as replicates. The seeds were either maintained in their raw state or heated in an air-draft oven (dry heating) or autoclave (moist heating) for 60 min at 120°C or by microwave irradiation (MIR) for 5 min. Compared with raw seeds, moist heating decreased ( $P < 0.05$ ) soluble protein (SP) content ( $56.5 \pm 5.55$  to  $25.9 \pm 6.16\%$  crude protein) and increased ( $P < 0.05$ ) rumen undegraded protein (RUP) content ( $36.0 \pm 5.19$  to  $46.9 \pm 2.72\%$  CP) and intestinal digestibility of RUP ( $61.0 \pm 2.28$  to  $63.8 \pm 2.67\%$  RUP). Dry heating did not alter ( $P > 0.05$ ) the protein subfraction profile and rumen degradation kinetics, whereas MIR increased ( $P < 0.05$ ) the RUP content from  $36.0 \pm 5.19$  to  $40.4 \pm 4.67\%$  CP. The MIR and dry heating did not alter ( $P > 0.05$ ) the amide I to amide II ratio, but moist heating decreased ( $P < 0.05$ ) both the amide I to amide II ratio and  $\alpha$ -helix-to- $\beta$ -sheet ratio. Regression equations based on protein molecular spectral intensities provided high prediction power for estimation of heat-induced changes in SP ( $R^2 = 0.62$ ), RUP ( $R^2 = 0.71$ ), and intestinal digestibility of RUP ( $R^2 = 0.72$ ). Overall, heat-induced changes in protein nutritive value and digestion were strongly associated with heat-induced alteration in protein molecular structures.

**Key Words:** heat processing method, protein molecular structure, protein subfraction

**T479 Investigation of protein digestion kinetics in vitro using <sup>15</sup>N-labeled timothy and red clover.** Merko Vaga\*, Kerstin Huss-Danell, Mårten Hetta, and Pekka Huhtanen, Dept. of Agricultural Research for Northern Sweden, Swedish University of Agricultural Sciences, Umeå, Sweden.

An in vitro method based on <sup>15</sup>N-labeled forage N was developed to study ruminal protein degradation of timothy and red clover. Timothy and red clover were grown on the same field with 2 replicate plots, 2 m<sup>2</sup> each. One replicate plot was fertilized with <sup>15</sup>N-enriched NH<sub>4</sub>NO<sub>3</sub> while the other received non-enriched fertilizer. Labeled timothy and red clover had average enrichment of 2.37 and 1.23 atom % excess

<sup>15</sup>N, respectively. Forages from the first-cut were preserved either as hay (TH, RCH), untreated (T, RC) or formic acid-treated silage (TF, RCF). Rumen fluid was collected from 2 Swedish red cows fed on grass silage:concentrate diet (60:40 DM basis). Samples of 1 g were incubated in 60 mL of buffered rumen fluid at 39°C for 48 h. For non-labeled forages NH<sub>3</sub>-N in the inoculum was labeled with <sup>15</sup>N enriched (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>. Vessels containing labeled forages received the same amount of non-enriched (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>. Liquid samples were collected every hour at 0 to 10 h, after 12, 16, 24, 30 and 48 h, and later analyzed for NH<sub>3</sub>-N and <sup>15</sup>N. Degraded CP was calculated as a sum of appearance of <sup>15</sup>N from labeled forages and disappearance of <sup>15</sup>N from labeled NH<sub>3</sub>-N. Degradation parameters were estimated by the 2 pool exponential model using the SAS NLIN procedure. Timothy forages had higher rapidly degradable fraction (b1). Slowly degradable fraction (b2) was smaller in formic acid treated silages and in red clover silages. Hay had slower degradation rates (kd) than silages. Total degraded CP (a+b) was higher for timothy and for hay. It is concluded that incubating <sup>15</sup>N labeled feeds or feed fractions simultaneously with corresponding unlabeled feeds by labeling ammonia pool with <sup>15</sup>N can be a useful tool in investigating kinetics of ruminal protein degradation.

**Table 1 (Asbtr. T479).** CP concentration and degradation parameters of timothy (T) and red clover (RC)

	TF	RCF	T	RC	TH	RCH
CP, g/kg DM	155	147	166	133	144	137
CP degradation <sup>1</sup>						
b1, g/kg	495	418	335	334	473	391
b2, g/kg	380	362	594	507	490	452
kd1, 1/h	0.696	1.250	1.263	1.200	0.815	0.818
kd2, 1/h	0.189	0.132	0.139	0.161	0.055	0.091
a+b, g/kg	875	780	929	841	963	843

<sup>1</sup>b1 = rapidly degradable, b2 = slowly degradable, kd1 = degradation rate of b1, kd2 = degradation rate of b2.

**Key Words:** in vitro, <sup>15</sup>N, protein

**T480 Effects of yam (*Dioscorea opposita*) supplementation on in vitro digestibility and rumen fermentation characteristics of ground corn and perennial ryegrass.** Jin Yeon Park\*<sup>1</sup>, Tea Hyeon Kim<sup>1</sup>, Hyuk Jun Lee<sup>2</sup>, Young Ho Joo<sup>2</sup>, Sardar M. Amanullah<sup>2</sup>, Dong Hyeon Kim<sup>2</sup>, In Hak Choi<sup>3</sup>, and Sam Churl Kim<sup>1,2</sup>, <sup>1</sup>Department of Animal Science, Gyeongsang National University, Jinju, Gyeongsangnam-do, South Korea, <sup>2</sup>Division of Applied Life Science (BK21Plus, Insti. of Agric. & Life Sci.), Gyeongsang National University, Jinju, Gyeongsangnam-do, South Korea, <sup>3</sup>Department of Companion Animal & Animal Resources Sciences, Joongbu University, Geumsan, Chungcheongnam-do, South Korea.

This study was carried out to investigate the effect of yam (*Dioscorea opposita*) supplement levels on in vitro digestibility, gas production and rumen fermentation characteristics of ground corn and perennial ryegrass using a novel wireless automated gas production system. Rumen fluid was collected from 2 cannulated Hanwoo heifers, filtered by 2 layers of cheese cloth and mixed with Van Soest medium at 1:2 ratio. The rumen fluid mixture (40 mL) added to the incubation bottles containing ground corn or perennial ryegrass (0.5 g) with 4 levels of Yam supplement (0, 5, 25 and 50 mg). Four replicates in each treatment with 2 blanks were incubated at 39°C for 48 h and 72 h for ground corn and perennial ryegrass, respectively. After incubation, the bottle content was used for the analyses of in vitro digestibilities of dry matter (IVDMD) and neutral detergent fiber (IVNDFD), pH, ammonia-N and

volatile fatty acid (VFA). Data were analyzed using the GLM procedure of SAS (SAS Inst., Cary, NC). The pH (Linear,  $P < 0.001$ ) and the concentrations of iso-butyrate (Quadratic,  $P = 0.025$ ) and iso-valerate (Quadratic,  $P < 0.001$ ) of ground corn decreased with increasing yam supplementation level, while the concentrations of total VFA (Linear,  $P = 0.01$ ) and butyrate (Quadratic,  $P = 0.001$ ) increased. However, the fermentation kinetic of ground corn was not affected by Yam supplementation level. With increasing yam supplementation level, IVDMD (Linear,  $P < 0.001$ ), IVNDFD (Linear,  $P = 0.002$ ), the potentially fermentable fraction (Linear,  $P = 0.057$ ) and the total fermentable fraction (Linear,  $P = 0.018$ ) of perennial ryegrass increased. Results indicated that yam (*Dioscorea opposita*) supplementation can increase not only total VFA concentration of ground corn, but also in vitro digestibilities of perennial ryegrass.

**Key Words:** digestibility, rumen fermentation, yam (*Dioscorea opposita*)

**T481 In vitro study of yeast cell-wall  $\beta$ -glucans behavior in ruminal fluid.** Nadia Yacoubi<sup>1</sup>, Jean Philippe Marden<sup>3</sup>, and Corine Bayourthe<sup>\*2</sup>, <sup>1</sup>INRA UR1268 Biopolymers Interactions Assemblies, Nantes, France, <sup>2</sup>Université de Toulouse, INRA, UMR 1388 INRA-INPT GenPhySE, Castanet-Tolosan, France, <sup>3</sup>Phileo Lesaffre Animal Care, Marcq en Baroeul, France.

Behavior of  $\beta$ -(1,3/1,6)-glucans (BG) of yeast cell-wall (*Saccharomyces cerevisiae*) in ruminal milieu was evaluated in vitro. The solubility of BG, pH, and production of VFA were measured after 4, 8, and 16 h of incubation in ruminal fluid. Two yeast cell-wall products were used: brewer's yeast (BrYBG) and baker's yeast (BaYBG), containing 15.1 and 26.3% DM of BG, respectively. Ruminal fluid was collected 3h post-feeding from a ruminally fistulated Holstein dairy cow fed a hay-based diet and strained through a metal sieve (2 mm). Within 45 min of sampling, 60 mL of ruminal fluid and 60 mL of degassed buffer solution (pH 7, 39°C) were added to 250-mL flasks, containing 5 g of yeast cell-wall product, flushed with O<sub>2</sub>-free CO<sub>2</sub>, and capped. For each incubation time, 5 flasks were prepared: 2 replicates each for BrYBG, and BaYBG, and one without product, used as control. Flasks were kept from light and air at 39°C in a waterbath rotary shaker. The pH was recorded at the start of incubation (0 h), 4 h, 8 h, and 16 h. Once the pH was measured, batch culture was centrifuged (4,500 × g for 10 min). Supernatants were taken to determine VFA contents. Total VFA produced was calculated by subtracting the initial total VFA concentration from the final concentration. The sediment BG content was determined (enzymatic kit, Megazyme). BG solubility was calculated by subtracting residual BG in sediment at the end of each incubation time from the starting concentration at 0 h. This batch culture experiment was repeated on 3 different days. At 4 h, the percentage of soluble BG was 3 times higher ( $P < 0.0001$ ) for BaYBG than for BrYBG. At 16 h, it reached 96.4% and 92.5 for BaYBG and BrYBG, respectively. Amount of soluble BG increased ( $P < 0.0001$ ) linearly with time ( $r^2 = 0.916$  for BrYBG and 0.993 for BaYBG). Total VFA contents at 16 h differ ( $P = 0.0007$ ): 95.2 vs. 110.7 mM, for BaYBG and BrYBG, respectively. The pH fall was less important ( $P < 0.0001$ ) for BaYBG than for BrYBG. The decrease in pH and increase in total VFA production suggested that ruminal microflora would be potentially capable of degrading some soluble components in the yeast cell-wall products. But at 16h, 70% of DM BaYBG and 56% of DM BrYBG escape the ruminal degradation suggesting bioavailability in post-ruminal digestive tract.

**Key Words:** beta-glucan, yeast, in vitro

**T482 Effect of different sources of glycerol on in vitro fermentation parameters of corn silage.** E. H. C. B. van Cleef<sup>\*1,2</sup>, E. S. Castro Filho<sup>1</sup>, M. T. C. Almeida<sup>1</sup>, J. R. Paschoaloto<sup>1</sup>, I. Monsignati<sup>1</sup>, S. F. B. Buzinaro<sup>3</sup>, and J. M. B. Ezequiel<sup>1</sup>, <sup>1</sup>São Paulo State University, Jaboticabal, São Paulo, Brazil, <sup>2</sup>FAPESP, São Paulo, Brazil, <sup>3</sup>University of São Paulo, Pirassununga, São Paulo, Brazil.

Two studies were conducted to evaluate the effect of sources of glycerol on in vitro fermentation, and dry matter and neutral detergent fiber digestibilities of corn silage. In the first trial, rumen content was collected from 2 ruminally cannulated male sheep (68.5 kg BW) fed diet composed of 40% corn silage and 60% concentrate (corn, soybean hulls, soybean meal, urea, and minerals). Substrates tested were (1) corn silage, (2) corn silage + 20% crude glycerin (86% glycerol), (3) corn silage + dry glycerin (68% glycerol), and (4) corn silage + pure glycerin (99% glycerol). Substrates (200 mg) and buffered rumen fluid (20 mL McDougall's buffer and 10 mL rumen fluid) were placed into 60-mL bottles (n = 20), purged with helium gas and sealed. Gas production, pH, ammonia nitrogen (NH<sub>3</sub>-N) and DM disappearance were evaluated after 24 h of incubation at 39°C. In the second trial, Ankom Daisy<sup>II</sup> fermenter was used to evaluate in vitro digestibility of DM and NDF. Filter bags (n = 25) were filled with substrates, and incubated for 48 h (39°C) into vessels containing a solution composed of 400 mL rumen fluid, 1330 mL buffer A and 266 mL buffer B. After 48-h incubation, 40 mL of 6 N HCl and 8 g of pepsin was added to each digestion vessel, and incubated for another 24 h. Substrates and residues were evaluated for DM and NDF content. All the procedures were replicated for both trials. There was no effect of treatments on 24-h pH (6.1) and NH<sub>3</sub>-N (38.4 mg/dL), nor for production of total gas (51.2 mL), CH<sub>4</sub> (8.9 mL), and CO<sub>2</sub> (39.7 mL). When DM disappearance. When the dry matter disappearance was taken into account, corn silage produced more gas (total gas) and more CO<sub>2</sub> than glycerol treatments ( $P = 0.01$  and  $P = 0.003$ , respectively), and no difference was observed among treatments for CH<sub>4</sub>, and among glycerol treatments for CO<sub>2</sub>. Glycerol, regardless the source, increased IVDMD of corn silage ( $P < 0.0001$ ), and no changes were observed among glycerol treatments (71.7%). IVDMD was unaffected by treatments (41.7%). All sources of glycerol (crude, dry or pure) do not alter neither rumen fermentation nor in vitro digestibility, when inoculated at 20% with corn silage, thus any of these sources are suitable to improve IVDMD of corn silage.

**Key Words:** glycerol, in vitro dry matter digestibility, methane

**T483 Quality evaluation of corn silage prepared with microbial inoculants.** Luiz Keller<sup>\*1,4</sup>, Marcos Aronovich<sup>3</sup>, Christiane Perali<sup>2,4</sup>, Eliane Rodrigues<sup>3</sup>, Airton Castagna<sup>3</sup>, and Carlos Rosa<sup>2,4</sup>, <sup>1</sup>Universidade Federal Fluminense (UFF), Rio de Janeiro, RJ, Brazil, <sup>2</sup>Universidade Federal Rural do Rio de Janeiro (UFRRJ), Rio de Janeiro, RJ, Brazil, <sup>3</sup>Empresa de Desenvolvimento Agropecuário do Estado do Rio de Janeiro (PESAGRO-RJ), Rio de Janeiro, RJ, Brazil, <sup>4</sup>Conselho Nacional de Pesquisas Científicas (CNPq), Belo Horizonte, MG, Brazil.

Silage is a common widespread practice to preserve forages in Brazil. One of the main problems encountered is the high value of rations and raw materials, so use mainly corn, sorghum and agro-industrial sub products. Being reviewed mycobiota present in the ensiled material and mycotoxins levels are an indicator of quality, because the poor storage conditions can lead contamination and toxins production. The research evaluated the quality of silages preserved with biological inoculants on mycobiota, mycotoxins levels and nutritional assessment parameters. Two dosages of commercial inoculant product, with 10 replications was used on corn silage and evaluated at 0, 30, 60 and 90 d. Fungal



counts were done by surface-spread method and toxigenic ability of isolates strains was evaluated with in vitro conditions. Aflatoxins (AFs), ochratoxin A (OTA), fumonisin B1 (FB1) and deoxivalenol (DON) natural contamination was determined with immunoaffinity columns extracts in HPLC. Total fungal counts were generally high (range  $1.0 \times 10^6$  to  $1.0 \times 10^2$  cfu g<sup>-1</sup>). *Aspergillus flavus*, *Penicillium citrinum*, and *Fusarium verticillioides* were the prevalent toxigenic strains isolated. Mycotoxins levels differed ( $P < 0.001$ ) from pre and post-fermentation samples, probably due to mold growth. Dry matter, carbohydrates, lipids, protein, volatile fatty acids, and fiber content not differed ( $P < 0.001$ ) from pre and post-fermentation samples and were not different from literature. The inoculants does not helped in reducing the count of fungal propagules but decreased the biodiversity of the toxigenic strains presents in the treated silos. So, the use of microbial inoculant on silage production should be recommended to reduce some toxigenic strains contamination. However, each product must be evaluated and the applying process must be carefully conducted. The mycotoxin binding and nutritional quality increase related on literature was not observed on the present study.

**Key Words:** aflatoxin, corn silage, feedstuffs

**T484 Utilization of equations to predict carbohydrate fractions in some tropical grasses.** Romualdo S. Fukushima\*, Carolina B. Bacha, Adriana P. Fuzeto, Ana C. R. Port, Valdo R. Herling, and Alejandro V. Velasquez, *Sao Paulo University, Pirassununga, SP, Brazil.*

The chemical composition of 5 tropical grasses, divided into stalk and leaf, at 3 maturity stages, was used to predict carbohydrate fractions by equations of the Cornell Net Carbohydrate and Protein System (CNCPS) or equations from our research group. Carbohydrate fraction A is a rapidly fermented pool that is primarily composed of sugars, some organic acids and short oligosaccharides. Fraction B1 is constituted mainly of starch and pectin. Fraction B2 has a slower rate of degradation and is available cell wall carbohydrates. The C pool is unavailable cell wall, which includes lignin. These carbohydrates fractions are estimated based on NDF analysis. However, NDF does not recover pectic substances and other ND soluble oligosaccharides such as  $\beta$ -glucans, fructans, galactans, etc. that are part of the cell wall matrix. Structurally, NDF is not complete plant cell wall. Then, a crude cell wall (CW) preparation, which represents plant cell wall matrix more completely because it preserves those carbohydrates that otherwise would be solubilized by the ND solution, was used in equations to predict carbohydrate fractions. By substituting NDF for CW, it was found that pectin and other ND soluble oligosaccharides (soluble fiber - SF) actually appeared in the fraction A of CNCPS, the rapidly degradable carbohydrate pool, instead of fraction B1, as proposed in the original CNCPS model. However, location of SF in the fraction A seems inadequate because degradation rate of SF is lower than fraction A components; thus, an alternative could be to place SF in a specific carbohydrate fraction (B2). This B2 fraction, soluble fiber, can be estimated by subtracting NDF from CW preparation. Because in the original CNCPS model the slowly degradable cell wall carbohydrates were assigned as fraction B2, we suggest naming this carbohydrate pool a new fraction, B3. With this arrangement, the fraction B1 would be constituted only by starch. These fractions are expressed on total carbohydrate basis, here suggested as: CHO = 100 - (CP + EE + MM + Lignin). This equation excludes lignin from the CHO compartment.

**Key Words:** cell wall, Cornell, soluble fiber

**T485 Analysis of microbial populations in Rusitec fermenters fed diets of variable composition.** Ivan Mateos<sup>2</sup>, Maria Jose Ranilla<sup>\*2,3</sup>, Cristina Saro<sup>2</sup>, Alexey Díaz<sup>2</sup>, Maria Gracia De Garnica<sup>2</sup>, Jairo Garcia<sup>2</sup>, and Maria Dolores Carro<sup>1</sup>, <sup>1</sup>*Technical University of Madrid, Madrid, Spain*, <sup>2</sup>*University of León, León, Spain*, <sup>3</sup>*IGM (CSIC-ULE), Grulleros, León, Spain.*

Fermenters are widely used to study ruminal fermentation, but information on microbial populations developing in fermenters over the incubation period is limited. Four Rusitec fermenters were fed 2 diets representative of those administered to dairy sheep (DAI; 50:50 alfalfa hay:concentrate) and fattening lambs (FAT; 15:85 barley straw:concentrate) in a crossover design with 2 14-d incubation periods to assess the evolution of the microbial populations. There were 4 fermenters per diet. The fermenters received daily 30 g of diet DM and samples from liquid (LIQ) and solid (SOL) digesta were taken on d 3, 8 and 14, and stored frozen at  $-80^{\circ}\text{C}$  until DNA extraction. Concentrations of bacterial and protozoal DNA and relative abundance of fungi and methanogenic archaea to total bacterial DNA concentration were determined by real time PCR using previously validated primers and DNA from bacteria and protozoa isolated from sheep rumen as standards. Data were analyzed as a mixed model with repeated measures using the PROC MIXED of SAS. The model included diet, incubation run, time, and diet  $\times$  time as fixed effects, and fermenter as a random effect. Diet  $\times$  sampling time interactions ( $P > 0.05$ ) were detected for bacterial and protozoal DNA concentrations in both digesta phases. The bacterial DNA concentrations in SOL did not change ( $P = 0.002$ ) over the incubation period, whereas concentrations in LIQ increased ( $P < 0.001$ ) by 1.5 and 1.8 times for DAI and FAT diets by the end of the incubation, respectively. Protozoal DNA concentrations on d 14 were 37.8 and 8.0 times lower ( $P < 0.001$ ; means across diets) than those on d 3 for SOL and LIQ phases, respectively. Relative abundance of fungi decreased ( $P < 0.05$ ) with time in both phases, and that of methanogenic archaea remain unchanged in LIQ and increased ( $P = 0.021$ ) in SOL. Concentration of bacterial and protozoal DNA and the relative abundance of methanogenic archaea were greater in the fermenters fed the DAI diet ( $P < 0.05$ ) compared with FAT diet. The results show that microbial populations in Rusitec fermenters are affected by the incubated diet and change over the incubation period.

**Key Words:** Rusitec fermenter, microbial populations, real-time PCR

**T486 Influence of inoculum preparation method on in vitro methane production by ruminal microorganisms.** Mireia Ramos<sup>1</sup>, Ivan Mateos<sup>2</sup>, Cristina Saro<sup>2</sup>, Alexey Díaz<sup>2</sup>, Maria Jose Ranilla<sup>\*2,3</sup>, and Maria Dolores Carro<sup>1</sup>, <sup>1</sup>*Technical University of Madrid, Madrid, Spain*, <sup>2</sup>*University of León, León, Spain*, <sup>3</sup>*IGM (CSIC-ULE), Grulleros, León, Spain.*

The characteristics of the inoculum are recognized as one of the most relevant factors influencing the results of in vitro fermentations in batch cultures of ruminal microorganisms. Four rumen-fistulated sheep fed a 66:34 alfalfa hay:concentrate diet were used as donors to investigate the effect of rumen contents' processing on in vitro methane (CH<sub>4</sub>) and volatile fatty acid (VFA) production from 3 substrates of variable composition. Rumen contents were sampled from each individual sheep and subjected to the following treatments: SQ) squeezed through 4 layers of cheesecloth; FIL) SQ treatment and further filtration through a 100- $\mu\text{m}$  nylon cloth; STO) treated with a Stomacher for 3 min at 230 rev min<sup>-1</sup> and followed by SQ. The resulting fluids were used as inoculum for batch cultures containing alfalfa hay, concentrate, or a 50:50 mixture of both feeds. Cultures were incubated at 39°C for 8 and 24 h, and CH<sub>4</sub> and VFA production was measured. There were no treatment  $\times$  substrate

interactions ( $P > 0.05$ ) for any variable at any incubation time, excepting for the molar proportion of acetate at 24 h ( $P = 0.019$ ). The method of processing the rumen contents did not affect ( $P > 0.05$ ) either total VFA and  $\text{CH}_4$  production or molar proportions of individual VFA at any time. At both incubation times, increasing the amount of concentrate in the substrate increased  $\text{CH}_4$  production ( $P < 0.001$ , quadratic) and molar proportion of butyrate ( $P < 0.001$ , linear), but decreased acetate proportion ( $P < 0.001$ , quadratic) without affecting ( $P > 0.05$ ) proportions of propionate. Whereas total VFA production was linearly decreased ( $P = 0.007$ ) by increased amounts of concentrate in the substrate at 8 h of incubation, it was quadratically increased ( $P < 0.001$ ) after 24 h of incubation. There were clear differences in  $\text{CH}_4$  and VFA production among inocula from different sheep, which persisted across substrates. The results show that the tested methods of processing rumen contents did not affect in vitro fermentation characteristics of good quality substrates, but studies analyzing their possible influence on fermentation of low-quality substrates are required.

**Key Words:** Rumen content treatment, methane, volatile fatty acids

**T487 Microbial rDNA sequences as markers to determine microbial synthesis in Rusitec fermenters: A comparison with  $^{15}\text{N}$ .** Cristina Saro<sup>2</sup>, Maria Jose Ranilla<sup>\*2,3</sup>, Ivan Mateos<sup>2</sup>, Alexey Díaz<sup>2</sup>, Jairo Garcia<sup>2</sup>, Maria Gracia de Garnica<sup>2</sup>, and Maria Dolores Carro<sup>1</sup>, <sup>1</sup>Technical University of Madrid, Madrid, Spain, <sup>2</sup>University of León, León, Spain, <sup>3</sup>IGM (CSIC-ULE), Grulleros, León, Spain.

Microbial rDNA sequences have been proposed as potential internal markers to determine microbial synthesis in the rumen. The objective of this experiment was to compare values of microbial growth determined using  $^{15}\text{N}$  as external marker with concentrations of microbial DNA in fermenters, and to assess if both procedures detected similar differences between diets and solid (SOL) and liquid (LIQ) digesta phases. Four Rusitec vessels were used in a crossover trial with 2 14-d periods. In each period, 2 fermenters received a 50:50 alfalfa hay:concentrate diet (MC) and 2 were fed a 15:85 barley straw:concentrate diet (HC). A solution of  $^{15}\text{NH}_4\text{Cl}$  was infused for 5 d before taken samples of SOL and LIQ digesta and isolation of bacterial pellets from both digesta phases to estimate microbial protein synthesis. Samples of SOL and LIQ digesta were simultaneously taken for DNA extraction and analysis of concentrations of total bacterial and protozoal DNA by quantitative PCR. Total microbial N (TMN) was calculated from the  $^{15}\text{N}$  enrichment in digesta and isolated bacterial pellets, and total microbial DNA (TMDNA) was calculated as the sum of bacterial DNA and protozoal DNA in both digesta fractions. There were no diet  $\times$  digesta phase interactions ( $P > 0.05$ ) with any marker. Both TMN and TMDNA were greater ( $P < 0.001$ ) in MC-fermenters than in HC-fermenters (1.5 and 2.0 times greater for TMN and TMDNA, respectively). Values of TMN were greater ( $P = 0.004$ ) in SOL than in LIQ digesta (108 and 89.7 mg N, respectively), whereas the opposite was found for TMDNA (3.37 and 13.1 mg DNA, respectively). There was no difference between diets ( $P > 0.05$ ) in the contribution of SOL digesta to TMN (53.8 and 55.7% for MC and HC diets, respectively), but contribution of SOL digesta to TMDNA was greater in MC than in HC diet ( $P = 0.039$ ; 24.5 and 11.5%, respectively). There was no relationship ( $P > 0.05$ ) between TMN and TMDNA values, but a significant relationship was observed when only values in the LIQ digesta were considered ( $P = 0.024$ ;  $r = 0.821$ ). In summary, both markers detected similar differences between diets, but not between digesta phases.

**Key Words:** microbial growth,  $^{15}\text{N}$ , qPCR

**T488 Comparison of Roche 454 and Ion Torrent Personal Genome Machine sequencing on the rumen bacterial profiles of dairy cows.** Nagaraju Indugu<sup>\*</sup>, Sanjay Kumar, Bonnie Vecchiarelli, and Dipti Pitta, *Department of Clinical Studies, School of Veterinary Medicine, New Bolton Center, University of Pennsylvania, Kennett Square, PA.*

Next generation sequencing (NGS) is a widely accepted technology used by microbial ecologists for metagenomic analysis of complex microbial communities. As technologies continue to improve, it is necessary to compare data sets obtained from different platforms and analyze their effect on community structure. In the present study, we compared the 454 data set with that of Ion Torrent Personal Genome Machine (PGM) on the same DNA samples ( $n = 14$ ) obtained from the rumen of dairy cows during their transition period. Despite the substantial difference in number of reads and length of reads, the platforms provided a similar community structure. The weighted UniFrac distances between the samples that were sequenced on both 454 and PGM were significantly correlated, as determined by Procrustes analysis of principal coordinate matrices ( $M2 = 0.319$ ; Monte Carlo  $P < 0.001$ ). Though similar major abundant phyla were detected by both platforms, PGM recovered 4 additional phyla. At the genus level, there was no substantial variation between the 454 and PGM data sets for each animal except for *Prevotella*, *Cyanobacteria* YS2 and *Succiniliclasticum* ( $P < 0.05$ ; chi-squared test). However, there was variation in the abundance of genera between different animals, irrespective of the platform used. Collectively, the present study will be useful for microbiologists/ecologists to compare the microbial community structure obtained from different platforms; particularly with the expectation 454 will be completely replaced by PGM and/ or Illumina.

**Key Words:** Ion Torrent PGM, microbial community, 454-Roche

**T489 Effect of chitosan in dairy cows diets on ruminal fermentation and milk yield and composition.** Carlos Eduardo Cardoso Consentini<sup>1</sup>, Elmeson Ferreira de Jesus<sup>2</sup>, Pablo Gomes de Paiva<sup>\*2</sup>, Tiago Antonio Del Valle<sup>1</sup>, Gustavo Ferreira de Almeida<sup>1</sup>, Artur Gabriel Brao Vilas Boas Costa<sup>1</sup>, Fernanda Carolina Ramos dos Santos<sup>1</sup>, Victor Chiaroni Galvão<sup>1</sup>, and Francisco Palma Rennó<sup>1</sup>, <sup>1</sup>School of Veterinary Medicine and Animal Science of USP, Pirassununga, São Paulo, Brazil, <sup>2</sup>School of Agricultural and Veterinary Sciences of UNESP, Jaboticabal, São Paulo, Brazil.

In this study, we evaluate the effects of chitosan level for cows in late lactation on ruminal fermentation and milk yield and composition. Eight Holstein cows cannulated in the rumen ( $215.4 \pm 60.9$  DIM,  $22.07 \pm 5.32$  kg of milk yield and  $641.6 \pm 41.06$  kg of BW) in replicated  $4 \times 4$  Latin squares, were fed the following diets: (CTR) control diet without addition of chitosan, CHI75 mg/kg, CHI150 mg/kg and 225 mg/kg of chitosan addition per kg BW. Each period had 14 d adaptation and 7 for collection data. Ruminal fluid was collected on 20th day of each period at 7 times to evaluate the effect of the diets on ruminal fermentation. Sampling of milk was done on 16 to 18th day of each period to evaluate the composition. The results of milk composition was subjected to ANOVA, while ruminal fermentation data were analyzed as repeated measures by PROC MIXED of SAS. Chitosan changed ruminal fermentation profile, increasing rumen propionate ( $P < 0.05$ ) without shift acetate and total VFA concentrations ( $P > 0.05$ ). However, chitosan decreased linearly acetate:propionate ratio ( $P < 0.05$ ) similarly to butyrate and AGCR concentrations ( $P < 0.05$ ). Chitosan increased milk yield ( $P < 0.05$ ). Furthermore, dietary chitosan linearly increased protein and lactose milk production ( $P < 0.05$ ). Chitosan as feed additive

improve animal performance and ruminal fermentation with increased ruminal propionate concentration.

**Key Words:** additive, antimicrobial, performance

**T490 Method to measure production of volatile fatty acids and gases in vitro.** Latisha M. Judd\* and Richard A. Kohn, *University of Maryland, College Park, MD.*

In vitro methods have been developed to measure digestibility, but such methods may not accurately estimate methane or volatile fatty acid (VFA) production. Methane emissions are stoichiometrically linked with VFA profiles. For example, a shift from acetate to propionate may decrease CO<sub>2</sub> and H<sub>2</sub> production, and in turn decrease conversion of CO<sub>2</sub> and H<sub>2</sub> to methane. The objective of this study was to determine the effect of different conditions of in vitro procedures on VFA and gas profiles in comparison with in vivo measurements. Experimental design was a 4 × 2 × 2 factorial CRD with 4 replicates. Treatments were 4 ratios of rumen fluid to buffer by volume (95/5, 75/25, 50/50, 25/75), 2 concentrations (w/v) of added timothy hay (0.5% or 1%), and with or without sodium acetate addition (50 mmol final concentration). Statistical analysis was conducted using a mixed model that included all fixed effects and interactions. Total volume of broth (rumen fluid and buffer) was 10 mL per tube. Measurements of gas production and VFAs were recorded at 0, 4, 16, 24, and 48 h. Total gas was proportioned into CO<sub>2</sub> and non-CO<sub>2</sub> after collection at 48 h. Higher hay concentration averaged more ( $P < 0.0001$ ) total gas than the lower hay concentration (12.1 vs. 6.7 mL gas/tube; SE = ± 0.23), and more ( $P < 0.0001$ ) non-CO<sub>2</sub> gas (0.28 vs. 0.15 mL; SE = ± 0.046). Total gas production increased ( $P < 0.001$ ) with higher buffer concentration, and averaged 6.1, 9.1, 11.2 and 11.2 mL per tube (SE = ± 0.32 mL) as buffer concentration increased. The acetate/propionate (A/P) concentration decreased over time ( $P < 0.0001$ ). The initial rumen fluid A/P ratio was 3.7 but the A/P ratio of produced VFA averaged 2.5 (SE = ± 0.14). The addition of acetate did not affect gas production or A/P ratio of produced VFA. This could mean that adding acetate to a system does not necessarily shift production away from acetate. A/P ratio differed for VFA produced in vitro compared with initial rumen fluid, but no tested treatments were identified as a cause of the difference.

**Key Words:** in vitro digestion, volatile fatty acids, methane

**T491 A rapid mold and yeast enumeration technique is comparable to a conventional technique for animal feedstuffs.** Lauren Meyer\*<sup>1</sup> and John Goesser<sup>1,2</sup>, <sup>1</sup>Rock River Laboratory, Watertown, WI, <sup>2</sup>University of Wisconsin-Madison, Madison, WI.

Feedstuff and TMR yeast and mold enumerations (cfu/g) have grown in popularity to diagnose opportunities on farm. Turnaround time with conventional enumeration (CON) limits utility, requiring 5 d incubation, extending total time from sampling to reporting to 7d or more. More recent human food grade, rapid yeast and mold enumeration techniques (RAP) offer faster turnaround and may have utility for production agriculture. The objective here was to determine if RAP, tested under 2 incubation lengths, was equivalent to CON. Corn silage (n = 17), TMR (n = 3), alfalfa silage (n = 15), high moisture corn or snaplage (n = 6), small grain silages (n = 6), and concentrate (n = 6) samples submitted for CON in late February 2015 were further assayed using RAP, with both 48-h and 5-d incubation. When samples arrived, roughly 5g feed was blended and stored at 1C for later plating. At plating, 1g wet feed was subsampled and diluted to 100mL in sterile buffer, shaken and then serially diluted 1:1000, 1:10,000 and 1:100,000 for most prob-

able number enumeration. For CON, subsamples of each dilution were taken with sterile glass pipette and plated on potato-dextrose agar using spread-plate method. For RAP, subsamples of each dilution taken with an electronic pipette were plated on Petrifilm, using a Petrifilm flat spreader (3M, St. Paul, MN). For RAP plates were aerobically incubated at 28C for both 48h and 5d and CON for 5d. Post incubation, enumeration was done by direct microscopy. Yeast and mold count mean/median across feeds and techniques were  $1.69 \times 10^6/1 \times 10^3$  and  $2.53 \times 10^5/1 \times 10^4$ , respectively. Raw and log-transformed data were determined not normally distributed, hence data were fit using one-way analysis option of SAS JMPv11.0. Technique (CON, RAP-48h, and RAP-5d) and feed main effects were compared using non-parametric Wilcoxon/Kruskal-Wallis test. Significance was declared if resulting Chi-squared statistic p-value was < 0.05. For both mold and yeast enumeration, techniques did not differ ( $P > 0.05$ ) while feed types differed ( $P < 0.01$ ). Our results suggest both yeast and mold enumeration results are comparable across the techniques tested here.

**Key Words:** feed, mold, yeast

**T492 Comparison of acetyl bromide lignin with acid detergent lignin and relationship with in vitro forage degradability.** Romualdo S. Fukushima\*<sup>1,2</sup>, Monty Kerley<sup>2</sup>, Marcelo H. Ramos<sup>2</sup>, James H. Porter<sup>2</sup>, and Robert L. Kallenbach<sup>2</sup>, <sup>1</sup>Sao Paulo University, Pirassununga, SP, Brazil, <sup>2</sup>University of Missouri, Columbia, MO.

The spectroscopic acetyl bromide lignin (ABL) and the gravimetric acid detergent lignin (ADL) methods were compared with in vitro forage dry matter degradability (IVDMD) and neutral detergent fiber degradability (IVNDFD) assays of 73 grass and legume samples, and a conjecture was made with the lignin component of the Cornell Net Carbohydrate and Protein System (CNCPS) equations. The slopes and intercepts of regressions were declared different when there was an interaction effect among forages within each lignin method (MIXED procedure of SAS). Regression curves of ADL values with forage IVDMD (grass:  $y = -7.929x + 901.8$ ; legume:  $y = -3.663x + 853.6$ ) and IVNDFD (grass:  $y = -3.289x + 916.0$ ; legume:  $y = -1.051x + 697.8$ ) revealed different slopes, with steeper curves for grasses. Grass and legume samples assayed with the ABL procedure, exhibited similar slopes, with parallel lines for both IVDMD (grass:  $y = -3.847x + 886.7$ ; legume:  $y = -3.638x + 789.8$ ) and IVNDFD (grass:  $y = -3.636x + 1117.7$ ; legume:  $y = -3.454x + 889.9$ ) assays. Steeper inclination of curve for grasses relative to legumes in the ADL method has been attributed to grass lignin being more inhibitory to degradation than legume lignin. Similar and parallel curves of ABL method suggests that grass lignin is no more inhibitory than legume lignin. However, the steeper inclination may be attributed to partial loss of grass lignin during the ADL procedure. We hypothesize that this loss is around 2.4, that is, the residual ADL multiplied by 2.4 would yield the actual lignin content. This number is the same used in the Cornell equations to estimate the B2 and C carbohydrate fractions (NDF x Lignin x 2.4). When we multiplied the grass ADL values by 2.4, forage IVDMD regressions were: (grass:  $y = -3.690x + 934.7$ ; legume:  $y = -3.663x + 853.6$ ), which originated parallel lines and were strikingly similar to the ones obtained with the ABL method. After correcting IVNDFD, the regressions were: (grass:  $y = -1.744x + 962.9$ ; legume:  $y = -1.737x + 805.8$ ), also yielding parallel curves. At this moment we can speculate that grass and legume lignins have the same effect on cell wall degradation and that ABL method seems a promising procedure for lignin quantification.

**Key Words:** lignin methods, Cornell carbohydrate equations



**T493 What roles do viruses play in the rumen?** Christopher Anderson, Galen Erickson, and Samodha Fernando\*, *University of Nebraska, Lincoln, NE.*

Viruses are the most abundant biological entity on earth. However, the role of viruses within ecosystems are poorly understood. As an attempt to better understand the role and functional relationships of viruses and their influence on rumen bacterial communities, we investigated viral and bacterial community relationships using culture independent metagenomic approaches under 4 different dietary conditions (55% corn silage, 27% condensed distillers plus solubles (CDS), 40% modified distillers grains plus solubles (MDGS), corn based control diet) in a Latin-square design with 5 fistulated steers. Rumen samples were collected after total rumen evacuation and mixing following a 21-d adaptation period. Tangential flow filtration was performed to enrich for viral particles from the rumen sample. The enriched viral metagenome and the total metagenome were sequenced using the Ion Torrent Personal Genome Machine (PGM) to identify species composition, interactions between viruses and bacteria, and to identify the role of

virus auxiliary genes within rumen metabolism. The metagenome analysis displayed the total metagenome contained 118 differentially abundant genes and the viral metagenome 309 differentially abundant genes based on diet. Interestingly, the metagenomes and metaviromes contained different metabolic profiles. To better understand the role of the virome in ecosystem function, we mapped the genes identified to a community metabolic network. Using the metabolic networks we compared topological features of enzyme nodes to identify the roles of differentially abundant genes. The nodes that were differentially abundant in the total metagenome and virome had significantly higher betweenness centrality ( $P < 0.05$ ) and a lower average shortest path length compared with non-differential genes in the network ( $P < 0.05$ ). In addition, differential viral genes had a significantly higher total degree and in-degree compared with non-differential genes and the differential genes in the total metagenome ( $P < 0.05$ ). Currently, we are applying network approaches to understand the ecological roles of viruses within energy metabolism in the rumen environment.

**Key Words:** metagenome, virome, metabolic network

## Small Ruminant II

**T494 Effects of short-term inclusion of mixed fish and sunflower oils in finishing diet on carcass characteristics and performance of fat tailed Afshari lambs.** H. R. Mirzaei Alamouti\*, T. Khademi, M. H. Shahir, and M. Hajilo, *Department of Animal Science, University of Zanjan, Iran.*

An experiment was conducted to determine the effects of unsaturated fatty acids supplementation on growth performance and carcass characteristics of finishing lambs. Thirty-two finishing Afshari lambs (initial weigh  $58 \pm 5$  kg and 8 mo age) were used in a completely randomized design with 4 treatments (slaughtered in time series) and 8 lambs/treatment for the final 30 d before slaughter. The slaughter times were d 0 (first day after adaptation period), 10, 20 and 30 after beginning of experiment. The lambs individually fed to the basal diet (80% concentrate; 65% barley grain and 11% soybean meal and 4% mineral and vitamin supplement, and 20% alfalfa hay) and daily fed with 60 g sunflower oil and 40 g fish oil. Body weight changes and daily feed intake were recorded. Blood samples, ultrasonography (back fat and Longissimus muscle thickness) and rumen fluid was taken on the last day of each period. Cold and warm slaughter weight, dressing percentage, fat yield, pH, moisture loss, chemical composition, and visceral organs mass were measured. The results of this study were revealed that body weight, back fat and Longissimus muscle thickness, for d 0, 10, 20 and 30 after allocating to high oil supplemented diets was 59.20, 62.45, 65.26, and 69.07 kg; 4.8, 5.09, 5.36, and 4.79 mm; 24.41, 24.28, 25.90, and 30.38 mm; respectively. Warm and cold carcass weight and rumino-reticulum mass increased during time. Oil supplementation decreased tail weight to carcass weight ratio from 0.17 to 0.12 and also improved feed conversion ratio. Plasma concentration of cholesterol was highly increased by oil supplementation and was significantly different among the experimental diets ( $P < 0.01$ ). This experiment showed that short-term inclusion of mixed fish and sunflower oils in final days of finishing lambs diet can improve carcass characteristics and performance of lambs.

**Key Words:** unsaturated fatty acid, carcass characteristic, finishing lamb

**T495 Growth, hepatic enzymes and carcass characteristics of lambs fed diets containing increasing levels of crude glycerin.**

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Crude glycerin is a high energy by-product of biodiesel production. However, some impurities may be present in it including methanol and salts. Methanol can impair liver function. The objectives of this trial were to determine the effects of partial replacement of corn by crude glycerin on growth, hepatic enzymes and carcass characteristics of lambs fed high-concentrate diets. Fifty (40 males and 10 females) White Dorper  $\times$  Santa Ines lambs were assigned to a randomized complete block design, defined by initial BW, sex and age. Lambs were penned individually and fed, during 56 d (2 periods of 28-d), a TMR composed of 90% concentrate. Diets were isonitrogenous ( $15.7 \pm 0.3\%$  CP, DM basis) and the increasing levels of crude glycerin (83.6% glycerol and 397 mg of methanol/kg, DM basis) were 0, 5, 10, 15 or 20%. Lambs were weighted and blood samples were collected on d-0, d-28 and d-56. Average daily gain (ADG), feed efficiency (FE) and dry matter intake (DMI) were determined in each period. At the end of the 56-d, all ram lambs were

slaughtered for carcass characteristics evaluation. Data were analyzed using the MIXED procedure (SAS Inst. Inc.). Orthogonal polynomials for diet responses were determined by linear and quadratic. Replacing corn by crude glycerin did not affect ADG (0.29, 0.29, 0.30, 0.31 and 0.28 kg), DMI (1.03, 1.02, 1.04, 1.08, 1.00 kg/d), FE (0.28, 0.28, 0.29, 0.28, 0.28) and body weight on d-28 (29.3, 29.3, 29.3, 29.2, 28.4 kg) and d-56 (38.9, 38.7, 39.0, 39.2, 38.2 kg). There was a diet and period interaction for DMI. In the second period (d28-d56) a quadratic effect for DMI (1.11, 1.15, 1.18, 1.25, 1.11, kg/d) was observed. Hepatic enzymes (aspartate aminotransferase and gamma glutamyltransferase) activity and carcass characteristics were not affected by crude glycerin addition to the diet. Crude glycerin can be used to replace up to 20% of corn (DM basis) in high-concentrate diet of lambs without detrimental effects on performance, hepatic enzymes and carcass characteristics.

**Key Words:** co-product, glycerol, sheep

**T496 Effect of supplementation with dried citrus pulp, urea, and DDGS on reproductive performance of goats.** Mayra A. Liñan Gonzalez<sup>1</sup>, Hugo Bernal Barragan<sup>\*1</sup>, Fernando Sanchez Davila<sup>1</sup>, Rogelio A. Ledezma Torres<sup>1</sup>, Miguel Cervantes Ramirez<sup>2</sup>, and Braulio Valles de la Mora<sup>3</sup>, <sup>1</sup>*Universidad Autonoma de Nuevo Leon, San Nicolas de los Garza, Nuevo Leon, Mexico,* <sup>2</sup>*Universidad Autonoma de Baja California, Mexicali, Baja California, Mexico,* <sup>3</sup>*Universidad Nacional Autonoma de Mexico, Martinez de la Torre, Veracruz, Mexico.*

The objective of this study was to evaluate the effect of supplementation with 2 agroindustrial byproducts and urea, on body weight, serum glucose and BUN, as well as reproductive performance and blood progesterone of 56 goats of a commercial farm in Northeastern Mexico, grazing mixed thornscrub vegetation, including browse species, grass, forbs and *Opuntia* spp. (3.6 to 15.5% crude protein (CP) and 27.0 to 69.3% NDF), with daily access to shelter. Goats were blocked by breed (Nubian, Alpina and Saanen), age (1.5 to 2.5 years), parity (1 to 3), and body weight, and were randomly assigned among blocks either to T1: No supplementation; or to individually receiving 300 g/d of T2: dried citrus pulp (DCP, 4.9% CP, 20.0% NDF); or T3: DCP+Urea+DDGS (17.7% CP, 26.0% NDF); or T4: DCP+DDGS (15.4% CP, 29.2% NDF; n = 14 goats/treatment). Supplementation lasted 28 d in July–August 2014 (outside the natural breeding season). Beginning on d 13, a 9-d oestrus synchronization protocol was conducted applying 50  $\mu$ g GnRH on d 13 and 22, and 75  $\mu$ g PGF2 $\alpha$  on d 20. Oestrus was observed utilizing teaser bucks, and goats were fixed-time trans cervically inseminated 16 h after the second GnRH dose, using fresh processed and evaluated semen of 2 randomly assigned sires. Blood samples were collected by venipuncture of jugular vein on d 1, 6, 12, 20, 22 and 26. Glucose and BUN were colorimetrically analyzed. Progesterone was measured by ELISA. Results were analyzed by ANOVA and Chi-squared. Supplemented goats lost less BW ( $P < 0.05$ ) than control animals. Serum glucose ( $42.3 \pm 0.7$  mg/dL) was similar ( $P > 0.05$ ) among treatments. Goats in T1 and T4 had higher BUN values than T2 (mean 27.6 vs 25.2 mg/dL;  $P < 0.05$ ). No differences were detected among treatments in oestrus appearance. Fertility rate of T2 and T3 (53 – 57%) was numerically better than in T1 and T4 (35 – 43%). Goats in T1 had on d 22 higher ( $P < 0.05$ ) progesterone values than others. In conclusion, supplementing 300 g

of DCP and DCP+Urea+DDGS during 28 d helped avoid loss of BW and improved fertility of goats.

**Key Words:** goat, dried citrus pulp, dried distillers grains with solubles

**T497 Effect of feed restriction on protein metabolism of Saanen goats of different sexes.** Nhayandra C. D. Silva\*, Izabelle A. M. A. Teixeira, Carla J. Härter, Simone P. Silva, Amélia K. Almeida, Diogo C. Soares, and Kléber T. Resende, *Unesp Univ Estadual Paulista, Jaboticabal, São Paulo, Brazil.*

The objective of this study was to evaluate the effect of feed restriction on protein metabolism of 84 Saanen goats (26 intact males, 27 castrated males and 31 females) with initial BW of  $30.3 \pm 0.87$  kg. At the beginning of the experiment 8 intact males, 9 castrated males and 13 females were slaughtered with 30 kg of BW to estimate their initial body composition. The remaining 18 goats of each sex were assigned to 3 levels of feed restriction (ad libitum, 25% and 50% of feed restriction), with 6 goats per sex-feeding level ( $3 \times 3$  factorial). Animal sets (1 goat per sex-feeding level) were slaughtered when BW of ad libitum goats was 45 kg. Blood samples were collected every 14 d, in a total of 7 blood collections (98 d of experimental period). In these samples we evaluated total protein, albumin, urea, creatinine, triiodothyronine (T3) and IGF-1. Data were analyzed using Proc Mixed of SAS. Intact males presented lower body protein retention (kg) regardless the level of feed restriction ( $P = 0.020$ ). Intact males fed ad libitum and with 50% of feed restriction had greater serum levels of creatinine in the blood (0.88 mg/dL and 1.28 mg/dL respectively;  $P = 0.014$ ). Females fed ad libitum had lower urea levels ( $42.08 \pm 1.83$  mg/dL) than intact ( $51.1 \pm 1.83$  mg/dL) and castrated males ( $48.2 \pm 1.81$  mg/dL) however, when subjected feed restriction (25% and 50%), females increased serum levels of urea, whereas males had an opposite behavior. The feed restriction and sex did not affect serum albumin and total protein levels ( $P > 0.05$ ). Intact males fed ad libitum and 25% feed restriction showed greater plasma levels of IGF-1 than females and castrated males ( $P = 0.018$ ), but when fed 50% restriction, the IGF-1 concentration were similar between all sexes ( $85.0 \pm 9.89$  ng/mL). Plasma levels of T3 were similar in females and castrated males (1.57 ng/mL) and greater than in intact males (1.43 ng/mL;  $P = 0.001$ ). The levels of IGF-1 and T3 in the blood showed a linear decrease with the increase of feed restriction ( $P < 0.0001$ ). The feed restriction changed the protein metabolism in females and males. Pubertal males were not able to keep protein synthesis during feed restriction.

**Key Words:** dairy goat, gender, metabolic profile

**T498 Female goat kids change their energy metabolism when subjected to feed restriction.** Nhayandra C. D. Silva\*, Izabelle A. M. A. Teixeira<sup>1</sup>, Carla J. Härter<sup>1</sup>, Fernanda O. M. Figueiredo<sup>1</sup>, Rafael F. Leite<sup>1</sup>, Moaceli M. Freire<sup>2</sup>, and Kléber T. Resende<sup>1</sup>, <sup>1</sup>Unesp Univ Estadual Paulista, Jaboticabal, São Paulo, Brazil, <sup>2</sup>Universidade Federal de Alagoas, Maceió, Alagoas, Brazil.

The objective of this study was to evaluate the effect of sex and feed restriction on energy metabolism of 72 Saanen goat kids (24 intact males, 24 castrated males and 24 female) with initial BW of  $15.8 \pm 0.17$  kg. At the beginning of the experiment 6 animals of each sex were slaughtered with 15 kg of BW to estimate their initial body composition. The remaining 18 kids of each sex were assigned to 3 levels of feed restriction (ad libitum, 25% and 50% of feed restriction), with 6 kids per sex-feeding level ( $3 \times 3$  factorial). Animal sets (1 kid per sex-feeding level) were slaughtered when BW of ad libitum kids was 30 kg. Blood samples were collected from all animals every 10 d, in a total

of 7 blood collections (70 d of experimental period). In these samples, we evaluated glucose, cholesterol, NEFA,  $\beta$ -hydroxybutyrate (B-HB) and aspartate aminotransferase (AST). Data were analyzed using Proc Mixed of SAS. Females presented greater fat deposition ( $31.6 \pm 0.36\%$  EBW) than castrated males ( $24.7 \pm 0.36\%$  EBW) and intact males ( $11.5 \pm 0.36\%$  EBW) regardless the level of feed restriction ( $P < 0.0001$ ). Serum glucose was similar in intact and castrated males throughout the experiment ( $67.4 \pm 4.59$  mg/dL). As the feed restriction level increased, serum AST activity of castrated males decreased ( $P = 0.0251$ ) in a ratio of 4.3% for those subjected to 25% of feed restriction and 15.8% for those under 50% feed restriction compared with castrated fed ad libitum. In females, AST activity was higher in those subjected to 50% feed restriction ( $83.83 \pm 4.96$  U/L), whereas in intact males remained unchanged regardless of the restriction regimen ( $80.9 \pm 4.95$  U/L). The highest B-HB concentration was observed when animals were subjected to the maximum level of feed restriction (0.129 mmol/l) followed by those fed ad libitum (0.103 mmol/L) and restricted by 25% (0.090 mmol/L;  $P < 0.0149$ ). Sex and feed restriction did not influence NEFA and cholesterol blood levels ( $P > 0.05$ ). Goat kids of different sex act physiologically different when subjected to feed restriction. Females changed all their glycolytic metabolism to keep the fat deposition even when they are subjected to feed restriction.

**Key Words:** growth, metabolic profile

**T499 Effects of restricted diet access on intake and performance by dairy goats in mid- to late lactation.** Nhayandra C. D. Silva\*<sup>1,2</sup>, Ryszard Puchala<sup>1</sup>, Terry A. Gipson<sup>1</sup>, Yoko Tsukahara<sup>1</sup>, Tilahun Sahlul<sup>1</sup>, and Arthur L. Goetsch<sup>1</sup>, <sup>1</sup>American Institute for Goat Research, Langston University, Langston, OK, <sup>2</sup>UNESP, Universidade Estadual Paulista, Department of Animal Science, Jaboticabal, SP, Brazil.

Restricting periods of dietary access of lactating dairy goats could influence level or efficiency of production and offer different management options. Therefore, objectives of the experiment were to determine effects of offering feed at different times of the day and for various lengths on intake and milk yield and composition of 50 Alpines (15, 25, and 10 of parity 1, 2, and  $\geq 3$ , respectively) with initial BW of 55.2 kg (SE = 0.95) and 125 d-in-milk (SE = 3.0). A 40% forage diet (20% alfalfa pellets, 10% cottonseed hulls, and 10% coarsely ground grass hay) was given free-choice in Calan gate feeders during a 2-wk covariate period and subsequent 12-wk experiment. Treatments were feed access continuously (C), during the day for 8 h (D) or night for 16 h (N), and for 2 or 4 h/d with equal lengths after milking in the morning and afternoon (2H and 4H, respectively) (10 animals/treatment). Neither DMI (2.05, 1.87, 2.08, 1.91, and 1.87 kg/d; SE = 0.107) nor milk yield (1.77, 1.75, 1.67, 1.64, and 1.68 kg/d for C, D, N, 2H, and 4H, respectively; SE = 0.098) were influenced by treatment ( $P > 0.05$ ), with milk yield (1.83, 1.84, 1.60, and 1.54 kg/d in periods 1, 2, 3, and 4, respectively; SE = 0.051) but not DMI differing among periods. Treatment also did not influence ADG (32, 22, 49, 9, and 20 g; SE = 13.0) or body condition score during the study (2.35, 2.32, 2.24, 2.26, and 2.34; SE = 0.052) and at the end (2.49, 2.39, 2.32, 2.33, and 2.42; SE = 0.054). However, there were treatment effects on milk concentrations of fat (3.78, 3.64, 3.54, 3.75, and 3.21%; SE = 0.126) and protein (2.91, 2.88, 2.88, 2.84, and 2.58% for C, D, N, 2H, and 4H, respectively; SE = 0.049). Energy-corrected milk (3.5% fat, 3.2% protein) in kg/d (1.70, 1.66, 1.58, 1.53, and 1.52 kg/d; SE = 0.101) and relative to DMI (0.79, 0.84, 0.78, 0.81, and 0.81 kg/kg for C, D, N, 2H, and 4H, respectively; SE = 0.073) were similar among treatments. In conclusion, dairy goats in mid- and late lactation possess considerable flexibility in eating behavior that may



allow for incorporation of limited feed access regimens in management systems for most efficient facility utilization.

**Key Words:** dairy goat, feed access, milk production

**T500 Effect of sodium monensin on rumen metabolism in lambs fed high-forage diets.** Mariana F. Westphalen<sup>\*1</sup>, Daniel M. Polizel<sup>2</sup>, Marcelo H. Santos<sup>2</sup>, Renan G. Silva<sup>2</sup>, Analisa V. Bertoloni<sup>1</sup>, Gabriela B. Oliveira<sup>1</sup>, Thiago S. Andrade<sup>2</sup>, Vinicius N. Gouvea<sup>2</sup>, Marcos V. Biehl<sup>2</sup>, and Alexandre V. Pires<sup>1,2</sup>, <sup>1</sup>University of São Paulo, Piracicaba, São Paulo, Brazil, <sup>2</sup>University of São Paulo, Pirassununga, São Paulo, Brazil.

The objectives of this trial were to determine the effects of increasing doses of sodium monensin on rumen metabolism of lambs fed high-forage diets. Five wethers (BW 70.5 ± 2.8 kg), cannulated in the rumen, were used in 5 × 5 Latin Square design. Animals were fed daily and diet was composed of coastcross hay. Sodium monensin were offered twice a day and doses were 0 (control), 8, 16, 24 or 32 mg/kg DM, corresponding to 0, 40, 80, 120 and 160 mg of Rumensin 200. The delivery vehicle of the set dosage of monensin was 20 g of ground corn per 1 kg DM intake. Every period of experiment lasted 20 d and rumen fluid was collected in the last day, every 3 h, starting prior feeding, 3, 6, 9 and 12 h after feeding. Short-chain fatty acids (SCFA) and pH were analyzed as repeated measures over time. Protozoa concentration was determined only 3 h after feeding. Data were analyzed using MIXED procedure (SAS Inst. Inc.) and LSMEANS option was used to generate individual means. Orthogonal polynomials for diets responses were determined by linear and quadratic effect. There was a quadratic response for acetate (78.22, 78.25, 78.65, 77.62, 76.27 mM/100 mM,  $P = 0.03$ ), propionate (14.81, 14.59, 15.07, 15.60, 16.90 mM/100 mM,  $P = 0.02$ ), isobutyrate (0.66, 0.59, 0.53, 0.56, 0.64 mM/100 mM,  $P = 0.02$ ) and acetate:propionate ratio (5.30, 5.35, 5.24, 5.02, 4.54,  $P = 0.02$ ). However, there was an interaction ( $P < 0.05$ ) between diet × hour for acetate:propionate ratio. The inclusion of sodium monensin decreased acetate:propionate ratio during 9 and 12 h. There was a linear response for valerate (0.76, 0.66, 0.59, 0.60, 0.59 mM/100 mM,  $P = 0.03$ ) and pH (6.57, 6.61, 6.46, 6.43, 6.49,  $P = 0.05$ ). Butyrate (4.42 ± 0.06 mM/100 mM), isovalerate (1.01 ± 0.04 mM/100 mM), total SCFA (85.58 ± 1.14 mM/L) and protozoa concentration (1.88 ± 0.22 × 10<sup>5</sup>/mL) were unaffected ( $P > 0.05$ ) by the experimental diets. Sodium monensin doses above 16 mg/kg DM provided an increase in the molar proportion of propionate over acetate, consequently reducing the acetate-to-propionate ratio and pH.

**Key Words:** ionophore, propionate, acetate:propionate ratio

**T501 Either intramuscular or submucous vulvar administration of HCG positively affects the reproductive outcomes of anovulatory Alpine goats in Northern Mexico.** Karen Isabel Tapia-Robles<sup>\*1</sup>, Cesar Alberto Meza-Herrera<sup>2</sup>, Jessica Maria Flores-Salas<sup>1</sup>, Alan Sebastian Alvarado-Espino<sup>1</sup>, Vicente Homero Gonzalez-Alvarez<sup>1</sup>, Evaristo Carrillo-Castellanos<sup>3</sup>, Juan Manuel Guillen-Muñoz<sup>1</sup>, Francisco Gerardo Veliz-Deras<sup>1</sup>, and Rafael Rodriguez-Martinez<sup>1</sup>, <sup>1</sup>Universidad Autonoma Agraria Antonio Narro, Torreon, Mexico, <sup>2</sup>Universidad Autonoma Chapingo, Unidad Regional Universitaria de Zonas Aridas, Bermejillo, Durango, Mexico, <sup>3</sup>Instituto Tecnológico de Torreon, Torreon, Mexico.

This study was performed to determine whether vulvar administration HCG, either intramuscular (IM) or submucous (SM), influence the reproductive response of goats during the natural anestrus season in Northern Mexico. Anovulatory Alpine goats (n = 30) under intensive

conditions were divided in 3 homogeneous groups (n = 10, each) based on body weight (BW; 37.36 ± 8.48 kg) and body condition (BC, 1.96 ± 0.32). On June 29th, 20 mg of progesterone was applied to all of the goats; 24-h later (d 0), 7.5 mg of PGF<sub>2α</sub> IM was also applied. Group 1; (GC) received 0.5 ml of isotonic saline solution IM, group 2 (GIM) received 100 UI of hCG of IM and group 3 (GSM) received 100 UI of HCG SM. Estrous activity was registered twice per day from d-0 to d-5 using a male; females accepting mating were considered to be in estrus. Ovulation activity was assessed on d-10 by detecting the presence of at least one corpus luteum while gestation was determined on d-45 both throughout a transrectal ultrasonographic scanning (7.5 MHz). The percentages of females in estrous, ovulating and pregnant were compared with chi<sup>2</sup>. While the control group did not depict any response in either variable, treated groups depicted an important reproductive response without differences ( $P > 0.05$ ) between treated groups regarding estrus behavior (GIM 100% vs GSM 100%), ovulatory activity (GIM 70% vs GSM 100%), and pregnancy rate (GIM 70% vs GSM 100%). To conclude, regardless of the administration route of 100 IU of HCG, either i.m. or s.m., both treatments were equally efficient to induce sexual activity with significant reproductive outcomes in Alpine goats during the natural anestrus season in northern Mexico (25°N).

**Key Words:** anestrus goats, HCG, administration route

**T502 Performance of Santa Inês meat lambs receiving cactus pear (*Nopalea cochenillifera*) in substitution of Tifton hay with or without access of water.** Alma V. Cordova Torres<sup>1</sup>, Leonardo S. Knupp<sup>2</sup>, Antonello Cannas<sup>\*2</sup>, Giustino Gaspa<sup>2</sup>, José T. Araújo Filho<sup>1</sup>, Ariosvaldo N. Medeiros<sup>1</sup>, Neymar L. Alves<sup>3</sup>, and Roberto G. Costa<sup>1</sup>, <sup>1</sup>Programa de Pós Graduação em Ciência e Tecnologia de Alimentos, Universidade Federal da Paraíba, João Pessoa, Paraíba, Brasil, <sup>2</sup>Dipartimento di Agraria, University of Sassari, Sardinia, Italy, <sup>3</sup>Departamento de Zootecnia, Universidade Federal de Alagoas, Rio Largo, Alagoas, Brasil.

This work aimed to study the performance of Santa Inês meat lambs receiving different levels of inclusion of cactus pear (*Nopalea cochenillifera*) in substitution of Tifton (*Cynodon dactylon*) hay. The trial was conducted using 48 Santa Inês male lambs at 100 d of age, with average initial weight of 18.75 ± 0.46 kg. Diets were formulated to be isoproteic and isoenergetic. Animals had ad libitum access to feed. The experiment was carried out as a completely randomized factorial design (3 × 2), with 3 levels of substitution of Tifton hay with cactus pear (30%, 50% and 70%), subdivided in 2 groups, with or without access of water, and a control treatment. DMI significantly differed among treatments ( $P < 0.001$ ) and the interaction among levels of cactus and access of water were not significant for all variables ( $P > 0.05$ ). The equations to estimate DMI and the ADG in the diets with cactus pear were: DMI = 1572.3 - 17.8x - 112.8y + 0.07x<sup>2</sup> + 1.78xy ( $R^2 = 0.63$ ); ADG = 145.3 + 1.2x + 15.5y - 0.02x<sup>2</sup> - 0.10xy ( $R^2 = 0.29$ ), being x = level of inclusion of cactus pear in the diet (30, 50, 70%) and y = animals with (1) or without (0) access of water. In contrast, the equations to predict the DMI and the ADG including the control treatment with the group that had access of water were: DMI = 897.3 + 7.8x - 0.16x<sup>2</sup> ( $R^2 = 0.40$ ); ADG = 109.2 + 3.2x - 0.04x<sup>2</sup> ( $R^2 = 0.43$ ), being x = diet control and levels of cactus (0, 30, 50, 70%). The maximum value of DMI was 1.1 kg/day and the minimum was 0.7 kg/day, obtained for the group receiving 30% of cactus without access of water and 70% without access of water, respectively. The ADG was different among treatments ( $P < 0.05$ ) ranging between 177.8 g to 106.8 g for the group with 30% of cactus in the diet with access of water and the control treatment, respectively. Feed efficiency was affected only by treatments, being the group that received 70% of

cactus in the diet with access of water the most efficient (0.19 g). In conclusion, cactus pear is recommended at 30% (with access of water) or 50% (without access of water) of substitution of Tifton hay, resulting in ADG of 178 or 157 g/day, respectively.

**Key Words:** cactus, dry matter intake, weight gain

#### **T503 Influence of supplementing lamb with dried algae + live yeast product on growth and blood metabolites during summer.**

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Endocrine and immune system stimulation by Algae is an active area of research. Thus, the current case conducted to examine the impact of an dried algae (*Schizochytrium limacinum*) and live yeast product (*Saccharomyces cerevisiae* CBS 493.94;  $3.0 \times 10^{10}$  cfu) on lamb performance and blood metabolites during summer (mean temperature humidity index 75.5). Lambs were blocked by weight and randomly assigned to receive either control ration (n = 20) based on wheat grain and sunflower meal or a algae derived supplement with live yeast product (n = 20) in a crossover design with two 21-d experimental periods. Supplement top dressed 1×/d at a rate of 5 g/head/d. Daily pen dry matter intake were recorded and lambs were weighed weekly throughout the trial to determine daily weight gain and feed efficiency. Blood samples were collected from the individual animals during both periods at the beginning of the treatment period (d 0) and on the last day (d 21) of trial. Data were analyzed with the repeated measures PROC MIXED procedure of SAS (SAS Inst. Inc. Cary NC). Overall, Algae+ live yeast supplement increased final live weight and average daily gain ( $P < 0.05$ ). However, no differences were noted between treatments for average dry matter intake ( $P = 0.24$ ) or feed efficiency ( $P = 0.23$ ). Plasma glucose (98.47 vs. 84.97 mg/dl) and insulin (64.14 vs. 29.26 ng/mL) concentrations were higher ( $P < 0.05$ ) while total plasma cholesterol concentration tended to be lower ( $P = 0.09$ ; 58.22 vs. 61.68 mg/dL) in Algae+ live yeast-fed lambs relative to control animals. However, plasma non-ester fatty acid ( $P = 0.68$ ), total protein ( $P = 0.55$ ), blood urea nitrogen ( $P = 0.58$ ), lipopolysaccharide binding protein ( $P = 0.89$ ), diamine oxidase ( $P = 0.17$ ), malondialdehyde ( $P = 0.49$ ) and glutathione peroxidase ( $P = 0.44$ ) concentrations were unaffected by supplementation. Results of this study indicate that algae supplement influences productivity and alter bioenergetics in growing lambs during summer. This study was supported by the Scientific Research Project Council of Uludag University, Project Number: KUAP(Z)-2013/46.

**Key Words:** algae, blood metabolite, lamb performance

**T504 Net energy and protein requirements for growth of Moxotó goats grazing in the semiarid region of Brazil.** Marcos J. Araújo\*<sup>1</sup>, Ariosvaldo N. Medeiros<sup>2</sup>, Carlo A. T. Marques<sup>1</sup>, Roberto G. Costa<sup>2</sup>, Francisco F. R. Carvalho<sup>3</sup>, and Jacira N. C. Torreão<sup>4</sup>, <sup>1</sup>Federal University of Piauí, Bom Jesus, Piauí, Brazil, <sup>2</sup>Federal University of Paraíba, Areia, Paraíba, Brazil, <sup>3</sup>Federal Rural University of Pernambuco, Recife, Pernambuco, Brazil, <sup>4</sup>Colégio Técnico de Bom Jesus, Bom Jesus, Piauí, Brazil.

Indigenous goats play an important role in the semiarid region of Brazil as a biological resource with great genetic variability and historical value. They are raised in the traditional manner because they are rustic animals, commonly used for milk and meat production. Therefore, understanding their nutritional requirements is important for a success-

ful nutrition system because animals fed properly will more efficiently convert the nutrients ingested in products. The objective of this study was to determine net energy (NE<sub>g</sub>) and protein (NP<sub>g</sub>) requirements for growth of 36 male Moxotó goat kids (15.69 ± 0.78 kg initial BW), grazing in the semiarid region of Brazil. Four kids were slaughtered at the beginning of the experiment (baseline group, 15.37 ± 0.30 kg BW) and the remainder (n = 32) were allocated randomly to one of the 4 levels of concentrate supplementation (treatments groups: 0, 0.5, 1.0 and 1.5% BW), with 8 kids per group. When the animals in the 1.5% BW treatment group reached 25 kg BW, the animals in the other treatment groups were also slaughtered. The individual whole empty body was weighed, ground, mixed and sampled for chemical analyses. We used the comparative slaughter method to assess body composition and calculate the nutritional requirements. Linear regressions were used to determine the relationship between the shrunk BW and the empty body weight (EBW). The allometric equations were calculated using the relationship between the amount of fat, energy and protein and the EBW. Body composition varied from 156.40 to 171.52 g of protein, from 65.20 to 138.44 g of fat and from 1.56 to 2.51 Mcal of energy per kg of EBW. The ratio of EBW/BW was 0.81. The NE<sub>g</sub> requirements to gain ranged from 2.85 to 4.58 Mcal/kg EBW gain and NP<sub>g</sub> to gain ranged from 181.36 to 198.78 g/kg EBW gain for the castrated indigenous goat kids weighing between 15 and 25 kg. This study has indicated that castrated Moxotó goat kids have different protein and energy requirements in relation to those values commonly recommended by feeding system for other breeds.

**Key Words:** body composition, caatinga, comparative slaughter

#### **T505 Reproductive outcomes of anovulatory females exposed to males treated with either i.m. or s.c. testosterone.**

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The aim of this study was to evaluate the reproductive response of anovulatory goats exposed to bucks treated with either intramuscular (i.m.) or subcutaneous (s.c.) testosterone in April in northern Mexico (25°N, 103°W). Anovulatory female goats (n = 60) were divided in 3 homogeneous groups regarding body weight and condition (20 goats each) and exposed to 2 of 6 multiracial bucks. Males were randomly selected to receive different treatments: (1) 50 mg testosterone i.m. (TIM, n = 2), (2) 50 mg testosterone s.c. (TSC, n = 2) and (3) 0.5 mL of physiological saline (CONT, n = 2). Treatments lasted 21 d, with treatments every 3 d. Thereafter, each group of males was exposed to 20 females to evaluate the male effect. Response variables considered the latency (interval between introduction of the males and estrus initiation) and sexual activity of females twice daily (0700 and 1900 h), for 10 d. The percentages of females in heat, ovulated (10-d post-male introduction) and pregnant (45-d post-treatment) were also considered. Ovulation and pregnancy response were evaluated by transrectal ultrasonographic scanning (7.5 MHz, HS-2000, Honda Electronic CO, LTD). The proportions of females showing estrus, ovulation and pregnancy were compared using an exact Fisher probability test, while latency considered the Student *t*-test (SYSTAT 5.03 software). Results suggest that males treated with either SC or IM testosterone depicted similar reproductive outcomes, with the TIM depicting the highest

estrus response ( $P < 0.05$ ) yet without differences regarding ovulation and pregnancy rates (Table 1).

**Table 1 (Abstr. T505).** Reproductive outcomes in anovulatory goats exposed to the male effect with mix-breed bucks treated either with s.c or i.m. testosterone during the anovulatory season in northern Mexico (May, 25°N)

Groups	Latency	Estrus %	Ovulation %	Gestation %
CONT	50 ± 3.8 <sup>a</sup>	65 (14/20) <sup>a</sup>	50 (10/20) <sup>a</sup>	10 (2/20) <sup>a</sup>
TIM	38.4 ± 5.4 <sup>a</sup>	100 (20/20) <sup>b</sup>	65 (14/20) <sup>a</sup>	55 (12/20) <sup>b</sup>
TSC	42 ± 3.9 <sup>a</sup>	65 (14/20) <sup>a</sup>	47.5 (9/20) <sup>a</sup>	45 (8/20) <sup>b</sup>

Values with different superscripts within column differ ( $P < 0.05$ ).

**Key Words:** seasonal reproduction, female goat, testosterone

**T506 Mineral requirements for growth and maintenance of F<sub>1</sub> Boer × Saanen male kids.** Izabelle A. M. A. Teixeira<sup>\*1</sup>, Carla J. Härter<sup>1</sup>, José M. Pereira Filho<sup>2</sup>, Américo G. Silva Sobrinho<sup>1</sup>, and Kleber T. Resende<sup>1</sup>. <sup>1</sup>UNESP, Universidade Estadual Paulista, Department of Animal Science, Jaboticabal, SP, Brazil, <sup>2</sup>Universidade Federal De Campina Grande, Patos, PB, Brazil.

The objective of this study was to determine the net requirements of minerals for the growth and maintenance of intact male F<sub>1</sub> Boer × Saanen goat kids in the initial phase of growth. The following 2 experiments were performed: Exp.1 was performed to determine the net growth requirements of Ca, P, Mg, Na and K of F<sub>1</sub> Boer × Saanen goat kids from 5 to 25 kg of BW; Exp.2 was performed to determine the maintenance requirements for F<sub>1</sub> Boer × Saanen goats from 15 to 25 kg BW. In Exp.1, 32 intact male goat kids were distributed in a completely randomized design and its mineral body composition were fitted by an allometric equation in the form of nonlinear model. To determine the mineral requirements for maintenance of Exp. 2, 21 intact male goat kids were distributed in a randomized-block design, where the goat kids were subjected to 3 levels of feed restriction (0, 30, and 60% feed restriction). At the onset of the Exp.2, 7 goat kids were harvested and used to estimate the initial body composition (15 kg BW). Initial body composition was used to calculate the retention of minerals. The maintenance requirements were estimated by regressions obtained from the retention of minerals in the empty body and the intake of the mineral. The concentration of Ca, P, Na, and K in the empty BW decreased by 11, 13, 26, and 23% with the increase in BW from 5 to 25 kg ( $P < 0.01$ ). As a consequence, our results showed that net requirements of Ca, P, Mg, Na, and K for weight gain decreased by 27.5, 27.8, 4.25, 43.2, and 39.7%, respectively, with the increase in BW from 5 to 25 kg ( $P < 0.01$ ). The net requirements (in g/kg of ADG) decreased from 9.7 to 7.0 for Ca, 6.5 to 4.7 for P, 0.38 to 0.36 for Mg, 0.88 to 0.50 for Na, and 1.9 to 1.2 for K when BW increased from 5 to 25 kg. The daily net requirements for maintenance per kg of BW were 38 mg of Ca, 42 mg of P, 1.6 mg of Mg, 5.0 mg of Na, and 19 mg of K. These results for the nutritional requirements of minerals may help to formulate more balanced diets for F<sub>1</sub> Boer × Saanen goat kids in the initial growth phase.

**Key Words:** body composition, comparative slaughter, crossbred goat

**T507 Analysing the diversity of five Spanish sheep breeds by combining massive genotyping and RNA-seq data.** Antonia Noce<sup>1</sup>, Arianna Manunza<sup>1</sup>, Ángela Cánovas<sup>1</sup>, Silvia Adán<sup>2</sup>, Luis A. Bermejo<sup>3</sup>, Juan Capote<sup>4</sup>, Juan Vicente Delgado<sup>5</sup>, Jordi Jordana<sup>6</sup>, Vincenzo Landi<sup>5</sup>, Agueda Pons<sup>7</sup>, Armand Sánchez<sup>1</sup>, Oriol Vidal<sup>8</sup>, Amparo Martínez<sup>5</sup>, Marcel Amills<sup>1</sup>, Joaquim Casellas<sup>\*6</sup>, <sup>1</sup>Centro de Investigación en Agrigenómica, Bellaterra, Spain, <sup>2</sup>Federación

de Razas Autóctonas de Galicia, Coles, Spain, <sup>3</sup>Universidad de La Laguna, San Cristóbal de la Laguna, Spain, <sup>4</sup>Instituto Canario de Investigaciones Agrarias, San Cristóbal de la Laguna, Spain, <sup>5</sup>Universidad de Córdoba, Córdoba, Spain, <sup>6</sup>Universitat Autònoma de Barcelona, Bellaterra, Spain, <sup>7</sup>Serveis de Millora Agrària i Pesquera, Son Ferriol, Spain, <sup>8</sup>Universitat de Girona, Girona, Spain.

The objective of this research was to characterize the genetic diversity in 5 Spanish sheep breeds from high throughput data. We have used the OvineSNP50 Genotyping BeadChip to genotype 54,241 single nucleotide polymorphisms (SNPs) in 5 endangered Spanish ovine breeds (Xisqueta, Ripollesa, Roja Mallorquina, Canaria de Pelo o Pelibuey and Gallega) with the intention of investigating their genetic relationships and population structure. Multidimensional scaling plot and Admixture analyses revealed that the 2 insular breeds, i.e., Canaria de Pelo (Canary Islands) and Roja Mallorquina (Majorca Island), are the 2 most differentiated populations. African influences and prolonged geographic isolation may partly explain these observations. In contrast, the 3 peninsular breeds showed a weak level differentiation ( $F_{ST} = 0.01-0.03$ ). We have also investigated the variability of the 5 Spanish breeds mentioned above through a RNA-seq approach. More specifically, we sequenced muscle mRNA from pools of 10 individuals by using a HiSeq 2000 platform (Illumina). Our main goal was to find out which percentage of the variation of these populations is breed-specific. Around 300 million reads were obtained per breed, and approximately 70% of them were successfully mapped to the latest version of the sheep reference genome. The total number of SNPs retrieved per breed ranged from 147,262 SNPs (Gallega) to 368,661 SNPs (Xisqueta). Obviously, genotyping experiments in a broader sample of individuals will be necessary to confirm the population-specificity of these SNPs. As a whole, our data suggest that ovine breeds share a substantial amount of variability, probably due to the combined effects of recent divergence and gene flow. However, we have also obtained evidence that thousands of variants appear to segregate specifically in particular breeds, a finding that reinforces the need of making every effort to conserve these unique genetic resources.

**Key Words:** genetic diversity, RNA sequencing, sheep breed

**T508 Body condition as a reference for slaughter of feedlot lambs fed sunflower cake.** Flavio Monção<sup>2</sup>, Euclides Oliveira<sup>1</sup>, Andréia Gabriel<sup>1</sup>, Rodrigo Sousa<sup>1</sup>, Jefferson Gandra<sup>\*1</sup>, Mariana Santos<sup>1</sup>, Lais Moura<sup>1</sup>, Luiz Henrique Silva<sup>1</sup>, Leandro Silva<sup>1</sup>, Loan Silva<sup>1</sup>, Thais Pereira<sup>1</sup>, and Vadim Carbonari<sup>1</sup>, <sup>1</sup>Faculdade de Ciências Agrárias, Universidade Federal da Grande Dourados, Dourados, MS, Brazil, <sup>2</sup>Universidade Estadual Julio de Mesquita, Jaboticabal, SP, Brazil, Jaboticabal, SP, Brazil.

The aim of this study was to evaluate the qualitative and quantitative carcass traits of lambs fed different levels of sunflower cake. For this experiment 28 lambs, crossbred Suffolk, non-castrated males, with an average weight of 21 kg, aging 4 mo were used. The animals were divided into weight classes (4 treatments) and 7 animals per treatment in a randomized block design. The lambs stayed in the feedlot for a period 70 d. The study treatments were: T 1 - Treatment control, 0% of sunflower cake (84.56% DM; 17.45% CP; 60.22% NDF; 1.34% Fat; 56.60% TDN); T 2 - Treatment with 10% sunflower cake; T 3 - Treatment with 20% sunflower cake; T 4 - Treatment with 30% of sunflower cake. The roughage: concentrate ratio of 50:50 was used on the dry matter basis (DM). When the lambs from each treatment reached the body condition in the range of (2.5 to 3.5), the animal was slaughtered and determined the following parameters: SW- slaughter weight; HCY - Hot carcass yield; CCY- Cold carcass yield; ICL- Internal carcass length; SFT- Subcutaneous fat thickness; TEX- Texture; MARB- Marbling.



The statistical analyses were performed by the SAEG program. In this trial the correlation analysis of the variables allowed to verify that SW (slaughter weight) of lambs was not significantly correlated with HCY, CCY, ICL, SFT, COLOR, MARB and AEL. However, it displayed a negative correlation with TEX (texture) of the meat. The HCY was highly correlated with the CCY, in other words, the greater the HCY lower losses occurred during cooling of the carcass, the better CCY. The CCY also showed a positive correlation with TEX and AEL, ICL also showed a positive correlation with the SFT. It was verified that the color and marbling of the meat showed no significant correlations with the other traits. Therefore, it was expected that animals with higher slaughter weight presented positive correlation with marbling, which did not happen in this study that could be justified by the period of confinement of the animals. About the color and texture, no significant difference was noticed. In conclusion, the sunflower cake can be used in supplements for lambs in intensive production systems in partial substitution of soybean meal up to 20%.

**Key Words:** body condition, sunflower cake, lamb

**T509 Performance of Santa Inês meat lambs receiving cactus pear (*Opuntia ficus indica* Mill) in substitution of forage with or without access of water.** José Matias Porto Filho<sup>1</sup>, Leonardo S. Knupp<sup>2</sup>, Antonello Cannas<sup>\*2</sup>, Alberto S. Atzori<sup>2</sup>, Ariosvaldo N. Medeiros<sup>1</sup>, George R. B. Cruz<sup>1</sup>, Genilson B. Silva<sup>1</sup>, and Roberto G. Costa<sup>1</sup>, <sup>1</sup>Programa de Pós Graduação em Ciência e Tecnologia de Alimentos, Universidade Federal da Paraíba, João Pessoa, Paraíba, Brazil, <sup>2</sup>Dipartimento di Agraria, University of Sassari, Sardinia, Italy.

This work aimed to study the performance of Santa Inês meat lambs receiving different levels of inclusion of cactus pear (*Opuntia ficus indica* Mill) in substitution of Tifton (*Cynodon dactylon*) hay. The trial was conducted using 48 Santa Inês male lambs at 180 d of age, with average initial weight of 21.64 ± 0.48 kg. Diets were formulated to be isoproteic and isoenergetic. Animals had ad libitum access to feed. The experiment was carried out as a completely randomized factorial design (3 × 2) with 3 levels of substitution of Tifton hay with cactus pear (30%, 50% and 70%) subdivided in 2 groups, with or without access of water and a control treatment. DMI significantly differed among treatments ( $P < 0.001$ ) and the interaction among levels of cactus and access of water were not significant for all variables ( $P > 0.05$ ). The equations to estimate DMI and the ADG in the diets with cactus pear were:  $DMI = 1776.9 - 26.3x + 96.6y + 0.14x^2 - 2.08xy$  ( $R^2 = 0.86$ );  $ADG = 34.4 + 7.9x - 106.5y - 0.09x^2 - 1.74xy$  ( $R^2 = 0.57$ ), being  $x$  = level of inclusion of cactus pear in the diet (30, 50, 70%) and  $y$  = animals with (1) or without (0) access of water. In contrast, the equations to predict the DMI and the ADG including the control treatment with the group that had access of water were:  $DMI = 1094.3 + 5.3x - 0.19x^2$  ( $R^2 = 0.87$ );  $ADG = 242.3 + 1.77x - 0.047x^2$  ( $R^2 = 0.76$ ), being  $x$  = diet control and levels of cactus (0, 30, 50, 70%). The maximum value of DMI was 1.1 kg/day and the minimum was 0.5 kg/day, obtained for the group receiving 30% of cactus with access of water and 70% with access of water, respectively. The ADG was different among treatments ( $P < 0.05$ ) ranging between 245.1 g to 134.4 g for the group with 30% of cactus in the diet with access of water and those that received 70% of cactus with access of water, respectively. Feed efficiency was affected by treatments, being the group that received 50% of cactus in the diet more efficient (0.27 g). In conclusion, cactus pear is recommended at 30% (with access of water) or 50% (without access of water) of substitution of Tifton hay, resulting in ADG of 253 or 205, respectively.

**Key Words:** cactus, dry matter intake, weight gain

**T510 Effects of oral glycerol dosage on short duration transportation shrink in goats.** Amy L. Bax<sup>\*1</sup>, James D. Caldwell<sup>1</sup>, Taylor N. Drane<sup>1</sup>, Kelsey L. Basinger<sup>2</sup>, Haley L. Bartimus<sup>2</sup>, Jessica K. Clark<sup>2</sup>, Cindy A. DeOrnellis<sup>1</sup>, Jeri D. Rippetto<sup>1</sup>, Abbey J. Kemper<sup>1</sup>, Mikel J. Thompson<sup>1</sup>, Blake E. Koelling<sup>1</sup>, and Bruce C. Shanks<sup>1</sup>, <sup>1</sup>Lincoln University, Jefferson City, MO, <sup>2</sup>University of Arkansas, Fayetteville, AR.

The use of various osmolyte products as a means for increasing blood glucose and mediating effects of shrink have been shown to be effective in different livestock species, but have been examined very little in goats. Therefore, the objective was to evaluate the effects of oral glycerol dosage on short duration transportation shrink in goats. On January 19, 2015, a total of 27 (23.4 ± 0.4 kg BW) intact and castrated male goat kids were stratified by sex and BW, and allocated randomly to 1 of 3 treatments: 1) control (C; n = 9); 2) 1 mL oral dose of glycerol per 0.45 kg of BW (n = 9); or 3) 2 mL oral dose of glycerol per 0.45 kg of BW (n = 9). Goats had ad libitum access to mixed-grass pasture and water until initiation of the study. At 0830, goat kids were gathered, bled, weighed, and dosed with glycerol according to treatment. Goats were then transported in a livestock trailer for approximately 4.5 h with an average ambient temperature of 12°C. After transport, goats were unloaded, bled, re-weighed, and returned to their pasture for the night. After a 24-h recovery time, goats were bled and weighed a final time. Data were analyzed using PROC MIXED of SAS with 2 preplanned orthogonal contrast statements: (1) the mean of control vs the mean of 1 mL and 2 mL, and (2) the mean of 1 mL vs 2 mL. Beginning weight, final weight, 24-h recovery weight, shrink, and recovery weight change did not differ ( $P \geq 0.31$ ) across treatments. Beginning white blood cell, final monocyte, basophil, and platelet, and 24-h monocyte counts were higher ( $P \leq 0.05$ ) and beginning neutrophil and eosinophil, and final red blood cell and white blood cell counts and mean corpuscular volumes tended ( $P \leq 0.10$ ) to be higher for 2 mL vs 1 mL. The 24-h neutrophil counts were higher ( $P = 0.05$ ) and 24-h lymphocyte counts and white blood cell counts change tended ( $P \leq 0.08$ ) to be higher for C vs 1 mL and 2 mL. There were no differences ( $P \geq 0.13$ ) in the other blood measurements. Therefore, oral glycerol dosage may affect some blood measurements, but ultimately kid goats in this study did not shrink; thus, the effects of this treatment on short duration transportation shrink in goats were undetermined.

**Key Words:** glycerol, goat, shrink

**T511 Effects of breed and resistance classification of sire on progeny growth performance and response to artificial infection with *Haemonchus contortus* in a central performance test.** Yoko Tsukahara<sup>\*1</sup>, Terry A. Gipson<sup>1</sup>, Steven P. Hart<sup>1</sup>, Lionel J. Dawson<sup>1,2</sup>, Zaisen Wang<sup>1</sup>, Ryszard Puchala<sup>1</sup>, Tilahun Sahlu<sup>1</sup>, and Arthur L. Goetsch<sup>1</sup>, <sup>1</sup>American Institute for Goat Research, Langston University, Langston, OK, <sup>2</sup>Center of Veterinary Health Sciences, Oklahoma State University, Stillwater, OK.

Fifteen Dorper (D; 3.8 mo of age, 29 kg), 14 St. Croix (C; 3.9 mo, 18 kg), 14 Kiko (K; 4.0 mo, 19 kg), 13 Boer (B; 3.2 mo, 22 kg), and 17 Spanish (S; 3.1 mo, 18 kg) males were used to investigate effects of classification for resistance to *Haemonchus contortus* of sire and among and within breed differences in the second year of a central test at Langston University (LU) for growth performance and response to artificial infection with infective larvae. In the first year of the test, males were randomly selected from 4 commercial farms in KS, MO, and OK and LU B and S goat herds. Animals used in this study were progeny of the sires (i.e., High and Moderate, with no progeny of susceptible males) selected in the first year. For both years, the test entailed an adjustment period of

2 wk followed by 8 wk of data collection. Animal groups were housed separately in adjacent pens with automated feeders allowing free-choice access to a 15% CP (DM) and 50% concentrate pelletized diet. During adaptation, anthelmintic treatment resulted in low fecal egg count (FEC; < 600/g), after which 10,000 larvae were administered orally. Packed cell volume (PCV) was measured weekly and FEC was determined 4 times in wk 6–8. For analysis, initial BW, PCV, and FEC were covariates, and the logarithmic transformation  $\ln(x + 2,000)$  was used for mean FEC. Breed affected ( $P \leq 0.01$ ) ADG (307, 286, 159, 247, and 142 g; SEM = 13.8), DMI (2.2, 1.6, 1.3, 1.5, and 1.3 kg; SEM = 0.12), FEC (2,098, 1,278, 1,419, 1,335, and 716 eggs/g, original scale; SEM = 80.9), and PCV (27.2, 31.7, 31.6, 28.1, and 25.6%; SEM = 0.76 for D, C, K, B, and S, respectively). Means of resistance classification of sires were similar ( $P > 0.10$ ) for FEC, PCV, ADG, and DMI. Correlation coefficients of sire and progeny FEC within breed were nonsignificant ( $P > 0.10$ ). In conclusion, with only one generation of selection, there was no detectable relationship in resistance to internal parasite between selected sires and progeny based on FEC after an artificial challenge with *H. contortus* larvae in a standardized environment.

**Key Words:** goat, internal parasitism, sheep

**T512 Growth performance and resistance to internal parasitism of small ruminant males from the south-central US in a centralized test.** Yoko Tsukahara\*<sup>1</sup>, Terry A. Gipson<sup>1</sup>, Steven P. Hart<sup>1</sup>, Lionel J. Dawson<sup>1,2</sup>, Zaisen Wang<sup>1</sup>, Ryszard Puchala<sup>1</sup>, Tilahun Sahlu<sup>1</sup>, and Arthur L. Goetsch<sup>1</sup>, <sup>1</sup>American Institute for Goat Research, Langston University, Langston, OK, <sup>2</sup>Center of Veterinary Health Sciences, Oklahoma State University, Stillwater, OK.

Various breeds of young male sheep and goats from commercial farms in Arkansas, Kansas, Missouri, and Oklahoma and of Langston University (LU) were used in a centralized test at LU, which included artificial infection with *Haemonchus contortus*, to investigate growth performance and genetic resistance to internal parasitism. Year 1 included 2 Katahdin flocks (KS-A, n = 17, 3.5 mo of age, 35 kg; KS-B, 18, 4.0 mo, 19 kg), 20 Dorper (DS; 8.2 mo, 45 kg), 13 St. Croix (CS; 4.4 mo, 21 kg), 2 Boer herds (BG-A; 16, 3.8 mo, 21 kg; BG-B, 17, 19 kg) 16 Kiko (KG; 3.1 mo, 20 kg), and 14 Spanish (SG; 4.4 mo, 19 kg). In year 2, animals were progeny from breeding groups classified in year 1 as of high and moderate resistance, with 15 DS (3.8 mo, 29 kg), 14 CS (3.9 mo, 18 kg), 14 KG (4.0 mo, 19 kg), 13 BG-A (3.2 mo, 22 kg), and 17 SG (3.1 mo, 18 kg). There was 2 wk for adaptation and an 8-wk test period, with automated feeders allowing free-choice access to a 50% concentrate pelletized diet. During adaptation, anthelmintic treatment resulted in low fecal egg count (FEC; < 600/g), after which 10,000 infective larvae were administered orally. Packed cell volume (PCV) was measured weekly and FEC was determined 4 times in wk 6 to 8. Breed affected ( $P \leq 0.01$ ) FEC in year 1 (1,512, 2,196, 3,072, 1,229, 1,069, 2,713, 3,575, and 1,182 eggs/g for KS-A, KS-B, DS, CS, BG-A, BG-B, KG, and SG, respectively; SE = 100.0) and year 2 (2,621, 1,368, 1,413, 1,669, and 884 eggs/g for DS, CS, BG-A, KG, and SG, respectively; SEM = 48.1). Animals were placed in 3 categories of resistance (i.e., high, moderate, low) within flocks/herds based primarily on FEC but also considering residual feed intake and ADG using cubic clustering criterion. Resistance category means were similar ( $P > 0.05$ ) for ADG and ADG:DMI in both years. In conclusion, based on FEC after an artificial challenge with *H. contortus* larvae in a standardized environment, there was considerable variability among flocks/herds of small ruminants in resistance to internal parasitism due to multiple factors such as species, breed, and genetic differences within breed.

**Key Words:** goat, internal parasitism, sheep

**T513 The *FecGE* allele of the ovine *GDF9* gene in the Pelibuey breed in México and its effects on prolificacy.** Felipe A. Rodríguez-Almeida\*, Claudia P. Pérez-Camacho, María E. Burrola-Barraza, and Joel Domínguez-Viveros, *Facultad de Zootecnia y Ecología, Universidad Autónoma de Chihuahua, Chihuahua, México.*

The *FecGE* allele of the ovine *GDF9* gene, reported previously in the Santa Inés breed in Brazil, is segregating in Pelibuey, another hair type sheep breed in México. To determine allelic and genotypic frequencies, as well as effects on prolificacy and potential for marker assisted selection, 160 Pelibuey ewes from 4 flocks in different regions of México [Jalisco 1 (n = 34), Jalisco 2 (n = 62), Tabasco (n = 30) and Chihuahua (n = 34)] were genotyped by RT-PCR (StepOnePlus) using the FAMACTTCAAACAGTTTCTTTTTMGBNFQ and VICTCAAA-CAGTGTCTTTTTMGBNFQ probes (Applied Biosystems). Pearson's Ji-squared was used to test genotypic frequencies for Hardy-Weinberg equilibrium. After discarding data of lambings that resulted from synchronized estrous with hormones, 631 records of the Poisson variable prolificacy were analyzed using a generalized linear model with PROC GLIMMIX of SAS (SAS Institute Inc., Cary, NC.). The model included fixed effects of year, season, flock, genotype, 2-way interactions and age as a covariate, as well as the random effect of ewe within flock and genotype. The average frequency of the *FecGE* allele was 0.48 (0.48 to 0.54 for the flocks in Jalisco and Chihuahua, and 0.35 for the flock in Tabasco, which is in most challenging environment). Genotypic frequencies were as expected under Hardy-Weinberg equilibrium ( $P > 0.10$ ). Back transformed estimated means for litter size for ewes of the different genotypes were 1.43 for +/+, 1.73 for +/E, and 1.78 for E/E ( $P < 0.05$ ). For genotypes +/+, +/E, and E/E, frequencies of litters with more than 2 lambs were 0.5, 11.4, and 15.2%, respectively; frequencies of litters with a single lamb were 56.0, 37.6, and 35.6%, respectively. Homozygous E/E ewes reproduced normally, in agreement with results from Brazil, but in contrast to the sterility of homozygous ewes reported for other *GDF9* mutations in European breeds. Potential for marker assisted selection exists, especially to produce homozygous rams to be used in low-prolificacy commercial flocks and(or) in crossbreeding with low-prolificacy well-adapted maternal breeds under harsh environments to obtain F<sub>1</sub> ewes.

**Key Words:** fecundity gene, hair sheep, litter size

**T514 Methane concentration and degradation profile of broom sorghum based-diets for sheep.** M. A. Cerrillo-Soto<sup>1</sup>, A. L. Abdalla<sup>2</sup>, R. C. Lucas<sup>2</sup>, A. Estrada-Angulo<sup>3</sup>, F. G. Rios-Rincón<sup>3</sup>, and M. Guerrero-Cervantes\*<sup>1</sup>, <sup>1</sup>Universidad Juárez del Estado de Durango, Durango, Dgo, México, <sup>2</sup>Centro de Energía Nuclear na Agricultura, Piracicaba, São Paulo, Brasil, <sup>3</sup>Universidad Autónoma de Sinaloa, Culiacán, Sin., México.

Broom sorghum (BS) represents a suitable alternative in sheep nutrition practices in Northwest Mexico. Moreover, concerns on rumen methane production support the search for feeds to minimize environmental impacts. This in vitro assay evaluated rumen methanogenesis and degradability characteristics of mixed sheep diets with increasing levels (0, 20, 40, 60 and 80%) of broom sorghum substituting sorghum grain. Diets included soybean meal, Sudan and molasses as well. A semi-automated system for gas production (GP) using a pressure transducer was used. Ground samples (500 mg DM) were weighed into filter bags and further placed into 160-mL glass bottles. A mixture of incubation medium- rumen inoculum was added to the bottles and sealed. Nine Santa Inés cannulated sheep fed tropical grass were used as donors. Head space gas pressure was measured at 4, 8, 12 and 24 h. The CH<sub>4</sub> concentration was determined using gas chromatography. The truly

degraded organic matter (TDOM) was estimated after 24 h incubation by refluxing the filter bags with NDF solution. The partitioning factor was calculated as the ratio of TDOM (mg) and gas volume (mL). Data were analyzed using ANOVA (GLM, SAS). Values for CP, NDF and ADF of the diets varied ( $P < 0.05$ ) from 11 to 13%, 54 to 66% and 16 to 39%, respectively. Gas production (GP) varied among treatments ( $P < 0.05$ ). Increasing levels of BS promoted a reduction in GP from 171 mL/g DM in the 0% (control) to 125 mL/g DM in the 80% treatment. Methane production calculated per unit of TDOM varied from 9.4 to 7.5 mL/g TDOM, although no differences were detected ( $P > 0.05$ ). Nonetheless, methane production calculated per unit of degraded NDF resulted in significant variations ( $P < 0.05$ ). Treatment containing no broom sorghum (0%) resulted in the highest methane production (4.7 mL/g DNDF), whereas 60% treatment showed the lowest (1.9 mL/g DNDF). No differences were detected in TDOM among treatments (mean = 351 g/kg DDM). Similarly, no effect was found in the partitioning factor (mean = 1.24). Results suggested that increasing levels of BS did not affect OM digestibility, although that might promote a reduction in methane concentration. However, further studies are to be performed to elucidate stronger effects.

**Key Words:** sheep, methane concentration, broom sorghum

**T515 Effect of timing of PGF<sub>2α</sub> administration in a short-term progesterone-based estrous synchronization protocol on fertility in ewes.** Callayn D. Paul\*, Erin N. Greenleaf, Adam K. Redhead, Abiodun E. Adebisi, and Marlon Knights, *West Virginia University, Morgantown, WV.*

Traditionally, prostaglandin F<sub>2α</sub> (PGF<sub>2α</sub>) has been included in short-term progesterone-based estrous synchronization (STPBES) protocols but its inclusion has been associated with a reduction in fertility at the synchronized estrus. To determine if there is an effect of timing of prostaglandin administration relative to the progesterone pre-treatment on fertility, ewes ( $n = 442$ ) on 4 farms located in WV and PA were randomly assigned to receive controlled internal drug-releasing devices (CIDR-g, 0.3 g progesterone) for 5 d alone ( $n = 123$ ; treatment 4), in combination with PGF<sub>2α</sub> (5 mL Lutalyse; 25 mg Dinoprost) at CIDR insertion ( $n = 103$ ; treatment 1) or removal ( $n = 100$ ; treatment 2), or 25 mg PGF<sub>2α</sub> alone ( $n = 116$ ; treatment 3) before being joined with sexually mature rams. Data were analyzed using ANOVA with the model consisting of the main effects of treatments, farms and their interactions and additionally, least squares means for treatment effects were determined. Ewes receiving only PGF<sub>2α</sub> had a lower estrous response than other treatments ( $58.7 \pm 3.7\%$  vs.  $74.6 \pm 37.6\%$ ;  $P < 0.0001$ ) but tended to have a higher conception rate than ewes receiving combination treatments of PGF at insertion ( $78.8 \pm 8.9\%$  vs.  $61.2 \pm 4.9\%$ ;  $P = 0.08$ ) or removal ( $78.8 \pm 8.9\%$  vs.  $59.8 \pm 5.0\%$ ;  $P = 0.06$ ). PGF<sub>2α</sub> ewes also had a significantly lower pregnancy rate to 1st service than ewes receiving the combination treatment with PGF<sub>2α</sub> at insertion ( $41.6 \pm 4.4\%$  vs.  $54.1 \pm 4.5\%$ ;  $P < 0.05$ ) and ewes receiving CIDR only ( $41.6 \pm 4.4\%$  vs.  $58.5 \pm 4.1\%$ ;  $P < 0.005$ ). There were no effects of treatment on any other measures of reproductive performance. In conclusion, inclusion of PGF<sub>2α</sub> at the beginning or end of progesterone pretreatment did not enhance synchrony of estrus or other reproductive outcomes. Furthermore, synchronization of estrus with a 5 d treatment of progesterone was sufficient to synchronize estrus with high fertility in ewes.

**Key Words:** progesterone, prostaglandin, fertility

**T516 Method of zilpaterol hydrochloride supplementation on meat quality of feedlot lambs.** Horacio Davila-Ramos\*, Karla

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Zilpaterol hydrochloride (ZH) is a  $\beta$ -adrenergic agonist was approved in México (2002) and the United States (2006) to promote growth and carcass dressing in beef cattle. Forty Dorper  $\times$  Katahdin ( $45.4 \pm 4.5$  kg) crossbred male lambs were used in a 27-d feeding trial (5 pens/treatment) to evaluate the influence of method of zilpaterol supplementation on growth performance and carcass characteristics. Lambs were fed twice daily with a cracked corn based (64%) experimental diet (1.39 Mcal/kg of NE<sub>g</sub> and 14.0% CP). The animals were allotted to individual pens (6 m<sup>2</sup>) with full shade and ad libitum water. Treatments were (1) control (CON), no zilpaterol supplementation; (2) total dose of zilpaterol was offered only in morning feeding (AM), (3) total dose of zilpaterol was offered in morning and afternoon feeding (PPM), and (4) total dose of zilpaterol consisted in offering one day and suspended the next day, like intermittent supplementation (INT). Zilpaterol was supplemented at a rate of 0.20 mg/kg of live weight d<sup>-1</sup> (as zilpaterol chlorhydrate, Zilmax). The lambs were slaughtered in a local slaughterhouse. The data were analyzed at complete randomized design, with orthogonal contrast and means compare with LSD. Zilpaterol supplementation (CON vs AM, PPM) decreased the redness index ( $a^*$ ) (10.9%;  $P < 0.04$ ), yellowness index ( $b^*$ ) (15.5%;  $P < 0.03$ ) and chromaticity (13.6%;  $P < 0.02$ ). Shear force values of zilpaterol supplementation tended to be greater ( $P = 0.08$ ) than shear force values of CON. There were not differences on pH values between treatments. Zilpaterol intermittent supplementation (CON vs INT) reduced luminosity ( $L^*$ ) (10.3%;  $P = 0.05$ ), no statistical difference was found with the remaining variables ( $P \geq 0.12$ ), similarly, the contrast (INT vs. AM) showed no significant differences ( $P \geq 0.33$ ) in all variables evaluated.

**Key Words:** zilpaterol supplementation, lamb, meat quality

**T517 Effect of weaning on rapid rebreeding in Katahdin ewes.** Erin N. Greenleaf\*, Callayn D. Paul, Abiodun E. Adebisi, Kyle J. Powell, Adam K. Redhead, and Marlon Knights, *West Virginia University, Morgantown, WV.*

To increase the frequency of lambing, ewes must be mated during seasonal anestrus and while lactating. The present study investigated the effect of lactation and season on the fertility of Katahdin ewes rebred while lactating. Fall-lambing ewes ( $n = 57$ , 2 mo postpartum, experiment 1) and spring-lambing ewes ( $n = 40$ , 3 mo postpartum, experiment 2) were randomly assigned within birth type to be weaned approximately 3 weeks before breeding or to continue to suckle their lambs for an additional 3 mo. In experiment 1, all ewes were treated with a controlled internal drug releasing (CIDR-g; 0.3 g progesterone) device for 5 d and introduced to rams. In experiment 2, half of the ewes in each lactation status group were treated with an estrus induction protocol consisting of a pre-treatment with a CIDR device for 5 d and an injection with 3 mL P.G. 600 (240IU eCG and 120 IU hCG) at insert removal or received no further treatment. In experiment 1, the mean estrous response, conception rate, pregnancy rate to the first and second service period, proportion of ewes lambing and prolificacy was 64.9, 89.2, 57.9, 91.2, 67.3 and 129% respectively and was not affected by lactation status. In experiment 2, estrous response tended to be higher in suckling compared with weaned ewes (50 vs 11.1%,  $P = 0.08$ ) but lactation status did not affect any of the other variables measured. Ewes receiving the estrous induction protocol had a higher ( $P < 0.01$ ) estrous response, conception rate, pregnancy rate to the first and second service period, proportion of ewe lambing to the first service period and overall and lambing rate (84.2 vs 33.3%; 81.3 vs 0; 80 vs 0; 94 vs 23.8; 61.1 vs 0; 72.2 vs 30 and



111.1 vs 40%, respectively). Lambs suckling their dams gained more weight and grew faster (19.6 vs 14.1 kg;  $13.1 \pm 0.68$  vs  $11.3 \pm 0.85$  kg and  $183 \pm 0.6$  vs  $131 \pm 0.6$  g/d;  $128.2 \pm 0.7$  vs  $110.4 \pm 0.8$  g/d, ( $P < 0.001$  and  $P < 0.1$  for experiments 1 and 2, respectively). In conclusion, weaning depressed growth of lambs and did not improve the ability of Katahdin ewes to rebreed in the postpartum period. However, treatment with an estrous induction protocol is necessary for early rebreeding of Katahdin ewes during seasonal anestrus.

**Key Words:** Katahdin, postpartum period, estrous

**T518 Effect of supplementation with propylene glycol and lactation period on energy metabolism of lactating ewes.** Simone Pedro da Silva\*<sup>1</sup>, Gilberto de Lima Macedo Junior<sup>2</sup>, Rogério Pereira dos Santos<sup>3</sup>, Jhone Talisson Lira de Sousa<sup>3</sup>, Marina Elizabeth Barbosa de Andrade<sup>4</sup>, Érica Beatriz Schultz<sup>2</sup>, Luciano Fernandes de Sousa<sup>3</sup>, Adriano Santana Crozara<sup>2</sup>, and Nhayandra Christina Dias e Silva<sup>4</sup>, <sup>1</sup>Instituto Federal Goiano, Hidrolândia, Goiás, Brazil, <sup>2</sup>Universidade Federal de Uberlândia, Uberlândia, Minas Gerais, Brazil, <sup>3</sup>Universidade Federal do Tocantins, Araguaína, Tocantins, Brazil, <sup>4</sup>Universidade Estadual Paulista, Jaboticabal, São Paulo, Brazil.

The objective of this study was to evaluate the effect of supplementation with propylene glycol (PG) in water and the lactation period on energy metabolism of 19 lactating ewes Santa Ines with initial body weight (BW)  $58.67 \pm 0.80$  kg, from calving to weaning (0–90 d), organized into 4 treatments (0.0, 1.5, 3.0 and 4.5% PG) in a completely randomized in a split plot design. The experiment was conducted at sheep sector in Federal University of Uberlândia, Minas Gerais, Brazil. The animals were fed a total mixed ration consisting relation roughage concentrate 67:32. Blood samples were collected every 7 d, totaling 12 blood collections. In these samples we evaluated triglycerides, cholesterol, very low density lipoprotein (VLDL), high density lipoprotein (HDL), low density lipoprotein (LDL), LDL/HDL,  $\beta$ -hydroxybutyrate and enzyme gamma glutamyl transferase (GGT). The supplementation with propylene glycol not affected the levels serum of triglycerides, VLDL, LDL, and LDL/HDL in the blood ( $P > 0.05$ ). The triglycerides and VLDL levels in the blood showed a linear decrease with the advancing lactation ( $P < 0.01$ ). The cholesterol and HDL levels in the blood showed quadratic response with maximum cholesterol concentration at 61, 70, 68 e 53 d of lactation and maximum HDL concentration at 58, 86, 58 e 85 d of lactation according with supplementation of 0.0, 1.5, 3.0 and 4.5% propylene glycol, respectively. The  $\beta$ -hydroxybutyrate level showed a linear decrease with the increase of propylene glycol ( $P < 0.05$ ). There was no effect of lactation period on the levels serum  $\beta$  hydroxybutyrate on the blood ( $P > 0.05$ ). The level of enzyme gamma glutamyl transferase (GGT) in the blood was not affected by the inclusion of propylene glycol ( $P > 0.05$ ), but was affected by lactation period ( $P < 0.01$ ). With 36 d of lactation, sheep showed the maximum concentration of GGT (80.36 IU/L). Supplementation with propylene glycol improved the energy status of the sheep and, thus, caused a reduction in the levels of ketone bodies. We concluded that supplementation with propylene glycol reduces the adverse effects of negative energetic balance in sheep during the lactation.

**Key Words:** energetic balance, metabolic profile

**T519 Crude glycerin added into low-starch diets improved fatty acid profile of lamb meat.** V. B. Carvalho\*, J. M. B. Ezequiel, R. F. Leite, M. T. C. Almeida, J. R. Paschoaloto, H. L. Perez, E. A. Oliveira, A. C. Homem Junior, E. B. Carvalho, and E. S. Castro

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The objective was to evaluate replacement of corn with crude glycerin (CG) in high-concentrate, low starch diets on fatty acid profile of meat. Forty Santa Ines lambs ( $23.5 \pm 1.35$  kg BW) were assigned to a randomized block design with 5 treatments: 0, 7.5, 15, 22.5, and 30% of CG (DM basis). The diet with 30% of CG promoted total replacement of corn. The diets consisted of Tifton-85 hay, corn, CG (83% glycerol), corn gluten meal, corn oil, urea, sunflower meal, soybean hulls and mineral. All diets had around 18% roughage and 82% concentrate. The animals were housed in individual pens and fed ad libitum twice daily at 0700 and 1600 h. The experimental period had  $72 \pm 7$  d of duration and animal harvest was performed when the animals had reached 38 kg BW. Orthogonal contrasts were used to determine the linear and quadratic effects of CG and also contrast of 0% CG vs. CG treatments was used. Odd-chain fatty acids (C15:0, C17:0 and C17:1), oleic (C18:1), palmitoleic (C16:1), total monounsaturated, total unsaturated fatty acids, and their ratios to saturated fatty acids increased linearly within increasing CG. The concentration of all these fatty acids and the ratios, except C16:1, were greater in animals fed any concentration of CG compared with animals fed without CG ( $P < 0.01$ ). The CLA tended to increase in glycerin-fed lambs ( $P = 0.06$ ). The CG decreased linearly the stearic (C18:0), palmitic (C16:0), transvaccenic (C18:1 t11) and total saturated fatty acids ( $P < 0.01$ ). The myristic acid (C14:0) tended to decrease quadratically with increasing CG ( $P = 0.06$ ). The concentration of C18:0, C16:0, C14:0 and total saturated fatty acids decreased when CG was added, regardless of concentration ( $P \leq 0.02$ ). The C18:1 t11 tended to be present in greater concentrations in animals fed without CG compared with animals fed CG ( $P = 0.09$ ). Crude glycerin can be a viable alternative as an energy source in the diet of lambs fully replacing corn into low-starch diets, providing meat with healthier fatty acid profile.

**Key Words:** corn, glycerol, high-concentrate

**T520 Detection of gene expression and location of receptors activated by the oral administration of lithium chloride for conditioned taste aversion in sheep.** Katariina Vara<sup>1</sup>, Ahmed K. K. Salama<sup>1,2</sup>, Carmen L. Manuelian<sup>1</sup>, Maristela Rovai\*<sup>1</sup>, Juan J. Lóor<sup>3</sup>, Elena Albanell<sup>1</sup>, Xavier Such<sup>1</sup>, and Gerardo Caja<sup>1</sup>, <sup>1</sup>Group of Ruminant Research (G2R), Universitat Autònoma de Barcelona, Bellaterra, Spain, <sup>2</sup>Animal Production Research Institute, Dokki, Giza, Egypt, <sup>3</sup>Department Animal Science, University of Illinois, Urbana, IL.

The emetic system activation (nausea and vomiting) plays a key role in the negative postingestive feedback. Lithium chloride (LiCl) is a nonlethal gastrointestinal drug that stimulates the chemoreceptor trigger zone, activating the emetic center and generating conditioned taste aversion (CTA) without other side-effects. There is scarce information on the pathways involved in the CTA mechanism by using LiCl. The aim of this study was to determine changes in gene expression of 4 genes (*CNRI*, *GCG*, *GLPIR*, *HTR3A*) involved in food intake and nausea. A total of 9 Manchega dry ewes, orally dosed with 225 mg LiCl/kg BW, were allocated into 3 euthanasia groups (0, 12 and 24 h after LiCl administration). Brain (area postrema), distal small intestine and colon tissue samples were collected, snap-frozen in liquid nitrogen, and stored at  $-80^{\circ}\text{C}$  until analysis. The total RNA was extracted and purified using miRNeasy Mini Kit, and concentration and integrity determined by NonoDrop ND-1000 Spectrophotometer and Agilent 2100 Bioanalyzer, respectively. Gene expressions were determined by RT-qPCR with designed primers and 3 internal control genes. Data were normalized using the geometric mean of internal control genes,

and expression values calculated using base-10 logarithm. As results, expressions of CNR1 and GLP1R were detected in all 3 tissues, and GCG was only detected in the distal small intestine and colon. The overall expressions of CNR1 and GLP1R differed by time and tissue, whereas GCG differed by time points. The CNR1 expression showed a linear regression in distal small intestine (decrease) and colon (increase) when comparing expression levels before and after Li administration. The GCG expression increased in distal small intestine and GLP1 expression decreased in colon 24 h after euthanasia. In conclusion, LiCl administration for a CTA treatment revealed changes in the expression of the genes *CNR1*, *GCG* and *GLP1R* in brain, distal small intestine and colon. Acknowledgments: Spanish Plan Nacional I+D+I (Project AGL2010-22178-CO2-01).

**Key Words:** *CNR1*, *GCG*, food aversion

**T521 Conditioned taste aversion generalization by aroma in sheep.** Carmen L. Manuelian<sup>1</sup>, Elena Albanell<sup>1</sup>, Maristela Rovai<sup>\*1</sup>, Ahmed K. K. Salama<sup>1,2</sup>, and Gerardo Caja<sup>1</sup>, <sup>1</sup>Group of Ruminant Research (G2R), Universitat Autònoma de Barcelona, Bellaterra, Barcelona, Spain, <sup>2</sup>Animal Production Research Institute, Dokki, Giza, Egypt.

Neophobia is an innate protective mechanism that allows animals to learn from postingestive consequences of eating a new and potentially toxic feed before being harmed by it. Small ruminants usually begin to eat an edible feed that they have not previously encountered by ingesting small amounts; thereafter, if there is no negative feedback, they will gradually increase the intake. On the other hand, if there is a negative feedback, they will associated feed characteristics (taste, odor, texture and sight) with the gastrointestinal discomfort and develop rejection for that feed (food aversion, AV). Despite odor, taste and flavor have been used to increase feed intake; however, few studies have examined its effect in AV. The AV occurs to both the taste and odor of a food (flavor); however, if odor is conditioned in compound with taste, potentiation of the odor will occur. Therefore, when the odor is tested by itself, it becomes a highly potent cue for AV. The aim of this study was to evaluate the ability of 2 breeds of sheep (Manchega, n = 10; Lacaune, n = 10) to generalize the induced AV against a concentrate with strawberry odor to barley and grass flavored with the same aroma. The odor was selected after a preliminary test of acceptability between chocolate and strawberry odors with no taste cues. Ewes were allocated into 2 groups/ breed and treatment consisted in: Control (C, water) and Aversion (AV, 225 mg LiCl/kg BW). For AV induction, 100 g of concentrated with strawberry odor was offered individually during 5 min and thereafter orally administered the treatment (water or LiCl). The AV induction lasted 3 d, and redosing when necessary (intake > 75 g). On d 6, 100 g of concentrated, barley and grass with strawberry odor were offered individually during 5 min each food. The 80% of the animals needed  $\geq 2$  LiCl doses to establish the AV. No differences between breeds were observed in the AV behavior. Generalization of the AV toward barley and grass with strawberry odor was not observed. In conclusion, the use of a flavor instead of an odor could be more effective in the AV generalization. Acknowledgments: Spanish Plan Nacional I+D+I (Project AGL2010-22178-CO2-01) and Lucta (Montornés del Valles, Spain).

**Key Words:** lithium chloride, flavor, neophobia.

**T522 Microbial population and in vitro gas production of sheep fed diets with starch and neutral detergent-soluble fiber.** Josemir S. Gonçalves<sup>\*1</sup>, Jane M. B. Ezequiel<sup>1</sup>, Eric H. C. B. Van Cleef<sup>1</sup>, Antonio C. Homem Junior<sup>2</sup>, and Raquel L. Salgado<sup>3</sup>, <sup>1</sup>UNESP

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Six ruminally cannulated male sheep (Santa Ines  $\times$  Dorper;  $35 \pm 0.9$  kg BW) were used to evaluate the effects of diets containing different levels of starch and neutral detergent-soluble fiber on protozoa and bacterial fractions, as well as methane and carbonic gases productions. Lambs were assigned to a replicated  $3 \times 3$  Latin square design. Each experimental period consisted of 20 d adaptation and 1 d sampling. Diets consisted of 30% corn silage and 70% concentrate (corn grain, citrus pulp, sunflower meal, soybean hulls, sunflower oil, urea and minerals), providing one diet with high starch content (26.7%), one with same content of starch and neutral detergent-soluble fiber (18.4%) and one with high neutral detergent-soluble fiber (24.3%). Ruminal content were sampled on d 21 of each experimental period, at 1.5; 6.5 and 11.5 h after the morning feeding. Samples were strained through a nylon filter to separate liquid and solid phases. Liquid-associated protozoa (LAP) and bacteria (LAB) were obtained using series of filtrations, dilution with salt solution and centrifugations. Particle-associated bacteria (PAB) were obtained using agitation with a salt solution, as well as filtrations and centrifugations. Data were analyzed using the MIXED procedure of SAS, with repeated measures. In vitro fermentation was performed using 250-mL flasks in a completely randomized design with 3 treatments and 6 replicates to evaluate the methane and carbonic gasses production. Samples of diets were added at 1.25 g DM/flask, and consisted of same diets fed to ruminal fluid donor. Data were analyzed using the GLM procedure of SAS. There were no effect ( $P > 0.05$ ) of diets on LAP (980.4 mg/kg DM), LAB (858.9 mg/kg DM) or PAB (1237.7 mg/kg DM). The methane production was unaffected by diets (7.1 L/kg DM;  $P > 0.05$ ) but the high-starch diet decreased carbonic gas production ( $P < 0.05$ ). The starch or neutral detergent-soluble fiber diets do not affect ruminal microbial populations nor methane production.

**Key Words:** bacteria, fermentation, methane

**T523 Effects of replacement of soybean meal with handmade fish meal on productive performance of Pelibuey ewes and their suckling kids.** Jose L. Loya-Olguin<sup>\*1</sup>, Yissel S. Valdes-Garcia<sup>1</sup>, Lilia E. Nuñez-Gonzalez<sup>1</sup>, Alberto Barreras<sup>2</sup>, Alejandro Plascencia<sup>2</sup>, Francisco Escalera-Valente<sup>1</sup>, and Alejandro A. Gomez-Danes<sup>1</sup>, <sup>1</sup>Posgrado en Ciencias Biológico Agropecuarias/Unidad Académica de Medicina Veterinaria y Zootecnia de Universidad Autónoma de Nayarit, Tepic, Nayarit, Mexico, <sup>2</sup>Instituto de Investigaciones en Ciencias Veterinarias, Universidad Autónoma de Baja California, Mexicali, Baja California, Mexico.

Thirteen Pelibuey ewes ( $38 \pm 7$  kg) were used to evaluate the effect of replacing soybean meal (SBM) with a handmade product of fish (HMF). The experimental period lasted 6 weeks which comprised the last 3 weeks of gestation, where the change of body weight in pregnancy was individually evaluated, and the first 3 weeks of lactation, where changes in body weight, feed intake and milk production and composition were evaluated in sheep, and changes in body weight was evaluated on 22 of their kids (initial wt =  $2.52 \pm 0.05$  kg). Control diet and the test diet used in both phases were formulated to be isonitrogenous and isocaloric. The level of inclusion of SBM and HFM in diets was 7 and 3.5%, respectively. HFM was elaborated with marine fish by-products (whole fish, heads, tails, and backbone). The chemical composition of HMF averaged:  $50.8 \pm 7.7\%$  CP, fat  $9.4 \pm 0.8$  and  $22.7 \pm 5.9\%$  ash. There were no differences on DM intake, BW changes and milk production;

however, as a result of an increase ( $<0.01$ ) in the percentage of protein in milk with HFM treatment, was obtained greater final weight (19%,  $P < 0.01$ ) of their suckling kids. It was concluded that HFM is a suitable substitute of SBM in gestating and lactating ewes. Replacing of SBM with HFM did not alter DM intake and milk performance in ewes. However, changes in the concentration of protein in milk in ewes that received HFM may be reflected in a greater weight gain of lambs in the first 21 d of age.

**Key Words:** lactating ewes, soybean meal, fish

**T524 Ruminal kinetics on sheep fed diets with starch and neutral detergent-soluble fiber.** Josemir S. Gonçalves<sup>\*1</sup>, Jane M. B. Ezequiel<sup>1</sup>, Eric H. C. B. Van Cleef<sup>1</sup>, Antonio C. Homem Junior<sup>2</sup>, and Raquel L. Salgado<sup>3</sup>, <sup>1</sup>UNESP - Univ Estadual Paulista, Faculdade de Ciências Agrárias e Veterinárias, Jaboticabal, São Paulo, Brazil, <sup>2</sup>UNESP-Univ Estadual Paulista, Faculdade de Engenharia, Ilha Solteira, São Paulo, Brazil, <sup>3</sup>UFERSA-Federal University of Semi-arid, Mossoró, Rio Grande do Norte, Brazil.

The objective of this study was to evaluate the effects of different soluble carbohydrates (starch and neutral detergent-soluble fiber - NDSF) types and proportions on ruminal degradability and fermentation parameters in crossbred sheep. Six ruminally cannulated male sheep (Santa Ines × Dorper;  $35 \pm 0.9$  kg BW) were housed in metabolism cages, assigned to a replicated  $3 \times 3$  Latin square design. During 63 d, lambs were fed with 3 isonitrogenous and isoenergetic diets (30% corn silage and 70% concentrate): D1 - high starch concentration (26.6%) and low NDSF concentrations (13.4%); D2 - similar concentrations (18.4%) of starch and NDSF, D3 - high NDSF concentration (24.3%) and low starch concentration (8.3%). To formulate concentrates were used corn, citrus pulp (only orange), soybean hulls, soybean meal, sunflower meal, sunflower oil and urea. The dried ground samples were incubated in nylon bags ( $7 \times 14$  cm) into the rumen for 3, 6, 12, 24, 48 and 72 h. The fraction "a" was determined by washing the bags in water and the residue after 72 h incubation was considered the fraction "c." The potential and effective degradabilities were calculated with the models:  $P = a + b(1 - e^{-kt})$ , and  $P = a + b * [k / (k + K_p)]$ , respectively. Ammonia nitrogen (AN), volatile fatty acids (VFA), and pH were determined at 1 h pre-prandial and 0, 1, 2, 4, 6, and 8 h after morning feeding. Data were analyzed using the MIXED procedure of SAS, with repeated measures. The degradation parameters were not affected by diets ( $P > 0.05$ ). The corn OM presented potential degradability of 81.7% and degradation rate of 10.7%/h, while citrus pulp OM showed 90.5% and 8.4%/h, respectively. Total VFA, acetic, propionic and butyric acids were unaffected ( $P > 0.05$ ) by dietary treatments (79.3, 50.9, 18.8, and 9.6 mM, respectively). The diets did not alter ruminal pH values (averaging 6.3;  $P > 0.05$ ), but AN concentrations (15.87%) were higher ( $P < 0.05$ ) in D3. Diets with different types and proportions of soluble carbohydrates do not affect neither ruminal degradation kinetics nor VFA concentrations, however the ruminal AN concentration increases in NDSF diets.

**Key Words:** fermentation, rumen, soluble carbohydrate

**T525 Relationship between body condition score and body fat depots in Pelibuey ewes.** Gamaliel Antonio-Molina<sup>1</sup>, Alfonso Chay-Canul<sup>\*1</sup>, Juan Ku-Vera<sup>2</sup>, Armando Gomez-Vazquez<sup>1</sup>, and Aldenamar Cruz-Hernandez<sup>1</sup>, <sup>1</sup>División Académica de Ciencias Agropecuarias, Universidad Juárez Autónoma de Tabasco, Tabasco, Mexico, <sup>2</sup>Facultad de Medicina Veterinaria y Zootecnia, Universidad Autónoma de Yucatán, Yucatán, Mexico.

Twenty-eight nonlactating and non-pregnant adult Pelibuey ewes, ranging in body condition score (BCS) from 1 (thin) to 5 (obese) were used to study the relationship between body condition score and body fat depots. The animals were humanely slaughtered following the Mexican Official Norms; before slaughter, shrunk BW and BCS were measure after feed and water deprivation for 24 h. The BCS was evaluated using palpation by 2 trained evaluators. Data recorded at slaughter were weights of viscera and carcass. Internal fat (IF, internal adipose tissue) was dissected, weighed and grouped as mesenteric (MF), omental (OF) and kidney knob and channel fat (KKCF). Carcass was then split at the dorsal midline in 2 equal halves, weighed, and chilled at 1°C during 24 h. After refrigeration, the left half of the carcass was completely dissected into subcutaneous and intermuscular fat (carcass fat, CF), muscle, bone and each component weighed separately. Dissected tissues of the left carcass were adjusted as whole carcass. The relationships between BCS and with body fat depots were investigated using regression analysis, with PROC REG of SAS. The BW and BCS at slaughter were highly correlated ( $r = 0.92$ ). The regression equation for estimating BW from BCS was  $BW$  (kg) =  $23.46 (\pm 1.62) + 6.34 (\pm 0.54) \times BCS$  ( $r^2: 0.84$ ; RSD: 3.41;  $P: < 0.0001$  and  $n = 28$ ). All equations for relationship between BCS and body fat depots were significant ( $<0.0001$ ) and the  $r^2$  ranged from 0.87 for CF (RSD = 1.016 kg) to 0.96 for IF (RSD = 1.206). Because the intercept of equations that involved BCS and IF, OF, MF and KKCF were no significant, we fitted a linear regression trough the origin. The regression equation for estimating total body fat (TBF) from BCS were  $TBF$  (kg) =  $-2.64 (\pm 0.911) + 4.29 (\pm 0.302) \times BCS$  ( $r^2: 0.89$ ; RSD: 1.916;  $P: < 0.0001$  and  $n = 28$ ). These results indicated that BCS could be used as a predictor of the main body fat reserves in Pelibuey ewes.

**Key Words:** body condition, body energy reserves, energy balance

**T526 The sexual behavior of male goats treated with exogenous testosterone is affected by the feeding level.** Ma de Santiago Miramontes<sup>\*1</sup>, J. F. Alvarado-Espinosa<sup>1</sup>, F. G. Véliz-Deras<sup>1</sup>, O. Ángel-García<sup>1</sup>, A. Gonzalez-Tavizón<sup>1</sup>, M. G. Calderón-Leyva<sup>1</sup>, L. I. Vélez-Monroy<sup>2</sup>, J. D. Hernández-Bustamante<sup>1</sup>, and M. Mellado<sup>1</sup>, <sup>1</sup>Universidad Autónoma Agraria Antonio Narro, <sup>2</sup>Instituto Nacional de Investigaciones Agrícolas y Pecuarias.

A successful male effect is supported on sexually active bucks, therefore the aim and the present study were to determine the effect of feeding level + testosterone ( $T_4$ ) treatment, on search and consummation behaviors in bucks (mixed-breed in arid region of Mexico; 26° N) during the sexual rest. Four groups were used ( $n = 5$  c/u; Body condition Score,  $2.5 \pm 0.12$  points, 1–4 scale). On February 2nd, one group:  $NT_4$  (Normal feeding +  $T_4$ ) received 0.5 kg of alfalfa hay, 1.1 kg of oaten hay and 0.2 kg of molasses/animal + 25 mg of  $T_4$  IM; NC group (Normal feeding, no  $T_4$ ) received a same feeding + 1 mL of NaCl (as placebo);  $LT_4$  group (Low feeding +  $T_4$ ) received 0.3 kg of alfalfa hay, 0.5 kg of oaten hay and 0.15 kg of molasses per animal + 25 mg of  $T_4$ , the LC group (Low feeding, no  $T_4$ ) received a same feeding + 1 mL of NaCl.  $T_4$  and NaCl was applied every 3rd day for 3 weeks. On April 14, the sexual behaviors (search; ano-genital sniffs, approaches, vocalizations, flehmen. Consummation; attempts to mount, mounts, mounts with a scabbard, mounts with ejaculation) were individually registered using a estrogenized female. The behaviors were compared with a  $\chi^2$  test (MYSTAT 12). The Body Condition Score at the end of the study was:  $LT_4$  &  $LC = 1.7 \pm 0.2$  and:  $NT_4$  &  $NC = 2.4 \pm 0.13$ . The search behaviors showed statistical differences ( $P < 0.05$ ) for all groups ( $NT_4$ , 53%;  $LT_4$ , 24%;  $LC$ , 15%;  $NC$ , 8%). Likewise, on consummation ( $P < 0.05$ ) for all groups ( $NT_4$ , 54%;  $LT_4$ , 32%;  $LC$ , 10%;  $NC$ , 4%). We conclude that an appropriate



feeding, improves the sexual behaviors of search and consummation in male goats treated with exogenous testosterone. These sexually active bucks would stimulate through the “male effect” the estrous behavior in mixed-bred anovulatory goats in the arid region of northern Mexico.

**Key Words:** goat, testosterone, male effect

**T527 Pre-weaning and post-weaning growth performance of F<sub>1</sub> intact male Kiko × Boer goat kids from does divided into high and low lines for parasite resistance—One year summary.**

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Goats are becoming increasingly popular because of their potential economic benefits and their ability to compliment other livestock enterprises. However, gastrointestinal parasites are a serious constraint in goat production; one solution may be selection for parasite resistance. Therefore, the objective of this study was to evaluate pre-weaning and post-weaning growth performance of F<sub>1</sub> intact male Kiko × Boer goat kids from does divergently grouped based on parasite resistance. Intact male F<sub>1</sub> Kiko × Boer progeny (n = 57; 3.0 ± 0.70 kg birth weight) from 2 lines of does with either a high resistance to internal parasites (HL; n = 34) or low resistance to internal parasites (LL; n = 23) were compared. Goat kids were born and managed on pasture with ad libitum access to creep feed until weaning at approximately 100 d of age. Kids were then placed in a drylot with ad libitum access to water, commercial goat mineral, a high-concentrate finishing diet, and were offered mixed-grass hay at approximately 10% of their total diet daily. Pre- and post-weaning weights did not differ ( $P \geq 0.16$ ) between HL and LL; however, weights across pre- and post-weaning weigh periods collectively tended ( $P = 0.10$ ) to be heavier for LL than HL. As expected, weight differed ( $P < 0.0001$ ) across all weigh periods. Weight was regressed over all weigh periods using linear equations to graphically display differences in kid growth rate. Kids from LL had lower birth weights than HL, but, by weaning, weights of LL kids tended to increase at a greater rate (slope difference,  $P = 0.15$ ) than that of HL kids (pre-wean weight<sub>LL</sub> = 2.99776 + [0.13235 × weigh d],  $R^2 = 0.45$  vs. weight<sub>HL</sub> = 4.3369 + [0.1045 × weigh d],  $R^2 = 0.46$ ). Yet, after weaning, weights of LL and HL kids increased at virtually the same rate (slope difference,  $P = 0.81$ ). Maternal contributions to weaning weight differences are typically attributed to dam's milking ability; thus, these preliminary results indicate that HL does may not have the same milk production as LL does when grazing parasite-infected pastures.

**Key Words:** goat, parasite, growth

**T528 Effect of adding zeolite (clinoptilolite) on growth performance and carcass characteristics in hair lambs fed a finishing diet.**

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Mexican sheep meat production covers only 56.52% of its demand; thus, an alternative to the national sheep production shortage is intensive completion based on the supply of potential of byproducts, additives

and minerals that could be included in the diets for intensive fed sheep to reduce the finishing time. Clinoptilolite zeolite are hydrated alkaline aluminosilicates with known capacity of increasing digestion time, thus increasing the digestive efficiency of feed nutrients. To determine the effect of the addition of 4 increasing levels of zeolite (0, 1.5, 3.0, 4.5%) in completely mixed diets for sheep on growth performance and carcass characteristics, 40 male sheep 1/4 Kathadin × 3/4 pelibuey were used. According to a randomized complete block design, sheep were grouped into 5 blocks with 4 pens each and each treatment randomly sorted into each block. The test lasted 75 d and weighings were carried at 28, 56 and 75 d respectively. The treatments used were: T1) 0% Zeolite (16.5% CP and 1.38 Mcal / kg ME), T2) 1.5% Zeolite (16.0% CP and 1.35 Mcal / kg ME), T3) 3% Zeolite (16.5% CP and 1.32 Mcal / kg ME) and T4) 4.5% zeolite (15.1% CP and 1.29 Mcal / kg ME). The feed was offered in twice a day at 8:00 and 14:30, adjusted to 3.0% of their initial body weight amount and gradually adjusted based on the excess or shortage of existing food the next day. Feed was served in a 40:60 ratio in the morning and afternoon respectively. At the end of the trial animals were killed, and 24 h after slaughter carcass data were obtained. In the productive performance data, no effect in final weight ( $P = 0.50$ ), daily weight gain ( $P = 0.50$ ) and dry matter intake ( $P = 0.08$ ) was observed due to the effect of addition of zeolite. Also, no effect owed to zeolite added to diet was observed in HCW ( $P = 0.98$ ) or rib-eye area ( $P = 0.64$ ). It is concluded that clinoptilolite zeolite can be replaced up to 4.5% in substitution of corn and soybean, ingredients of high economic impact, with no significant effect in productive of carcass variables.

**Key Words:** additive, mineral, ruminant nutrition

**T529 Indices of enzyme activities and atherogenicity of lamb meat fed high levels of crude glycerin.**

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The objective was to evaluate the effects of corn replacement by crude glycerin (CG) in diets with high-concentrate diets and low starch on enzyme activities and atherogenicity index of lamb meat. Forty Santa Ines lambs (23.5 ± 1.35 kg BW) were assigned to a randomized block design with 5 treatments: 0, 7.5, 15, 22.5, and 30% of CG on dry matter basis. The diet with 30% of CG promoted total replacement of corn. Diets consisted of Tifton-85 hay, corn, crude glycerin (83% glycerol), corn gluten meal, corn oil, urea, sunflower meal, soybean hulls and mineral. All diets had around 18% roughage and 82% concentrate. The Animals were housed in individual pens and fed ad libitum twice daily at 0700 and 1600 h. The experimental period had 72 ± 7 d of duration and the animals were slaughtered when reached 38 kg BW. The indices of enzyme activities  $\Delta^9$ -desaturase on C16 and C18 fatty acids and enolase, as well as atherogenicity index (indicator of the risk of cardiovascular disease) were calculated using the concentration of fatty acids from *Longissimus* muscle. Orthogonal contrasts were used to determine the linear and quadratic effects of CG and also contrast of 0% CG vs. CG treatments were used. The enzymatic activity indexes for elongase and  $\Delta^9$  desaturase 16 and 18 increased linearly with increasing dietary crude glycerin ( $P \leq 0.02$ ). Activity indexes were also higher in the meat from animals fed CG, regardless of concentration, when compared with meat from animals fed without CG ( $P \leq 0.04$ ). Increasing CG linearly reduced the meat atherogenicity index ( $P < 0.01$ ). Moreover, the index was lower in animals fed CG, regardless of concentration, in comparison to animals fed CG ( $P < 0.01$ ). The CG can provide higher activity of

enzyme linked to production of CLA and oleic fatty acid in lamb meat. The CG has potential to decrease the risk of cardiovascular disease.

**Key Words:** cardiovascular disease, CLA, glycerol

**T530 Performance of lambs fed corn stalk silages plus pig excreta, poultry litter and urea, or cane molasses and bakery by-products.**

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This experiment was carried out to evaluate the effect of corn stalk (CS) silage diet (145 g CP/kg DM; 2.4 Mcal/kg DM) plus 3 nitrogen sources (pig excreta, PE; poultry litter, PL; urea, UR) and 2 energy sources (sugar cane molasses, CM; bakery by-products, BBP). Composition (g/kg DM) of silages was (1) 384. Six CS; 384.6 PL; 230.8 CM or BBP; (2) 294.1 CS; 529.4 PE; 176.5 CM or BBP; (3) 630.0 CS; 30 UR; 340 CM or BBP. Diet contained (g/kg DM) silage 400:600 supplement (corn grain, soybean meal, wheat bran, fish meal, vitamins and minerals). The

experimental design was completely randomized, using 30 Criollo lambs (24.05 ± 3.68 kg initial BW) housed in individual cages during 60 d. Data were analyzed with PROC MIXED for average daily gain (ADG) and dry matter intake (DMI) utilizing initial body weight as covariable, whereas PROC GLM and Tukey test ( $P < 0.05$ ) were used for carcass variables. Qualitative variables (color of meat and fat) were analyzed by U-Mann-Withney test. No differences ( $P > 0.05$ ) were found for ADG (161, 175, 163, 160, 161, 150 g/day), DMI (792, 856, 894, 962, 961, 767 g DM/day) for PL-CM, PL-BBP, PE-CM, PE-BBP, UR-CM and UR-BBP, respectively. Rib eye area was larger for lambs fed PE-BBP (14.26 cm<sup>2</sup>), as compared with UR-CM (9.34 cm<sup>2</sup>). Empty BW was lower ( $P < 0.05$ ) for lambs fed corn stalk silage plus UR and CM (27.7 kg), as compared with those fed CS, PE and CM (34.8 kg). Corn stalk silage plus UR and CM decreased ( $P < 0.05$ ) carcass width (23.04 cm), as compared with corn stalk silage plus PE and BBP. No differences were found for the qualitative variables among treatments ( $P > 0.05$ ). Therefore, it may be concluded that lambs fed corn stalk silage plus pig excreta and bakery by-products showed better carcasses.

**Key Words:** corn stalk, waste and by-product, lamb

## Teaching/Undergraduate and Graduate Education

**T531 Instructors' perceptions of the importance and adequacy of "high-impact educational practices" in the animal science undergraduate curriculum in the United States.** Sanjeeva D. Ranathunga<sup>\*1</sup>, Michel A. Wattiaux<sup>1</sup>, and Peter Crump<sup>2</sup>, <sup>1</sup>*Department of Dairy Science, University of Wisconsin, Madison, WI*, <sup>2</sup>*Department of Computing and Biometry, University of Wisconsin, Madison, WI*.

High-impact educational practices (HIP) correlated positively with students' educational outcomes and can be an approach to address achievement gaps. A survey was conducted in 2012 and 2013 to evaluate the perception of animal sciences instructors on the importance (1 = not at all to 5 = a great deal) and adequacy (need more, good as it is, need less) of 16 educational practices which included 7 HIP: internships (INT), capstone courses or projects (CCP), collaborative assignments and projects (CAP), undergraduate research (URS), writing-intensive courses (WIC), diversity/global learning (DGL), and service learning, community-based learning (SCL). The analyzed data set included 148 instructors from 68 academic institutions. Rankings and scores (mean  $\pm$  SD) for the 7 HIP were INT (3, 4.44  $\pm$  0.64), CCP (4, 4.37  $\pm$  0.79), CAP (5, 4.26  $\pm$  0.68), URS (6, 4.25  $\pm$  0.79), WIC (7, 4.21  $\pm$  0.76), DGL (10, 3.88  $\pm$  0.88), and SCL (15, 3.54  $\pm$  0.86). Except for DGL, demographic factors affected HIP scores as follows: types of colleagues (animal scientists vs. others; INT,  $P = 0.010$ ), academic position (assistant, associate, full professor, others; INT,  $P = 0.008$ ; CAP,  $P = 0.026$ ), graduate program completed in the US (yes or no; CAP,  $P = 0.011$ ), teaching experience (no, limited, some experience, taught full-semester courses; CAP,  $P = 0.031$ ), type of university (Carnegie basic classification, 2010; URS,  $P = 0.034$ ), survey year (2012 vs. 2013; WIC,  $P = 0.006$ ), first generation to attend college (yes or no; SCL,  $P = 0.022$ ), and ethnicity (minority vs. Caucasian; SCL,  $P = 0.040$ ). Percentage of respondents who indicated "need more" of the HIP in the curriculum in their institutions were: WIC:55, URS:53, CCP:53, SLC:53, CAP:48, INT:46, and DGL:46%; whereas the percentages for "good as it is" were 34, 38, 34, 37, 41, 43, and 47%, respectively. Multiple demographic factors influenced animal science instructors' perceptions of the importance of HIP. Furthermore, most instructors recognized the need to include more HIP in the curriculum.

**Key Words:** high-impact educational practices, survey, curriculum

**T532 Effect of volunteer activities on the students' understanding of equine assisted therapy.** Molly Nicodemus\*, Torea Bova, and Jennifer Lockhart, *Mississippi State University, Mississippi State, MS*.

The Professional Association of Therapeutic Horsemanship International certification requires applicants not only to have a strong background in horsemanship activities, but also to have a history of volunteering at an equine-assisted therapy program. Nevertheless, the equine assisted therapy course, ADS 3233, at Mississippi State University has only consisted of a laboratory dedicated to developing horsemanship skills. This school year, volunteer hours performed at a local equine-assisted therapy program were required of students, and thus, the study objective was to evaluate whether volunteering assisted in the development of the students' understanding of equine-assisted therapy. Researcher-developed, 22 forced-choice question survey was given at the start (S) and end (E) of the semester to students taking ADS 3233 in 2013 ( $n = 15$ ) and 2014 ( $n = 15$ ). While in both years students participated in a riding laboratory, only students in 2014 were required to volunteer a minimum of 10 h with a local equine-assisted therapy program. Each

question on the survey was a statement concerning activities associated with working in various aspects of equine-assisted therapy programs with students rating from 1 to 5 their agreement to the statements. Lower scores indicated a stronger disagreement with the statement given concerning the aspect of the equine-assisted therapy program covered in the statement, while a higher score indicated a stronger agreement. Differences between S and E answers were calculated for each year and these differences were compared between years using a 2-sample Student's  $t$ -test ( $P = 0.05$ ). No question resulted in a score of 5. While both years demonstrated improvements on all 22 questions after completing the course, 5 of the questions demonstrated significant difference between years ( $P < 0.05$ ). These questions focused on the ability of the students to work with various experts in the industry and setting up riding programs for specific groups. By volunteering, students were able to get a more thorough understanding of equine-assisted therapy programs by working firsthand with those participating in these programs.

**Key Words:** equine-assisted therapy, volunteering

**T533 Students' perceptions of learning effectiveness in a capstone dairy management course.** Lisa A. Holden\*, *Pennsylvania State University, University Park, PA*.

Animal Science 410 is an advanced dairy management course in a capstone series for graduating seniors. The course is team-taught by a group of instructors, has an on-farm evaluation component, and integrates production information with financial and business management concepts. For the past 4 years, students were asked to provide course feedback and complete a short supplemental survey about "learning effectiveness." This survey was in addition to the regular online course evaluation data gathered for all courses. The objective of this supplemental survey was to better understand the impact of a team taught course with real world examples had on the quality of student learning. From 2011 to 2014, a total of 80 students completed the course and 76 responses were gathered in the short supplemental survey about 3 key areas of course structure: team taught format, farm evaluation experience and integration of production and financial data. Students were asked to rank the learning effectiveness of each area on a Likert scale of 1 to 5 where 1 = poor for learning and 5 = excellent for learning. Students were also asked to provide feedback about course structure, either verbally with an instructor or as written comments as part of the short supplemental survey. Average ratings for the team taught format were 3.8 ( $n = 74$ ) with a range of 2 to 5, for farm evaluation 4.2 ( $n = 76$ ) with a range of 2 to 5 and for integration of production and financial information 4.7 ( $n = 75$ ) with a range from 3 to 5. Key areas for improvement from student comments included information about "linking" classroom concepts to the farm evaluation, "standardizing" information and testing across instructors, and the need for "real world" financial data. Key areas that appeared to positively affect student learning were the use of classroom examples to explain concepts, interactive classroom discussion, and the quality of data available from farm evaluations. Based on results of this data set, consistency of information among instructors in a team-taught course and the use of high quality data in classroom examples can have positive benefits on student learning.

**Key Words:** student, learning, effectiveness



**T534 Ever wonder what they're thinking? Using a reflective academic journal to gauge learning in a content heavy classroom.**

Douglas Vincent\*, *University of Hawaii at Manoa, Honolulu, HI.*

As instructors of content classes, we often lack real-time data on how our students are progressing in class. The objective is to present a teaching technique, Reflective Academic Journal, which provides for a 2-way conversation between student and instructor. The student uses the journal to summarize their learning in class; to ask questions they might not in the classroom. The instructor benefits because he/she receives real-time feedback on the content being presented. Learning involves 3 components: (1) the actual knowledge—what it is you know; (2) your skills or ability to apply the knowledge in a particular context; and (3) your feelings—what you think about (1) and (2). Students in junior/senior classes in animal physiology and reproduction are required to write weekly reflective journals beginning after the third week of the semester. A journal can include a description of what happened in class; a constructive criticism of the learning media; a summary of what the student learned that week; a personal response to a class discussion; a reflection on what the student enjoyed about the class and even an expression of fear about an upcoming exam. In practice, the students benefit because they can open up a conversation with the instructor in a non-threatening manner, ask questions, and clear up misconceptions. The instructor benefits because they can obtain real time data on the student learning in that particular week. Questions raised in the journal can be addressed in the next class period. If general misconceptions exist among students, the instructor can trace back and repeat critical information. Student feedback from final journals and end-of-semester course evaluations indicate that writing journals help student learning. Reflective academic journals are an effective tool to improve student learning.

**Key Words:** learning, academic journal, assessment

**T535 Retention of concepts related to beef palatability from classroom experience of an informal consumer sensory panel in conjunction with discussion.** Jay A. Daniel\*<sup>1</sup>, George R. Gallagher<sup>1</sup>, and T. Dean Pringle<sup>2</sup>, <sup>1</sup>*Berry College, Mount Berry, GA*, <sup>2</sup>*University of Georgia, Athens, GA.*

We have previously reported use of an informal consumer sensory panel in conjunction with discussion as an effective means of teaching concepts related to beef palatability. In this experience, students evaluate steak samples for tenderness, juiciness, beef flavor and overall desirability. The samples serve to demonstrate the impact of cut and degree of doneness on beef palatability. After sampling the steak, results of the students' evaluation and other factors affecting beef palatability are discussed. To evaluate retention of concepts related to beef palatability, students (n = 37) enrolled in ANS 422 Beef Systems were asked to complete a 10 question quiz on concepts related to beef palatability. Five questions on the quiz were related to the informal consumer sensory panel experience (experience), and 5 were related to only the discussion which followed (lecture). All students had completed the informal consumer sensory panel (steak lab) as part of ANS 120 Introduction to Animal Science. In addition to the 10-question quiz, students were asked when they had completed ANS 120 (year), who their instructor was for ANS 120 (instructor, n = 2), and if the steak lab affected their steak purchasing habits. Overall scores on the quiz were tested for effect of instructor, year and instructor × year interaction by ANOVA with JMP software (version 10, SAS Inst. Inc., Cary, NC). There was no effect of instructor ( $P = 0.31$ ), year ( $P = 0.60$ ) or instructor × year interaction ( $P = 0.56$ ) on quiz score. Furthermore, student scores on experience questions were compared with scores on lecture questions using ANOVA. Student scores

on experience questions were greater than scores on lecture questions ( $81 \pm 2\%$  vs.  $60 \pm 2\%$  respectively,  $P < 0.0001$ ). Additionally, 65% of students agreed or strongly agreed that the steak lab influenced their choices in steak purchases. These results indicate use of an informal consumer panel is an effective means of teaching concepts related to beef palatability.

**T536 Relationship between course performance and graduation rates of animal science majors.** M. J. Anderson\*, J. L. Leatherwood, M. M. Beverly, K. J. Stutts, and S. F. Kelley, *Sam Houston State University, Huntsville, TX.*

Student success in completing a degree and graduating is dependent on course performance. Additionally, courses in a student's major will not only affect graduation, but also determine the aptitude of that student for their chosen career path. The objective of this study was to investigate the relationship between graduation rates and the grade earned in specific animal science courses. To accomplish this, course grades and graduation rates of students receiving those specific grades were collected for animal science courses over a 10-year period at Sam Houston State University. The courses included Animal Science, Anatomy and Physiology of Domestic Animals, Animal Nutrition, Meat Science, Equine Science, Animal Breeding and Genetics, Range Management, Animal Reproduction, and Animal Feeds and Feeding. Animal Science is an introductory course for freshmen-level students and prerequisite for other courses evaluated. Anatomy and Physiology of Domestic Animals, Equine Science, and Animal Nutrition are commonly composed of sophomore-level students, while the remaining courses are generally composed of upperclassmen. When graduation rates are reviewed across courses, there is a steady decline in graduation rate of students in the Animal Science course as the final course grade moves from a B (74%), to a C (51%), to a D (30%), and, last, to an F (0%). This pattern does not continue with the sophomore- and upper-level courses. In those courses, over 77% of students graduate when earning a final course grade of C or better. This illustrates that the introductory Animal Science course at Sam Houston provides the necessary rigor to the program and adequately prepares students for the advanced courses in animal science. This also demonstrates that students who receive a lower grade in the introductory Animal Science course may be able to continue in the program, but their likelihood of graduating is significantly lower. Therefore, the introductory Animal Science course at Sam Houston provides an early indicator of student success in reaching graduation.

**Key Words:** graduation rate, education, animal science

**T537 Fine Focus: A new international undergraduate microbiology research journal.** John L. McKillip\*, *Ball State University, Muncie, IN.*

*Fine Focus* is the first product-based course of its kind at Ball State University, and is poised to meet the AAAS recent call to action for transformative learning in biology. In utilizing the skill sets of 12 undergraduate students spanning 4 departments, *Fine Focus* has developed a peer-reviewed academic digital and print journal whose mission is to publish findings of undergraduate research internationally. We hypothesize that this "immersive learning" course provides participating students direct experience in double-blind peer-reviewed manuscript management, copy editing, and marketing/advertising available nowhere else. Participating students gain a multitude of experiences through collaborations with professionals from the American Society for Microbiology (ASM), National Institutes of Health (NIH), and the Council on Undergraduate Research (CUR). Such experiences include acquisition

of a working knowledge on scientific writing, editing, peer review, graphic design, and advertising, and professional correspondence with authors and reviewers, as they relate to dissemination of microbiological research data through an academic journal with an international scope. This multifaceted learning impact is assessed through student surveys, progress report notebooks, and self-evaluation exercises, which consistently indicated that *Fine Focus* offered exposure to technical aspects of manuscript review and data analyses not available in traditional content courses. After 4 semesters, *Fine Focus* has established an Editorial

Board of more than four dozen experts in microbiology and is publishing the first issue in January 2015 that includes 6 published articles on original undergraduate research. Students leave the course having also established permanent career-relevant contacts in varied subdisciplines of microbiology worldwide, and the leadership development skills necessary to serve their profession in the realm of peer-reviewed research, a vital skill in today's community of scientists.

**Key Words:** undergraduate research, microbiology, journal

# ADSA Production Division Symposium: Production efficiency of the dairy cow

## 349 **Genetics of productive life.** Chad Dechow\*, *Penn State University, University Park, PA.*

Historic selection for yield has improved the efficiency of dairy production for individual cows and current interest in direct selection for feed utilization aims to further increase productive efficiencies. However, herd efficiency will improve only marginally if selection practices reduce cow fitness levels and increase herd replacement rates. US genetic evaluations for productive life were introduced in 1994 and remain a robust general indicator of cow fitness levels. Productive life credits are limited to a cow's lactation cycle with early lactation weighted more heavily than late lactation. There is considerable variation in herd-life with a genetic standard deviation of 5 mo despite a relatively low heritability (8%). Factors that influence herd life vary across herds and have shifted over time as herd management has evolved. The relationship of productive life with body size has become increasingly antagonistic over time, whereas the relationship with yield has gone from a moderately favorable to a low association. Given the current US Holstein population structure, productive life is strongly correlated with higher cow fertility and lower somatic cell score. Deriving the economic value of longer productive life is complicated by shifts in milk price, heifer rearing and replacement costs, and cull cow value. This has led to varying degrees of emphasis on productive life in different countries and across time. The evaluation of productive life is also complicated by the necessity of a cow's life-cycle to be completed before her true productive life is known. Despite such challenges, higher sire productive life has been demonstrated to be associated with lower rates of daughter mortality and early lactation culling across a range of management systems. As dairy cattle breeders continue to emphasize productive and economic efficiencies, the need to consider traits related to cow fitness levels are of increasing importance to ensure that selection for cow-level efficiencies do not diminish productive efficiency at the herd level.

**Key Words:** productive life, genetic, efficiency

## 350 **Economics of production efficiency: Nutritional grouping.** Victor E. Cabrera\*, *University of Wisconsin-Madison, Madison, WI.*

Nutritional grouping of lactating cows under TMR feeding systems has been discussed in the literature since the 70s. Most of the studies have concluded that using multiple, more-homogeneous TMR feeding groups is economically beneficial because of either or both nutrient cost savings or improved productivity. Nonetheless, there is not yet an absolute consensus or wide adoption. Latest studies using optimal basis for grouping and optimal diet specifications are reporting consistently greater income over feed cost (\$/cow per yr) with multiple TMR groups compared with 1 TMR (3 TMR = 46 to 77 and 2 TMR = 21 to 45). Critical factors that determine the economic value of nutritional grouping are (1) basis for grouping, (2) diets' specifications, (3) effects on milk production, and (4) additional costs. It has been strongly documented that grouping cows according to their simultaneous nutritional requirements (a.k.a., cluster grouping) is optimal. Cluster grouping is superior to other methods such as grouping by DIM, milk production, or production and BW combined. However, the dairy industry still uses less than optimal grouping bases. Using cluster grouping would enhance the positive economic impacts of multiple TMR. Next, groups diets' specifications seem not to be optimal either. The concept of lead factors, which are only based on group average milk production are heavily used. Nonetheless, diets should be more

precise following overall group nutrient requirements. Providing more precise diets will also be in favor of grouping economics. Next, an area that requires further attention is the potential negative effects of grouping on milk production because of either or both social interactions or diet concentration changes. Although the literature is inconclusive on this, latest studies indicate that multiple TMR largely outperform economically 1 TMR even after considering plausible potential milk losses of grouping. Finally, additional costs of management, labor, facilities, and equipment required for grouping are farm specific. The few studies that integrated these factors in their analyses found that multiple TMR would still be economically superior to 1 TMR.

**Key Words:** nutritional feeding, feed efficiency, cluster

## 351 **Potential benefits of nutrition on reproductive performance of high-efficiency dairy cows.** Milo Wiltbank\*<sup>1</sup>, Paulo Carvalho<sup>1</sup>, Alex Souza<sup>1</sup>, Paul Fricke<sup>1</sup>, Mateus Toledo<sup>1</sup>, Roberto Sartori<sup>2</sup>, Jose Santos<sup>3</sup>, Guilherme Pontes<sup>2</sup>, Daniel Luchini<sup>4</sup>, Francisco Penagaricano<sup>3</sup>, Hasan Khatib<sup>1</sup>, Katherine Hackbart<sup>1</sup>, and Randy Shaver<sup>1</sup>, <sup>1</sup>*University of Wisconsin-Madison, Madison, WI*, <sup>2</sup>*University of Sao Paulo, Piracicaba, SP, Brazil*, <sup>3</sup>*University of Florida, Gainesville, FL*, <sup>4</sup>*Adisseo, Alpharetta, GA.*

During the last century, increasing milk production has been associated with decreasing reproductive performance. However during the last decade, there has been a dramatic improvement in reproduction even as milk production continues to increase. The reasons for improving reproduction are multifactorial with dramatic improvements in reproductive management programs, advances in cow comfort and health management programs, and a turn-around in the genetics of reproduction underlying some of these gains. In addition, older research and many recent studies indicate that gains in reproductive performance require optimized nutritional programs including 4 specific areas that will be emphasized in this presentation. First, nutritional deficiencies in the prepartum diet can affect reproduction. As an example, recent research indicates that supplementation during the last month before calving with vitamin E in marginally-vitamin E deficient dairy cattle decreased retained placenta and stillbirths and increased subsequent reproductive performance. Second, nutritional deficiencies near calving and during the first 21 d after calving can have substantial effect on subsequent reproduction. In a recent retrospective study and consistent with previous results, we observed changes in BCS during the first 21 d after calving were associated with dramatic differences in fertility in high-producing lactating dairy cows. Third, increased insulin during the week before AI, potentially due to diets with high non-fiber carbohydrates, can have negative effects on fertilization and reproductive performance of ruminants. Thus, reducing insulin by targeted but subtle changes in feed intake or energy composition of the diet could be used to improve reproduction. Fourth, optimization of amino acid composition of diets may improve reproductive efficiency. Our recent research demonstrates that supplementing rumen-protected methionine altered gene expression in early preimplantation embryos and reduced subsequent pregnancy loss in lactating dairy cows. Thus, inadequate nutritional programs can reduce reproductive performance and optimized nutrition may augment reproduction even in herds with enhanced genetics and reproductive management strategies.

**Key Words:** fertility, nutrition



**352 Providing facilities to improve health, welfare, and productive life.** Trevor J. DeVries\*<sup>1</sup>, Marina A. G. von Keyserlingk<sup>2</sup>, and Daniel M. Weary<sup>2</sup>, <sup>1</sup>*Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada,* <sup>2</sup>*Animal Welfare Program, University of British Columbia, Vancouver, BC, Canada.*

Poorly designed and managed facilities cause injuries and increase the risk of health problems, including lameness and other infectious and metabolic diseases, whose high prevalence is a welfare challenge facing the dairy industry. Massive capital expenditures are made on housing systems for dairy cattle, with the aim of providing a comfortable environment for these animals—one that ensures adequate rest, protection from climatic extremes, and free access to an appropriate, well-balanced diet. Despite these laudable aims, housing systems do not always function well from the perspective of the cow—poorly designed and maintained facilities can reduce feeding and lying time, increase competitive interactions, cause injuries, and increase the risk of health disorders. This review will focus on examples from empirical work on the feeding, standing, and lying areas utilized by dairy cattle, and show how these can be better designed and managed to meet the behavioral needs of dairy cattle, and thus improve welfare, health, and in some cases, productivity. Much of the research reviewed has evaluated housing systems from the cow's perspective by asking how the housing affects cow health, what elements of the housing environment the cow prefers, and how these elements affect behavior and welfare. This presentation will focus primarily on free-stall housing and will first, identify science-based solutions that result in improved health and welfare of dairy cows and, second, identify gaps where further research is needed.

**Key Words:** dairy cow, housing, welfare

**353 Precision dairy monitoring technologies as tools to improve dairy production efficiency.** J. M. Bewley\*, R. A. Russell, A. E. Stone, B. A. Wadsworth, K. A. Dolecheck, M. R. Borchers, M. E. Weatherly, L. M. Mayo, I. C. Tsai, M. C. Hardy, and J. M. Klefot, *University of Kentucky, Lexington, KY.*

Technologies are changing the shape of the dairy industry across the globe. In fact, many of the technologies applied to the dairy industry

are variations of base technologies used in larger industries such as the automobile or personal electronic industries. Undoubtedly, these technologies will continue to change the way that dairy animals are managed. This technological shift provides reasons for optimism for improvements in both cow and farmer well-being moving forward. Many industry changes are setting the stage for the rapid introduction of new technologies in the dairy industry. Dairy operations today are characterized by narrower profit margins than in the past, largely because of reduced governmental involvement in regulating agricultural commodity prices. The resulting competition growth has intensified the drive for efficiency resulting in increased emphasis on business and financial management. Furthermore, the decision making landscape for a dairy manager has changed dramatically with increased emphasis on consumer protection, continuous quality assurance, natural foods, pathogen-free food, zoonotic disease transmission, reduction of the use of medical treatments, and increased concern for the care of animals. Lastly, powers of human observation limit dairy producers' ability to identify sick or lame cows or cows in heat. Precision dairy management may help remedy some of these problems. Precision dairy management is the use of automated, mechanized technologies toward refinement of dairy management processes, procedures, or information collection. Precision dairy management technologies provide tremendous opportunities for improvements in individual animal management on dairy farms. Although the technological "gadgets" may drive innovation, social and economic factors dictate technology adoption success.

**Key Words:** precision dairy, monitoring, technology

## Animal Health: Transition cow health

**354 Characterizing critical thresholds of subclinical ketosis using the in-line milk monitoring system Herd Navigator.** Elizabeth R. Ellis\*, Tom C. Wright, John P. Cant, and Vern R. Osborne, *Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada.*

Sub-clinical ketosis (SCK) refers to a state of elevated  $\beta$ -hydroxybutyrate (BHBA) without visual signs of acute disease. With a prevalence of 26 to 60% (dependent on BHBA level used to define disease), this costly metabolic disorder often goes undetected. Innovative in-line milk testing technology, Herd Navigator (DeLaval), enables automated milk BHBA analysis of individuals in a herd. The objective of this study was to characterize SCK as it affects milk production using Herd Navigator technology. The study comprised 3 hundred and 56 Holstein cows from 5 dairy farms in Ontario, operating herd-specific SCK treatment protocols based on BHBA thresholds ranging from 1.2 to 1.7 mmol/L. Individual cow data were collected from June 2013 to July 2014 for daily milk yield, days to peak BHBA level, days to peak milk, peak milk yield; total predicted milk yield to 60 d and 305 d were generated for all cows. Based on the maximum BHBA level attained within 60DIM, cows were assigned to 1 of 5 groups (G1  $\geq$  1.7 mmol/L, G2  $\geq$  1.5 mmol/L, G3  $\geq$  1.4 mmol/L, G4  $\geq$  1.2 mmol/L, G5  $<$  1.2 mmol/L) for analysis. Data were analyzed by ANOVA using the maximum BHBA value as covariate and farm as a random effect. Results showed that first-lactation cows in G1 had higher milk yield at 60 DIM (165 kg  $\pm$  91,  $P <$  0.05) than first-lactation cows in G5. In second-lactation cows, there were no differences in milk yield between groups. Third-lactation cows in G2 had higher milk yield at 60 DIM than those in G4 (431 kg  $\pm$  153,  $P <$  0.05) G5 (295 kg  $\pm$  125,  $P <$  0.05). Predicted milk yield at 305 DIM of third lactation cows in G1 was lower (-2505 kg  $\pm$  1318,  $P <$  0.05) than in G2. Across farms, elevated ( $\geq$  1.4 mmol/L) milk BHBA values were associated with higher milk production for mature cows at 305DIM, but not for first- and second-lactation cows. Results of this study indicate that farm-level milk BHBA data from Herd Navigator coupled with available on-farm milk production records could be used to refine the effectiveness of SCK treatment protocols and identify different BHBA treatment thresholds by parity to improve milk yield.

**Key Words:** ketosis, milk production, Herd Navigator

**355 Monitoring rumination in transition dairy cows for the early detection of subclinical ketosis.** Emily I. Kaufman\*<sup>1</sup>, Stephen J. LeBlanc<sup>2</sup>, Brian W. McBride<sup>1</sup>, Todd F. Duffield<sup>2</sup>, and Trevor J. DeVries<sup>1</sup>, <sup>1</sup>*Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada,* <sup>2</sup>*Department of Population Medicine, University of Guelph, Guelph, ON, Canada.*

The objective of this study was to characterize the relationship between rumination and subclinical ketosis (SCK) in transition dairy cows. A study was conducted on 4 commercial dairy farms in Eastern Ontario, Canada. A total of 339 dairy cows (107 primiparous and 232 multiparous) were monitored for rumination activity and SCK from 14d before calving until 28d after calving. Rumination was recorded daily using an automated monitoring system. A blood sample was taken from the coccygeal vein of each cow for the measurement of  $\beta$ -hydroxybutyrate (BHBA) 1x/wk throughout the 6-wk observation period. Cows with a BHBA concentration  $\geq$  1.2 mmol/L postpartum were considered to have SCK. Cases of retained placenta, metritis, milk fever, or mastitis during the study period were also recorded. Cows were categorized into 1 of

3 groups: healthy (H) cows had no SCK or any other health issue (n = 139); SCK (K) cows with no other health problems during transition (n = 97); or ketotic plus (K+) cows that had SCK and one or more other health problems (n = 53). Data were summarized by wk and analyzed in a repeated measures general linear mixed model. From 2 wk before (-2) calving to 4 wk after calving (+4), there was no difference in daily rumination time (409  $\pm$  9.8 min/d; mean  $\pm$  SE) among H, K, and K+ cows in their first lactation ( $P = 0.5$ ). Multiparous cows in H spent an average of 459 min/d ruminating from wk -2 to wk +4. Multiparous K cows ruminated 25  $\pm$  12.8 min/d less ( $P = 0.05$ ) than H cows, while K+ cows ruminated 44  $\pm$  15.6 min/d less ( $P = 0.005$ ) than H cows. The largest differences in rumination time between H and K+ cows were seen during wk -1, +1 and +2, when K+ cows ruminated 48  $\pm$  17.2 min/d, 73  $\pm$  16.0 min/d, and 65  $\pm$  19.4 min/d less ( $P \leq 0.005$ ) than H cows, respectively. These results suggest that rumination monitoring across the transition period might contribute to identification of SCK and other health issues in multiparous cows.

**Key Words:** transition cow, rumination, subclinical ketosis

**356 Use of a rumination and activity monitoring for the identification of dairy cows with health disorders.** Matias L. Stangaferro\*, Robert Wijma, Cristian E. Quinteros, Miranda M. Medrano, Magdalena Masello, and Julio O. Giordano, *Department of Animal Science, Cornell University, Ithaca, NY.*

Objectives were to evaluate: 1) the ability of a commercial rumination (Rum) and activity (Act) monitoring system (HR Tags, SCR Dairy) to identify cows with health disorders and 2) the interval between the day of diagnosis of disease and day of alert by the HR system (HR). Holstein cows (n = 1,118; 449 nulliparous and 669 multiparous) were fitted with an HR tag from -28 to 80 DIM. Every 12 h after 1 DIM, an individual cow Health Index (HI) was generated based on Rum and Act. Cows with a HI value  $<$  86 points were flagged by HR. Farm personnel examined cows for signs of clinical disease (CDZ) daily. From 1 to 10 DIM, personnel evaluated: appetite, rectal temperature, ketone bodies in urine, rumen fill and movements, vaginal discharge, daily milk weights, and conductivity. Data from 1,099 cows was available. Number of CDZ events included was: displaced abomasum (DA) 41, ketosis (KET) 57, indigestion (IND) 9, metritis (MET) 360, and mastitis (MAST) 74. Sensitivity (Se) of HR to flag cows with CDZ (farm personnel diagnosis as gold standard) and the interval between day of CDZ diagnosis and the day a cow was flagged by HR was evaluated with PROC FREQ and PROC TTEST of SAS, respectively. The Se of HI was: 97.6% (CI 93-100%) for DA, 84.2% (CI 75-94%) for KET, 88.9% (CI 68-100%) for IND, 45.6% (CI 40-51%) for MET and 49.7% (CI 42-57%) for all MAST. For all DA, KET, and IND combined, Se of HR was 89.7% (CI 84-95%). Sensitivity of HR by MAST pathogen was 66.7% (CI 50-83%) for *E. coli* and *Klebsiella*, 52.6% (CI 37-68%) for *Streptococcus* and *Staphylococcus* spp. combined, and 22.2% (CI 0-49%) for *Staph. aureus*. Mean and 95% CI for interval between day of CDZ to day flagged by HR (cows flagged only) was: -3 (-3.7- -2.3;  $P <$  0.01), -1.6 (-2.3- -1.0;  $P <$  0.01), -0.5 (-1.5- 0.5;  $P = 0.28$ ), -0.8 (-1.2- -0.44;  $P <$  0.01), -0.8 (-1.2- 0.3;  $P <$  0.01), for DA, KET, IND, MET, and all MAST, respectively. We conclude that the HR system is most effective to identify cows suffering metabolic and digestive disorders. A relatively lower Se to identify cows with MET and MAST might be explained by less severe systemic illness and type of mastitis-

causing pathogen. The HR system identified cows with DA, KET, MET and MAST earlier than farm personnel.

**Key Words:** rumination, activity, dairy cow

### 357 Development of a ketosis prevalence tool in Holstein dairy cows based on milk component data and cow test-day information.

Tawny L. Chandler<sup>\*1</sup>, Ryan S. Pralle<sup>1</sup>, Garrett R. Oetzel<sup>1</sup>, Robert H. Fourdraine<sup>2</sup>, and Heather M. White<sup>1</sup>, <sup>1</sup>University of Wisconsin-Madison, Madison, WI, <sup>2</sup>AgSource Cooperative Services, Verona, WI.

Subclinical ketosis affects between 40 and 60% of dairy cows and negatively affects cow productivity and health. Although cow-side ketone testing strategies are available, many lack sufficient accuracy, are labor-intensive, and can be costly. The objective of this study was to validate the use of multiple regression models to predict blood  $\beta$ -hydroxybutyrate (BHBA) from milk components and continuous test-day variables in early lactation cows for determining ketosis prevalence. Blood samples were collected on the same day as milk test from 658 Holstein cows 5 to 20 DIM on 10 dairy farms. Blood serum was analyzed for concentration of BHBA by colorimetric assay (Stanbio, Boerne, TX). Milk samples were analyzed for milk BHBA and acetone concentrations by Fourier transform infrared spectrometry (FOSS Analytical A/S, Eden Prairie, MN), in addition to standard milk analysis variables. Continuous test-day variables were collected from DairyComp305 (Valley Agricultural Software, Tulare, CA) records. Models were built in the REG procedure of SAS 9.4 (SAS Institute Inc., Cary, NC) using stepwise, forward selection by excluding variables with a  $P$ -value  $< 0.15$  and selection criterion of Akaike's information criterion. Data interrogation justified development of separate models for primiparous and multiparous cows, as well as cows 5 to 11 DIM and cows 12 to 20 DIM. Additionally, disease etiology allowed for unique models for the 5 to 11 and 12 to 20 DIM ranges. Significant variables were milk BHBA, acetone, and fat:protein ratio; parity, previous days dry, previous lactation length, and age at first calving; and DIM and milk production on test day. Overall, model accuracy was 88% for multiparous cows 5 to 11 DIM ( $R^2 = 0.57$ ), 83% for multiparous cows 12 to 20 DIM ( $R^2 = 0.67$ ), 96% for primiparous cows 5 to 11 DIM ( $R^2 = 0.74$ ), and 97% for primiparous cows 5 to 20 DIM ( $R^2 = 0.66$ ). These results suggest that modeling blood BHBA based on milk component data and cow test-day information can serve as a valuable diagnostic tool for monitoring herd-level ketosis prevalence.

**Key Words:** ketosis, linear regression, model

### 358 Evaluation of recurrence of frequent diseases and disorders in early postpartum dairy cows.

Achilles Vieira-Neto<sup>\*1</sup>, Ana Carolina Parize<sup>3</sup>, Carlos A. Risco<sup>2</sup>, José Eduardo P. Santos<sup>1</sup>, and Klíbs N. Galvão<sup>2</sup>, <sup>1</sup>Department of Animal Sciences, University of Florida, Gainesville, FL, <sup>2</sup>Department of Large Animal Clinical Sciences, University of Florida, Gainesville, FL, <sup>3</sup>Escola de Veterinária e Zootecnia, Universidade Federal de Goiás, Goiânia, GO, Brazil.

Objective was to evaluate if having disease in one lactation (Lac1) would affect the risk of developing disease and calving related problems in the subsequent lactation (Lac2). Holstein cows ( $n = 1351$ ) from 2 herds in North Florida were used to create 2295 lactation pairs from 4590 lactations occurring between 2006 and 2013. Data were collected within 60 DIM. Data were evaluated using the LOGISTIC procedure of SAS. Having dystocia in Lac1 increased the risk of having it in Lac2 (37.0 vs. 23.7%;  $P < 0.01$ ). Dystocia in Lac2 was also increased by induced parturition and twins in Lac2. Having twins in Lac1 increased

the risk of having it in Lac2 (4.6 vs. 2.4%;  $P < 0.02$ ). Twins in Lac2 tended ( $P = 0.09$ ) to be increased in parity  $> 2$  vs. parity 2 in Lac2. Having stillbirth in Lac1 did not affect the risk of having it in Lac2 (5.7 vs. 3.2%;  $P = 0.12$ ). Stillbirth in Lac2 was increased ( $P < 0.05$ ) by dystocia and twins in Lac2. Having milk fever in Lac1 increased the risk of having it in Lac2 (38.9 vs. 2.6%;  $P < 0.01$ ). Milk fever in Lac2 was also increased ( $P < 0.05$ ) by dystocia and parity  $\geq 2$  in Lac2, but was decreased by induced parturition in Lac2. Having retained placenta in Lac1 increased the risk of having it in Lac2 (13.1 vs. 6.2%;  $P = 0.01$ ). Retained placenta in Lac2 was also increased ( $P < 0.05$ ) by twins and induced parturition in Lac2. Having metritis in Lac1 increased the risk of having it in Lac2 (20.4 vs. 10.4%;  $P < 0.02$ ). Metritis in Lac2 was also increased ( $P < 0.05$ ) by dystocia, induced parturition, twins, stillbirth, retained placenta and ketosis in Lac2. Having ketosis in Lac1 increased the risk of having it in Lac2 (41.9 vs. 17.0%;  $P = 0.01$ ). Ketosis in Lac2 was also increased ( $P < 0.05$ ) by induced parturition, metritis, and parity  $> 2$  in Lac2. Having displaced abomasum in Lac1 had a tendency to increase the risk of having it in Lac2 (10.5 vs. 2.1%;  $P = 0.06$ ). Displaced abomasum in Lac2 was increased ( $P < 0.05$ ) by ketosis in Lac2. Having mastitis in Lac1 had a tendency to increase the risk of having it in Lac2 (22.9 vs. 9.3%;  $P = 0.09$ ). Mastitis in Lac2 was increased ( $P < 0.05$ ) by parity  $> 2$ . In conclusion, with the exception of stillbirth, disease in Lac1 affected the incidence of disease in Lac2.

**Key Words:** dairy cow, postpartum diseases, disease recurrence

### 359 Early lactation disease incidence in Holstein cows across multiple US regions.

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The objective was to assess the incidence of transition cow diseases in Holstein cows ( $n = 10,959$  in 16 herds) calving during the warm (WS: May–Aug) and cool (CS: Oct – Jan) seasons. Cows were enrolled at parturition and monitored weekly for multiple diseases until 60 DIM. Disease included retained fetal membranes (RFM), metritis (MET,  $7 \pm 3$  DIM; foul-smell, watery, brownish vaginal discharge), subclinical ketosis (SKT,  $7 \pm 3$  DIM; serum BHBA  $> 1.0$  mmol/L), mastitis (MAS; farm records), left displaced abomasum (LDA), pneumonia (PN; farm records), clinical endometritis (CE  $28 \pm 3$  DIM; from mucopurulent to fetid vaginal discharge) and lameness (LAM  $35 \pm 3$  DIM; score  $> 3$ ). Study locations included the Northeast (NE; 4 herds), Midwest (MW; 6 herds), Southeast (SE; 1 herd), and the Southwest (SW; 5 herds) regions. Disease incidence was estimated by region and season (top part of Table 1). Associations between disease occurrence and region, calving season, and parity ( $1, \geq 2$ ) were tested by logistic regression. The effect of region on disease occurrence was only significant for LAM ( $P < 0.05$ ). Contrarily, season of calving and parity were significantly associated with the risk of transition diseases in all the disorders analyzed (bottom part of Table 1). This project was funded by USDA-NIFA-AFRI (2013–68004–20361).

*Contd.*



**Table 1 (Abstr. 359).** Incidence (%) of transition cow diseases by region and season of calving (top) and odds ratios (OR) for the risk of disease by region, season of calving, and parity (bottom); references are SW, WS, and parity  $\geq 2$

Item	NE		MW		SE		SW					
	WS	CS	WS	CS	WS	CS	WS	CS				
RFM	8.0	5.9	7.4	5.4	15.0	7.6	4.3	2.9				
MET	21.7	23.8	19.5	20.2	19.7	18.5	27.6	24.8				
SKT	41.8	18.7	25.9	15.5	24.9	20.1	31.3	14.6				
MAS	26.1	16.0	6.1	5.5	18.0	21.3	12.0	8.1				
LDA	3.0	5.6	2.9	1.4	6.0	4.0	1.0	1.0				
PNM	1.1	1.5	1.7	1.8	3.8	13.4	7.1	3.5				
CLE	15.4	32.5	25.9	20.4	23.4	42.9	24.3	26.1				
LAM	11.3	2.6	2.1	8.1	1.7	12.1	5.4	2.0				

  

Region	RFM		MET		SKT		MAS		CLE		LAM	
	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI
MW	1.9	0.7-5.0	0.8	0.5-1.4	0.7	0.2-2.1	0.6	0.2-2.0	1.0	0.6-1.7	3.7	1.3-10.5
NE	2.4	0.8-7.1	1.0	0.6-1.8	1.2	0.4-4.3	2.4	0.8-6.9	1.1	0.6-2.0	0.9	0.3-2.5
SE	4.2	0.8-23.0	0.8	0.3-2.0	1.0	0.1-7.6	2.9	0.5-16.3	1.7	0.7-4.5	1.4	0.3-6.3
Season												
CS	0.6	0.5-0.7	1.1	1.0-1.2	0.4	0.3-0.4	0.7	0.7-0.9	1.4	1.2-1.5	2.5	1.8-3.4
Parity												
1	0.4	0.4-0.5	1.8	1.6-2.0	0.6	0.5-0.7	1.3	1.1-1.5	1.1	1.0-1.2	0.2	0.1-0.3

**Key Words:** health, transition, Holstein

**360 Association between dry matter intake pre- and postpartum and postpartum diseases in dairy cows.** Johanny Perez Baez\*<sup>2</sup>, Carlos A. Risco<sup>2</sup>, Jorge A. Hernandez<sup>2</sup>, Gabriel C. Gomes<sup>2</sup>, Leandro F. Greco<sup>1</sup>, Sha Tao<sup>1,3</sup>, Izabella Thompson<sup>1,4</sup>, Bruno do Amaral<sup>1,5</sup>, and Charles Staples<sup>1</sup>, Jose Eduardo P. Santos<sup>1</sup>, and Klibs N. Galvão<sup>2</sup>, <sup>1</sup>Department of Animal Sciences, University of Florida, Gainesville, FL, <sup>2</sup>Department of Large Animal Clinical Sciences, University of Florida, Gainesville, FL, <sup>3</sup>Department of Animal and Dairy Science, University of Georgia, Tifton, GA, <sup>4</sup>Agriculture and Agri-Food Canada, Ottawa, Ontario, Canada, <sup>5</sup>Land O'Lakes, Inc., St. Paul, MN.

The objectives of this retrospective observational study were to determine the association between dry matter intake (DMI) pre- (-14 d to -1) and postpartum (1 to 28 d) and postpartum diseases [retained placenta (RP), metritis (MET), mastitis (MAST), ketosis (KET), and displaced abomasum (DA)] within 28 d postpartum; and to estimate the risk of disease postpartum based on DMI prepartum. Data involving 294 cows from 7 studies were collected. The data were analyzed with the MIXED and GLIMMIX procedures of SAS. Random and repeated variables were cow and day relative to calving, respectively. Models were adjusted for parity, BCS, treatment, study, and interaction between disease and other covariates. Variables with  $P \leq 0.05$  were considered significant. Cows that had RP ate less on d -3 ( $RP \times day P = 0.01$ ) and on d 5, 7, 8, 12, and 19 ( $RP \times day P = 0.10$ ). Cows that had MET ate less on d -3 and -2 ( $MET \times day P = 0.03$ ) and ate less postpartum ( $P < 0.01$ ). Cows that had MAST ate less prepartum ( $P = 0.02$ ) and in the first 14 d postpartum ( $MAST \times day P < 0.01$ ). Cows that had KET ate less pre- and postpartum ( $P < 0.01$ ). Intake of cows that had DA did not differ prepartum ( $P = 0.70$ ) but they ate less postpartum ( $P < 0.01$ ). Cows with at least one disease ate less pre- and postpartum ( $P < 0.01$ ). Moreover, for each kg decrease in DMI in the last week prepartum, there was an increase of 28% in the odds of having KET (OR = 1.28; CI = 1.15-1.41;  $P < 0.01$ ), and 24% in the odds of having at least one disease (OR = 1.24;

CI = 1.13-1.34;  $P < 0.01$ ) postpartum. However, DMI was not associated with the odds of having MAST (OR = 1.09; CI = 0.95-1.23;  $P = 0.23$ ), RP (OR = 1.04; CI = 0.86-1.22;  $P = 0.66$ ), MET (OR = 1.08; CI = 0.97-1.19;  $P = 0.14$ ), or DA (OR = 1.07; CI = 0.82-1.32;  $P = 0.6$ ). Collectively, these data suggest that there is an association between DMI pre- and postpartum and postpartum diseases, and that a reduction in DMI prepartum predisposes cows to disease postpartum.

**Key Words:** postpartum disease, dry matter intake, dairy cow

**361 Laboratory validation of a prototype cow-side instrument for the measurement of blood ionized calcium concentrations in dairy cattle.** Rafael C. Neves\*, Tracy Stokol, and Jessica A. A. McArt, Department of Population Medicine & Diagnostic Sciences, Cornell University, Ithaca, NY.

There is currently no efficient and inexpensive method for field measurement of blood calcium concentrations. Ionized calcium (iCa) is the homeostatic form of the mineral and is thought to have greater biological relevance over that of total calcium. The objective of this study was to evaluate the linearity and precision of a prototype cow-side instrument (Horiba, Japan) for measuring blood iCa concentrations. Blood (300 mL) was collected from the right jugular vein of a multiparous dairy cow (4 DIM) into lithium heparin tubes immediately before (T0) and 5 min after (T5) intravenous administration of 500 mL of 23% calcium borogluconate. The iCa concentrations were determined using a blood-gas analyzer (ABL-800 FLEX, Radiometer) as a gold-standard. The T0 sample was diluted using 0.9% saline to create a sample with low iCa (reference interval = 1.10 to 1.35  $\mu\text{mol/L}$ ). The diluted T0 sample was then mixed with the T5 sample in different ratios (100/0, 75/25, 50/50, 25/75, 0/100) to obtain 5 levels of iCa concentrations (0.69, 1.0, 1.28, 1.58, and 1.82  $\mu\text{mol/L}$ ). Each mixture was then analyzed in triplicate using 3 different prototypes under one-point (1P) and 2-point (2P) calibration with the means compared with results from the blood-gas

analyzer. Cumulative sum tests for linearity from Passing and Bablok regressions showed no deviation from linearity for the combined results of all 3 prototypes under 1P vs. the gold-standard ( $P = 0.19$ ) and under 2P vs. the gold-standard ( $P = 0.19$ ). Instrument precision (coefficient of variation; CV) was determined by 10 repeat measurements of the diluted T0 sample, T0, and T5 samples under 1P and 2P calibrations. The CV ranged from 1.3 to 5% for the 3 prototypes. Laboratory results indicate good accuracy and precision for a cow-side instrument at the tested iCa concentrations. Investigation of the instrument under field conditions is warranted.

**Key Words:** ionized calcium, cow-side instrument, dairy cattle

**362 Assessment of daily activity patterns in lactating dairy cows diagnosed with metritis.** Santiago Bas\*, Adrian A. Barragan, Juan M. Piñeiro, Gustavo M. Schuenemann, Päivi J. Rajala-Schultz, and Troy A. Brick, *Department of Veterinary Preventive Medicine, College of Veterinary Medicine, The Ohio State University, Columbus, OH.*

Metritis (MET) is a prevalent uterine disease that affects dairy cows, and causes substantial economic losses due to reduced milk yield, delayed pregnancy, cost of treatments, and increased culling and death rates. The objective was to assess changes in daily activity patterns (i.e., number of steps, number of lying bouts, standing time and lying time) of lactating dairy cows diagnosed with MET using activity monitors (IceQube, IceRobotics, Edinburgh, UK). Lactating dairy cows ( $n = 60$ ) from one commercial dairy herd were enrolled. Primiparous (PRIM;  $n = 12$ ) and multiparous (MULT;  $n = 48$ ) cows were housed in the same pen and were milked 4 times daily. Cows diagnosed with MET ( $n = 30$ ) were matched by lactation number and DIM to cows without MET (noMET;  $n = 30$ ). On study d 1, MET was confirmed (using a metrichk device) by the presence of watery, reddish or brownish foul smelling vaginal discharge. In addition, activity monitors were placed on the hind legs of MET and noMET cows and were kept until study d 7. The daily number of steps (n/d), number of lying bouts (n/d), standing time (min/d) and lying time (min/d) were recorded. Cows showing any other signs of disease (e.g., lameness, mastitis) were not included. Data were analyzed using the MIXED procedure of SAS. Preliminary results showed no difference in the number of steps ( $P = 0.30$ ), number of lying bouts ( $P = 0.93$ ) standing time ( $P = 0.89$ ), or lying time ( $P = 0.89$ ) between MET and noMET cows. However, a different pattern of behavioral activity was observed between PRIM and MULT cows regardless of the MET status. PRIM cows had more steps ( $P < 0.0001$ ; PRIM = 1921; MULT = 1728), more lying bouts ( $P < 0.0001$ ; PRIM = 14; MULT = 11), spent more time standing ( $P = 0.01$ ; PRIM = 860; MULT = 779) and less time lying ( $P = 0.01$ ; PRIM = 580; MULT = 660) than MULT cows. Previous studies have reported differences in activity patterns between cows with and without MET. In the present study, PRIM and MULT cows were housed together and were milked 4 times daily; thus, on-farm management may affect daily behavioral activity patterns of lactating dairy cows regardless of the uterine health status postpartum.

**Key Words:** dairy, metritis, activity

**363 The effect of ketosis on milk production in early lactation.** Khaled Gohary\*, Todd Duffield, and Stephen LeBlanc, *Department of Population Medicine, Ontario Veterinary College, University of Guelph, Guelph, Ontario, Canada.*

The objective of this study was to evaluate the effect of ketosis on milk production in early lactation. Data from 1,156 cows enrolled in a previous study from 5 commercial dairy herds in Southern Ontario were analyzed. Blood samples from all cows were obtained 1 week before calving and once a week for the first 2 weeks following calving. Sera were submitted to the laboratory at the University of Guelph to measure blood analytes including  $\beta$ -hydroxybutyrate (BHBA). Results were categorized from 1.0 to 2.0 mmol/L in 0.2 mmol/L increments. Daily milk production for all cows was recorded until 63 d in milk (DIM). Using weekly milk production as an outcome, for each of the selected thresholds, 3 linear mixed models that accounted for repeated measures within cow were fitted to test the effects of ketosis in wk 1 or wk 2 postpartum, or both. The analyses were performed with farm as a random effect or as a fixed effect. Overall, as the concentration of BHBA increased, milk yield was lower in cows with ketosis compared with cows without ketosis. Ketotic cows produced less total milk to 63 DIM when ketosis persisted for the first 2 weeks after calving than if present in wk 1 or 2 (at BHBA  $\geq 1.4$  mmol/L: -97, -37, and -62 kg, for first 2 weeks after calving, wk 1, and wk 2 after calving, relative to cows without ketosis, respectively,  $P < 0.0001$ ). Cows ketotic at wk 2 only produced more milk in wk 1 than cows without ketosis (at BHBA  $\geq 1.4$  mmol/L:  $25.9 \pm 1.3$  kg/d and  $23.9 \pm 1.2$  kg/d, respectively,  $P = 0.004$ ). When farm was included as a fixed effect, there was a significant interaction between farm and ketosis in wk 2 alone for BHBA  $\geq 1.2$  mmol/L, and in wk 1 and 2 for BHBA  $\geq 1.6$  mmol/L. The effect of ketosis on yield varied among farms from no significant difference to 7.3 kg/d ( $P < 0.0001$ ). The degree and duration of ketosis were negatively associated with milk yield in early lactation, but the timing of onset of ketosis affects the association. The threshold at which ketosis reduces milk production in early lactation may vary among farms and this should be explored in a large data set from many herds.

**Key Words:** dairy cattle, ketosis, milk production

**364 Estimating glucose requirements of an activated immune system in lactating Holstein cows.** Sara K. Stoakes\*, Erin A. Nolan, David J. Valko, Mohannad Abuajamieh, Edith J. Mayorga, Jake Seibert, Maria V. Sanz Fernandez, Patrick J. Gordon, and Lance H. Baumgard, *Iowa State University, Ames, IA.*

Activated immune cells are obligate glucose utilizers; thus, study objectives were to estimate the quantity of whole body glucose utilization during an IV endotoxin challenge. Fasting lactating Holstein cows ( $718 \pm 16$  kg;  $169 \pm 7$  DIM) were jugular catheterized and assigned 1 of 3 bolus treatments: control (CON; 5 mL saline;  $n = 6$ ), lipopolysaccharide infused (LPS;  $1.5 \mu\text{g/kg BW}$ ; *E. coli* 055:B5;  $n = 6$ ), and LPS + euglycemic clamp (LPS-Eu;  $1.5 \mu\text{g/kg BW}$ ; 50% dextrose infusion;  $n = 6$ ). After infusion, blood glucose was determined every 10 min and dextrose infusion was adjusted in LPS-Eu cows to maintain euglycemia. Blood samples were obtained 3, 6, 9, and 12 h post-bolus for further analysis. Cows were milked 6 and 12 h post-bolus. Milk yield decreased in LPS and LPS-Eu cows relative to CON (80%,  $P < 0.01$ ). Milk SCC was increased in LPS relative to CON (48%,  $P = 0.02$ ) while LPS-Eu did not differ from either treatment. LPS and LPS-Eu cows were hyperglycemic for 3 h post-bolus, but thereafter glucose content decreased in LPS relative to LPS-Eu and CON cows (30%,  $P < 0.01$ ). Circulating insulin was and tended to be increased in LPS-Eu (80%,  $P = 0.01$ ) and LPS (72%,  $P = 0.06$ ) relative to CON. Plasma NEFA, BHBA, and Ca were decreased ( $P < 0.01$ ) in LPS and LPS-Eu relative to CON (46, 53, and 46%, respectively). Plasma haptoglobin and L-lactate were increased ( $P < 0.01$ ) in LPS and LPS-Eu cows relative to CON (72 and 62%, respectively). Serum amyloid A was increased in LPS (47%,  $P$

= 0.01) and tended to be increased in LPS-Eu (34%,  $P = 0.09$ ) compared with CON cows. White blood cells were decreased in LPS and LPS-Eu relative to CON cows ( $P < 0.01$ ). Total glucose deficit during the 12 h post-bolus was calculated as the decrease in the amount of glucose required to synthesize milk (relative to pre-infusion levels) plus the amount of glucose infused to maintain euglycemia (in LPS-Eu cows only). Glucose deficit for CON, LPS, and LPS-Eu cows was 483, 1259, and 1553 g, respectively. If this model is a proxy of glucose demands during an immune response, our data indicates an intensely activated immune system uses at least 90 g glucose/h and maintaining euglycemia does not rescue the decrease in milk synthesis.

**Key Words:** lipopolysaccharide

### 365 DNA methylation patterns in peripheral blood leukocytes as a marker of uterine function.

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The objective of this study was to assess the suitability of using DNA methylation patterns in peripheral blood leukocytes as a marker of uterine function in the dairy cow. Peripheral blood and endometrial tissues were obtained at 29 d postpartum from cows with subclinical endometritis (SCE,  $n = 6$ ) and control cows (CON,  $n = 6$ ). DNA was extracted from peripheral blood leukocytes and endometrial tissues and DNA methylation was measured using Methylated DNA immunoprecipitation (MeDIP) in combination with microarrays. Genome-wide

DNA methylation was assessed using a custom 400K Agilent microarray (GPL16270). The agreement between DNA methylation measured in leukocytes and in uterine tissue was assessed using Pearson correlation between log ratio in leukocytes and log ratio in intercaruncular and caruncular endometrium. Correlations were considered high if either of the 2 correlation coefficients was greater than 70%. All analyses were performed using SAS 9.2. Genes that were highly correlated between leukocytes and endometrial tissue were submitted for pathway enrichment analysis using PANTHER (Protein Analysis THrough Evolutionary Relationships). 9,733 DNA methylation probes had high (>70%) correlation between blood and endometrial tissue. These probes mapped to a total of 3,329 genes. Enrichment analysis identified several pathways of which the top 5 pathways were; gonadotropin releasing hormone receptor pathway (1.85 fold enrichment,  $P = 1.5 \times 10^{-4}$ ), oxytocin receptor mediated signaling pathway (2.7 fold enrichment,  $P = 3.17 \times 10^{-3}$ ), heterotrimeric G-protein signaling pathway-Gq  $\alpha$  and Go  $\alpha$  mediated pathway (1.9 fold enrichment,  $P = 4.2 \times 10^{-3}$ ), PI3 kinase pathway (2.5 fold enrichment,  $P = 4.3 \times 10^{-3}$ ), and Wnt signaling pathway (1.5 fold enrichment,  $P = 5 \times 10^{-3}$ ). DNA methylation in the blood is highly correlated with DNA methylation status in endometrial tissue. In genes that were highly correlated, there was significant enrichment for several biological pathways that regulate the reproductive and immune systems. This study provides support for the efficacy of peripheral blood leukocytes as a marker of uterine function. Further research will establish if any of these markers are associated with improved reproductive function.

**Key Words:** epigenetics, reproduction



# ARPAS Symposium: Reproductive efficiency of beef cows— Current status and new technologies

## 366 Nutrition and management of cows—Supplementation and feed additives.

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Reproductive efficiency is the primary factor affecting profitability of a cow/calf enterprise. The objective is to review how nutrition affects reproduction in beef females and subsequent calf performance. Nutrition has profound effects on reproduction in beef females. Body condition is an indicator of nutritional status and when used in conjunction with BW change can provide a useful method to assess reproduction. Body energy reserve at calving is the most important factor influencing pregnancy rate in beef females. Energy and protein are the nutrients required in the greatest amounts and are the first priority in nutritional programs to optimize reproduction. Beef females underfed and/or in poor body condition lack ovarian activity as a result of suppression of pulsatile release of LH under the control of GnRH. Factors affecting the postpartum interval and pregnancy rate include breed type, suckling status, age, dystocia, energy and protein supplementation pre and post calving, and BCS pre and post calving. Using management strategies to influence when a beef female calves during the calving season affects future productivity of both dam and offspring. Feeding an ionophore results in earlier return to estrus postpartum. The effect of feeding fat pre or postpartum on reproductive performance in beef females has been extensively researched but results are inconclusive. Recent research has evaluated how energy restriction after AI affects embryo development and survival. Nutritional considerations and effects on reproduction have focused on postnatal development; however, prenatal nutrition appears to have potential effects on subsequent reproductive performance in beef cattle. No feed ingredient exists that will compensate for a diet deficient in any nutrient or poor body condition score.

**Key Words:** beef cow, nutrition, reproduction

**367 Selection of a calving season.** R. N. Funston\*<sup>1</sup>, E. E. Grings<sup>2</sup>, A. J. Roberts<sup>3</sup>, and B. T. Tibbitts<sup>4</sup>, <sup>1</sup>University of Nebraska West Central Research and Extension Center, North Platte, NE, <sup>2</sup>South Dakota State University, Brookings, SD, <sup>3</sup>Fort Keogh Livestock and Range Research Laboratory, Miles City, MT, <sup>4</sup>University of Nebraska West Central Research and Extension Center, North Platte, NE.

Calving date affects cost and timing of production events. Due to the polyestrous nature of beef females, producers can choose a calving date that fits their production system and geographic region. Any time an entire production system is considered, decision making becomes complex. Any calving system, regardless of date, should address the relationship between nutritional requirements of beef females and the quality and quantity of available feed. Nutritional status of beef females is influenced by stage of production, and the environment, including; length of growing season, forage species, day length, topography, forage quality and availability, ambient temperature, annual rainfall, and weather extremes. These differences cause grazing and feeding strategies to vary across regions. Ideally, high nutrient demand at parturition and peak lactation overlaps with optimal weather conditions and seasonal peaks in forage quality, and lowest nutrient demand overlaps with lowest quality forage, to minimize supplemental feed cost. Calving systems that do not match nutritional demand with forage quality must address

potential nutrient deficits faced by breeding females, likely occurring in late gestation and early lactation. Alternative calving systems with higher feed costs need to justify alternative dates through increased revenue generated from higher market value, increased calf performance, or improved reproductive performance. Heat stress, resulting from high temperature and humidity, can reduce calf performance and negatively affect reproductive performance in both the male and female. Hot and humid regions may favor a breeding season during seasonally lower temperatures to minimize poor reproductive performance. Additionally, regions prone to freezing temperatures, heavy snowstorms, or other severe weather events, must consider such risks when choosing a calving date. Many differences exist across regions in regard to environment, production systems, and marketing strategies that contribute to the complexity of choosing a calving date; therefore, beef producers must make site-based decisions according to conditions present on their operation.

**Key Words:** calving date, calving season, reproduction

## 368 Advantages of current and future reproductive technologies for beef cattle production.

G. Cliff Lamb\*, Vitor R. G. Mercadante, Darren D. Henry, Pedro L. P. Fontes, and Nicolas DiLorenzo, North Florida Research and Education Center, University of Florida, Marianna, FL.

The refinement of current and development of new technologies aimed at increasing the productivity of the resources while minimizing the environmental impact will be critical to meet the global food demand in the near future. During the past 50 years assisted reproductive technologies have been developed and refined to increase the prolificacy and quality of calves from beef females. Artificial insemination (AI), estrous synchronization and fixed-time AI (TAI), semen and embryo cryopreservation, multiple ovulation and embryo transfer (MOET), in vitro fertilization, sex determination of sperm or embryos, and nuclear transfer are technologies that are used to enhance the production efficiency of beef systems. Development and implementation of these technologies is responsible for significant changes to world production of beef. Sales of beef semen for AI increased from 3.3 to 13.0 million units between 1993 and 2010 in Brazil, whereas that in the US has increased from 2.9 to 4.4 million units during the same period, likely as a result of the development of practical TAI systems that have allowed beef producers the opportunity to eliminate detection of estrus in their AI programs with a high degree of success. Similarly, the quantity of in vivo produced embryos transferred worldwide has increased from 361,000 in 1997 to 506,000 in 2012. In addition, during the last 15 years the transfer of in vitro produced embryos has increased more than 300%. Incorporating applied reproductive technologies continues to affect beef cattle production systems by providing producers opportunities to enhance genetics, reduce transfer of disease, advance fertility, and ultimately increase offspring value. Improvements in fertility and technology, reductions in cost, and improvements in ease of application will ensure that more cattle producers will adopt applied reproductive technologies in future years. However, incorporation of applied reproductive technologies into production systems will vary worldwide depending on cattle markets, infrastructure, production systems, and climate.

**Key Words:** beef cattle, reproductive technology, fertility

### 369 Use of ultrasonography to make management decisions.

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Transrectal ultrasonography has been available for making management decisions since the mid 1980s. This technology allows for the real-time visualization of internal structures (i.e., ovary and fetus) that are otherwise difficult to evaluate. The use of this technology in making reproductive management decisions can be divided into 3 key areas: (1) selection of animals to be kept in the breeding herd, (2) increasing the likelihood of reproductive success, and (3) pregnancy determination. When selecting replacement animals, ultrasonography can be utilized to assess antral follicle counts in females. Antral follicle counts have a direct impact on animal fertility. Around the start of the breeding season transrectal ultrasonography can be utilized to determine puberty status, reproductive tract score, and estrous cycling status. In addition it can be used to determine response to synchronization protocols (presence of a dominant follicle, ovarian cyst, and follicle diameter), or if other assisted

reproductive technologies are being utilized it can be used to predict success (superovulation response or presence of a CL for embryo transfer). Following insemination this technology can be used to determine pregnancy status as early as 28 d after insemination, fetal age and sex, and presence of multiple offspring. The largest limitations to the use of ultrasonography include the time and technical skills required, and the strain on the arm and shoulder. While handles can be used for pregnancy diagnosis exams, measurements on the ovary require consistent placement of the transducer that to date can only be accomplished manually. Future improvements that may overcome some of these limitations include better imaging software, larger hard drives allowing the storage of video clips, and improved diagnostic capabilities of the machine (i.e., software that will be able to use pixel density to categorize ovaries). Color doppler images are being explored for earlier pregnancy diagnosis by blood flow to CL. Thus transrectal ultrasonography has and will have a role in the successful reproductive management of cattle herds. USDA is an equal opportunity provider and employer.

**Key Words:** ultrasonography, reproductive management

# Beef Species Symposium: Keeping beef in the center of the plate— Meeting consumer demand in a period of reduced cattle numbers and increased prices

**370 Reducing antibiotic use in cattle: Making healthier cattle starting at conception.** Daniel Thomson\*, *Kansas State University, Manhattan, KS.*

Antibiotic use creates antibiotic resistance. Claims and concerns suggest that modern animal agriculture is dependent on antibiotic usage to compensate for poor hygiene, poor health and poor management of animals. Regardless, there is still much debate in the scientific communities on how antibiotics in food animal production contribute to antibiotic resistance in humans. Many countries have increased antibiotic regulation in hopes of decreasing antibiotic usage to decrease antibiotic resistance. Producers and veterinarians will need to make a paradigm shift from antibiotic residue avoidance alone to prevention of both antibiotic residues and antibiotic resistance in our production units. The beef industry will need to develop antibiotic stewardship programs that monitor antibiotic usage on farms, antibiotic resistance in target pathogens and antibiotic resistance in foodborne pathogens. Complete removal of antibiotics from use in animal agriculture would not be prudent or practical for animal health and well-being. The antibiotics used in food animal medicine and important for human medicine recognized as potential resistance issues are macrolides, cephalosporins and fluoroquinolones. The most important foodborne pathogens in antibiotic resistance in human medicine are *Salmonella*, *E. coli* and *Campylobacter* species. The most common reasons for use of antibiotics in beef and dairy beef cattle production are bovine respiratory disease, neonatal calf scours, mastitis, foot rot and pink eye. The most common use of feed-grade antibiotics in beef cattle is prevention of liver abscesses and control of bovine respiratory disease. In the end, strategies that improve animal health will decrease antibiotic usage, which by theory will decrease antibiotic resistance. Antibiotic usage can be decreased in modern beef production systems if focus is applied to improvements in pre- and perinatal nutrition, neonatal calf housing and management, weaning calf management, marketing systems, transportation, receiving calf programs and nutritional management of finishing cattle.

**Key Words:** beef cattle, health, antibiotics

**371 Can we produce more with less? A critical look at technology in the feedlot sector.** Clint R. Krehbiel\*<sup>1</sup>, Casey L. Maxwell<sup>1</sup>, Bryan C. Bernhard<sup>2</sup>, Blake K. Wilson<sup>1</sup>, Cathy L. Haviland<sup>1</sup>, Michelle S. Calvo-Lorenzo<sup>1</sup>, Sara E. Place<sup>1</sup>, Deb L. VanOverbeke<sup>1</sup>, Gretchen G. Mafi<sup>1</sup>, Chris J. Richards<sup>1</sup>, and D. L. Step<sup>1</sup>, <sup>1</sup>*Oklahoma State University, Stillwater, OK*, <sup>2</sup>*Texas Tech University, Lubbock, TX.*

Adoption of technologies has enhanced beef cattle production and efficiency. With the increasing world population, use of technologies that are economically, environmentally, and socially sustainable is needed to meet protein demand. Long-term use of growth-enhancing technologies (implants, ionophores,  $\beta$ -adrenergic agonists) in the feedlot sector has proven that compounds enhance lean-tissue deposition, and changes in performance result in an economic benefit to both consumers and producers. Land needed and environmental impact is decreased when growth technologies are used. We determined the effects of feedlot production systems with the use of growth technologies compared with an all-natural program on cattle performance and carcass characteristics.

Treatments consisted of an all-natural treatment (NAT; no technologies), a conventional treatment (CONV; ionophore and implant), and a CONV treatment with the addition of a  $\beta$ -adrenergic agonist (CONV-Z). There was a 35.7% increase in carcass gain and a 32.6% improvement in carcass efficiency for CONV-Z steers compared with NAT steers. Hot carcass weight was increased by 46 kg for CONV-Z steers compared with NAT steers. An increase in yield grade (YG) 1 and a decrease in YG 3 carcasses was observed for CONV-Z compared with CONV steers. Based on per capita beef consumption of 37.2 kg, added HCW for a single CONV-Z steer compared with a NAT steer would feed 1.2 more US Citizens per year. As society has concern over technologies used in animal production, it is imperative to communicate how increased animal productivity, reduced environmental impact and improved animal well-being are interrelated. Our results suggest that growth promoting products do not affect behavior, mobility, or the overall observed health and well-being of finishing beef steers. Food security depends on developing technologies for improving production efficiencies of beef cattle while adapting to climate change, protecting animal health, and improving the nutritional quality and safety of meat products for consumers. Developing technologies that can improve beef cattle production while minimizing environmental impact, enhancing animal well-being, and ensuring the health and safety of consumers should be our goal.

**Key Words:** beef cattle, growth-enhancing technologies, production

**372 Keeping the eating experience enjoyable: Postmortem management of heavy carcasses.** Chris Calkins\*, *University of Nebraska, Lincoln, NE.*

Tenderness is one of the most important palatability traits for beef. As beef prices rise, the importance of palatability also increases. Consumer demand is a reflection of perceived value (including eating quality) compared with price. Although demand is increasing, consumers indicate that beef does not always perform on tenderness. Thus, postmortem changes that influence tenderness are critical to understand and control. The improvement in tenderness that occurs during cooler aging is well known and cannot be overlooked. Identifying carcasses with desirable tenderness characteristics offers the opportunity to assure consumers of beef quality. Many studies have shown the consumers are willing to pay a premium for beef that performs in tenderness. The USDA recently approved a marketing claim for beef tenderness, reflecting increased consumer interest. The ability to identify beef that meets consumer expectations is of increasing interest to the industry and may hold the key to sustained customer satisfaction. Branded beef is an attempt to provide that assurance. Similarly, optimal use of individual muscles increases the likelihood of a desirable eating experience. Projects such as muscle profiling have helped to identify potential value upgrade opportunities. Intermediately priced cuts provide the chance for consumers to continue to eat beef in the face of rising costs. Success is most likely when the marketplace puts a premium on tenderness and systems are in place to reward its presence. Clear links between consumer demands and producer compensation help to encourage production of high-quality, tender beef.

**Key Words:** beef, palatability, tenderness



# Breeding and Genetics: Application and methods—Dairy I

**373 Montbéliarde × Holstein and Viking Red × Holstein crossbreds compared with pure Holsteins during first lactation in eight high-performance dairies in Minnesota: I. Production.** A. R. Hazel\*<sup>1</sup>, B. J. Heins<sup>2</sup>, and L. B. Hansen<sup>1</sup>, <sup>1</sup>University of Minnesota, St. Paul, MN, <sup>2</sup>West Central Research and Outreach Center, Morris, MN.

Montbéliarde × Holstein crossbred cows (MH, n = 484) and Viking Red × Holstein crossbred cows (VH, n = 510) were compared with pure Holstein cows (HO, n = 887) for 305-d milk, fat, and protein production and somatic cell score (SCS) during first lactation. A designed crossbreeding study was initiated in 8 commercial dairy herds in Minnesota in 2008. The first generation cows (MH, VH, and HO) calved for the first time from December 2010 to November 2013. Best Prediction was used to calculate production and SCS for 305-d lactations with adjustment for age at calving, and records less than 305 d were projected to 305 d. Independent variables for statistical analysis of all traits included the fixed effects of herd-year-season of calving, breed, interaction of herd-year-season and breed group, and random sire effect nested within breed. The MH, VH, and HO did not differ significantly for age at first calving (23.8 mo, 23.7 mo, and 23.8 mo, respectively). The MH and HO did not differ significantly ( $P = 0.81$ ) for milk volume (11,304 kg vs. 11,272 kg, respectively); however, VH had significantly less ( $P < 0.01$ ) milk volume (−495 kg) than HO. Fat solids production was similar ( $P > 0.17$ ) for MH (429 kg) and VH (423 kg) compared with HO (423 kg). However, the MH had significantly greater ( $P < 0.01$ ) protein production (355 kg vs. 344 kg) and fat plus protein production (784 kg vs. 767 kg) compared with HO. The protein production and fat plus protein production of VH (344 kg and 767 kg, respectively) were similar ( $P > 0.94$ ) to pure HO. The MH (2.2), and VH (2.2) were also similar ( $P > 0.27$ ) to HO (2.1) for SCS. The results have potential effect on the calculation of lifetime profit of cows because revenue from milk solids is the largest component of total revenue in many regions of the world.

**Key Words:** crossbreeding, Montbéliarde, Viking Red

**374 Montbéliarde × Holstein and Viking Red × Holstein crossbreds compared with pure Holsteins during first lactation in eight high-performance dairies in Minnesota: II. Fertility, survival, and conformation.** A. R. Hazel\*<sup>1</sup>, B. J. Heins<sup>2</sup>, and L. B. Hansen<sup>1</sup>, <sup>1</sup>University of Minnesota, St. Paul, MN, <sup>2</sup>West Central Research and Outreach Center, Morris, MN.

Montbéliarde × Holstein crossbred cows (MH) and Viking Red × Holstein crossbred cows (VH) were compared with pure Holstein cows (HO) for fertility, survival, and conformation during first lactation. A designed crossbreeding study was initiated in 8 commercial dairy herds in Minnesota in 2008. The first-generation cows (MH, VH, and HO) calved for the first time from December 2010 to November 2013. Cows (n = 451, 487, and 794 for MH, VH, and HO, respectively) were required to have at least 250 DIM for days open (DO), and cows with DO greater than 250 d were truncated to 250 d. For survival to second calving (n = 415, 450, and 697 for MH, VH, and HO, respectively), cows had at least 20 mo of opportunity to calve a second time. Conformation was scored once between 4 and 150 d on a linear, 9-point scale. Independent variables for the analysis of DO and conformation traits included herd-year-season, breed, interaction of herd-year-season and breed, and random sire effect nested within breed. Chi-squared was used to test survival to second calving. The MH (116 d) had significantly fewer DO ( $P < 0.05$ ) than

HO (126 d); however, VH (122 d) were similar to HO for DO. The MH (86%) had significantly greater ( $P < 0.01$ ) survival to second calving compared with HO (80%), but VH (83%) were similar to HO. The MH and VH had greater udder depth than HO (Table 1); however, culling for udder conformation did not differ for breed groups. The MH and VH had less stature, angularity, and body depth than HO (Table 1), indicating they had smaller body size with more body condition.

**Table 1 (Abstr. 374).** Least squares means of conformation scores for Montbéliarde × Holstein crossbred cows (MH) and Viking Red × Holstein crossbred cows (VH) compared with pure Holstein cows (HO) in 8 high-input dairies in Minnesota

Trait	HO (n = 1,137)	MH (n = 523)	VH (n = 564)
Stature (9 = tall)	5.4	4.7**	3.9**
Angularity (9 = angular)	5.6	2.6**	4.2**
Body depth (9 = deep)	5.1	4.1**	4.4**
Foot angle (9 = steep)	5.6	6.6**	5.5
Udder depth (9 = shallow)	6.9	5.4**	6.2**

\*\*Difference from HO ( $P < 0.01$ ).

**Key Words:** crossbreeding

**375 Milk production and fertility performance of Holstein, Friesian, Jersey, Holstein x Jersey, and Friesian x Jersey crossbred cows on commercial Irish farms.** E. L. Coffey\*<sup>1,2</sup>, B. Horan<sup>1</sup>, R. D. Evans<sup>3</sup>, K. M. Pierce<sup>2</sup>, and D. P. Berry<sup>1</sup>, <sup>1</sup>Teagasc Moorepark, Fermoy, Co. Cork, Ireland, <sup>2</sup>School of Agricultural and Food Science, UCD, Dublin, Ireland, <sup>3</sup>Irish Cattle Breeding Federation, Bandon, Co. Cork, Ireland.

There is renewed interest in dairy crossbreeding as a means to further augment farm productivity and profitability. Research experiments have demonstrated the benefits of crossbreeding on animal performance and financial efficiency, exploiting both additive and non-additive genetic effects. The objective of this study was to compare the biological performance of Holstein, Friesian, Jersey, Holstein × Jersey and Friesian × Jersey crossbred cows using commercial dairy herds practicing crossbreeding over a 5-year period. Milk production and fertility information from the national database on 11,808 cows from 40 spring calving dairy herds that adopted crossbreeding between Holstein and Jersey breeds from 2008 to 2012 inclusive were available. Obvious data errors were removed. Least squares means for traits of interest were estimated for purebred and crossbred animals using linear mixed models. Holstein-Jersey first cross cows produced more ( $P < 0.001$ ) milk solids (heterosis = 6.5%), calved earlier ( $P < 0.001$ ) as heifers (heterosis = 1.6%), had a shorter ( $P < 0.001$ ) calving interval (heterosis = 2.1%) and had a higher ( $P < 0.05$ ) submission rates in the first 21 d of the breeding season (heterosis = 4.5%) relative to their purebred parent breeds. Friesian-Jersey first cross cows had a lower ( $P < 0.001$ ) somatic cell count (heterosis = 4%), a shorter ( $P < 0.05$ ) calving interval (heterosis = 2.5%) and a higher ( $P < 0.01$ ) submission rate in the first 21 d of the breeding season (heterosis = 1.2%) relative to their purebred parent breeds. Results were consistent with findings from controlled experiments. Breed complementarity and heterosis attainable from crossbreeding resulted in superior performance in crossbreds relative to their purebred parents.

**Key Words:** crossbreeding, Jersey, heterosis

**376 Fertility and production of 3-breed and third-generation Holstein-sired crossbreds compared with pure Holstein cows in a seasonal pasture production system.** Bradley J. Heins<sup>1,2</sup>, Amy R. Hazel<sup>2</sup>, and Leslie B. Hansen<sup>2</sup>, <sup>1</sup>University of Minnesota, West Central Research and Outreach Center, Morris, MN, <sup>2</sup>University of Minnesota, Saint Paul, MN.

Three-breed crossbred cows (Montbéliarde × Jersey × Holstein (HO) and Jersey × Montbéliarde × HO, n = 64); and HO-sired crossbred cows HO × Montbéliarde × Jersey × HO and HO × Jersey × Montbéliarde × HO, n = 87) were compared with pure HO cows (n = 138) for 305-d milk, fat, and protein production; SCS; and days open (DO) during their first 3 lactations. Cows were housed at the University of Minnesota West Central Research and Outreach Center, Morris, Minnesota, a grazing herd, and calved from March 2007 to May 2014. Best Prediction was used to calculate actual production (milk, fat, protein, and SCS) for 305-d lactations with adjustment for age at calving, and records less than 305 d were projected to 305 d. The DO was measured as actual DO for cows and cows were required to be at least 250 d in milk and those with greater than 250 DO were truncated to 250 d. Independent variables for statistical analysis were the fixed effects of season of calving, genetic group (HO, versus 3-breed crossbred versus Holstein-sired crossbred), parity nested within genetic group, and cow nested within genetic group, which was a random effect. Across the first 3 lactations, the 3-breed crossbred (485 kg) cows and Holstein-sired crossbred (499 kg) cows were not different ( $P > 0.05$ ) from pure HO (495 kg) cows for fat plus protein production. Across lactations, 3-breed crossbred cows (2.78) and HO-sired crossbreds (2.92) were similar to pure HO cows (2.90) for SCS. The 3-breed crossbred cows (111 d) and HO-sired crossbreds (125 d) had significantly ( $P < 0.05$ ) fewer DO than pure HO (150 d) cows during first lactation. During second lactation, the DO for the pure HO cows increased to 162 d, and the 3-breed crossbred ( $P < 0.05$ ) and Holstein-sired crossbreds ( $P < 0.10$ ) had a fewer DO (130 d and 136 d, respectively) compared with the pure HO cows. Mating 3-breed crossbreds to HO bulls may have advantages in a rotational crossbreeding system. Three distinct breeds is likely optimum for a rotational crossbreeding system.

**Key Words:** crossbreeding, days open, pasture

**377 Relationships between yield, fitness, and type traits in US Brown Swiss.** Keith D. Gibson\* and Chad D. Dechow, *The Pennsylvania State University, State College, PA.*

Genetic parameters have often been estimated for the US Holstein population, but infrequently for Brown Swiss. The objective of this research was to evaluate heritabilities and correlations of fitness with yield and type traits for Brown Swiss cattle born in 2000 and later. The data consisted of 105,164 first through fifth lactation yield and somatic cell score (SCS) records from 45,464 cows with 82,761 d open (DO) and 31,105 productive life (PL) records. Approximately 52,250 of the records had values for 17 type traits and 41,074 had values for milking speed. These data were analyzed using a series of 3 trait models in ASREML that included fixed effects for lactation, age, and herd-calving cluster. Random effects were animal, permanent environment, and residual. The heritabilities of milk, fat, and protein yield were 0.30, 0.20, and 0.20, respectively. The heritabilities of PL, DO, SCS, and milking speed were 0.11, 0.03, 0.12, and 0.26, respectively. Yield traits had strong positive correlations with PL (0.63 to 0.71), whereas DO (−0.69) and SCS (−0.60) had strong negative correlations with PL. Milking speed had a moderate positive correlation with PL (0.26) and DO (0.19) and a slight negative correlation with SCS (−0.10). Yield traits had a slight unfavorable genetic correlation with DO (0.11 to

0.18). However, rear udder width (0.55), final score (0.45), body depth (0.43), dairy form (0.43), rear udder height (0.32), and stature (0.27), were more unfavorably correlated with DO than was yield. Yield was positively correlated with rear udder height (0.19 to 0.25) and rear udder width (0.34 to 0.48), and negatively correlated with udder depth (−0.40 to −0.24) and fore udder attachment (−0.21 to −0.04). However, udder depth (−0.26) and fore udder attachment (−0.14) were more favorably correlated with SCS than rear udder height and rear udder width (−0.02 and 0.18, respectively). Of the udder traits, fore udder attachment, rear udder width, rear udder height, and udder cleft were most favorably correlated with PL (0.31 to 0.44). These updated genetic parameters will allow for improved genetic selection in the Brown Swiss breed.

**Key Words:** Brown Swiss, correlation, productive life

**378 Genetic disorders in Indian dairy cattle.** Prasad Dinkar Deshpande\*, Marimuthu Swaminathan, Jayant Ramchandra Khadse, Hemant Dasharath Kadam, Ashok Bhaskarrao Pande, Sumit Prakashrao Totade, and Priyanka Shivaji Garad, *BAIF Development Research Foundation, Central Research Station, Urulikanchan, Pune, Maharashtra, India.*

The advancements in cattle genetics and genomics have widened the scope of understanding number of hereditary diseases in cattle, leading to the development of early diagnostic techniques to detect presence of lethal or mutant allele for a specific phenotype. Bovine Leukocyte Adhesion Deficiency decreases host resistance and increases susceptibility to fatal illnesses. Citrullinemia is a disorder of urea cycle and this condition leads to increase levels of ammonia accumulation in blood and tissues, and subsequent death. Deficiency of uridine monophosphate synthase (DUMPS) causes prenatal embryonic mortality before d 40 of gestation. Factor XI results in a bleeding disorder and the affected animals (homozygotes) have hemophilia-like disorder. Although the frequency of these alleles in the population is usually very low, it can be easily increased if heterozygous (carrier) sires are used for large scale breeding through artificial insemination program resulting in increase in the proportion of lethal alleles. As per minimum standard protocol laid down by Government of India, it is mandatory to screen each stud bull before its entry to semen collection to rule out genetically transmitted diseases. All the stud bulls were screened by single nucleotide polymorphism (SNP) genotyping for BLAD, citrullinemia, DUMPS by Custom Designed Assays from Life Technologies (a brand of Thermo Fisher Scientific) and temperature melting curve for Factor XI. BAIF Development Research Foundation has a regular genetic disorder-screening program for its own stud bull farm. To date, 400 stud bulls have been screened and not a single male carrier for such genetic disorders has been found (Table 1).

**Table 1 (Abstr. 378).** Screening for disorders in Indian stud bulls

Sr. No.	Breed	Tests carried out	No. of stud bulls screened
1	Indigenous cattle and buffaloes	Factor XI, BLAD, citrullinemia	150
2	HF and HF crossbreds	Factor XI, BLAD, citrullinemia, DUMPS	150
3	Jersey and Jersey crossbreds	Factor XI, BLAD, citrullinemia	100

**Key Words:** genetic disorder, stud bull

**380 SNP-analysis solves questions on the genetic background of the rare Dutch breed of Red Friesian cattle.** Kor Oldenbroek\*<sup>1</sup>, Ina Hulsegge<sup>2,3</sup>, Jack Windig<sup>2,3</sup>, and Mario Calus<sup>2,3</sup>, <sup>1</sup>*Centre for Genetic Resources the Netherlands, Wageningen, the Netherlands*, <sup>2</sup>*Animal Breeding and Genomics Centre, Wageningen, the Netherlands*, <sup>3</sup>*Wageningen UR Livestock Research, Wageningen, the Netherlands*.

The Red Friesian cattle population went through a severe bottleneck around 1990. It revived due to the availability in the Dutch genebank of semen from bulls born between 1960 and 1970. Nowadays, it counts approximately 500 females and together with the bulls in the genebank, some 50 bulls are available for breeding. Three questions were put forward by the breeding organization: (1) is the Red Friesian cattle a unique breed or is it strongly related to the Black and White Dutch Friesian? (2) Is it relevant to distinguish the 5 different sire lines within the breed? (3) Are 2 herds of non-registered Red Friesian Cattle related to the registered population? DNA samples were extracted from 25 genebank bulls and 43 females from the 5 sire lines and 2 non-registered herds from semen and hair roots. These were analyzed with the BovineSNP50 BeadChip. After editing 42552 SNP's remained from the 54609 observed. Genetic relationships were evaluated by analyzing admixture using STRUCTURE and analyzing the average relatedness within and between lines. These revealed that (1) the Red Friesian Cattle is a breed in its own right, strongly related to the Black and White Dutch Friesian, (2) a large overlap in genetic variation exists between the 5 sire lines, and (3) the 2 non-registered herds although highly related to the registered population, contain genetic diversity not present in the 5 lines. For one of these herds this diversity originated from some Holstein Friesian sires used in the past. The other herd had been bred in isolation from the rest of the breed leading on the one hand to a high amount of inbreeding (25%) and associated loss of diversity, but on the other hand to conservation of diversity not present in the rest of the breed.

**Key Words:** SNP, genetic variation, genetic relatedness

**381 Genetic variation and breeding values of casein in Holstein cattle using novel FTIR predictive modeling.** Herwin Eding<sup>1</sup>, Gerben de Jong\*<sup>1</sup>, Hiemke Knijn<sup>1</sup>, Jan Rademaker<sup>2</sup>, and Nisha Shetty<sup>2</sup>, <sup>1</sup>*CRV, Arnhem, the Netherlands*, <sup>2</sup>*Qlip, Zutphen, the Netherlands*.

Based on reference method measurements and Fourier transform infrared (FTIR) spectra, prediction equations for casein in raw milk were calculated. Subsequently casein content was predicted from multiple milk samples of 1.3 million cows from March 1, 2012, until February 23, 2013. After editing 8.5 million test-day records were available for analysis. The average casein content observed in milk was 2.77% (m/m), 77 percent of the total milk protein, with a standard deviation of 0.37% (m/m). Genetic parameter estimation was carried out on 61 000 Holstein cows, with 480 000 test days. The found heritability was 0.30, 0.34 and 0.39 for first, second and third lactation, respectively. The genetic correlation among the lactations ranged from 0.52 to 0.60. Genetic correlation with overall protein was 0.982. The genetic standard deviation for casein:protein ratio was 1.36 percent, with an heritability of 29 percent. The genetic correlation between casein:protein ratio and casein and protein yield was 0.23 and 0.35 respectively, while the phenotypic correlation was 0.01 lower, showing the ratio trait is independent from the yield traits. A genetic evaluation for protein yield and content, casein yield and content and casein:protein ratio was carried out on the whole population. This showed that selection for casein:protein ratio increases casein content but decrease casein and protein yield. In conclusion, casein yield showed to be a promising parameter in a cheese merit index.

Efficient assessment of casein using the novel FTIR prediction equation allowed for access to sufficient data being available. This research was supported by the EU and the province of Gelderland, the Netherlands.

**Key Words:** dairy cattle, FTIR, infrared

**382 Use of infrared spectroscopy to enhance technological and nutritional quality of milk: Genetic components of FTIR spectra and breeding values estimates in Italian Simmental cattle.** Valentina Bonfatti\*<sup>1</sup>, Daniele Vicario<sup>2</sup>, Lorenzo Degano<sup>2</sup>, Andrea Lugo<sup>3</sup>, and Paolo Carnier<sup>1</sup>, <sup>1</sup>*Department of Comparative Biomedicine and Food Science, University of Padova, Legnaro, Italy*, <sup>2</sup>*National Simmental Cattle Breeders Association, Udine, Italy*, <sup>3</sup>*Friuli Venezia Giulia Milk Recording Agency, Codroipo, Italy*.

The aim of this study was to develop a general procedure for routine estimation of breeding values in the Italian Simmental cattle population for several innovative FTIR-predicted traits, using a direct prediction method, where breeding values estimation is directly conducted on milk FTIR spectral variables instead of FTIR-predicted traits. Breeding values for all new traits are then derived from the genetic component of the spectra. The investigated traits were: detailed milk fatty acid composition measured by 2D-GC, milk coagulation properties assessed by renneting meter analysis, pH, contents of major casein and whey protein fractions measured by RP-HPLC, cheese yield, curd composition and recovery rates of protein and fat in the curd, measured in individual micro-cheese making procedures, contents of major minerals by ICP-OES and of lactoferrin quantified by ELISA. FTIR calibrations were developed using more than 1,000 milk samples for all traits, with the exception of minerals (820 samples) and lactoferrin (635 samples). The calibration set included approximately 90% Simmental and 10% Holstein Friesian cows. A total of 100,272 milk spectra from 11,216 Italian Simmental cows were collected during the routine milk recording and stored. Principal component analysis of the unprocessed spectral data resulted in 8 latent traits that explained 99% of the total spectral variability. Restricted maximum likelihood was used to estimate (co)variance components of the latent variables. Heritability for these 8 latent traits ranged from 0.09 to 0.40. Variance ratios of the permanent environmental effect were between 0.10 and 0.25 and residual variance ratios ranged from 0.39 to 0.81. Variance components and the breeding values of original spectral transmittances were obtained by back transformation. Breeding values for the new milk traits were then predicted through FTIR calibrations as values correlated with the genetic component of the FTIR spectra. The procedure yielded estimates of breeding values for all the investigated traits for more than 10,000 Simmental animals.

**Key Words:** spectroscopy, breeding value, milk quality

**383 Association between *INHA* A192G polymorphism and dairy traits in Holstein cattle of Antioquia-Colombia.** Stephania Madrid\*, Albeiro López, and Julián Echeverri, *Universidad Nacional de Colombia, Medellín, Antioquia, Colombia*.

Productive and reproductive performance are some of the most important parameters in dairy cattle because they affect profitability of these systems. Marker-assisted selection is a useful tool to improve efficiency of dairy herds, because it allows identification of genetic differences related to a higher productive potential. Inhibin protein belongs to the transforming growth factor  $\beta$  super family, and its function is to modulate the secretion and action of the FSH. The goal of this research was to determine the effect of A192G polymorphism on productive and reproductive traits in Antioquia Holstein cows. The PCR-RFLP technique



was used to amplify a segment of 249bp of the bovine inhibin  $\alpha$  gene (*INHA*), and then digested with the enzyme *MspI*. It was analyzed 1240 lactations from 356 dairy cows. The effect of the *INHA* genotypes on the traits was determinate by a mixed linear model and Tukey test was used to establish significant differences between means for the 3 genotypes. The effect of allelic substitution was made through a linear regression model where the genotypes AA, AG and GG were transformed into a quantitative scale of 0, 1 and 2, according to the number of G alleles. In Antioquia Holstein cattle the most common genotype was AG (0.529), followed by AA (0.269) and GG (0.201). Allele frequencies were 0.534 for A and 0.466 for G. The *INHA* genotypes did not exerted a significant effect on the principal productive parameters, except for fat percentage, where GG animals presented the highest value for this trait. Despite it was not a significant effect of this polymorphism on reproductive parameters, Tukey test showed that G allele tended to have a detrimental effect on female fertility, increasing the calving interval and the number of services per conception, decreasing the reproductive efficiency of the herds. Further studies are required to determine whether this SNP may be used as a molecular marker.

**Key Words:** inhibin, PCR-RFLP, SNP

### **384 Genome-wide association studies for fertility traits in Brown Swiss cattle using single SNP regression and Bayesian approaches applied to high-density SNP array information.**

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Large-scale, in-depth, genome-wide analyses of various economically important traits are now possible due to the availability of medium (50K; 54,609 SNP) and high-density (HD; 777,962 SNP) SNP arrays. Genome-

wide association studies (GWAS) provide a powerful tool for identifying associations between phenotypes and variants in the underlying genome. Here we present GWAS results for 5 fertility traits in Brown Swiss cattle. Traits analyzed included non-return-rates in heifers (NRR\_H) and cows (NRR\_C), days to first service (DFS), and intervals between first and last insemination in heifers (IFL\_H) and in cows (IFL\_C). Imputation from 50K to HD was carried out with FImpute. After filtering out SNPs with minor allele frequency <0.05, a total of 654,847 imputed SNPs on 29 bovine autosomes remained. Deregressed breeding values of 1,502 (IFL\_H) to 3,379 (NRR\_C) progeny-tested bulls were used as phenotypes. Single SNP GWAS (M1) were conducted with the SNP & Variation Suite package GoldenHelix. The Efficient Mixed-Model Association expedited (EMMAX) algorithm was applied to account for population stratification. Additionally, bulls with more than 75% Original Braunvieh pedigree background were excluded. Significance levels were determined by false discovery rates. Multi-SNP GWAS (M2) using Bayesian methodology implemented in GenSel were also conducted. In those analyses, all SNPs were fitted simultaneously. A 1 Mb non-overlapping window approach that accumulated contributions of adjacent SNPs was used to identify associated genomic regions. The M1 analyses using genome-wide significance levels showed that regions on BTA3, 17, and 25 were associated with NRR\_C, a region on BTA29 was associated with DFS, and a region on BTA8 was associated with IFL\_C. The significant SNPs in these regions were also represented in the 1 Mb windows accounting for the largest proportion of genetic variance in the corresponding traits from M2. The promising results presented here provide evidence that specific regions of the genome are associated with fertility traits in Brown Swiss cattle.

**Key Words:** GWAS, fertility, dairy cattle

# Cell Biology Symposium: Regulation of growth through amino acid sensing

## 385 Role of amino acid transporters in amino acid sensing.

Peter M. Taylor\*, *College of Life Sciences, University of Dundee, Dundee, UK.*

Amino acid (AA) transporters have functional importance in nutrient sensing as well as in delivering tissue nutrient supplies. These transmembrane proteins mediate AA transfer and exchange between extracellular and various intracellular fluid compartments. AA transporters at the cell surface, particularly those for large neutral AA such as leucine, interact functionally with intracellular AA sensors and nutrient-signaling pathways which regulate metabolism; for example, the mTORC1 pathway which promotes cell growth and the GCN pathway activated by AA starvation. Upregulated expression of these AA transporter; for example, the leucine transporter SLC7A5 (System L1; LAT1), is required under some circumstances to initiate AA-dependent activation of the mTORC1 pathway. Transporter activity for leucine and other essential AA may be an important determinant of baseline insulin-sensitivity. Gastrointestinal-endocrine interactions contribute to dietary regulation of AA transporter expression and activity in the intestine. Certain AA transporters (e.g., SLC38A2, SLC38A9) may have dual receptor-transporter functions, operating as “transceptors” to sense extracellular (or intracellular) AA availability upstream of intracellular signaling pathways. SLC38A2 (System A; SNAT2) at the cell surface provides a repressive signal for gene transcription during AA sufficiency, thus echoing AA sensing by transceptor orthologs in yeast (e.g., Gap1). Expression and activity of SLC38A2 is upregulated by AA starvation by a mechanism dependent on both SLC38A2 gene transcription and enhanced stabilization of SLC38A2 protein. This forms part of an integrated cellular stress response to availability of nutrients, including unsaturated fatty acids which promote SLC38A2 protein degradation via the ubiquitin-proteasome system. SLC38A9 at the lysosomal membrane may act as the endosomal AA sensor for mTORC1 activation by glutamine and arginine. New opportunities for nutritional therapy may include targeting of AA transporters (or mechanisms which regulate their expression) to promote protein-anabolic signals for growth or retention of lean-tissue mass. Research funded by the Wellcome Trust, BBSRC UK and RERAD (Scottish Government).

**Key Words:** amino acid, membrane transport, cell signaling

## 386 Integration of amino acid signaling and metabolism in the mTORC1 pathway.

John Blenis\*, Gwen Buel, Anders Mutvei, Alfredo Csibi, Jing Li, Gina Lee, Sang Gyun Kim, and Andy Choo, *Sandra and Edward Meyer Cancer Center, Department of Pharmacology, Weill Cornell Medical College, New York, NY.*

The mTOR complex 1 (mTORC1) signaling pathway has evolved to sense and respond to amino acid availability, cellular energy status, surrounding oxygen concentrations and stress conditions. In addition, mTORC1 can be further activated by mitogen- and hormone-stimulated kinases including Akt, ERK, and RSK, and suppressed by mTORC1-regulated S6K1 via a variety of negative feedback loops. The integration of these multiple inputs control the strength and duration of downstream signaling, which is important in differentially regulating mTORC1-dependent processes such as protein synthesis and cellular metabolism. Importantly, amino acid-mediated regulation of mTORC1 is reportedly dominant over mitogen-dependent activation of mTORC1 signaling and therefore has attracted much interest as a therapeutic target in mTORC1-driven diseases such as obesity, diabetes, aging, and cancer.

The molecular basis for amino acid signaling is complex but many gaps in our knowledge of this regulatory system remain. We will discuss our recent observations regarding amino acid-dependent regulation of mTORC1 and the ability of mTORC1 to regulate cellular metabolism.

**Key Words:** mTOR complex, amino acid signaling, cellular metabolism

## 387 Integration of signals generated from nutrients, hormones, growth factors, and exercise.

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The stimulation of protein synthesis that occurs in skeletal muscle in response to either nutrient intake or a bout of resistance exercise requires activation of the protein kinase known as the mechanistic (a.k.a. mammalian) target of rapamycin in complex 1 (mTORC1). The activation state of mTORC1 is controlled by several upstream signaling pathways that function in a combinatorial manner to integrate positive (e.g., re-feeding and/or resistance exercise) or negative (e.g., inactivity or glucocorticoid treatment) inputs to the kinase. For example, amino acids and insulin act through independent pathways to activate mTORC1 in an additive manner. In contrast, hindlimb immobilization attenuates the stimulatory effect of leucine on mTORC1 activity in muscle. A growing body of evidence implicates a protein known as REDD1 (regulated in development and DNA damage response 1) as a critical regulator of mTORC1 signaling, and in a variety of studies REDD1 expression has been shown to be inversely proportional to the activation state of mTORC1. For example, REDD1 abundance is reduced and mTORC1 signaling is elevated after a bout of resistance exercise, but the opposite effects are observed after endurance exercise. Moreover, compared with freely fed rats, REDD1 abundance is elevated and mTORC1 signaling is reduced after an overnight fast, and re-feeding rapidly reverses both effects. The important role played by REDD1 in controlling mTORC1 activity is emphasized in studies demonstrating that both re-feeding and muscle contraction activate mTORC1 signaling to a significantly greater extent in muscle of REDD1 deficient mice compared with wildtype mice. This presentation will focus on the signaling pathways through which amino acids, insulin, and resistance exercise act to activate mTORC1 as well as the role of REDD1 in governing the stimulatory effect of these inputs on the kinase. Work in the author's laboratory is supported by NIH grants DK13499, DK15658, and DK094141.

**Key Words:** mTOR, amino acid, insulin

## 388 Distributed nutrient sensing in the integrated control of energy balance.

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The presence of nutrients in the gut following ingestion of a meal gives rise to multiple neural and humoral signals that depend on the mechanical and chemical properties of the ingested food. Specialized sensors within the gut are able to transduce these properties to activate a central brainstem-forebrain network that includes the brainstem dorsal vagal complex as well as forebrain hypothalamic and striatal sites. Both extrinsic nerves supplying the gut as well as nutrient stimulated, gut secretory factors mediate the transmission of gut of meal-related stimuli to this central network. Once activated, this network engages multiple effector systems that drive behavioral and physiological responses to

peripheral gut nutrient availability to determine energy balance. These effectors include the negative feedback control of food intake, hepatic glucose production, thermogenesis, and food seeking behavior for palatable diets. Activity in effector pathways can limit body weight gain and adiposity, as well as improve glucose tolerance and insulin sensitivity. The responsiveness of this sensory-motor network to meal-related signals is modulated by the neuroendocrine metabolic context in which signals are received and processed, depending on the adiposity of the organism. We have identified important roles for vagal and non-vagal gut afferents linking gut nutrient availability to activation of the brainstem-forebrain network. Furthermore, growing evidence suggests that local nutrient sensing at both hindbrain and forebrain sites is acutely

engaged to drive behavioral and physiological effectors. Results from neuroanatomical and neurochemical studies of each effector response suggest distinct yet overlapping neurohumoral circuits linking the gut to the brain network can be identified to modulate each distinct effector systems. As gut nutrient stimuli drive each of these pathways simultaneously during gut-meal related meal stimulation, major challenges for the field are (1) to understand the temporal and spatial patterns of neural activity during simultaneous activation of these pathways, and (2) how or whether they interact during a meal to coordinate effector responses.

**Key Words:** feeding, nutrient sensing, neurobiology



## Companion Animals: Nutrition and behavior

**389 Evaluation of pelleted diets targeted for grazing ruminants housed in zoological institutions.** Katherine R. Kerr<sup>\*1,2</sup>, Marcos Zenobi<sup>2</sup>, Rodrigo Gardinal<sup>2</sup>, Jorge Zuniga<sup>2</sup>, Adegbola Adesogan<sup>2</sup>, Charles Staples<sup>2</sup>, and Eduardo Valdes<sup>1</sup>, <sup>1</sup>Disney's Animal Kingdom, Lake Buena Vista, FL, <sup>2</sup>University of Florida, Gainesville, FL.

Objective was to evaluate the apparent digestibility of 2 commercially prepared pellets targeted for herbivores housed in zoological institutions (Mazuri Wild Herbivore Hi-Fiber Diet, Purina Mills International, Brentwood MO). The pellets, differing primarily in ether extract concentration, were designated low fat (LF; DM basis: 91% OM, 29% ADF, 46% NDF, and 5.9% EE) and high fat (HF; DM basis: 92% OM, 30% ADF, 48% NDF, and 10% EE). Dietary treatments consisted of 30% chopped Bermuda grass hay (*Cynodon dactylon*) and 70% commercial pellets. Using a crossover design, 13 male lambs (45.0 ± 4.8 kg BW) served as a model for ex situ ruminant herbivores. Lambs were offered diets at 90% of ad libitum intake for a 7-d adaptation and the subsequent 7-d fecal collection period. Ten percent of wet feces were retained, dried at 55 C, ground, and chemically analyzed. Daily intake of pelleted feed tended ( $P = 0.06$ ) to be greater in sheep fed LF pellets (1040 vs. 1003 g DM/d); however, intake of total DM did not differ due to treatment (1434 ± 183 g DM/d). Apparent digestibility coefficients of DM (57 ± 1.8%) and OM (57 ± 1.8%) did not differ ( $P > 0.10$ ) due to treatment. Apparent digestibility coefficients of NDF, ADF, and EE were greater ( $P < 0.01$ ) for lambs fed HF pellets (42, 45, and 94%, respectively) compared with those fed LF pellets (36, 42, and 90%, respectively). Blood was collected via jugular vein approximately 4 h after feeding. Plasma concentrations of glucose (70 ± 2.0 mg/dL) and urea nitrogen (21.3 ± 1.1 mg/dL) were not affected ( $P > 0.10$ ) by diet. To provide proper care and ensure optimal health of ex situ wildlife species, a clear understanding of the diet utilization by these species is necessary. Use of appropriate model species (i.e., closely related domestic counterparts) allows researchers to measure physiological data that might be otherwise unattainable due to research limitations when working with ex situ wildlife species (e.g., low numbers, control of variation, and sampling limitations). Herein, we utilized the sheep as a model for ruminants housed in zoological institutions.

**Key Words:** captive exotic, fiber digestibility, herbivore

**390 Serum metabolite profiling to identify biomarkers and mechanistic insight into the metabolic changes associated with weight loss in overweight cats.** Marissa R. Pallotto<sup>\*1</sup>, Maria R. C. de Godoy<sup>2</sup>, Kirk L. Pappan<sup>3</sup>, Preston R. Buff<sup>4</sup>, and Kelly S. Swanson<sup>1,2</sup>, <sup>1</sup>Division of Nutritional Sciences, University of Illinois, Urbana, IL, <sup>2</sup>Department of Animal Sciences, University of Illinois, Urbana, IL, <sup>3</sup>Metabolon, Durham, NC, <sup>4</sup>The Nutro Company, Franklin, TN.

Previous studies have shown that obese and overweight models have increased oxidative stress and lipid metabolism dysregulation. Thus, the objective of this study was to identify metabolite changes and potential biomarkers during weight loss that could be used as indicators of reestablished health. During a 4-wk baseline period, 8 adult neutered male cats were fed to maintain BW. For 18 wk following baseline, cats were fed to lose weight at a targeted rate of 1.5% BW/wk. As expected, mean BW (7.7 ± 0.4 vs. 6.2 ± 0.4 kg) and mean BCS (7.6 vs. 6.0) decreased ( $P < 0.05$ ) at wk 18 vs. wk 0. Daily food intake, twice-weekly BW, and

weekly BCS were recorded throughout the study. Fasted (~15 h) blood samples were collected at wk 0, 1, 2, 4, 8, 12, and 16. Serum was stored at -80°C until GC-MS and LC-MS/MS analyses. A total of 535 named biochemicals were identified, with up to 269 metabolites altered ( $P$ - and  $q$ -values < 0.05) at wk 16 vs. wk 0. Principal component analysis showed a continual shift as weekly weight loss progressed. Component 1 and 2 explained 14.3% and 10.3% of the variability, respectively. There was a significant and dramatic reduction of bile acids (cholate; taurocholate; deoxycholate) with weight loss. A reduction in numerous free fatty acids (FFA) and an increase in ketones (acetoacetate; 3-hydroxybutyrate) and monoglycerides suggest a shift toward lipolysis and hepatic FFA oxidation. Decreased markers of inflammation and oxidative stress were indicated by reduced pro-inflammatory oxylipids, eicosanoids, and oxidized biomarkers following weight loss. Markers of collagen degradation, such as proline-hydroxyproline and proline-glycine, and muscle protein turnover, such as 1-methylhistidine, increased. Mevalonate was significantly decreased after wk 8 compared with baseline, which agrees with the reduced bile acids without altering cholesterol. This may suggest upregulation of hepatic ketogenesis through activation of HMG-CoA lyase. Considering these results, global metabolomics was useful to potentially identify biomarkers of systemic changes in response to caloric restriction of a high-protein, high-fiber diet.

**Key Words:** feline, metabolomics, obesity

**391 Behavior assessment of dogs fed soybean hulls.** Mariana Scheraiber<sup>\*1</sup>, Tabyta Tamara Sabchuk<sup>1</sup>, Tatiane Ramos<sup>1</sup>, Juliana Regina da Silva<sup>2</sup>, Lidiane Priscila Domingues<sup>1</sup>, Ana Vitória Fischer da Silva<sup>1</sup>, and Ananda Portella Felix<sup>1</sup>, <sup>1</sup>Federal University of Paraná, Curitiba, Paraná, Brazil, <sup>2</sup>Federal University of Santa Catarina, Florianópolis, Santa Catarina, Brazil.

The behavior of dogs is influenced by dietary effects on satiety. Dietary fiber may prolong the gastric emptying time, slow the absorption of nutrients and dilute the energy of the diet. Thus, the development of strategies to increase satiety may be desirable to help to increase socialization and decrease undesirable behaviors of the animals. Soybean hulls (SH) is an alternative fiber source for dog food, due to the soluble and insoluble fiber portions resulting in benefits in the animal's health. Thus, this study aimed to evaluate the behavior of dogs after ingestion of diets without (0%SH) and with 16%SH. Twelve adult Beagle dogs (6 males and 6 females) in a completely randomized design were used. Dogs were individually housed in concrete kennels with solarium (2 m wide × 5 m long) and received the diets for 28 d. The behavior of dogs was evaluated on the 28th day, for 24 h continuously, at intervals of 10 min between each observation. The behaviors assessed were: alert, walking, scratching, sleeping, stereotypies, hygiene, lying idle, standing idle, sitting leisure and socialization. The results were expressed as the behavior occurrence frequency (%) (number of observations of the behavior / total number of observations × 100). Data were analyzed using the Mann-Whitney-Wilcoxon test, with  $P < 0.10$ . There was a decrease in the behavior of scratching ( $P < 0.05$ ; 9.3% 0%SH to 3.7% 16%SH) and stereotypy ( $P < 0.10$ ; 8.3% 0%SH to 4.8% 16%SH) in dogs fed 16%SH, in comparison with the 0%SH diet. The other behaviors did not differ ( $P > 0.05$ ) between diets. Diets with SH, as fiber source, can decrease stereotypy behaviors of dogs.

**Key Words:** dog nutrition, feeding behavior, fiber

### 392 Evidence for a cat pheromone that modulates kitten scratching.

John J. McGlone\*<sup>1,2</sup> and Rebekkah R. Plummer<sup>2</sup>,  
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Inappropriate scratching is a behavioral problem among kittens and cats in homes. Tools to modulate (increase or decrease) scratching would be valuable for cat owners and cats. Anecdotal literature suggests that cats have pheromones in their paws or claws, or in their saliva that they transfer to their claws. These pheromones are reportedly used to mark territory or objects. Through evaluation of cat scratchers, we noticed effects over time that suggested saliva, claw, or paw pheromones may modulate kitten scratching. We used the highly preferred S-shaped cardboard scratcher and 6 kittens (<3 mo of age) per study in 2 studies with the objective of finding evidence that supports pheromones may be present. In study 1, we compared kitten scratching with paired access to a previously used cat scratcher compared with a new cat scratcher. Using a 20-min evaluation period, kittens spent more ( $P < 0.05$ ) time interacting (scratching and playing) with a previously used cat scratcher than with a new cat scratcher ( $33.1$  vs.  $20.6 \pm 6.3$  s). In study 2, new S-shaped cardboard scratchers were evaluated alone or with catnip, catnip oil, or cat hair added (clipped from adult donor cats and allowed to fall between the cracks of the cardboard scratcher). Kittens spent more ( $P < 0.05$ ) time interacting with the S-shaped cardboard scratcher with added cat hair than with a control scratcher ( $16.0$  vs.  $8.3 \pm 2.7$  s). Adding dried catnip plants or catnip oil did not cause a change in scratching compared with the control ( $5.0, 10.2$  vs.  $8.3 \pm 2.7$  s). In conclusion, we provide evidence that kittens touching cardboard cause future kittens to interact more with that material. Second, we show that catnip plants and oil are not effective at increasing use of a scratcher by kittens. Finally, we show that cat hair (odor) induces enhanced interactions (scratching and play) with a cardboard scratcher. These findings can be used as a basis to discover new pheromones that modulate kitten and cat behavior.

**Key Words:** cat, behavior, scratch

### 393 The use of varying strengths of gelatin and the effect on physical properties of extruded pet food.

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Previous work with low-bloom gelatin demonstrated an increase in hardness and durability when incorporated into extruded kibbles. The objective of this experiment was to determine the effect of gelatin bloom strength on extruded kibble physical properties. Four nutritionally complete diets (30% protein) were formulated with no gelatin (control) or with 10% gelatin from 50, 175 and 250 bloom strength sources (Pro-Bind 100, PigSkin 175, and PigSkin 250) exchanged with chicken by-product meal. All diets were produced on a Wenger X-20 extruder through a circle die (3mm diameter) then a triangle die (5.3mm base  $\times$  4.9mm height). Products were analyzed for bulk and piece density, throughput, cross-sectional expansion, hardness, and pellet durability index (PDI). Experimental treatments were arranged as a factorial with main effects separated using statistical software (SAS 9.4). Kibble shape had an effect on hardness, cross-sectional expansion, and piece density, but this was likely due to a slight difference in die open area. The interaction of gelatin strength and kibble shape also had effects on hardness, PDI, and density, but these can be attributed to changes in bloom strength. Relative to the control, hardness increased with 50 bloom gelatin but decreased with 175 and 250 bloom gelatin ( $P < 0.05$ ; 5.93, 7.38, 4.57, 3.59kg, respectively). Relative to the control, PDI was the same for 50 bloom but decreased with both 175 and 250 bloom gelatin (88.49%, 87.57%, 64.55%, and 30.01%, respectively). The main effect means

of cross-sectional expansion and specific length increased with bloom strength (3.05, 3.54, 4.29, 4.92mm<sup>2</sup>/mm<sup>2</sup> expansion and 4.27, 4.26, 4.49, 4.74cm/g length for 0, 50, 175, 250 bloom, respectively). The main effect means for bulk and piece density became lighter with increasing bloom (0.56, 0.48, 0.39, 0.33g/cm<sup>3</sup> piece density and 347.6, 310.7, 242.4, 212.3g/L bulk density for 0, 50, 175, 250 bloom, respectively). There were no treatment effects on throughput. Low-bloom gelatin may improve expansion while increasing product hardness and durability, making it a potential structure-enhancing ingredient.

**Key Words:** pet food, gelatin, extrusion

### 394 Digestibility and palatability of diets containing crude corn oil in dogs.

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Crude corn oil (CCO) has large amount of linoleic acid, an essential fatty acid for dogs. Furthermore, CCO has less variation in composition and quality than animal fats. The objectives were to evaluate the apparent total-tract digestibility (ATTD), ME, and palatability of diets containing CCO in dogs. Four diets were evaluated: a control diet (with 12% poultry fat, PF), and 3 diets containing: 4, 8, and 12% CCO, replacing the PF. Diets were sprayed with the fat sources. Eight Beagle dogs were assigned in a replicated Latin square ( $4 \times 4$ ), totaling 8 replicates per treatment. The experimental diets were offered for a 5-d adaptation period, followed by 5 d of total fecal collection per period. The results were submitted to analysis of regression and orthogonal contrasts (PF vs. CCO). Palatability tests were performed using 2 pair-wise comparisons: 12% PF vs. 12% CCO, both diets with (Test 1) or without (Test 2) palatability enhancer (liquid chicken liver hydrolyzed). Palatability was determined by measuring intake ratio (IR) between the foods offered to 15 adult dogs. Each test was repeated for 3 d ( $n = 45$ ). The IR data were analyzed by Student's *t*-test. The ATTD of DM (80.0, 80.4, 78.2, and 79.2%), CP (80.8, 81.7, 79.4, and 80.0%), GE (84.2, 83.7, 82.3, and 83.5%), and ME (15.68, 15.58, 15.32, and 15.55 MJ/kg DM) of diets 12% PF, 4, 8, and 12% CCO, respectively, did not differ ( $P > 0.05$ ). The IR did not differ ( $P > 0.05$ ) between diets with palatability enhancer (0.53 to 12% CCO vs. 0.47 to 12% PF) and without palatability enhancer (0.52 to 12% CCO vs. 0.48 to 12% PF). The use of CCO as a lipid source in diets for dogs does not change the digestibility and the palatability of the diet, even in diets without palatability enhancer, when compared with the PF.

**Key Words:** companion animal nutrition, fat source, food preference.

### 395 Effects of androstenone on reduction of leash pulling in dogs.

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Behavioral problems are the primary reason owners relinquish dogs to animal shelters. Interomones are olfactory cues produced by one species that elicit a response in another species. The objective of this study is to determine the effects of androstenone acting as an interomone to reduce leash-pulling behavior using a model of dog walking. Androstenone is a swine sex pheromone capable of eliminating jumping and barking in dogs; as such, it may also reduce leash pulling. This study took place at a local animal shelter using adoption dogs surrendered by owners due to behavioral issues, providing an exceptional model for this study. A researcher walked each dog on a standard nylon neck collar and leash 53 m around an enclosed yard (average time  $76.0 \pm$

11.6 s). Leash pulling was defined as pressure exerted on the leash by the dog resulting in the researcher's inability to walk comfortably. Each dog was walked twice with 90 min of rest between walks. On the first walk the researcher did not respond to leash pulling (NOT). On the second walk the researcher responded to each leash pull by spraying the designated treatment 46 cm from the dog's head. Treatments were water (H<sub>2</sub>O, n = 8), androstenedione+water (ANH, n = 9), or InteroSTOP (Meridian Animal Health, Omaha, NE) (INT, n = 10). The study was designed so each dog was walked twice: once with NOT, and once with H<sub>2</sub>O, ANH or INT. Walks were video recorded and observed by researchers blind to the treatments. The observers counted number of leash pulls during each walk and noted reactions to sprays. Data were analyzed using a 2-tailed Student's *t*-test, with number of leash pulls during the walk with NOT as the control for each treatment. ANH and INT reduced the number of leash pulls by 84% and 86%, respectively, compared with H<sub>2</sub>O ( $P < 0.001$ ). There was no difference in reduction of leash pulling between ANH and INT ( $P > 0.05$ ). INT reduced leash pulling; however, the compressed air sound accompanying the spray frightened many dogs when sprayed from behind. These findings may be beneficial in developing behavioral modification training tools for dogs, thereby improving the human-animal bond and possibly reducing the desire of owners to relinquish ownership of dogs.

**Key Words:** behavior, canine, interomone

**396 Effects of graded dietary HP 300 concentrations on apparent total-tract macronutrient digestibility, fecal characteristics, and fecal fermentative end-products in healthy adult dogs.** Alison N. Beloshapka\*<sup>1</sup>, Maria R. C. de Godoy<sup>1</sup>, Katelyn B. Detweiler<sup>1</sup>, Ingmar S. Middelbos<sup>3</sup>, George C. Fahey Jr.<sup>1,2</sup>, and Kelly S. Swanson<sup>1,2</sup>, <sup>1</sup>University of Illinois at Urbana-Champaign Department of Animal Sciences, Urbana, IL, <sup>2</sup>University of Illinois at Urbana-Champaign Division of Nutritional Sciences, Urbana, IL, <sup>3</sup>Hamlet Protein Inc., Findlay, OH.

Animal proteins, such as poultry by-product meal (PBM), are commonly used in extruded dog foods. Plant-based proteins have a more consistent nutrient profile than animal sources but may contain anti-nutritional factors (ANF), including trypsin inhibitors and oligosaccharides. The test protein, HP 300, is a proprietarily processed soy-based product with very low ANF concentrations and high nutritional value. The objective was to evaluate if HP 300 can be an effective PBM replacement in pet food and if there are practical limits to its use. We tested the effects of 0% (control), 4%, 8%, 12%, 24%, and 48% HP 300 on apparent total-tract macronutrient digestibility, fecal characteristics, and fecal fermentative end products in healthy adult dogs. All 6 diets were formulated to meet AAFCO nutrient profiles with approximately 30% crude protein and 16% fat. Forty-eight healthy adult beagle dogs (20 females, 28 males; mean age: 3.4 y; mean BW: 10.0 kg) were studied in a completely randomized design. The treatment period consisted of a 10-d diet adaptation phase followed by a 4-d fresh and total fecal collection phase. Stool output (both on as-is and dry matter basis [DMB]) did not differ from control until 48% HP 300 ( $P < 0.01$ ). Fecal output per unit food intake (DMB) differed ( $P < 0.01$ ) from control only at the 24% and 48% HP 300 inclusion rates. Notably, no effect of HP 300 inclusion was found for stool consistency scores. Digestibility of DM, organic matter and energy did not differ from control at any inclusion rate, except for a decrease ( $P < 0.01$ ) at 48% HP 300, which is likely due to increased dietary fiber concentrations. Crude protein digestibility was not affected by treatment and ranged from 82.9 – 86.2%. Short-chain fatty acid production increased ( $P < 0.01$ ) compared with control only at 24% and 48% HP 300. Conversely, branched-chain fatty acids

were reduced ( $P < 0.01$ ) compared with control at inclusion rates of HP 300 of 8 – 48%. Overall, these data suggest that HP 300 is a suitable replacement for PBM in canine diets up to at least a 24% inclusion rate.

**Key Words:** canine, digestibility, soy protein

**397 Characterization of hindgut microbiota during the onset of obesity in adult domestic cats.** Maria R. C. de Godoy\*<sup>1</sup> and Kelly S. Swanson<sup>1,2</sup>, <sup>1</sup>Department of Animal Sciences, University of Illinois, Urbana, IL, <sup>2</sup>Department of Veterinary Clinical Medicine Sciences, University of Illinois, Urbana, IL.

In humans, gastrointestinal microbiota of obese subjects harbor a less diverse bacterial community and a lower ratio of Bacteroidetes:Firmicutes phyla. However, these relationships have not been examined in the domestic cat. Thus, the objective of this study was to characterize the hindgut microbiota of domestic cats transitioning from a lean to an obese phenotype. Nine domestic male cats (mean age = 8 ± 0.3 yr; mean BW = 4.5 ± 0.4 kg, mean BCS range = 5.0 ± 0.5) were ad libitum fed a complete and balanced experimental diet for 36 wk. Food intake was measured daily, while BW and BCS were measured weekly, fresh fecal samples were collected at 0, 6, 12, 24 and 36 wk. Amplification of fecal bacterial DNA was performed using targeted primers and a Fluidigm Access Array. Illumina sequencing was performed utilizing a MiSeq2000 using v3 reagents. High quality (quality value >25) sequence data derived from the sequencing process were analyzed using QIIME 1.8.0. Resulting sequences were clustered into operational taxonomic units (OTU) using closed-reference OTU picking against the Greengenes 13\_8 reference OTU using a 97% similarity threshold. Data were analyzed as repeated measurements, with post-hoc Tukey adjustment.  $AP < 0.05$  was considered statistically significant. A total of 9,145,663 reads were generated in the current data set, with an average of 203,236 reads per sample used to provide diversity estimates. Cats incrementally changed from a lean phenotype (mean BCS = 5.0) at baseline to an overweight phenotype with a mean BCS of 7.5 at 36 wk. Similar to overweight observed in humans, overweight cats had a lower ratio of Bacteroidetes (e.g., Bacteroides; Prevotella) to Firmicutes (e.g., Lactobacillus) phyla. However, microbial diversity and species richness was not altered by BW gain (~250 species). Body weight gain resulted in a gut microbiota shift depicted by unweighted UniFrac-based PCoA analysis between cats at baseline and 6 wk vs. 24 and 36 wk. Proteobacteria (e.g., Succinivibrionaceae) and Fusobacteria (Fusobacteriaceae) were decreased ( $P < 0.05$ ) by BW gain. These data demonstrate the transition from a lean to an overweight phenotype alters the GI microbiota in cats.

**Key Words:** feline, microbiome, obesity

**398 Evaluation of cat scratcher efficacy.** John J. McGlone\*<sup>1,2</sup> and Rebekkah R. Plummer<sup>2</sup>, <sup>1</sup>Laboratory of Animal Behavior, Physiology and Welfare, Texas Tech University, Lubbock, TX, <sup>2</sup>McGlone Enterprises Inc., Lubbock, TX.

Inappropriate scratching is a behavioral problem among kittens and cats in homes. Many models of cat scratchers are available but there is little objective work that evaluates efficacy of cat scratchers. A model was developed to evaluate cat scratching using kittens in a controlled setting. Four studies were conducted. The first study used 12 kittens (<3 mo of age) in a completely random design with 2 scratcher types per study. A power test revealed 6 kittens gave sufficient power for a scratcher evaluation; therefore, studies 2 to 4 used 6 unique kittens each. In a given study, a kitten was given simultaneous access to 2 cat scratchers for 20 min. Kittens spent an average of 95 s scratching and



playing with cat scratchers. Preference studies examined cat scratchers. The key measure was the time kittens spent interacting (scratching and playing) with each scratcher. In study 1a, kittens spent more ( $P < 0.05$ ) time with a cardboard bed than with a hemp post ( $99.3$  vs.  $14.9 \pm 15.9$  s) while in study 1b, a carpeted tall post and cardboard bed induced similar scratching ( $24.2$  vs.  $20.2 \pm 23.3$  s). In studies 2a-c, a screen was placed in either horizontal or vertical positions. In study 2a, kittens spent more ( $P < 0.05$ ) time with the screen in the horizontal compared with the vertical position ( $21.7$  vs.  $10.5 \pm 2.8$  s). In study 2b, kittens spent similar time with the horizontal screen compared with the cardboard scratcher ( $25.0$  vs.  $29.8 \pm 8.6$  s). In 2c, kittens preferred ( $P < 0.05$ ) the cardboard scratcher to horizontal bubble wrap ( $31.0$  vs.  $9.0 \pm 5.1$  s). In study 3a, kittens spent more ( $P < 0.05$ ) time with cardboard than with screen ( $37.5$  vs.  $7.8 \pm 7.9$  s) and in 3b more time with cardboard vs. carpet ( $37.5$  vs.  $5.0 \pm 6.7$  s). In studies 4a-c, kittens spent more ( $P < 0.05$ ) time with an S-shaped cardboard than with long-flat, rectangular-flat, or boat-shaped cardboard scratchers ( $20.6, 6.2, 6.3, 11.3 \pm 3.9$  s). In conclusion, kittens prefer cardboard and carpet over screen, bubble wrap, or hemp-type scratchers. Second, kittens had a clear preference to play and scratch with cardboard scratchers in an S-shape. Finally, scratchers vary considerably in their ability to interest kittens in scratching.

**Key Words:** cat, kitten, behavior

**399 Evaluation of pork and pork by-products in raw meat diets for African wildcats.** Cayla J. Iske\*<sup>1</sup>, Cheryl L. Morris<sup>1,2</sup>, and Kelly Kappen<sup>2</sup>, <sup>1</sup>Iowa State University, Ames, IA, <sup>2</sup>Omaha's Henry Doorly Zoo and Aquarium, Omaha, NE.

Primarily, only 2 protein sources (beef or horse) are commercially used for raw meat diets fed to carnivores managed in zoos. An additional protein source such as pork has potential to mitigate some gastrointes-

tinal issues and provide dietary variety. Concerns with high levels of microbial populations in raw meat diets and possible zoonoses also have been expressed. The objectives of this study were to determine if a pork-based diet had similar apparent total-tract macronutrient digestibility and fecal scores in African wildcats as standard zoological carnivore diets formulated with either horse or beef and to characterize the microbial counts among the diets. Four African wildcats were used (2 males, 2 females) in a randomized crossover design consisting of 4 raw meat diets: horse (H), beef (B), beef/horse blend (HB), and pork (P) all fed isocalorically to maintain body weight. Data were analyzed using the Mixed models procedure of SAS. All diets were highly digestible, especially for fat where digestibility values ranged from 98.58 (H) to 99.73% (HB) and were not different. Digestibility of OM was higher ( $P < 0.05$ ) when cats consumed HB (97.15%) compared with P (93.10%). Crude protein digestibility was highest ( $P < 0.05$ ) for HB (98.08%) and lowest for B (93.44%). Fecal scores (1 = hard, dry feces to 5 = liquid/watery feces) were higher ( $P < 0.05$ ) for cats fed B (2.63) compared with cats fed H (1.55) or P (1.91). Microbial counts varied and included E. Coli ranging from 110 to 10,000 cfu/g for HB and B diets, respectively; total coliforms from 150 to 28,000 cfu/g for HB and B diets, respectively; yeast from 20 to 4,000 cfu/g for P and HB, respectively; mold counts were not detectable for all diets except B that contained 10 cfu/g and aerobic plate counts ranged from 23,000 to 26,000,000 cfu/g for H and HB diets, respectively. *Staphylococcus aureus* was not detected in any of the diets. *Salmonella* was presumptive positive in the P and HB diets; however, these values were not enumerated or quantified for specific strains. In conclusion, a pork-based diet can be utilized by exotic small cats and included among dietary options for felid management. Raw meat diets do contain variable levels of microbes and should be handled and managed appropriately.

**Key Words:** raw meat diet, cat, exotic animal

## Dairy Foods: Microbiology

**400 Development of an anaphylactic model of buckwheat using B-type CpG oligodeoxynucleotides from lactic acid bacteria.** Yoshinari Yamamoto\*<sup>1</sup>, Suguru Shigemori<sup>1,2</sup>, Kazushi Oshiro<sup>3</sup>, Pengfei Wang<sup>1</sup>, Yeqin Wang<sup>1</sup>, Takashi Sato<sup>4</sup>, and Takeshi Shimomoto<sup>1,5</sup>, <sup>1</sup>Interdisciplinary Graduate School of Science and Technology, Shinshu University, Kamiina, Nagano, Japan, <sup>2</sup>Research Fellow of the Japan Society for the Promotion of Science (JSPS), Chiyoda, Tokyo, Japan, <sup>3</sup>Graduate School of Agriculture, Shinshu University, Kamiina, Nagano, Japan, <sup>4</sup>Graduate School of Medicine, Yokohama City University, Yokohama, Kanagawa, Japan, <sup>5</sup>Institute for Biomedical Sciences, Shinshu University, Kamiina, Nagano, Japan.

Anaphylaxis is an acute systemic allergic reaction (severe type I hypersensitivity) to a specific allergen and occurs in human and other mammals. Food anaphylaxis is most frequently triggered by peanut, buckwheat, milk, egg, and wheat allergens. Anaphylactic mouse models against ovalbumin and peanut allergens have been established by repeated exposure to small amounts of allergen with adjuvant via subcutaneous (s.c.) and intraperitoneal (i.p.) routes. When immunity is established, the animals are challenged with a large quantity of allergens via an i.v. route to induce anaphylaxis. However, i.v. sensitization is a relatively complicated procedure, and establishing an anaphylactic model requires considerable time. In addition, the i.p. method for allergen delivery is simpler than i.v. routes, has lower risk for failure, and can shorten sensitization duration. Here, we attempted to develop a simple anaphylactic shock model by exposing mice to a B-type CpG-oligodeoxynucleotide (ODN), termed MsST, which had the immune ability to induce IL-33 and IFN- $\gamma$  production from the *lac Z* gene of *Streptococcus thermophilus* ATCC19258 with allergen via an i.p. route. After the allergen challenge for inducing anaphylactic shock, IgG<sub>2a</sub>-production and IFN- $\gamma$ -positive cells were markedly increased in mice that received B-type CpG-ODN. In contrast, IL-33-positive cells in the model mice were only slightly increased. Generally, IFN- $\gamma$  supports Th1 cell responses that suppress allergic disease. However, it was previously reported that IFN- $\gamma$  enhances platelet-activating factor, which is a bioactive lipid mediator that plays a role in the onset of inflammatory diseases such as asthma, anaphylaxis and atherosclerosis, and is involved in the generation of CD11b<sup>+</sup> and CD11c<sup>+</sup> cells. The anaphylactic shock model described here is expected to contribute to the development of anaphylaxis treatment strategies.

**Key Words:** anaphylactic model, buckwheat, B-type CpG-oligodeoxynucleotide (ODN)

**401 Construction of genetically modified *Lactococcus lactis* expressing buckwheat protein Fagag1 with strong allergenicity.** Suguru Shigemori\*<sup>1,2</sup>, Yoshinari Yamamoto<sup>1</sup>, Kazushi Oshiro<sup>3</sup>, Pengfei Wang<sup>1</sup>, Yeqin Wang<sup>1</sup>, Takashi Sato<sup>4</sup>, and Takeshi Shimomoto<sup>1,5</sup>, <sup>1</sup>Interdisciplinary Graduate School of Science and Technology, Shinshu University, Kamiina, Nagano, Japan, <sup>2</sup>Research Fellow of the Japan Society for the Promotion of Science (JSPS), Chiyoda, Tokyo, Japan, <sup>3</sup>Graduate School of Agriculture, Shinshu University, Kamiina, Nagano, Japan, <sup>4</sup>Graduate School of Medicine, Yokohama City University, Yokohama, Kanagawa, Japan, <sup>5</sup>Institute for Biomedical Sciences, Shinshu University, Kamiina, Nagano, Japan.

Buckwheat (*Fagopyrum esculentum*) is consumed as a pseudocereal in Asia and Western countries, and is recognized as a functional food. However, several proteins from buckwheat have strong allergenicity

and cause severe symptoms, such as an anaphylaxis, in hypersensitive patients. Here, we engineered strains of *Lactococcus lactis* NZ9000 (NZ9000) that express Fagag1, a major allergenic storage protein of buckwheat, fused with or without green fluorescent protein (GFP). Codon-optimized sequences of Fagag1 and GFP-Fagag1 were individually cloned into the *L. lactis* expression vector pNSH, containing a nisin-inducible promoter and 6x histidine-tag sequence. The resulting plasmids were separately introduced into NZ9000, and nisin-dependent expression of recombinant Fagag1 (rFagag1) and rGFP-Fagag1 was confirmed by Western blotting and confocal laser scanning microscopy. rFagag1 and rGFP-Fagag1 were individually purified by immobilized metal-ion affinity chromatography techniques, and the allergenicity of the purified proteins was then evaluated in *in vitro* cultures of splenocytes isolated from buckwheat crude protein-immunized mice. Treatment of splenocytes with rFagag1 markedly induced the mRNA expression of interleukin (IL)-4, IL-13, and IL-17F, which are known mediators of allergic inflammation. Similar expression levels of IL-4 and IL-17F mRNA were observed in the splenocytes stimulated with purified rGFP-Fagag1; however, the increases from baseline were significantly lower than those observed in rFagag1-treated cells. Recent evidence suggests that the mucosal (i.e., oral or intranasal) application of lactic acid bacteria genetically modified to produce allergen is a promising strategy for allergy therapy. Therefore, our present results suggest that NZ9000 expressing immunoreactive rFagag1 or rGFP-Fagag1 may be a powerful candidate for the prevention and therapy of buckwheat allergy. However, the future application of prophylactic and therapeutic strategies based on NZ9000 strains first requires a clear demonstration of efficacy in *in vivo* trials.

**Key Words:** buckwheat, *Lactococcus lactis*, food allergy

**402 Production of recombinant  $\beta$ -lactoglobulin in *Lactococcus lactis* and generation of a bioactive peptide with incretin-inactivation activity.** Kazushi Oshiro\*<sup>1</sup>, Suguru Shigemori<sup>2,3</sup>, Yoshinari Yamamoto<sup>2</sup>, Pengfei Wang<sup>2</sup>, Yeqin Wang<sup>2</sup>, Takashi Sato<sup>4</sup>, and Takeshi Shimomoto<sup>2,5</sup>, <sup>1</sup>Graduate School of Agriculture, Shinshu University, Kamiina, Nagano, Japan, <sup>2</sup>Interdisciplinary Graduate School of Science and Technology, Shinshu University, Kamiina, Nagano, Japan, <sup>3</sup>Research Fellow of the Japan Society for the Promotion of Science (JSPS), Chiyoda, Tokyo, Japan, <sup>4</sup>Graduate School of Medicine, Yokohama City University, Yokohama, Kanagawa, Japan, <sup>5</sup>Institute for Biomedical Sciences, Shinshu University, Kamiina, Nagano, Japan.

$\beta$ -Lactoglobulin (BLG) is the most abundant protein in cow's milk whey. Peptides prepared from BLG digested with the gastrointestinal (GI) proteases pepsin and trypsin strongly inhibit dipeptidyl peptidase-IV (DPP-IV) activity. DPP-IV is a ubiquitously expressed serine protease that cleaves N-terminal dipeptides from incretins, which are insulinotropic GI hormones. Recently, genetically modified lactic acid bacteria have emerged as efficient cell factories for the production and safe delivery of heterologous proteins to the GI tract. Here, we engineered a strain of *Lactococcus lactis* to secrete recombinant BLG (rBLG) and evaluated the DPP-IV-inhibiting activity of trypsin-digested rBLG. To construct rBLG-producing *L. lactis*, a codon-optimized bovine BLG gene was inserted into the lactococcal secretion vector pNZ8148#2:SEC, containing a nisin-inducible promoter and signal peptide sequence from USP45 protein. The constructed rBLG secretion vector was transformed into *L. lactis* NZ9000 (NZ9000) as a host strain for gene expression. The

secretion of rBLG by NZ9000 was confirmed by Western blotting. The DPP-IV-inhibitory activity of trypsin-digested rBLG was examined by comparative analysis with trypsin-digested commercial BLG (cBLG) using a DPP-IV Drug Discovery Kit. Results from the assay revealed that the inhibitory activity of trypsin-digested rBLG against DPP-IV was similar to that of cBLG. In conclusion, we successfully engineered a strain of NZ9000 that efficiently secretes rBLG, and clearly demonstrated that trypsin-digested rBLG has strong inhibitory activity against DPP-IV enzyme. In future studies, we plan to evaluate the potential of rBLG-secreting NZ9000 as a therapeutic agent for type 2 diabetes.

**Key Words:**  $\beta$ -lactoglobulin, *Lactococcus lactis*, dipeptidyl peptidase-IV

#### **403 Sodium chloride induced stress responses in dairy probiotic bacteria.** Akanksha Gandhi\* and Nagendra P. Shah, *The University of Hong Kong, Hong Kong.*

The study focused on possible mechanisms of salt stress injury and responses of selected probiotic bacteria to salt stress. The effects of varying NaCl/KCl concentrations on viability, membrane integrity and metabolic activity of *Lactobacillus acidophilus*, *Lactobacillus casei* and *Bifidobacterium longum* were studied using flow cytometry. Furthermore, the changes in surface functional groups, morphology and membrane fatty acid composition of these bacteria were investigated using Fourier-transform infrared (FTIR) spectroscopy, transmission electron microscopy and gas chromatography. The effect of NaCl/KCl stress on the in vitro adhesion ability of stressed bacteria to Caco-2 cells was also evaluated. Cell viability as evaluated using conventional culture technique was compared with the findings from flow cytometric analysis on metabolic activities of the cells and it was revealed that there was a correlation between culturability and dye extrusion ability of *Lb. casei* and *B. longum*. However, a certain population of *Lb. acidophilus* was viable as per the plate count method but the efflux activity was compromised. The metabolic activity of *Lb. casei* was found to be highest among the 3-probiotic bacteria. The FTIR spectral analysis also highlighted the shifts that occurred mainly in the amide regions upon increasing the NaCl concentration. Significant changes in the morphology of all bacteria were observed at higher salt concentrations. Shrinkage of the cytoplasmic content and irregularities in cell wall on exposure to high NaCl concentrations (10% w/v) were observed in all bacteria. Membrane fatty acid composition was affected by salt stress, and the ratio of unsaturated to saturated fatty acids was altered on exposure to NaCl stress. Adhesion ability of stressed bacteria to Caco-2 cell lines was also reduced at higher NaCl concentrations (10% w/v). Comparing the responses among the selected bacteria, *Lb. casei* appeared to be most robust to NaCl stress. Overall, the study revealed the impact of salt stress on membrane characteristics and adhesion capability of selected probiotic bacteria.

**Key Words:** sodium chloride, lactic acid bacteria, cell membrane

#### **404 Potential role of *Bacillus* strains isolated from the dairy environment as defect-causing organisms in yogurt.** Dipakkumar Mehta\*<sup>1</sup>, Ashraf Hassan<sup>2</sup>, Brandon Nelson<sup>2</sup>, and Hasmukh Patel<sup>1</sup>, <sup>1</sup>South Dakota State University, Brookings, SD, <sup>2</sup>Daisy Brand, Garland, TX.

*Bacillus* spp. can cause spoilage of milk and dairy products due to their ability to produce heat resistant enzymes and spores. Milk proteins, fat, and stabilizers (such as gelatin, amylopectin (AP), xanthan, and pectin) play an important role in structure formation and stability of yogurt.

Possible sources of *Bacillus* spp. in these products include raw milk, ingredients such as dairy powders, and processing equipment. The objective of this study was to test the proteolytic, lipolytic, and phospholipase activities as well as stabilizer-degrading characteristics of 25 *Bacillus* strains isolated from the dairy environment. Proteolysis, lipolysis, and phospholipase activity were tested using skim-milk agar, spirit blue agar, and lecithin agar, respectively. A basic agar medium containing AP, xanthan, or pectin and gelatin agar were used to study the stabilizer-degradation activities of the various *Bacillus* strains. All experiments were conducted in triplicate at 42°C and under mesophilic conditions. Many of the tested *Bacillus* strains were highly proteolytic and could degrade various stabilizers (AP, pectin, xanthan and gelatin). It was found that the majority (92%) of *Bacillus* strains were able to degrade AP while only 20% showed lipolytic activities. All proteolytic strains showed gelatin degradation activities. The xanthan, phospholipid, and pectin degradation activities were found in 68%, 40% and 24% *Bacillus* strains, respectively. Generally, *Bacillus* spp. exhibited higher activities at 42°C than at mesophilic temperatures. Based on these results, it can be concluded that the order of susceptibility of the different tested components to *Bacillus* strains was AP > protein = gelatin > xanthan > phospholipids > pectin > fat, suggesting their ability to induce texture defects in yogurt.

**Key Words:** *Bacillus*, yogurt, stabilizer

#### **405 Inactivation of thermophilic sporeformers in milk by combined effect of cavitation and pasteurization.** Dikshi Bawa\*, Sanjeev Anand, Harsh Dahiya, and Hasmukh Patel, *South Dakota State University, Brookings, SD.*

Thermophilic sporeformers show resistance to the commonly applied thermal treatments and later produce spores during further processing of milk. They survive pasteurization and can cause spoilage defects in dairy foods. The objective of this study was to investigate the application of controlled hydrodynamic cavitation in a continuous mode, and its combination with thermal treatments on the inactivation of thermophilic sporeformers in milk. We hypothesized that the high shear created by the cavitation shock waves and the resulting friction would cause rupturing of the bacterial cell wall, resulting in cell death. Mid exponential phase vegetative cells of *Bacillus coagulans* (ATCC 12245) were inoculated in sterile skim milk at log 5 cfu/mL. Inoculated milk samples were passed at 60 Hz frequency and 200 L/h flow rate with 120 kPa back pressure, using APV Cavitator (supplied by SPX, Denmark) fitted with 4 row rotor in 6mm housing, resulting in exposure time of 22 s per pass. The inoculated milk at 10°C was recirculated until 6 passes or 6 cavitation effects (a total exposure time of 132 s) with an average inlet and outlet temperature rise up to 68°C and 82°C respectively after the 6th pass. Samples were kept in an ice bath during the treatments for temperature control. For studying the combined effect, post cavitation, samples were exposed to lab pasteurization (63°C for 30 min). Brain Heart Infusion Agar was used to plate the survivors. Experiments were conducted as replicates of 2, and were repeated thrice. Statistical significance of the data was determined using SAS software. A significant reduction ( $P < 0.05$ ) was observed in the bacterial counts after the treatments. The 6-pass cavitation effect alone, resulted in 1.56 log survivors. Additionally, the combined effect of cavitation and pasteurization further reduced the survivors to only 0.64 log. The results revealed that 6-pass cavitation effect alone and in combination with pasteurization was very effective in inactivating thermally resistant vegetative cells of *Bacillus coagulans* by 99.963 and 99.996% respectively.

**Key Words:** cavitation, thermophilic sporeformers, pasteurization



## Extension Education

**406 Optimal dairy farm management subject to greenhouse gas emissions constraints.** Di Liang\*, Thomas F. Rutherford, and Victor E. Cabrera, *University of Wisconsin-Madison, Madison, WI.*

Dairy farm feed allocation decisions seek to maximize farm profit. We present a nonlinear programming model that chooses a robust policy among a set of dairy farm management strategies. In the optimal policy, animal feed may be produced or purchased to meet nutrition and production demands of cow groups in the herd. Nutrition requirements are calculated according to the National Research Council equations, production level, cow group, and lactation number. Farm-produced feed quantity and quality (e.g., total DM, CP, NDF,  $NE_L$ , RDP) are simulated with the Integrated Farm System Model using daily weather data. The quality of purchased feeds is established from published research. Based on these, milk sales, the farm-produced feed costs, purchased feed costs and the greenhouse gas emissions from manure and enteric fermentation are calculated. The optimal solution addresses the dual objective of maximizing profit (milk income over feed cost) while limiting greenhouse gas emissions. Farm characteristics (e.g., breed, production level, culling rate, reproductive performance, cropping strategy), feeding strategies (e.g., high or low forage, grazing, CP content, grouping strategy, seasonal diets which address heat stress), and manure management options (e.g., direct field application, lagoon, manure pile) provide detailed control of the dairy management strategies, which characterize an optimal policy. Consequently, the optimal solution provides a list of best feeding strategies and manure management practices according to farm-specific characteristics that maximize profit and minimize greenhouse gas emissions.

**Key Words:** profitability and environment, feed allocation, optimization modeling

**407 A decision support tool for *Escherichia coli* bacterin mastitis vaccine use in dairy cows.** Amanda E. Stone\*, Tyler B. Mark, and Jeffrey M. Bewley, *University of Kentucky, Lexington, KY.*

The objective of this study was to create a producer-friendly decision support tool to evaluate the economic decision of implementing *Escherichia coli* (0111:B4) bacterin vaccination as a management practice. A partial budget analysis was conducted in Excel (Microsoft Corporation, Redmond, WA) and a producer-friendly dashboard was created (SAP America, Inc. Newtown Square, PA; the dashboard is available at <http://afsdairy.ca.uky.edu/J5MastitisVaccine>). Farm-specific inputs adjustable by the end user included herd size, milk price, milk yield, vaccine cost, labor cost, feed cost, culling rate from mastitis, coliform prevalence, replacement cow cost, and cull cow value. To demonstrate model sensitivity and utility, 3 example scenarios were evaluated. In all scenarios, vaccine cost was estimated at \$4.74 per cow and the rate of clinical mastitis in vaccinated cows was estimated at 8.6%. In the first scenario, a 100-cow herd was modeled with an average clinical mastitis prevalence (48%/year) and milk price (\$17.65, calculated from Food and Agricultural Policy Research Institute for years 2015 to 2025). In the second scenario, a 100-cow herd was modeled with an average clinical mastitis prevalence (48%/year) and greater than average milk price (\$25.70/cwt, calculated from years 2005 to 2015). In the third scenario, a 1,000 cow herd was modeled with an average clinical mastitis prevalence (48%/year) and average milk price (\$17.65/cwt). Labor costs, cull cow price, and replacement cow price were \$10/h, \$1,000/cow, and \$1,500/cow for all scenarios, respectively. The benefit:cost ratio of using a *Escherichia*

*coli* bacterin vaccine was \$7.52:\$1, \$8.51:\$1, and \$7.53:\$1, for scenarios 1, 2, and 3, respectively. The benefit was \$8,719/herd/year, \$11,552/herd/year, and \$87,191/herd/year, for scenarios 1, 2, and 3, respectively. All scenarios evaluated in this project were positive investments. Dairy producers considering investing in a coliform bacterin vaccine may use this as a decision support tool. This work was supported by a grant award from USDA-NIFA-AFRI (2013–68004–20424).

**Key Words:** economic dashboard, coliform mastitis vaccine, decision support tool

**408 The Missouri Show-Me-Select Replacement Heifer Program: Improving heifer development practices and increasing technology utilization through economic incentives.** Jordan M. Thomas\*, Brianne E. Bishop, Jillian M. Abel, Jared E. Decker, Scott E. Pooch, Douglas S. Brown, Michael F. Smith, and David J. Patterson, *University of Missouri, Columbia, MO.*

The Show-Me-Select Heifer Program has resulted in improvements in development programs for replacement beef heifers and increased utilization of technology among participating beef operations across Missouri. Enrollment of heifers in the program has increased steadily in recent years, from 3,020 heifers enrolled in 2010 to 5,867 heifers in 2014. Enrolled heifers undergo a prebreeding evaluation that includes pelvic measurement and a reproductive tract score (RTS). Producers may elect to expose heifers for breeding via AI or natural service (NS), and the proportion of heifers exposed for AI service has increased steadily. In 2010, 68% of enrolled heifers were exposed for AI rather than serviced exclusively with NS; whereas, AI serviced heifers accounted for 91% of enrolled heifers in 2014. Use of ultrasound (US), as opposed to palpation per rectum for pregnancy diagnosis, has also increased in the program. In 2010, 59% of heifers were diagnosed for pregnancy using US, increasing to 72% of heifers in 2014. Use of US facilitates a more accurate determination of whether a pregnancy resulted from AI or NS and may also allow for determinations of fetal sex. Lastly, differences in average sale price among Show-Me-Select heifers indicate a growing awareness among buyers of the value associated with genetically elite females. Heifers meeting the minimum requirements for enrollment are classified as Tier 1. Heifers may be further distinguished as Tier 2 if the sire of the heifer meets minimum accuracy requirements for specified traits at the time of sale, including: calving ease direct, calving ease maternal, weaning weight, carcass weight, and marbling. From 2010 to 2014, Tier 2 heifers carrying AI sired pregnancies (\$2,279) sold on average for \$213 more per heifer than Tier 1 heifers carrying NS sired pregnancies (\$2,066). In summary, continued growth in the Show-Me-Select Heifer Program highlights the importance of economic incentives to drive technology utilization and improve heifer development practices statewide.

**Key Words:** heifer development, reproductive management, beef cattle

**409 Engaging industry personnel in an agricultural education program.** Angela R. Mays\*, *F.L. Emmert Company, Cincinnati, OH.*

Typically inter-dependent departments usually exist within animal agricultural products businesses. Those personnel in billing, sales, maintenance, production and other areas may not have knowledge or understanding of end product usage in this industry. Consequently,

the F.L. Emmert Company recently initiated an agricultural education program, Emmert Educates, to engage and educate its employees. The program is voluntary, occurs once a month, and lasts for approximately one hour. Attendees are provided lunch, followed by a 10–15 min presentation, and then open discussion among the group. Supplemental printed materials and the opportunity for employees to suggest future topics have also been made available. A survey was recently conducted to evaluate the progress and staying power of the program, and allowed participants the opportunity to express themselves anonymously. The survey was conducted during the February 2015 Emmert Educates program and included all attendees, for a total of 10 participants. Of those surveyed, 100% were interested in the program continuing through 2015, with changes occurring in the frequency in which meetings are held. When asked if the program was informative and if the format was adequate 100% of those surveyed responded yes to both questions. Seventy percent of individuals surveyed did not feel as though the program interfered with their daily schedules, while 20% felt as though it did and 10% were unsure. The most impactful response came from the question addressing if employees had gained information from this program they would not have obtained elsewhere, with a 100% positive response rate. Therefore, the capability of the program to progress and evolve has been made available and confirmed through the positive responses from the survey. Overall, the ability to communicate and connect with employees through an agricultural education program has provided many benefits beyond the sharing of knowledge regarding this industry. Employees have been able to interact with one another in a work-free environment, learn from one another, and begin to understand their role as an employee in this industry, as well as a consumer of agricultural goods.

**Key Words:** education, industry, personnel

**410 Using video for consumer attitude inoculation about beef animal slaughter.** Katherine E. Powers and Traci L. Naile\*, *Oklahoma State University, Stillwater, OK.*

In the face of increasing consumer demands for transparency in agricultural production, decreasing literacy about agriculture, and increasing needs for trustworthy information sources for consumers, methods of communicating information about agriculture need to be examined for their effectiveness. Specifically, concerns about beef cattle slaughter need to be addressed through creative and effective channels of communication. However, few studies have been conducted to determine whether consumers can be inoculated through specific mediums against negative messages about slaughter. As visual media platforms have been demonstrated to increase learning, this study was conducted to determine the success of video in educating consumers about the slaughter process. To accomplish this purpose, an online questionnaire was administered to control and treatment groups drawn from a population of university faculty and staff. Both groups responded to series of questions designed

to measure their attitudes toward beef consumption and beef cattle slaughter. The treatment group also watched a video of the humane beef cattle slaughter process immediately after answering the first series of questions, which was designed to capture attitudes about raising beef cattle for consumption. The American Meat Institute originally produced the video for the Glass Walls Project and permitted its use in this study. Consumers in the treatment and control groups had positive attitudes toward raising cattle for human consumption, humane beef cattle slaughter, education about slaughter, and consumption of beef. Consumers who viewed the video had significantly ( $P < 0.05$ ) more positive attitudes about humane beef cattle slaughter, transparency about the slaughter process, and education about slaughter. These attitudes demonstrate that viewing a video of humane beef cattle slaughter is a successful method for educating consumers about the slaughter process. Using this method of communication is effective in inoculating consumers to negative messages that could influence consumers' attitudes about humane beef cattle slaughter.

**Key Words:** beef, slaughter, video

**411 A high percentage of beef bull pictures in semen catalogs have obscured feet and legs.** March K. Franks and Temple Grandin\*, *Colorado State University, Fort Collins, CO.*

To assist semen buyers in evaluating conformation, bull photos should have fully visible feet and legs. A total of 1,379 beef bull pictures were surveyed to determine visibility of feet and legs from 4 American semen company websites. Five different breeds were represented: Angus, Red Angus, Hereford (polled and horned), Simmental, and Charolais. In addition to visibility, other variables were surveyed to establish frequencies and correlations. These included breed, color, material that obscured visibility such as grass, picture taken at livestock show or outside, semen company, photographer, video, age of bull, and semen price. A visibility score was given to each bull picture. The results showed that 19.4% of the pictures scored a visibility score of one, where the bull's feet and legs were fully visible. In the rest of the pictures, the bull's feet and legs were hidden to some degree, 42.9% hid the hooves (visibility score 2), 32.5% hid both the hooves and the dewclaws (visibility score 3), 1.5% covered the entire legs up to the brisket (visibility score 4); and 3.8% hid 2 feet or legs while the other 2 were fully visible (visibility score 5). Correlation (Spearman's test) between the bull's age and the first 4 visibility scores was statistically significant  $P < 0.001$ . As age increased the feet and legs were more likely to be visible in the bull's picture. Semen price and visibility score correlation was also statistically significant  $P = 0.0143$ . Higher semen cost was associated with higher percentage of pictures with hidden feet and lower legs. The results show that many bull semen catalog pictures need to have greater leg and foot visibility.

**Key Words:** bull, cattle, hoof

## Food Safety

### 412 Residue of three fluoroquinolone (ciprofloxacin, norfloxacin, and ofloxacin) in commonly consumed meat products.

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The study was conducted to investigate residue of 3 fluoroquinolone (ciprofloxacin, norfloxacin, and ofloxacin) in commonly consumed meat products in the study area. Microbiological assay, followed by HPLC, was used to screen market-ready meat for residues of 3 fluoroquinolones. Three hundred twenty samples, comprising 80 each of beef, chicken, pork, and chevon (goat) were collected from open markets. Initial screening by microbiological assay revealed that 50%, 55%, 40% and 40% of beef, chicken, pork and chevon respectively were positive for residues of antibiotics to which *Escherichia coli* was susceptible. Further analysis revealed the presence of ciprofloxacin, norfloxacin and ofloxacin and varying concentrations in the different meat types. Overall mean concentration of ciprofloxacin was 242.50 µg/kg, which differed significantly ( $P < 0.05$ ) from the means of 80.86 µg/kg and 33.46 µg/kg for norfloxacin and ofloxacin respectively. Mean ciprofloxacin was highest in chevon (345.62 µg/kg); beef had the highest concentration of norfloxacin and ofloxacin at 173.40 µg/kg and 79.28 µg/kg respectively. Ofloxacin was the least in frequency and abundance in all meat types. Results obtained in this study have practical implications for public health and will lead to steps that will further enhance the safety of foods originating from animals to protect the animal production industry.

**Key Words:** fluoroquinolone, meat, residue

### 413 Abattoir waste management practices in Ibadan metropolis.

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The study evaluated waste management practices in 2 major abattoirs in Ibadan, South West, Nigeria, through investigative survey/questionnaire, to educate dwellers in the vicinity of the danger abattoir operations pose to health and to assess the ethical practices of abattoir operators. Water samples taken from well water around the abattoirs were assessed for selected physical, chemical and microbiological properties. General Linear Model (GLM) procedure was used for statistical analysis. The survey showed that 100% of abattoir operators in both abattoirs disposed waste manually using spade, 90% sweep and wash the waste into open drainage, eventually flowing into streams and rivers around, 90% do not treat waste in any way before dumping at sites. All these give rise to the offensive odor and stench characteristic of the abattoir environment, leading to eutrophication. Study further showed that though the residents of the two abattoirs were aware of the danger abattoir operations pose to their health, majority of them (63%) were not ready to move from this vicinity. Water samples from the wells taken for analysis for physical, chemical and microbiological properties showed that heavy metal concentration-Cu (0.00) and Pb (0.00) at both abattoirs, Fe-0.00 at Bodija, 0.05± 0.01 at Akinyele, Zn-0.03 ± 0.03 and 0.16 ± 0.03 at Bodija and Akinyele respectively were not significantly different from each other, and were below the maximum permissible limit. Turbidity, total dissolved solids (TDS) and total suspended solids (TSS) were significantly different from each other in the two abattoirs. Total aerobic count (TAC) and coliform count (TCC) were  $2.1 \times 10^6$  and  $6.3$

$\times 10^5$ , respectively, in the well water at Bodija, while TAC and TCC were  $1.7 \times 10^6$  and  $7.6 \times 10^5$  in wells at Akinyele. High biochemical oxygen demand (BOD) of  $7.05 \pm 0.25$ , dissolved oxygen (DO)  $6.30 \pm 0.14$  and TAC of  $2.8 \times 10^6$  obtained on Wednesdays in both locations, significantly differed from those of Fridays. The high microbial load and its health implications further confirm the need to treat abattoir waste rather than discharge them to the environment. Portable water should be provided for operators.

**Key Words:** abattoir, waste management, food safety

### 414 Bioaccumulation of heavy metals, phenol, and polycyclic aromatic hydrocarbons in differently singed, skin-on, red Sokoto buck goats.

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The safety of skin-on meat obtained from singed carcasses is a matter of public health concern. This study was therefore carried out to investigate the concentration of heavy metals [lead (Pb), cadmium (Cd), zinc (Zn), manganese (Mn) and copper (Cu)], phenol and polycyclic aromatic hydrocarbons (PAH) in red Sokoto buck goat carcasses singed using fire wood, kerosene, scrap tire and liquefied gas (LG). A total of 24 good grade red Sokoto buck goats weighing between 18 and 20 kg were randomly distributed into each of the 4 treatments in a completely randomized design. Each treatment was replicated 6 times. The total PAH and phenol contents were determined by HPLC, whereas the minerals were measured by atomic absorption spectroscopy (AAS). PAH levels were highest ( $P < 0.05$ ) in scrap tire singed carcasses (0.040 mg/kg) and least in LG singed carcasses (0.001mg/kg). Pb and Mn were below detectable limit in carcasses singed with LG while the concentrations were similar ( $P > 0.01$ ) in the other treatments. Cd was not detected in any of the treatments. Zn concentration was highest ( $P > 0.05$ ) in carcasses singed with kerosene (0.005mg/kg). The level of phenol ranged from 0.02 Gae/kg in LG singed carcasses to 0.38 Gae/kg when scrap tire was used. Material used in singeing was found to have profound effect on heavy metal, phenol, and PAH depositions on skin-on meat from red Sokoto buck goats.

**Key Words:** skin-on, singed carcass, heavy metal

### 415 Antibiotic resistance of bacteria from commercial silages in Israel.

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Antibiotic resistance has become a major issue in health care treatment with the emergence of multidrug resistant pathogens, which is attributed to the excessive use of antibiotics in medicine and in agriculture. Accumulating evidence suggests that agricultural sources such as sewage effluents, biosolids and animal manure are potential sources of antibiotic resistant bacteria and resistance genes. The objective of the current study was to determine the magnitude of antibiotic resistance in lactobacilli and enterobacteria in commercial wheat and corn/sorghum silages in Israel. Mixed corn-sorghum silage and wheat silage were sampled at 2 commercial cattle feeding centers in Israel. Grab samples were taken at a depth of 10 cm from the center (pH = 3.9 in both silages) and shoulders (pH = 6.8 and 4.4, respectively) of each bunker silo. Isolates



of *Enterobacteriaceae* and *Lactobacillus* were screened on ampicillin, tetracycline, ciprofloxacin, ceftriaxone, erythromycin kanamycin and vancomycin. Surprisingly, in both types of silage, the vast majority of lactobacilli isolates were resistant (based on EUCAST MIC clinical breakpoint values) to all the 7 tested antibiotics. These isolates are currently being screened for the presence of clinically relevant antibiotic resistance genes to determine the potential for horizontal-transfer to other bacteria. The resistance of the enterobacterial isolates was significantly lower than that of the lactobacilli, however all of the isolates

were resistant to at least 2 of the 5 tested antibiotics, and some isolates were resistant to all 5. Interestingly, isolates from the corn-sorghum silage (sewage irrigation) were significantly more resistant than those isolated from the wheat silage (rain irrigated) (resistant to 3.25 of the screened antibiotics vs.2.5, respectively). This study demonstrates the vast scope of antibiotic resistance in silage, which may contribute to antibiotic resistance propagation through the food chain.

**Key Words:** silage, antibiotic resistance

## Forages and Pastures Symposium: Implications of climate change on the resiliency of forage and pasture production systems

**416 Managed grassland resiliency to climate change: Shifting species composition buffers climate change effects on plant production and forage quality.** Rebecca L. McCulley\*<sup>1</sup>, A. Elizabeth Carlisle<sup>1</sup>, Allison L. Cooke<sup>2</sup>, Matthew M. Conley<sup>3</sup>, Bruce A. Kimball<sup>3</sup>, and Jim A. Nelson<sup>1</sup>, <sup>1</sup>University of Kentucky, Lexington, KY, <sup>2</sup>Metabolic Disease Institute, University of Cincinnati, Cincinnati, OH, <sup>3</sup>US Arid-Land Agricultural Research Center, USDA-ARS, Maricopa, AZ.

Managed grasslands cover significant acreage in the eastern half of the United States, and are dominated by non-native grass species that may or may not respond to climate change similarly to native species. It is important to understand how plant species composition, plant production, and forage quality will respond to predicted warming and alterations in precipitation because managed grasslands can improve environmental quality and provide the forage base for animal production in the region. We hypothesized that warming would cause species shifts (increased relative abundance of C<sub>4</sub> vs. C<sub>3</sub> species) and would reduce plant production and forage quality, but increases in precipitation would ameliorate these effects. We tested this hypothesis using a field-based, climate manipulation, located in central Kentucky. In a mixed species pasture, we established 5 replicate plots of 4 climate treatments: an ambient control, increased temperature, increased precipitation, and the combination of increased temperature and precipitation. Treatments were applied for 5 consecutive years (2009–2013), and species composition, plant biomass, and forage quality were measured seasonally in all years. Warming significantly increased the relative abundance of C<sub>4</sub> grasses, especially in summer and fall. However, in the spring, C<sub>3</sub> grasses remained the dominant plant functional type in all plots. Climate treatment effects on production varied by year, but when harvested biomass was summed across the 5-year experiment, no significant effect was observed. Effects of climate treatments on forage quality metrics (% crude protein and lignin) also varied by year and season, but were less dramatic than effects on plant production and species composition. Increased temperature and precipitation did not substantially alter plant production or forage quality, but did promote C<sub>4</sub> vs. C<sub>3</sub> grass dominance, illustrating the potential of species compositional changes to buffer ecosystem response to climate change. This result suggests that although dominant plants will differ, managed grasslands of the eastern United States will continue, from an animal forage production perspective, to function similarly to today.

**Key Words:** pasture, climate change, forage production

**417 Carbon sequestration potential for forage and pasture systems.** Vern S. Baron\*<sup>1</sup>, R. Howard Skinner<sup>2</sup>, and Gilles Bélanger<sup>3</sup>, <sup>1</sup>Agriculture and Agri-Food Canada, Lacombe, AB, Canada, <sup>2</sup>USDA-ARS, University Park, PA, <sup>3</sup>Agriculture and Agri-Food Canada, Quebec City, QC, Canada.

Grassland soils are a reservoir of organic and inorganic carbon. Regionally, grasslands are CO<sub>2</sub> sources or sinks depending on management, soil organic carbon (SOC) concentration and climate. Land management changes (LMC) affect SOC sequestration rate, duration and C-store at steady state. A common hypothesis is that higher grassland productivity increases SOC sequestration to a higher steady state. High SOC-sequestration rates occur for 5 to 10 yr after LMC, but continue slowly up to 50 yr. Permanent grasslands are at steady state for CO<sub>2</sub>

exchange. The most significant LMC for SOC accumulation is conversion from cropland to grassland where C-inputs from perennial roots and residues are higher than annual crops. Residue- and root-C inputs for Alberta perennial forage systems were 2.3 to 3 times greater, than barley silage. SOC sequestration rates for improved pastures on degraded soils in South-Eastern US were up to 1.4 Mg SOC ha<sup>-1</sup> yr<sup>-1</sup>. Applications of fertilizer-N, manure and using legumes increased SOC through higher root-C inputs, but not always permanently, due to the increased degradable-C fraction. Manure-C application effectively replaces surface soil-C on Quebec dairy farms. But, micro-meteorological studies on net ecosystem exchange (NEE) on old pastures in Pennsylvania, and Alberta (>6% soil organic matter), indicated that when harvested forage-C was removed from NEE, net Biome-C loss occurred in 90% of the years. Increasing productivity through N-application in Pennsylvania resulted in lost SOC, but a 5-species forage mixture increased productivity and sequestered more SOC than a 2-species mixture. Specific LMC in this grassland was less important to sequestration rate than initial SOC content. Quebec studies show that grass and legume species respond differently to increased atmospheric temperature and CO<sub>2</sub> concentration. Elevated temperature and CO<sub>2</sub> caused a higher root C: N ratio, resulting in a lower in vitro degradation rate, indicating that reduced SOC-degradation rates under future climates might be possible. How a concomitant change in species mixture affects SOC and what effects changing moisture and temperature regimens have on potential SOC-sequestration at a regional level can only be speculated.

**Key Words:** C-sequestration, forage management

**418 Climate-related risk management in agriculture: Its importance for coping with current and future climate changes in the southeastern United States.** B. V. Ortiz\*<sup>1</sup>, C. Fraisse<sup>2</sup>, D. Dourte<sup>2</sup>, W. Bartels<sup>2</sup>, D. Zierden<sup>3</sup>, and P. Knox<sup>4</sup>, <sup>1</sup>Auburn University, Auburn, AL, <sup>2</sup>University of Florida, Gainesville, FL, <sup>3</sup>Center for Ocean-Atmospheric Prediction Studies, Tallahassee, FL, <sup>4</sup>University of Georgia, Athens, GA.

Agricultural food and feed production is extremely important to the southeastern (SE) United States, because it not only provides quality food and feed for residents but also contributes to the region's economy. However, the stability and sustainability of the SE agriculture is affected by the interannual and seasonal variability of the climate, and could be at much higher risk under the projected future climate. Currently, El Niño Southern Oscillation (ENSO), especially El Niño and La Niña phases, makes important contributions to the year-to-year variations in conditions. ENSO has the strongest influence in the SE climate during the winter and spring months. During those months El Niño phase brings wet and cold conditions and La Niña phase results on dry and warm conditions. Several research studies have shown the relationship of ENSO phases and changes in row crops and forages yield, pest, and diseases; therefore the importance of the use of the ENSO forecast as a risk management strategy. The impact of weather and climate conditions on livestock farming (e.g., milk yield and composition, beef productivity) and, especially extreme events have been also documented. The current US National Climate Assessment released in 2014 report data of the effect of future climate scenarios on rainfall, ambient temperature, extreme events, among other variables. By 2050 in the SE, compared with the period 1971–2000, SE summer rainfall is expected to decline

up to 20%, the number of days per year with maximum temperature above 95°F, heat waves, is expected to be in the range of 25 to 35 d, summer ambient temperature could increase in the range of 4°F to 8°F. Projections of future precipitation patterns are less certain than projections for temperature increases; however, tropical storms are projected to be fewer in number globally, but stronger in force, and extreme precipitation events are expected to increase in number. Several climate research, education, and extension regional projects funded by USDA-NIFA are currently implemented to help stakeholders with adoption of new or adaptation of current management strategies to increase resilience and reduce potential impacts. The Southeast Climate Extension project (<http://www.agroclimate.org/seclimate/>), a multi-state and multi-institution project, is using a participatory approach toward engaging producers, extension specialists and agents, and farmers associations on the identification, development and evaluation of climate adaptation strategies. Hand-on workshops on the use of and awareness of web-based climate decision support tools hosted in [www.Agroclimate.org](http://www.Agroclimate.org) are also very popular among the SE clientele. Awareness of the future climate projections and potential adaptation strategies should be considered into the risk management package.

**Key Words:** adaptive capacity, climate variability, climate change

**876 Intensive grazing systems can enhance carcass production with the same methane emissions.** A. Berndt\*<sup>1</sup>, L. S. Sakamoto<sup>2</sup>, A. P. Lemes<sup>3</sup>, A. F. Pedroso<sup>1</sup>, J. R. M. Pezzopane<sup>1</sup>, T. C. Alves<sup>1</sup>, D. F. Vilas Boas<sup>4</sup>, R. Ruegger<sup>5</sup>, P. P. A. Oliveira<sup>1</sup>, <sup>1</sup>*Embrapa Southeast Livestock, Sao Carlos, Brazil*, <sup>2</sup>*Animal Productivity and Quality Program, FZEA/USP, Pirassununga, Brazil*, <sup>3</sup>*Veterinarian Medicine, FCAV/UNESP, Jaboticabal, Brazil*, <sup>4</sup>*FAPED, Brazil*, <sup>5</sup>*CAPES/EMBRAPA, Brazil*.

Enteric methane emissions are significant from countries with considerable livestock production, including Brazil. Various strategies exist for the mitigation of greenhouse gas emissions and, in general, the

intensification of production systems reduces emissions intensity; that is, emissions of CO<sub>2</sub> equivalents per unit of product (meat or milk). The objective of this trial was to measure CH<sub>4</sub> emissions and animal performance in beef cattle in 4 Brazilian grazing systems with different levels of intensification. The study was conducted at EMBRAPA Southeast Livestock, in São Paulo State, Brazil. Twenty-four Nelore steers were distributed across 4 representative grazing systems: (1) irrigated pasture with high stocking rate (IHS); (2) dryland pasture with high stocking rate (DHS), planted with *Panicum maximum*; (3) dryland pasture with moderate stocking rate (DMS); and (4) degraded pasture (DP), planted with *Brachiaria decumbens*. The IHS and DHS systems were composed of 12 paddocks each, and the DMS system consisted of 6 paddocks, all under rotational grazing. The DP system was managed under continuous stocking. Animals were kept in the same grazing systems from weaning until slaughter. Methane measurements were taken during 4 seasons using the SF<sub>6</sub> tracer technique and gases were analyzed by Shimadzu GC 2014. Data were analyzed using the SAS MIXED procedure and averages were compared using the Tukey test and considered significant at  $P < 0.05$ . The animals finished in the DP system presented the lowest ( $P < 0.01$ ) carcass weights ( $265.5 \pm 15.6$  kg), whereas those finished in the IHS system presented the highest ( $328.3 \pm 10.2$  kg). The average daily methane emissions were similar at  $202.7 \pm 38.6$  g/d, as were emissions per kilogram of live weight at  $409.1 \pm 59.7$  (g of CH<sub>4</sub>/kg of live weight). At the end of the experimental period, total methane emissions were similar at  $66.9 \pm 12.7$  kg, whereas carcass gain over the period was significantly lower ( $P = 0.0234$ ) for the DP group at  $73.2 \pm 18.0$  kg, versus  $105.8 \pm 25.0$  kg in the other intensified systems. The use of technologies that permit increased production favors animal performance, increases carcass production, and dilutes the emissions per kilogram of carcass produced.

**Key Words:** intensive grazing, Brazil, methane



# Growth and Development I

**419 Growth, intake, and health of Holstein heifer calves fed an enhanced diet pre-weaning with or without exogenous estradiol immediately post-weaning.** Adam J. Geiger<sup>\*1</sup>, Robert E. James<sup>1</sup>, Catherine L. Parsons<sup>1</sup>, Anthony V. Capuco<sup>2</sup>, and Robert M. Akers<sup>1</sup>, <sup>1</sup>Department of Dairy Science, Virginia Polytechnic Institute and State University, Blacksburg, VA, <sup>2</sup>United States Department of Agriculture-Agricultural Research Service, Beltsville, MD.

Enhanced feeding (HP) of heifers post-weaning but before puberty can reduce mammary gland (MG) development and milk yield. Better pre-weaning (PW) nutrition is likely beneficial. Our objectives were to determine effects of PW HP and exogenous estradiol (E<sub>2</sub>) on growth, intake, and health. Thirty-six Holstein heifer calves were reared on: 1) a control milk replacer (MR) fed at 454 g powder/day (CON; 20% crude protein [CP], 20% fat), or 2) an accelerated MR fed at 1135 g powder/day (ACC; 28% CP, 25% fat). MR feeding was reduced 50% at wk 8. Starter was offered after wk 4 but balanced between treatments. BW and frame measures were taken weekly with intakes and health monitored daily. At weaning a subset of calves were killed (n = 6/diet). Remaining calves received E<sub>2</sub> implants and were killed at wk 10. The 4 treatments were: 1) CON, 2) CON + E<sub>2</sub> (EST), 3) ACC, and 4) ACC + E<sub>2</sub> (ACCEST). Data were analyzed using PROC GLIMMIX in SAS and treatment differences were separated with the PDFIFF function. Significance was declared when  $P < 0.05$  and tendencies when  $0.10 > P > 0.05$ . ACC calves were heavier at weaning (75 vs 51 kg,  $P < 0.01$ ). E<sub>2</sub> did not influence BW. ACC calves had greater average daily gain (ADG) between wk 1 to 7 ( $P < 0.01$ ). Post-weaning ADG was similar between CON, ACC, and ACCEST calves, but greater in EST calves ( $P < 0.05$ ) than ACC calves. At first slaughter, ACC calves had greater carcass, MG, thymus, liver, and spleen weights ( $P < 0.01$ ). EST calves had heavier MG than CON calves ( $P < 0.01$ ) and ACCEST calves heavier reproductive tracts and uteri compared with ACC calves ( $P < 0.01$ ). ACC calves consumed more MR DM, CP, fat, and ash PW ( $P < 0.01$ ). CON calves consumed more starter DM, CP, fat, NDF, ADF, and ash PW ( $P < 0.01$ ). These differences were not observed post-weaning. Fecal scores (FS) were greater for ACC calves (1.7 vs 1.5;  $P < 0.01$ ) PW. FS were lower for EST calves post-weaning. Results show that PW HP increases weight and frame measures pre and post-weaning. PW HP increased ADG PW, but not post-weaning. E<sub>2</sub> may elicit beneficial responses in some calves. Analysis of collected samples will allow us to determine cellular and molecular processes responsible for tissue differences.

**Key Words:** mammary gland, estradiol, milk replacer

**420 Plane of nutrition affects Holstein bull calf growth, bone mineral density and organ size.** Meghan E. MacGhee<sup>\*</sup>, Sarah R. McCoski, Camilla H. K. Hughes, Sally E. Johnson, and Alan D. Ealy, Virginia Polytechnic Institute and State University, Blacksburg, VA.

Early postnatal milk replacer diet affects ADG and feed efficiency and can have long-term consequences on calf growth, health and productivity. This study examined how plane of nutrition affects calf body weight, height, bone mineralization, and organ size. On postnatal d 1, Holstein bull calves (n = 4–5/diet/time point) were assigned randomly to a low plane of nutrition (LPN; 20% CP, 20% fat; 441 g DM/d) or high plane of nutrition (HPN; 27% CP, 10% fat; 882 g DM/d during wk 1 and 1131 g DM/d thereafter). Starter grain (25% CP, 4% fat) was offered at 1% BW beginning at wk 3. At 2, 4, or 8 wk of age, calves were euthanized and bone mineralization and body composition were

determined with Dual Energy x-ray Absorptiometry (DEXA). Visceral organs were removed and weighed, and empty carcass weight (ECW) was recorded. Data were analyzed by least squares ANOVA. Final BW and ADG were greater ( $P < 0.05$ ) in calves fed HPN at each time point. Hip heights and bone mineral densities were greater ( $P < 0.05$ ) for HPN calves than LPN calves at 8 wk but not at 2 or 4 wk. ECW-adjusted organ weights also were affected by diet. Thymus weight was greater ( $P < 0.05$ ) in HPN calves than LPN calves at 4 and 8 wk (0.37% vs. 0.13% and 0.64% vs. 0.22%, respectively; SEM = 0.03). Spleen weight was greater ( $P < 0.05$ ) in HPN calves than LPN calves at 8 wk (0.93% vs. 0.46%; SEM = 0.05). Also, kidney and liver weights were greater ( $P < 0.05$ ) in HPN calves at 4 wk, and lung weights were reduced ( $P < 0.05$ ) in HPN calves at 2 wk. Weights of the heart and pancreas did not differ based on plane of nutrition within each time point. Calf ECW was greater ( $P < 0.01$ ) in the HPN group at each time point. In summary, deviations in growth rates, bone mineralization, and organ weights were evident between calves fed HPN and LPN diets. These outcomes are consistent with the contention that HPN feeding accelerates growth and improves health in young calves. The beneficial effects of HPN on bone mineral density and immune-centric organs provide new possibilities for understanding the effects of early nutrition on calf immune responses and productive lifespan of the cow.

**Key Words:** milk replacer, organ growth, bone density

**421 Daily growth rate in Holstein Friesian heifers is affected by fasting insulin levels as newborn calves.** M. Van Eetvelde<sup>\*</sup>, M. M. Kamal, H. Bogaert, and G. Opsomer, Department of Reproduction, Obstetrics and Herd Health, Faculty of Veterinary Medicine, Ghent University, Merelbeke, Belgium.

Body growth and reproduction traits were followed in 51 female Holstein Friesian calves at one herd in Flanders (Belgium). After an overnight fast, 3-d-old calves were weighed and blood samples were taken to determine fasting glucose (GLU<sub>F</sub>) and insulin (INS<sub>F</sub>) levels. All calves were kept indoors during the first year of life, and were fed according to their requirements for maintenance and growth. Every 3 mo until their first calving, body weight was recorded, as well as insemination dates and date of calving. Calves weighed  $39.1 \pm 4.26$  kg at birth and were classified in 3 categories dependent on their daily growth rate during the first 6 mo of life: slow (<750 g/day; n = 6); moderate (750 to 950 g/day; n = 40) and fast growing (>950 g/day; n = 5). Analysis of variance was used to compare Glu<sub>F</sub> and body weight between categories and the differences in Ins<sub>F</sub> were assessed by the Kruskal-Wallis test. GLU<sub>F</sub> was  $6.0 \pm 0.63$  mmol/L without significant differences between the 3 groups ( $P = 0.16$ ). INS<sub>F</sub> was  $7.2 \pm 4.76$  mU/L in moderately growing calves. Slow growing calves had higher INS<sub>F</sub> of  $12.9 \pm 9.10$  mU/L in comparison to the fast growing ones ( $4.3 \pm 2.39$  mU/L,  $P < 0.05$ ). Calves were inseminated at 14.9 mo. Heifers that were growing > 950 g/day weighed  $472 \pm 45$  kg at first insemination, which was significantly more than the moderately and slow growing calves (respectively  $414 \pm 42$  and  $378 \pm 29$  kg,  $P < 0.05$ ). The effect of early growth was still visible at first calving, as the fastest growing heifers weighed  $734 \pm 144$  kg, which was significantly more than the heifers with a moderate growth rate ( $615 \pm 72$  kg,  $P < 0.05$ ). In human medicine, low INS<sub>F</sub> is seen in newborns that are small for gestational age, which is indicative for an increased insulin sensitivity to glucose and resembles conditions of prolonged fasting. Although in our research, no difference in birth weight was observed, the association of low insulin levels with subsequently high daily

growth rates resembles the human situation of catch up growth. Based on these preliminary results, we suggest that low fasting insulin levels in newborns could be the result of a suboptimal environment during gestation and could be a forecast of catch-up growth during early life.

**Key Words:** catch-up growth, insulin, Holstein Friesian heifers

**422 Maternal obesity (MO) during ovine pregnancy leads to increased collagen content and cross-linking in the myocardium of adult F<sub>1</sub> but not F<sub>2</sub> offspring.** Adel Bashir Ghnenis\*, John F. Odhiambo, Richard J. McCormick, and Stephen P. Ford, *Department of Animal Science, University of Wyoming, Laramie, WY.*

There is accumulating evidence that MO increases offspring risk of obesity and cardiovascular disease in later life. Utilizing a well-established (J. Anim. Sci. 2010. 88:3546) ovine model of diet-induced MO from conception through gestation, we reported that gestational d 135 fetal heart left ventricle weight and collagen content were markedly increased by MO. Further, in a Langendorff system, the d 135 fetal heart from MO fetuses could not sustain high work levels (FASEB J. 2010. 24:2066). Here we examined the effect of MO on increasing collagen content and crosslinking in the adult F<sub>1</sub> and F<sub>2</sub> offspring myocardium. Ewes were assigned randomly as controls (CON) and fed at 100% of NRC recommendations or to an MO group fed 150% of NRC from 60 d before conception to term. Left ventricular free wall myocardial samples were collected from adult (22 mo old) male F<sub>1</sub> and F<sub>2</sub> offspring of CON and MO ewes at necropsy following a 12-week ad lib feeding challenge. Myocardial tissue was either snap frozen in liquid nitrogen for collagen analyses or fixed and paraffin embedded for histological evaluation. Collagen concentration was determined by colorimetry as hydroxyproline equivalents. Collagen crosslinking was also determined by measuring the amount of hydroxylysylpyridinoline by immunoassay (Microvue, PYD EIA Kit, Quidel, San Diego, CA). Data were analyzed by mixed procedures of SAS. Myocardial collagen concentration was greater ( $P < 0.05$ ) in MOF1 compared with CON F1 offspring ( $1.73 \pm 0.10$  vs.  $1.42 \pm 0.07$   $\mu\text{g}/\text{mg}$ ,  $n = 6$ ). The differences in OBF1 and CONF1 collagen concentration were confirmed by visualizing trichrome stained sections of myocardial tissue. Myocardial collagen crosslinking was 11% greater ( $P < 0.05$ ) in OBF1 vs. CONF1 offspring. In contrast, myocardial collagen content and crosslinking did not differ between MOF2 and CONF2 offspring. These data provide evidence for programming of increased myocardial fibrosis in adult MOF1 offspring, which potentially results from MO-induced changes in fetal life and might impair cardiac muscle function in postnatal life.

**Key Words:** maternal obesity, myocardium, collagen

**423 Growth and lactation during gestation decrease placental efficiency in cattle.** M. Van Eetvelde\*, M. M. Kamal, H. Bogaert, and G. Opsomer, *Department of Reproduction, Obstetrics and Herd Health, Faculty of Veterinary Medicine, Ghent University, Merelbeke, Belgium.*

In adult pregnancy, the gravid uterus receives a high priority status for nutrient partitioning. A shift in this hierarchy in favor of maternal tissues, as seen in adolescent pregnancy, compromises placental growth and fetal development. This situation occurs during gestation in growing heifers and lactating cows, as a high priority status of respectively maternal tissue growth and lactation is then observed. To assess the effect of dam lactation and growth on placental and fetal development, placentas of 49 multiparous (MP) Holstein Friesian (HF) cows and 43 growing heifers [27 HF and 16 Belgian Blue (BB)] were compared with afterbirths of

27 non-growing, nonlactating MPBB cows. Placentas were weighed and cotyledons were removed, after which their surface was calculated by the formula  $\text{area}(\text{ellips}) = \pi ab$ ; with  $a = \text{half of major diameter}$  and  $b = \text{half of minor diameter}$ . Placental efficiency (PE) was assessed by the calf weight:cotyledonary surface ratio. Analysis of variance was used to compare placental weight and cotyledonary surface, differences in cotyledon number and PE were assessed by the Kruskal-Wallis test. Placentas of MPBB dams weighed  $5.9 \pm 1.79$  kg with a cotyledonary surface of  $0.53 \pm 0.110$  m<sup>2</sup>. Placentas of MPHf dams were 0.9 kg lighter ( $P < 0.01$ ) but tended to have a larger total cotyledonary surface ( $0.58 \pm 0.106$  m<sup>2</sup>,  $P = 0.05$ ). Placentas of heifers were smaller ( $4.7 \pm 1.34$  kg and  $0.48 \pm 0.085$  m<sup>2</sup>,  $P < 0.05$ ), without significant breed effect. The number of cotyledons in MPBB placentas ( $91 \pm 27.2$ ) was lower than in placentas of heifers and MPHf (respectively  $114 \pm 29.0$  and  $115 \pm 28.2$ ,  $P < 0.05$ ). In addition, MPHf dams had a greater proportion of small (<60 cm<sup>2</sup>) cotyledons; as mentioned in nutrient-restricted ewes. MPBB cows had the highest PE of  $107 \pm 25.1$  kg/m<sup>2</sup>, compared with  $74 \pm 13.5$  kg/m<sup>2</sup> in MPHf dams ( $P < 0.01$ ). BB and HF heifers had a similar PE of  $96 \pm 23.0$  and  $84 \pm 18.0$  kg/m<sup>2</sup> respectively. These preliminary findings suggest maternal growth and lactation during gestation to have a similar nutrient restrictive effect on placental development, which may result in a lower PE. This could be, at least partly, responsible for the differences in size of the offspring. Further research is necessary to assess the influence on organ development and future health of the calves.

**Key Words:** growth, lactation, placental development

**424 Effects of GH and IGF-I on proliferation and apoptosis of bovine mammary epithelial cells.** Hongrong Wang\*, Yun Ji, Xueyan Pang, Qing Tian, Mengzhi Wang, and Lihuai Yu, *College of Animal Science and Technology, Yangzhou University, Yangzhou, Jiangsu Province, China.*

Growth hormone (GH) may exert its function through direct activation of the the growth hormone receptor (GHR) in mammary gland or through stimulating insulin-like growth factor-I (IGF-I). We hypothesized that the reduction of milk yield caused in part by apoptosis of mammary epithelial cells depends on the ratio of GH and IGF-I which could be influenced by nutrition, however, evidence to support this suggestion is lacking. Therefore, the purpose of this study was to investigate the effects of GH and IGF-I alone and in combination on proliferation and apoptosis of bovine mammary epithelial cells cultured in vitro. Mammary epithelial cells isolated from bovine mammary tissue were identified by cytomorphology, immunocytochemistry and specific gene expression. Subcultured and purified cells were treated with GH (100 ng/mL) and IGF-I (100 ng/mL) alone or a combination of GH and IGF-I (each 100 ng/mL) in growth medium without fetal calf serum for 24 h. The mRNA abundance was quantified by RT-qPCR, cell proliferation was determined with a CCK-8 kit, and apoptosis was identified by an FITC-Annexin V/PI kit. Statistical analysis was carried out by using ANOVA procedure of SAS 9.1 software with Duncan's multiple-range test. Cell proliferation was not enhanced by adding GH alone ( $P > 0.05$ ) while it was promoted by IGF-I at about 6 h ( $P < 0.05$ ). The combination of GH and IGF-I increased cell proliferation from 4 to 24 h ( $P < 0.05$ ), which had no significant difference with IGF-I group ( $P > 0.05$ ); The supplementation of GH or IGF-I individually had no significant effect on the early apoptosis rates of cells ( $P > 0.05$ ), however, the late ( $P < 0.01$ ) and total apoptosis rates ( $P < 0.05$ ) were reduced. The late and total cell apoptosis rates were decreased by adding GH and IGF-I together ( $P < 0.01$ ). In addition, adding GH or IGF-I alone tended to decreased the mRNA abundance of IGFBP-5 ( $P < 0.1$ ) compared with the control, and the inhibitory effect was strongest when GH was combined with IGF-I

( $P < 0.05$ ). This study demonstrated that GH and IGF-I can regulate the number and activity of bovine mammary epithelial cell through promoting cell proliferation and inhibiting cell apoptosis to stimulate lactation.

**Key Words:** mammary epithelial cell, growth hormone, insulin-like growth factor-I

**425 Effects of recombinant bovine somatotropin (bST) administration at breeding on the cow, conceptus, and subsequent offspring performance of beef cattle.** Vitor R. G. Mercadante\*, Francine M. Ciriaco, Darren D. Henry, Pedro L. P. Fontes, Danilo D. Demeterco, Pedro H. S. Pereira, Nicolas DiLorenzo, and G. Cliff Lamb, *North Florida Research and Education Center, University of Florida, Marianna, FL.*

To determine the effects of administration of a low dose of slow-release bST (Posilac, Elanco, Greenville, IN) on hormone concentration and conceptus development, a total of 190 suckled beef cows were exposed to the 7-d CO-Synch+CIDR fixed-time AI (TAI) protocol. Cows were blocked by days postpartum, BCS, breed and randomly assigned to receive one of the following treatments: (1) 2 injections of 325 mg bST, one at TAI and a second injection 14 d after TAI (D-bST,  $n = 40$ ); (2) 1 injection of 325 mg bST at TAI and a placebo (saline) injection 14 d after TAI (TAI-bST,  $n = 48$ ); (3) a placebo injection at TAI and one injection

of 325 mg bST 14 d later (14D-bST  $n = 49$ ); and (4) 2 injections of placebo, one at TAI and a second injection 14 d after TAI (Ctrl,  $n = 53$ ). Pregnancy was determined via transrectal ultrasonography 35 d after TAI and conceptus development was assessed by measuring crown to rump length (CRL) on d 35 and crown to nose length (CNL) on d 65 after TAI. Blood samples were collected on d 0, 7, 14, 21, 35 and 65 relative to TAI to determine concentrations of and IGF-1. Plasma concentrations of pregnancy-specific protein B (PSPB) were also assessed on d 35 and 65 after TAI. Individual calf birth weight and sex were determined at birth. Procedure GLIMMIX of SAS was used to analyze all data with the appropriate models. There were no differences ( $P = 0.767$ ) among treatments on pregnancy to TAI ( $48.7 \pm 0.5\%$ ). Administration of bST at TAI increased ( $P < 0.01$ ) plasma concentration of IGF-1 on d 7, 14 and 21. However, CRL and CNL ( $12.11 \pm 0.4$  mm and  $17.09 \pm 0.2$  mm, respectively) did not differ ( $P = 0.231$ ) among treatments. Concentration of PSPB did not differ ( $P = 0.18$ ) among treatments and between days ( $P = 0.30$ ;  $2.69 \pm 0.11$  ng/mL), and gestation length ( $282 \pm 9$  d) did not differ ( $P = 0.49$ ) among treatments. In addition, calf birth weight was similar ( $P = 0.52$ ) among treatments. We conclude that administration of 325 mg bST during the time of TAI to suckled beef cows enhanced concentrations of IGF-1, but failed to improve pregnancy rates, fetal size, PSPB concentrations, and had no effect on calf birth weight.

**Key Words:** bovine somatotropin, fetal programming, IGF-1



## Horse Species Symposium: Exercise physiology of the horse

### 426 The effect of oxidative stress during exercise in the horse.

Carey A. Williams\*, Rutgers, The State University of New Jersey, New Brunswick, NJ.

Oxidative stress is an imbalance of the oxidant to antioxidant ratio in the body. In the following presentation, I will highlight studies from my laboratory along with other pertinent studies of oxidative stress in exercising horses. An increase in oxidative stress and changes in anti-oxidant status has been shown during endurance, intense exercise, and eventing competition in horses. Antioxidants are vitamins, minerals, and protein that must be synthesized in the body or obtained from the diet. Therefore, exercise level and diet are both factors that play a role in influencing the oxidative stress and antioxidant status of the equine athlete. Along with exercise intensity, duration and diet, age and conditioning program also have affected oxidative stress in the horse. The “free radical theory of aging” states that long-term effects of the degenerative changes associated with aging may create oxidative stress. However, in old horses ( $22 \pm 2$  yr), the amount of lipid peroxidation and blood antioxidant concentrations were similar to those found in mature but younger ( $12 \pm 2$  yr) horses. Same as older horses that may require dietary intervention to help combat oxidative stress so might young growing exercising horses. Studies have shown that yearlings ( $18 \pm 2.4$  mo) did not begin their exercise training with higher levels of oxidative stress in muscle or blood than mature mares ( $13 \pm 2.1$  yr). Prior to exercise conditioning, yearlings had lower lipid peroxidation and higher antioxidants than mature mares. Conditioning reduced oxidative stress and improved antioxidant status in mares, while few effects were seen in yearlings. This suggests that age alone was the biggest defense against oxidative stress after exercise. Other studies during competition (endurance, jumping, eventing, and racing) have investigated the influence on oxidative stress with varying results. These results will be expanded upon during the presentation. Even though there have been many studies examining the levels of lipid peroxidation, antioxidant status and other related metabolites in the horse during exercise, we still have a long way to go before we fully understand the large variation in results both with and without antioxidant supplementation.

**Key Words:** equine, antioxidant, oxidative stress

### 427 Effects of aging on mitochondrial function in skeletal

**muscle of Quarter Horses.** Chengcheng Li\*<sup>1</sup>, Sarah H. White<sup>2</sup>, Lori K. Warren<sup>1</sup>, and Stephanie E. Wohlgenuth<sup>1</sup>, <sup>1</sup>Department of Animal Sciences, University of Florida, Gainesville, FL, <sup>2</sup>College of Health Sciences, University of Kentucky, Lexington, KY.

**Abstract.** Research in human and rodents has shown an age-associated decline in physical function, aerobic capacity and skeletal muscle mitochondrial function, which in humans begins around the age of 50 yr. On the other hand, many horses can still actively work or compete beyond 20 yr of age, an age equivalent to a 65-year-old human. The purpose of the present study was to determine the age-related changes in fiber type composition and mitochondrial function in equine skeletal muscle. Muscle biopsies of right gluteus medius and triceps brachii from young ( $1.8 \pm 0.1$  yr;  $n = 24$ ) and aged ( $20 \pm 5$  yr;  $n = 10$ ) Quarter Horses were compared. High-resolution respirometry was performed on freshly sampled and subsequently permeabilized muscle fibers. Remaining tissue was frozen in liquid nitrogen and stored at  $-80^\circ\text{C}$  for measurement of fiber type composition and enzyme activities. Statistical differences were analyzed using one-way ANOVA and Holm-Sidak post

hoc analysis (Sigmaplot 12.0). We found that aged horses had a higher percentage of oxidative type I myosin heavy chain (MHC) isoform in both gluteus ( $P < 0.001$ ) and triceps ( $P = 0.024$ ) compared with young horses. The proportion of glycolytic type IIX MHC isoform tended to decrease with advancing age, particularly in triceps ( $P = 0.061$ ). The proportion of intermediate fiber type IIA MHC isoform was not affected by age. Age had no effect on mitochondrial respiration in gluteus; but triceps from aged compared with young horses had greater leak respiration ( $P = 0.038$ ), electron transport system capacity ( $P = 0.032$ ), and a tendency for a lower respiratory control ratio ( $P = 0.076$ ). Cytochrome *c* oxidase activity in both triceps ( $P < 0.001$ ) and gluteus ( $P < 0.001$ ) was lower in aged compared with young horses. Using citrate synthase activity as a marker, mitochondrial density increased by 26.8% in the gluteus of aged horses ( $P = 0.034$ ), but was unaffected by age in the triceps ( $P = 0.183$ ). Our data suggest that aging resulted in an increased percentage of oxidative type I fibers, increased mitochondrial density, and impaired mitochondrial function in Quarter Horse skeletal muscle.

**Key Words:** myosin heavy chain isoform, cytochrome *c* oxidase, high-resolution respirometry

### 428 Bones and muscles in endurance horses—Physiology, pathology, and clinical issues. Katja F. Duesterdieck-Zellmer\*, Oregon State University, Corvallis, OR.

Adaptation of muscle or bone to endurance exercise in the horse is of great interest, as specific training and nutrition regimens may maximize performance and prevent injury. Medical manipulation of muscle or bone, while ethically questionable, may also impact the fitness of these tissues. The speed in endurance competitions has increased in recent years, as has the incidence of fractures in endurance horses. The bones of endurance horses experience repetitive loading, resulting in microdamage, which can be repaired or may accumulate into stress fractures that can be catastrophic. This phenomenon is well known in flat-racing horses, but has received less attention in endurance horses. Apparently, front limb fractures are more common than hind limb fractures and most fractures occur in the metacarpal/metatarsal or proximal phalangeal regions in endurance horses, most of which were metatarsal/metacarpal condylar fractures. Information about specific training regimens directed towards prevention of certain bony injuries is available only for flat-racing horses, indicating a need for research in this field to benefit endurance horses. Additionally, bisphosphonate medications that have the potential to significantly affect bony remodeling have been recently approved for use in equines in many countries. While there is no information on specific effects of bisphosphonates on the skeleton in endurance horses, they may be less safe for them, compared with other equine athletes. Genetics determine the predominant muscle fiber types, total fiber number and relative size of fast and slow twitch fibers in horses, favoring certain breeds over others to be successful endurance athletes. Elite endurance horses have a high ratio of type I (slow twitch) and IIA (fast-oxidative glycolytic) muscle fibers and endurance training stimulates transition from fast twitch (type IIX) to slower twitch fibers (type IIA and I). Endurance training also causes hypertrophy of type I and IIA fiber types, resulting in increased aerobic and decreased anaerobic capacity of the muscle. On a cellular level, these changes are induced by quantitative and qualitative alterations in gene expression of both regulatory and structural genes.

**Key Words:** horse, muscle, bone

**429 Biomechanics of the exercising horse.** Hilary Clayton\*<sup>1,2</sup>,  
<sup>1</sup>*Sport Horse Science LLC, Mason, MI*, <sup>2</sup>*Michigan State University,*  
*East Lansing, MI.*

Horses have long been used for athletic pursuits due to their superb physiological and biomechanical abilities. This talk explores structural and functional specializations of the equine limbs in relation to athletic performance. During locomotion the limbs cycle back and forth relative to the trunk moving primarily in the sagittal plane. Protraction and retraction are facilitated by morphological adaptations that reduce limb mass, especially in the distal limb, and allow a more proximal location of the moment of inertia. The joint angulations and muscle architecture of the hind limbs facilitate the generation of forward and upward propulsion of the center of mass, whereas the more strut-like forelimbs control speed and direction of movement. During a stride each limb has stance and swing phases. During swing the limb is rapidly protracted and then there is a final period of retraction that reduces hoof velocity relative to the ground at impact. During the first 50 ms after hoof contact with the ground, the hoof is rapidly decelerated and brought to rest with

the effects of deceleration being mitigated if the hoof can slide a little forward and sink downward into the footing. The forces associated with hoof impact are damped within the hoof and distal limb so their effects are progressively attenuated in the more proximal parts of the limb. Vibrations associated with impact are damped by the digital flexor tendons. During the stance phase the limb is loaded by the mass of the horse (and rider). It is during this phase of the stride that stretching and recoil of elastic tendons makes a significant contribution to the exchange between kinetic and potential energy in the gaits that have a suspension phase. In the terminal part of the stance phase, tension applied to the third phalanx by the deep digital flexor tendon eventually exceeds the counteracting ground reaction force causing the heels to lift. During locomotion dynamic balance is maintained by the interactions between momentum that carries the horse forward, gravity that pulls the body downwards, and the propulsive forces from the limbs. As each limb contacts the ground it 'catches' the body, raises it and provides propulsion to maintain the forward progression.

## Lactation Biology II

**430 Glucose activates translation factors in muscle but not in mammary glands of lactating dairy cows when essential amino acids are in excess supply.** Kelly Nichols\*<sup>1</sup>, Michelle Carson<sup>2</sup>, Julie J. M. Kim<sup>1</sup>, John A. Metcalf<sup>2</sup>, John P. Cant<sup>1</sup>, and John Doelman<sup>2,1</sup>, <sup>1</sup>Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada, <sup>2</sup>Nutreco Canada Agresearch, Guelph, ON, Canada.

To determine how glucose modulates protein synthesis when essential amino acids (EAA) are supplied in excess, 5 early-lactation Holstein cows (78 ± 13 DIM) were abomasally infused for 5 d with EAA and glucose in a 5 × 5 Latin square design, and abundances and phosphorylation states of ribosomal S6 kinase 1 (S6K1), eukaryotic initiation factor 4E binding protein-1 (4EBP1) and eukaryotic initiation factor 2α (eIF2α) in mammary tissue and longissimus dorsi were assayed. Treatments were saline, 844 g/d EAA in the profile of casein, 1126 g/d EAA, 844 g/d EAA + 1000 g/d glucose, or 1126 g/d EAA + 1000 g/d glucose. Cows were fed a diet containing 6.96 MJ/kg NE<sub>L</sub> and 12% crude protein. Measurements were subjected to ANOVA using PROC MIXED in SAS, where cow was a random effect. While no differences were observed between levels of EAA, compared with saline, infusion of EAA increased arterial concentrations of EAA 3- to 4-fold ( $P < 0.01$ ), increased mammary uptake of EAA from plasma 66% ( $P < 0.01$ ), and led to 256 g/d higher milk protein yield ( $P < 0.01$ ). The addition of glucose to EAA infusions decreased Ile, Leu and Val concentrations by 29% ( $P < 0.01$ ), but did not affect mammary uptake of any amino acids ( $P > 0.40$ ) or milk protein yield ( $P = 0.32$ ). Infusion of EAA increased the mammary abundance of S6K1 ( $P = 0.01$ ) and tended to increase phosphorylated S6K1 abundance ( $P = 0.09$ ), indicating activation of mRNA translation. Infusion of glucose tended to increase the mammary abundance of phosphorylated eIF2α ( $P = 0.09$ ) and decrease the total abundance of eIF2α ( $P = 0.07$ ), both of which are inhibitory to mRNA translation. In muscle tissue, EAA infusion increased the phosphorylation state of 4EBP1 ( $P = 0.02$ ), which is stimulatory to mRNA translation. When glucose was added, the phosphorylation state of 4EBP1 increased ( $P = 0.11$ ), and total S6K1 abundance tended to increase ( $P = 0.11$ ). Thus, EAA activated regulators of mRNA translation in both mammary glands and skeletal muscle, while the addition of glucose activated pathways of mRNA translation in muscle tissue but not in the mammary glands.

**Key Words:** protein synthesis, mammary, muscle

**431 Early postnatal plane of nutrition of Holstein calves has an effect on milk production and feed intake during their first lactation.** Steffi Wiedemann\*<sup>1</sup>, Patricia Holz<sup>2</sup>, Hans-Juergen Kunz<sup>3</sup>, and Martin Kaske<sup>4</sup>, <sup>1</sup>Animal Health, Institute of Animal Breeding and Husbandry, Kiel University, Kiel, Germany, <sup>2</sup>Clinic for Cattle, University of Veterinary Medicine Hannover Foundation, Hannover, Germany, <sup>3</sup>Chamber of Agriculture of Schleswig-Holstein, Blekendorf, Germany, <sup>4</sup>Department for Farm Animals, University of Zurich, Zurich, Switzerland.

The objective of the study was to assess the effect of 2 different feeding strategies during the very early postnatal life of Holstein calves on subsequent first lactation milk production and feed intake. During the first 4 wk of life, calves were fed either ad libitum (AdL; ad libitum feeding of whole milk during wk 1 and milk replacer ([MR], 160 g/L) from d 8–28; n = 38) or according to a restrictive feeding protocol (RES; 4 L milk/d during wk 1, 6 L MR (120 g/L) from d 8–28; n = 30). Feeding

was similar in both groups after the 4th wk of life. All animals were kept individually during the first wk of life and in groups thereafter. The feed intakes were analyzed during the first 10 wk of life in all animals and during first lactation in 37 animals. Daily milk yield and monthly milk composition were recorded. Total energy intake during the first 4 wk was higher in AdL-calves compared with RES-calves (16.6 MJ ME/d vs. 10.2 MJ ME/d, respectively;  $P < 0.01$ ). Thereafter, no difference in energy intake was observed until the 10th wk of life. In AdL-calves the average daily gain was higher compared with RES-calves during the first 4 wk of life (0.72 vs. 0.45 kg/d;  $P < 0.001$ ), while age at first calving did not differ (765 vs. 777 d;  $P = 0.30$ ). In the AdL- and RES-group, 21 and 26 animals remained on the farm for a full 305-d first lactation, respectively. The FCM yield was higher in AdL-animals compared with RES-animals (29.2 ± 0.4 vs. 28.0 ± 0.4 kg FCM/d;  $P < 0.05$ ); but milk composition did not differ. The higher FCM yield was accompanied by a higher feed intake in AdL-animals (19.3 kg DM/d vs. 18.8 kg DM/d;  $P < 0.01$ ). The results indicate that an increased feeding intensity during early life has positive long-term effects on the milk production potential in the first lactation, which could be the result of an improved girth or height or of an enhanced development of mammary parenchyma.

**Key Words:** calf feeding, feed intake, milk production

**432 Interrelation of somatic cell count, lactate dehydrogenase, and immunoglobulin G during mastitis caused by different pathogens.** Lorenzo E. Hernández-Castellano\*<sup>1</sup>, Samantha K. Wall<sup>1</sup>, Roger Stephan<sup>2</sup>, and Rupert Bruckmaier<sup>1</sup>, <sup>1</sup>Veterinary Physiology, Vetsuisse Faculty, University of Bern, Bern, Switzerland, <sup>2</sup>Institute for Food Safety and Hygiene, Vetsuisse Faculty, University of Zurich, Zurich, Switzerland.

Somatic cell count (SCC) is the most widely used mastitis indicator. However, immunoglobulin G (IgG), part of the specific immune system, is transferred from blood to milk, and the extent of this transfer appears to be pathogen specific. As IgG measurement is not available for farmers, milk lactate dehydrogenase (LDH) activity has been considered a suitable mastitis indicator and a marker for the presence of IgG in milk. This study aims to analyze the correlation and linear regression of the variables SCC, LDH and IgG in mastitis produced by different bacteria, and to determine if LDH can be used as an indicator for elevated IgG. Four quarters of 38 cows on 2 dairy farms with automatic milking systems were sampled. Selection criteria was based on the composite milk SCC (>100,000 cell/mL) from each cow. Samples were measured for SCC, LDH, IgG and cultured for bacteriology. SCC, LDH and IgG data were analyzed using the CORR and GLM procedures of SAS. When milk samples were not grouped by bacterial population, IgG and LDH were the highest correlated variables ( $r = 0.49$ ) compared with IgG and SCC ( $r = 0.41$ ) or LDH and SCC ( $r = 0.41$ ). Prediction equations for all variables had an R-square ≤ 0.24. After quarter milk samples were classified by bacteria (control, *S. aureus*, *C. bovis*, *E. coli*, other *Staphylococcus* spp. and *S. uberis*), different correlations and regression patterns were observed. Control and infected quarters showed a positive correlation between LDH and SCC, with the exception of those infected with *E. coli* and *S. uberis*. A positive correlation between SCC and IgG was observed in quarters where *C. bovis* was identified. LDH/SCC regression slopes differed from the control group. Several differences between LDH/SCC and LDH/IgG regression slopes were observed among bacterial infections. In conclusion, LDH appears to be a good indicator for some bacterial infections (namely *C. bovis*); however the use of both SCC and



LDH may increase the mastitis detection rate, particularly infections caused by *C. bovis*, other *Staphylococcus* spp. and *S. aureus*.

**Key Words:** mastitis, lactate dehydrogenase, SCC

**433 The innate immune response of bovine mammary epithelial cells to live or heat-inactivated *Mycoplasma bovis*.** Christina Zbinden<sup>\*1,3</sup>, Paola Pilo<sup>2</sup>, Joachim Frey<sup>2</sup>, Rupert M. Bruckmaier<sup>1</sup>, and Olga Wellnitz<sup>1</sup>, <sup>1</sup>Veterinary Physiology, Vetsuisse Faculty University of Bern, Bern, Switzerland, <sup>2</sup>Institute for Veterinary Bacteriology, Vetsuisse Faculty University of Bern, Bern, Switzerland, <sup>3</sup>Graduate School for Cellular and Biomedical Sciences, University of Bern, Bern, Switzerland.

Although *Mycoplasma bovis*, an emerging etiological agent of bovine mastitis, lack classical virulence factors, they induce an immune reaction and inflammation in the host. Because effects on the bovine mammary immune system are not yet well characterized, this study aimed to investigate the immunogenic effects of *M. bovis* on the mammary gland in an established primary bovine mammary epithelial cell (bMEC) culture system. Primary bMEC in 4th passage of 4 different cows were challenged with live or heat-inactivated *M. bovis* strain JF4278 isolated from acute bovine mastitis in Switzerland with a multiplicity of infection (MOI) of 108, as well as with the type strain PG45 with a MOI of 30. The immune response of bMEC was evaluated after a co-incubation with mycoplasmas for 6, and 24 h at 37°C by measuring the relative mRNA expression of important immune factors by quantitative PCR. Live JF4278 *M. bovis* triggered a considerable immune response in bMEC ( $P < 0.05$ ), reflected by the upregulation of relative mRNA expression ( $\Delta\Delta CT$ ) of tumor necrosis factor (TNF)- $\alpha$  (6 h:  $4.5 \pm 0.9$ ; 24 h:  $4.9 \pm 0.4$  threshold cycles [CT]), interleukin (IL)-1 $\beta$  (6 h:  $7.9 \pm 1.0$ ; 24 h:  $8.1 \pm 0.7$  CT), IL-6 (24h:  $2.1 \pm 0.4$ ), IL-8 (6 h:  $5.6 \pm 0.9$ ; 24 h:  $7.1 \pm 1.1$  CT), lactoferrin (after 24 h:  $4.4 \pm 1.0$  CT), Toll-like receptor (TLR)-2 (6 h:  $1.9 \pm 0.3$  CT), and serum amyloid A (SAA; 24 h:  $8.5 \pm 0.7$  CT). For live type strain PG45, very similar results were obtained; that is, a significant induction of TNF- $\alpha$  (6 h:  $4.9 \pm 0.4$ ; 24 h:  $4.4 \pm 0.5$  CT), IL-1 $\beta$  (6 h:  $7.9 \pm 1.1$ ; 24 h:  $9.2 \pm 0.5$  CT), IL-8 (6 h:  $5.6 \pm 0.3$ ; 24 h:  $7.6 \pm 1.0$  CT), lactoferrin (6 h:  $1.1 \pm 0.2$ ; 24 h:  $4.0 \pm 1.4$  CT), TLR-2 (24 h:  $2.3 \pm 0.7$  CT), and SAA (24 h:  $8.5 \pm 0.7$  CT). Interestingly, this cellular reaction was only observed in response to live, but not to heat-inactivated *M. bovis*. This study provides evidence that bMEC exhibit a strong inflammatory reaction in response to live *M. bovis*. The lack of a cellular response to heat-inactivated *M. bovis* may indicate that its intracellular localization or secondary metabolites are involved in *M. bovis* pathogenesis.

**Key Words:** bovine mastitis, *Mycoplasma bovis*, innate immune response

**434 Heat stress and amino acid supplementation affected dramatically the expression of genes related to mammary cell activity and number.** A. A. K. Salama<sup>\*1</sup>, M. Duque<sup>2</sup>, K. Shahzad<sup>3</sup>, and J. J. Loo<sup>3</sup>, <sup>1</sup>Grup de Recerca en Remugants (G2R), Departament de Ciència Animal i dels Aliments, Universitat Autònoma de Barcelona, Bellaterra, Spain, <sup>2</sup>Grupo de Investigación Biogénesis and GRICA, Facultad de Ciencias Agrarias, Universidad de Antioquia, Medellín, Colombia, <sup>3</sup>Department of Animal Sciences, University of Illinois, Urbana; IL.

Heat stress (HS) causes reductions in milk yield and content of fat and protein. It is not clear whether these losses are due to reduced number or activity (or both) of mammary epithelial cells. To test mechanisms

by which mammary metabolism is impaired by HS, MAC-T cells were incubated in different ambient temperature conditions: thermo-neutral (TN; 37°C) and heat stress (HS; 42°C). In both conditions, 3 mediums varying in amino acid (AA) concentrations were used. These media were: optimal amino acid profile as control (Con), control plus methionine (Met), and control plus arginine (Arg). Consequently, there were 6 treatment combinations: TN-Con, TN-Met, TN-Arg, HS-Con, HS-Met and HS-Arg. After incubation, cells were harvested and RNA was extracted for the study of gene expression by quantitative RT-PCR. Both HS and AA increased ( $P < 0.01$ ) the expression of heat shock proteins (HSP70A1A), transcription and translation factors (RPS6KB1, JAK2, AKT2), and AA transporters (SLC1A5, SLC7A1). The expression of PPARG (fat transcription regulation), FASN (de novo fatty acids synthesis), BCL2L1 (anti-apoptotic), and AKT1 (cell survival) decreased ( $P < 0.05$ ) with HS, but increased ( $P < 0.05$ ) with AA. Furthermore, HS downregulated ACACA (de novo fatty acids synthesis) and upregulated BAX (apoptotic). Supplementation with Met or Arg upregulated ( $P < 0.05$ ) genes related to transcription and translation (MAPK1, MTOR, SREBF1), cell proliferation (MKI67), and insulin signaling (IRS1). The expression of EIF4EBP1 (inhibitor of protein synthesis) was upregulated by HS, but downregulated by Met. Results suggest that heat stress exerts its negative effect on milk production at least in part by inhibiting mammary synthetic capacity as well as increasing apoptosis of mammary cells without affecting cell proliferation. Supplementation with AA (especially Met) increased mammary synthetic activity and had a positive effect on cell number by increasing proliferation and decreasing apoptosis. This raises the possibility that supplemental rumen-protected Met during heat stress might have a positive effect on mammary metabolism.

**Key Words:** mammary cells, gene expression, heat stress

**435 Effects of different lysine/methionine pattern and glucose level on expression of the key genes involved in milk protein transcription and translation in bovine mammary epithelial cells.** F. Wang<sup>1</sup>, J. Q. Wang<sup>1</sup>, D. P. Bu<sup>\*1,2</sup>, X. M. Nan<sup>1,3</sup>, and S. Lian<sup>1</sup>, <sup>1</sup>State Key Laboratory of Animal Nutrition, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>2</sup>CAAS-ICRAF Joint Laboratory of Agroforestry and Sustainable Animal Husbandry, World Agroforestry Centre, East and Central Asia, Beijing, China, <sup>3</sup>Synergetic Innovation Center of Food Safety and Nutrition, Harbin, China.

The synthesis of milk protein requires the availability of amino acids and a large supply of energy. Lysine, methionine and glucose regulate milk protein production of lactating dairy cows in vivo. Moreover, the content and ratio of lysine to methionine can regulate milk protein synthesis via cellular signaling pathways involving JAK2-STAT5 and mammalian target of rapamycin (mTOR). This study was conducted to investigate the effects of different lysine/methionine pattern and glucose level on milk protein synthesis in vitro, mainly focused on genes related to JAK2-STAT5 and mTOR. Primary bovine mammary epithelial cells (BMEC) were obtained from Holstein dairy cows and cultured in Dulbecco's modified Eagle's medium-F12 medium containing 10% fetal bovine serum. BMEC were subjected to 4 treatments arranged in a 2 × 2 factorial design with lysine/methionine ratio (3:1 vs. 2.3:1, namely balanced vs. unbalanced) and glucose level (17.5 mM vs. 2.5mM, namely high vs. low) as 2 factors. In this experiment, total casein content, cell proliferation and genes expression related to JAK2-STAT5 and mTOR pathways were measured. Compared with low level of glucose groups, casein content and cell proliferation increased in groups with high level of glucose ( $P < 0.05$ ). Casein content was also higher in lysine/methionine balanced groups than in unbalanced groups

( $P < 0.01$ ). Expression of *CSN1S2*, *CSN2*, *LALBA*, *STAT5*, *ELF5*, *mTOR* ( $P < 0.01$ ) and *CSN1S1* ( $P < 0.05$ ) were upregulated in groups with high level of glucose compared with low level groups. The upregulation of *CSN1S2*, *CSN2*, *LALBA*, *JAK2*, *STAT5*, *ELF5*, *mTOR* ( $P < 0.01$ ) and *CSN1S1*, *CSN3* ( $P < 0.05$ ) were also observed in Lysine/Methionine balanced groups compared with unbalanced groups, while *EIF4EBP1* was downregulated ( $P < 0.05$ ). In conclusion, proper ratio of Lysine to methionine and high level of glucose may directly accelerate the BMEC proliferation and regulate the expression of genes related to milk protein transcription and translation, which can increase milk protein synthesis.

**Key Words:** mammary epithelial cell, glucose, amino acid

**436 Trans-10,cis-12 CLA regulates SREBP1 activation in bovine mammary epithelial cells through proteasomal degradation of Insig1.** Liang Chen\*, Andrea Lengi, and Benjamin Corl, Virginia Tech, Blacksburg, VA.

*trans*-10,*cis*-12 conjugated linoleic acid (t10,c12-CLA) was linked to milk fat depression in dairy cows; transcription factor sterol response element binding protein-1 (SREBP1) regulates fatty acid synthesis. SREBP1 activation and migration to the nucleus requires the removal of Insig1, a protein that anchors SREBP1 in the endoplasmic reticulum membrane. The molecular basis orchestrating the effect of t10,c12-CLA on bovine SREBP1 activation has not yet been elucidated. We hypoth-

esize that t10,c12-CLA reduces SREBP1 activation through delay of Insig1 degradation. In the present study, we employed a bovine mammary epithelial cell line (Mac-T) and found that, mRNA and protein levels of SREBP1 declined over 56% when cells were treated with 60  $\mu M$  or greater t10,c12-CLA for 24 h ( $P < 0.05$ ). Similar dose effects were observed in the mRNA expression of SREBP1-regulated genes including FAS, SCD1, and Insig1. Compared with 0  $\mu M$  t10,c12-CLA, 60  $\mu M$  or higher CLA increased Insig1 protein expression over 2-fold in cells transfected with FLAG-tagged Insig1 ( $P < 0.05$ ). The effect was greater with t10, c12-CLA than other fatty acids including *cis*9, *trans*11-CLA, linoleic acid, or oleic acid when cells were treated with 75  $\mu M$  for 6 h. Further investigation revealed that increased FLAG-Insig1 was due to the inhibitory effect of t10,c12-CLA on the proteasomal degradation of Insig1. Cells treated with 75  $\mu M$  t10,c12-CLA or 10  $\mu M$  MG132, a proteasome inhibitor, for 6 h had 2.5-fold greater accumulation of FLAG-Insig1 compared with 0  $\mu M$  t10,c12-CLA ( $P < 0.05$ ). The degradation of FLAG-Insig1 was delayed when cells were treated with 75  $\mu M$  t10,c12-CLA for 6 h. These findings suggest that t10,c12-CLA plays a role in regulating SREBP1 activation by reducing proteasomal degradation of Insig1. We conclude that stabilized Insig1 retains SREBP1 in the ER preventing activation and migration to nucleus, thus reducing lipogenic gene transcription.

**Key Words:** conjugated linoleic acid, SREBP1, Insig1

# Milk Protein and Enzymes Symposium: High milk protein foods— Challenges and opportunities in structures and digestion

**437 The role of milk proteins in the development of high protein foods.** Harjinder Singh\*, *Riddet Institute, Massey University, Palmerston North, New Zealand.*

The global demand for milk protein has increased significantly in recent years due to better understanding of its nutritional value, physiological and bioactive properties. A growing body of research shows that a greater intake of protein may be beneficial in sports performance, weight management, lean muscle mass retention, satiety and general wellbeing. Much of the evidence regarding many of these health benefits focuses on milk proteins, particularly whey proteins. As a result, the demand for milk protein-enriched food and beverage products has increased enormously over the last 5 years. Several milk protein products, such as milk protein concentrates, whey protein concentrates and whey protein isolates, are now available and can be added to beverages, yogurt, soups, desserts and bars. However, some functional properties of milk proteins, such as aggregation and viscosity, pose challenges to developing acceptable food formulations at high protein concentrations. For example, the addition of protein will typically cause a food product to become excessively thick and will create stability issues during processing. In addition to maintaining suitable stability and texture, protein at high concentration has a marked impact on the taste and flavor profile of the product. Although some solutions to these problems have been developed by the food industry, we need to have better understanding of the fundamental behavior of proteins at high concentrations in various food formats. Recent studies indicate that beneficial effects of protein on health are not just dependent on sufficient protein intake; the rate at which amino acids are released and absorbed, as well as the generation of bioactive peptides influence biological potency. Thus, the interactions of milk proteins in the food and in the gastro-intestinal tract can play an important role in modulating the efficiency of protein digestion and the magnitude of its biological effects. In the future, it may be possible to design structures based on milk protein preparations to optimize the delivery of amino acids, hence modulating the postprandial physiological responses.

**Key Words:** milk protein, functionality, digestion

**438 Structuring food for improving nutrient bioavailability: The case of dairy gels.** Didier Dupont\*<sup>1</sup>, Florence Barbe<sup>1</sup>, Steven Le Feunteun<sup>2</sup>, Olivia Menard<sup>1</sup>, Yann Le Gouar<sup>1</sup>, Amelie Deglaire<sup>1</sup>, Juliane Floury<sup>1</sup>, Didier Remond<sup>3</sup>, and Beatrice Laroche<sup>4</sup>, <sup>1</sup>*INRA-Agrocampus Ouest, Rennes, France*, <sup>2</sup>*INRA-AgroParisTech, Grignon, France*, <sup>3</sup>*INRA-Université Clermont 1, Clermont-Ferrand, France*, <sup>4</sup>*INRA, Jouy-en-Josas, France.*

The food matrix structure is one of the key drivers to control the fate of food in the digestive tract and, consequently, the kinetics of nutrient release. Milk is “the” perfect raw material to build a wide variety of structures and can be really seen as a “Lego box” from which all the constituents can be separated (cracking) and re-assembled into different structures. The behavior in the gastrointestinal tract of milk protein matrices of identical composition but different micro and macrostructures was investigated in the present study. Six multi-cannulated and catheterized mini-pigs were fed 6 different dairy matrices (raw and heat-treated milks, acid and rennet gels). Effluents from the duodenum and mid-jejunum as well as plasma from the abdominal aorta were

collected over 7h. Structure was shown to drive the time of residence of food in the stomach. The liquid-gel transition led to a significant increase in the gastric emptying time. Liquid matrices generated a fast and intense peak of proteins in the intestine and amino acids in the bloodstream. Peptidomic analysis showed different patterns between intestinal samples. Finally, rennet gels had a different behavior than the acid ones forming a compact coagulum in the stomach that slowed down the gastric emptying and delayed the release of amino acids. A mathematical model of digestion was built from these data. Therefore, controlling the time of residence of food in the stomach by playing on its structure should allow to design products with a fast release of nutrients particularly adapted for elderlies, athletes etc... whereas foods persisting in the stomach should induce satiety and be dedicated to overweight people. As an example, 2 isocaloric yogurts with 3.3 and 8.4g of protein/100g respectively were manufactured and given to 11 pigs. Gastric emptying followed by scintigraphy was significantly slowed down for the protein-enriched yogurt leading to different kinetics of proteolysis. Ongoing experiments using biophysical methods will help us in understanding the mechanisms of gel particle breakdown in the stomach to design dairy products of new generation perfectly adapted to the nutritional needs of specific populations

**Key Words:** digestion, dairy, bioavailability

**439 Dairy protein and soluble fiber complexation: Effect on digestion and healthfulness of high protein foods.** Bongkosh Vardhanabhuti\*, *University of Missouri, Columbia, MO.*

Through controlled assembly of protein and polysaccharide, especially anionic soluble fibers, biopolymer particles with desirable functional properties can be created. The majority of research has focused on fabrication conditions under associative interaction (e.g., at pH < isoelectric point of protein) where they form complex coacervates. Much less attention has been given to their interactions under limited thermodynamic compatibility (e.g., at neutral or near neutral pH) where the positively charged patches of protein can form electrostatic attraction with negatively charged polysaccharides. Research has shown that unheated and heated soluble complexes between whey protein and anionic soluble fibers can be formed under limited thermodynamic compatibility. At optimum conditions, structuring of these complexes improves functional properties of whey protein, leading to enhanced texture and stability of food products. Using *in vitro* digestion model, recent studies have revealed that high protein beverages containing whey protein and anionic soluble fiber complexes can form intragastric gel when entering the simulated gastric environment. The transformation of liquid (e.g., beverages) to gel results in slow gastric emptying and slower protein degradation. Formation of intragastric gel also traps other ingredients including sugar, resulting in delayed sugar-release from the gel network. Potentially, intragastric gelation behavior could lead to the development of high protein food products with healthy blood sugar or high satiety claims. As the demand for high protein products continues to grow, complexes between dairy protein and soluble fibers could be utilized to improve the quality and healthfulness of high protein foods and beverages.

**Key Words:** dairy protein, digestion, soluble fiber



**440 Milk protein ingredients for controlling hardening of protein bars.** Thom Huppertz<sup>\*1,2</sup> and Sean Hogan<sup>3</sup>, <sup>1</sup>*NIZO food research, Ede, the Netherlands*, <sup>2</sup>*South Dakota State University, Brookings, SD*, <sup>3</sup>*Teagasc Dairy Products Research Centre, Moorepark, Fermoy, Ireland*.

Demand for high-protein snack bars as meal replacers and by consumers engaged in sports and dieting has grown significantly in recent years. These products provide healthy alternatives to conventional snacks because of the inclusion of high levels of protein (15–35%, w/w) and other nutritionally beneficial ingredients. Inclusion of high levels of protein in bars may result in adverse quality effects, in particular bar hardening, the degree of which is dependent on the ingredients and process used to produce the bars. Particularly water migration to the protein powder particles is a strong contributor to hardening of protein bars due to subtraction of water from different constituents, which can lead to hardening due to crystallization of carbohydrates. Protein bars containing whey protein isolate as the protein source generally developed less hardness than those containing caseinate. Part of these differences can be related to differences in moisture sorption of the protein powder particles in the water activity region of the protein bars (~0.5–0.7). For understanding protein functionality in bars, it is important to keep in mind that at these water activities, proteins should not be considered as fully hydrated proteins, but as partially hydrated. A further strong contributor to protein ingredient behavior in protein bars is powder particle size and microstructure. A broad particle size distribution and agglomeration were found to be beneficial to reduce development of hardness. Such effects can be attributed to the fact that less dense packing of powder particles is achieved in polydisperse systems, as a result of which hardness is reduced. The use of protein blends can also be beneficial from this perspective. Overall, initial hardness of protein bars can be described initially as a function of volume fraction of suspended powder particles and the efficiency of packing thereof. During storage, water migration and partial hydration and swelling of protein particles further contribute to hardness development. Hence, control of water sorption of powder particles and powder particle size and structure are prime routes to control the development of hardness in protein bars.

**Key Words:** whey protein, casein, protein bar hardening

**441 Technological challenges of high milk protein formulations.** Juan M. Gonzalez<sup>\*</sup>, *Global Technology at Mead Johnson Nutrition, Evansville, IN*.

The formulation and processing of high protein nutritional dairy products present several challenges. Two product formats to be discussed: powder products to be consumed as reconstituted beverages, and ready-to-eat (RTE) liquid products. Regarding powder products, the challenges are related to the consumer experience. Regarding the RTE liquid product, the challenges are related to processing and stability. High dairy protein powder products processed via powder blending face consumer experience challenges related to preparation, such as hydration, and to the sensory perception. The use of agglomeration technologies may provide a fast wetting of the powder. However, the hydration of the proteins may become a challenge as the proteins would compete for water available for hydration. The sugars and salts that may be present in the formulation would also compete for the bulk water available for hydration. Regarding the sensory experience, the presence of insufficient bulk water may lead to a perceived graininess in the beverage. The high protein content may produce a thick beverage more suitable to be spooned or chewed rather than swallow. RTE liquid products present challenges of hydration and thermal stability of the high protein matrix through processing. The processing stability of RTE high protein liquid beverages is related to the protein components. Whey based formulations are more stable than casein based formulations. The target pH, presence of salts, carbohydrates, and the buffering capacity of the high protein slurry would present formulation and processing challenges, as is in the case of concentrated infant formulas. The presence of reducing sugars at pH > 7 would encourage the formation of Maillard reaction color and flavor compounds that may or may not be desirable. This reaction also compromises the nutritional value of the protein as it makes unavailable amino acids, such as lysine, arginine and glutamine. The use of the shockwave steam injection technology allows for the processing of high protein dairy slurries. The viscosity observed in these shockwave-processed slurries is considerably lower than the one observed in traditionally heat-treated systems. Lower protein denaturation has been observed and this may be attributed to thermodynamic relationships within the process.

# Physiology and Endocrinology: Reproduction in cattle

**442 Comparison of TAI at GnRH injection and delayed insemination of non-estrus beef heifers.** Hazy R. Nielson\*<sup>1</sup>, Dan J. Kelly<sup>2</sup>, and Rick N. Funston<sup>1</sup>, <sup>1</sup>University of Nebraska, West Central Research and Extension Center, North Platte, NE, <sup>2</sup>Kelly Ranch, Sutherland, NE.

A study evaluated the effect of a 16-h delayed AI following GnRH administration in a hybrid estrus detection and time AI protocol in heifers not in estrus at the time of scheduled AI. Angus-based, crossbred heifers (n = 453) of the same origin were managed at the Kelly Ranch (Sutherland, NE) or the University of Nebraska West Central Research and Extension Center (North Platte, NE). Estrus was synchronized utilizing the melengestrol acetate (MGA)-PG protocol; heifers received MGA for 14 d. Nineteen d later, on d 33 of the protocol, heifers received a PG injection and estrus detection aids (Estroject) were applied. Heifers were considered to have expressed estrus when greater than 50% of the rub off coating was removed from the Estroject. Heifers (n = 319) were then removed from the herd and AI 12 h later (ESTRUS). Seventy-two hours following the PG injection, heifers whose Estroject were less than 50% activated were administered GnRH, and randomly assigned to 1 of 2 treatment groups; 1) immediately AI (n = 70, GNRH1) or 2) AI 16 ± 1 h following GnRH injection (n = 64, GNRH2). Data were analyzed using the GLIMMIX procedure of SAS (SAS Inst. Inc., Cary, NC) and the proportion of pregnant heifers was found using an odds ratio utilizing the ILINK function. Pre-breeding BW was similar ( $P = 0.58$ ) between ESTRUS, GNRH1, and GNRH2 (351, 346, and 349 ± 6 kg, respectively). Furthermore, pregnancy diagnosis BW among the treatments was similar ( $P = 0.48$ ; 376, 380, and 377 ± 6 kg; ESTRUS, GNRH1, and GNRH2, respectively). Heifers who were AI on their estrus, as determined by an activated Estroject, had significantly higher ( $P < 0.01$ ) pregnancy rate compared with heifers in both GNRH1 and GNRH2 groups (70 vs. 56, 47 ± 6%). Pregnancy rates did not differ ( $P = 0.56$ ) between GNRH1 and GNRH2 (56 vs. 47 ± 6%). Heifers in all groups reached a similar ( $P = 0.59$ ) percentage of mature BW before the breeding season (63 ± 1%). Final pregnancy rate was not different ( $P = 0.54$ ) for ESTRUS, GNRH1, and GNRH2 heifers (92, 89, 91 ± 4%). There was no significant benefit to delayed AI of non-estrus beef heifers compared with traditional timed AI at the time of GnRH injection.

**Key Words:** beef heifer, delayed insemination, estrus synchronization

**443 Bee Synch for synchronization and fixed-time AI of *Bos indicus*-influenced cows: An update.** Gary L. Williams\*<sup>1,3</sup>, Randy L. Stanko<sup>2</sup>, and Marcel Amstalden<sup>3</sup>, <sup>1</sup>Texas A&M AgriLife Research, Beeville, TX, <sup>2</sup>Texas A&M University-Kingsville, Kingsville, TX, <sup>3</sup>Texas A&M University, College Station, TX.

Synchronization methodologies that are successful in straight-bred English and Continental breeds have proven less successful in *Bos indicus*-influenced cattle, with fixed-time AI (FTAI) pregnancy rates of 35 to 40%. Because of these poor outcomes, few commercial cattlemen who utilize these types of cattle employ synchronization technologies and FTAI. Research has indicated that variation in the rate of maturation of the dominant follicle after CIDR removal in *Bos indicus*-influenced females is a major contributor to reduced pregnancy potential at FTAI. Herein, we provide an updated report on the use of a modification of the standard 5-day CO-Synch + CIDR protocol, the 5-day Bee Synch + CIDR (Bee Synch) which has been termed PG-5-Day + CIDR by

the Beef Reproduction Task Force. The protocol was designed to eliminate mature corpora lutea at the onset of synchronization, thus reducing circulating concentrations of progesterone, minimize variation in the rate of follicle maturation, and improve FTAI pregnancy rates. Initial studies evaluated the 5-Day CO-Synch + CIDR protocol alone (n = 100; Trial 1) or in comparison with Bee Synch (Bee Synch, n = 133; 5-Day CO-Synch, n = 136; Trial 2) at one location with mature cows. Subsequently, Bee Synch was evaluated at 4 additional locations utilizing Braford, Brangus, and Nelore crossbred cows. The 5-d protocol involved insertion of a controlled internal drug releasing device (CIDR) and i.m. injection of GnRH (100 µg; Factrel) on d 0. On d 5, the CIDR was removed and cows were treated i.m. with prostaglandin F<sub>2α</sub> (PGF; Lutalyse, 50 mg). Cows were inseminated at 66–72 h after CIDR removal and treated with 100 µg GnRH. Bee Synch employed a similar sequence except that all cows were treated with 25 mg PGF at the time of CIDR insertion. Pregnancy rate to FTAI in Trial 1 (5-Day CO-Synch + CIDR) was 33%. In trial 2, Bee Synch markedly improved FTAI pregnancy rates compared with the standard 5-day protocol (51.8 ± 0.9 vs 40.9 ± 5.7%;  $P < 0.04$ ). Cumulative FTAI pregnancy rates in cows (n = 702) treated with Bee Synch at 5 locations averaged 51.9 ± 3.1% (range = 40 to 59.3). Bee Synch appears to improve FTAI feasibility in *Bos indicus*-influenced cows.

**Key Words:** synchronization, *Bos indicus*-influenced, Bee Synch

**444 Effect of MGA versus CIDR estrus synchronization on estrus response and pregnancy rates in 311-d-old beef heifers.** Hazy R. Nielson\*<sup>1</sup>, Rosemary V. Anderson<sup>2</sup>, and Rick N. Funston<sup>1</sup>, <sup>1</sup>University of Nebraska, West Central Research and Extension Center, North Platte, NE, <sup>2</sup>Anderson Ranch, Whitman, NE.

A study compared the effect of melengestrol acetate (MGA)-PG and 14-d controlled internal drug release (CIDR)-PG estrus synchronization protocols on estrus response and pregnancy rates of 311-d-old Angus-based, crossbred heifers (n = 153). Fall-born heifers, at 10 mo of age, were randomly assigned to 1 of 2 estrus synchronization protocols in the spring (2 replications/treatment). Heifers in the MGA protocol received MGA for 14 d fed through the diet beginning on d 0 of the synchronization treatment period. Heifers in the CIDR treatment received the same diet as MGA heifers and were implanted with a CIDR (Eazi-breed CIDR) on d 2 of the treatment period and removed on d 16. Following estrus synchronization, heifers from both treatments were combined and received a single PG (Lutalyse) injection on d 32. Heifers with activated heat detection aids (Estroject) were AI 12 h following observation. All data were analyzed with the GLIMMIX procedure of SAS (SAS Inst. Inc., Cary, NC). Group BW was measured at weaning (198 kg) and before breeding (273 kg). Pre-breeding BW was 50.1% of predicted mature BW. Heifer age at breeding was not different ( $P = 0.12$ ) between MGA and CIDR treatment groups. Percentage of heifers demonstrating signs of estrus was similar ( $P = 0.42$ ) between synchronization treatment groups (CIDR vs. MGA, 71.5 vs. 77.4 ± 1.0%). Heifers not expressing estrus were not given an opportunity to become pregnant and removed from the herd. Pregnancy rates to AI of heifers expressing estrus (n = 115) were similar ( $P = 0.27$ ) between CIDR and MGA synchronization treatment (46.3 vs. 36.1 ± 6.8%). Bulls were placed with heifers at a 1:25 ratio 10 d following AI. Final pregnancy rate was also similar ( $P = 0.96$ ) between CIDR and MGA treatment groups (51.0 vs. 51.5 ± 7.4%). Heifer BW at pregnancy diagnosis was not different ( $P = 0.45$ ) between CIDR and MGA treatment groups (325 vs. 321 ± 3.4 kg). The

numerical 10% decrease in AI pregnancy rate in MGA compared with CIDR synchronization is not significant but is of interest. Approximately half of these 311 d old heifers exposed to AI and bulls became pregnant.

**Key Words:** beef heifers, estrus synchronization, heifer development

**445 Variation in timed-AI pregnancy rates in beef sires.** Bo R. Harstine<sup>\*1</sup>, Rodrigo A. C. Martins<sup>2</sup>, Adnan D. P. Rodrigues<sup>4</sup>, Leandro H. Cruppe<sup>1,3</sup>, Matthew D. Utt<sup>3</sup>, Lon D. Peters<sup>3</sup>, José L. M. Vasconcelos<sup>4</sup>, Mel DeJarnette<sup>3</sup>, and Michael L. Day<sup>1</sup>, <sup>1</sup>The Ohio State University, Department of Animal Sciences, Columbus, OH, <sup>2</sup>RM Reprodução Animal, Brasília, Brazil, <sup>3</sup>Select Sires Inc., Plain City, OH, <sup>4</sup>Faculdade de Medicina Veterinária e Zootecnia, UNESP, Botucatu, SP, Brazil.

Differences between timed-AI (TAI) pregnancy rates (PR) of beef bulls and collections within bull are difficult to determine due to limited number of females per breeding group. PR to TAI between 6 Angus bulls and between collections (combined 2 ejaculates) within bull were examined in large Brazilian beef herds. In 4 bulls, 3 collections obtained in a 2-week period were packaged in different colored straws. In 2 bulls, a single collection was packaged in 3 straw colors as controls. Straws contained 20 million total spermatozoa (0.5 ml). Nelore females (n = 6003) in 75 groups on 4 farms were synchronized for TAI. Every bull was represented in each group, and collection within bull was randomized across groups. Parity and body condition score (BCS) were recorded at TAI. Pregnancy was diagnosed approximately 35 d post-TAI (5614 complete records). Data were analyzed using GLIMMIX procedure of SAS with main effects of sire, collection(sire), farm, parity(farm), and BCS ( $\leq 2.75$  or  $\geq 3$  on a scale of 5). Estroject patches were used on a subset of cows (n = 3334; 2 farms) to assess estrus activity. PR was greater ( $P < 0.05$ ) when BCS was  $\geq 3$  and if females displayed estrus. PR differed ( $P < 0.05$ ) between sires and tended to differ between collections within sire ( $P = 0.09$ ; Table 1). The numeric range in PR was similar among straw colors for both individual collections and a control (Sire 3), illustrating that the impact of binomial variation, even within large sample sizes such as these, cannot be ignored. In conclusion, large and economically significant differences in fertility exist between bulls used for TAI, and knowledge of bull fertility is crucial for the consistency and success of beef TAI programs.

**Table 1 (Abstr. 445).** Pregnancy rates (PR; %) to timed-AI (TAI) for bulls and individual collections within bull

Bull	TAI PR per bull		SEM (%)	TAI PR by straw color		
	n	TAI PR		Purple	Red	Yellow
1	828	46.1 <sup>a</sup>	2.7	47.7	44.8	45.8
2	1,109	44.7 <sup>a</sup>	2.4	47.3	40.4	46.6
3 <sup>1</sup>	735	41.3 <sup>ab</sup>	2.9	42.3	44.7	36.8
4	1,026	39.2 <sup>ab</sup>	2.6	31.8	42.3	43.7
5 <sup>1</sup>	841	35.7 <sup>bc</sup>	2.6	35.5	35.8	35.7
6	1,075	30.6 <sup>c</sup>	2.1	26.5	34.5	30.7

<sup>a-c</sup>Means within the same column not sharing a common superscript differ significantly,  $P < 0.05$ .

<sup>1</sup>All straws from sire are from the same collection.

**Key Words:** fertility, AI, bull

**446 Effect of decreasing the duration of a PRID-synch protocol and addition of a second prostaglandin F<sub>2α</sub> treatment on fertility after resynchronization of ovulation in lactating Holstein cows.** V. G. Santos<sup>\*1,2</sup>, P. D. Carvalho<sup>1</sup>, C. Maia<sup>3</sup>, B. C. Carneiro<sup>3</sup>, A. Valenza<sup>4</sup>, E. M. Bettencourt<sup>2</sup>, and P. M. Fricke<sup>1</sup>, <sup>1</sup>Department of Dairy Science, University of Wisconsin-Madison, Madison, WI, <sup>2</sup>Departamento de Medicina Veterinária, Escola de Ciências e Tecnologia, Instituto de Ciências Agrárias e Ambientais Mediterrânicas, Instituto de Investigação e Formação Avançada, Universidade de Évora, Núcleo da Mitra, Évora, Portugal, <sup>3</sup>Diessen Servicos Veterinarios Lda, Évora, Portugal, <sup>4</sup>CEVA Sante Animale, Libourne, France.

Our objective was to evaluate the effects of (1) decreasing the interval between the first GnRH treatment (G1) and the PGF<sub>2α</sub> (PGF) treatment and (2) a second PGF treatment 24 h after the first during hormonal protocols for resynchronization of ovulation and timed artificial insemination (TAI). Lactating Holstein cows (n = 249) from 3 commercial dairy farms were randomly assigned to a nonpregnancy diagnosis to 1 of 3 hormonal protocols for resynchronization of ovulation: (1) a 7-d PRID-synch protocol with 1 PGF injection (7D1P: d 0; GnRH +PRID; d 7, PGF -PRID; 56 h, GnRH; 16 h, TAI); (2) a 7-d PRID-synch protocol with 2 PGF injections (7D2P: d 0, GnRH +PRID; d 7, PGF -PRID; 24 h, PGF; 32 h, GnRH; 16 h TAI); and (3) a 5-d PRID-synch protocol with 2 PGF injections (5D2P: d 0, GnRH +PRID; d 5, PGF -PRID; 24 h, PGF; 32 h, GnRH; 16 h, TAI). Ovaries of all cows were examined at G1 using transrectal ultrasonography, and cows were classified as either having or lacking a CL. Data were analyzed by logistic regression using PROC GLIMMIX of SAS. Pregnancy diagnosis was conducted 32 d after TAI using transrectal ultrasonography. Overall, P/AI 32 d after TAI did not differ ( $P = 0.83$ ) between primiparous and multiparous cows [47% (35/75) vs. 44% (77/174), respectively]. At 32 d after TAI, P/AI tended to differ ( $P = 0.09$ ) among treatments and was least for 7D1P cows, intermediate for 5D2P cows, and greatest for 7D2P cows [39% (35/89) vs. 44% (34/77) vs. 52% (43/83), respectively]. Furthermore, P/AI did not differ ( $P = 0.20$ ) between cows classified as having or lacking a CL at G1 of the resynchronization protocols [48% (83/174) vs. 39% (22/56), respectively]. Based on these preliminary data, addition of a second PGF injection to a 7-d PRID-synch protocol increased ( $P = 0.03$ ) P/AI 32 d after TAI by 13 percentage points (7D2P vs. 7D1P cows), whereas decreasing the interval between G1 and the PGF injection (5D2P vs. 7D2P cows) did not increase ( $P = 0.62$ ) P/AI 32 d after TAI in resynchronized Holstein cows. Supported by CEVA Sante Animale.

**Key Words:** timed AI, dairy cow, resynchronization

**447 Evaluating blood perfusion of the corpus luteum in beef cows during fescue toxicosis.** Garrett F. Cline<sup>\*1</sup>, Ashleigh M. Muth-Spurlock<sup>1</sup>, Benjamin E. Voelz<sup>2</sup>, Caleb O. Lemley<sup>1</sup>, and Jamie E. Larson<sup>1</sup>, <sup>1</sup>Mississippi State University, Mississippi State, MS, <sup>2</sup>Kansas State University, Manhattan, KS.

The aim of this study was to determine if fescue toxicosis altered blood perfusion in the corpus luteum (CL), and peripheral concentrations of progesterone (P4) in cattle. In 2 replicates, the estrous cycles of 36, non-pregnant Angus or Charolais cows were synchronized using the CO-Synch+CIDR protocol. Seven d after initiation of the protocol, cows were assigned (d 0) to 1 of 2 treatments: 2.5 kg of (1) Kentucky-31 endophyte-infected (E+; n = 14), or (2) MaxQ novel endophyte (NE; n = 12) tall fescue seed. On d 7 ovaries were examined using ultrasonography and only cows that had 1 CL present remained on the study (n = 26). Images of blood perfusion of CL, blood samples, rectal temperatures, and blood pressure of tails were collected on d 10, 13, 15 and 18. Images



of CL blood perfusion were analyzed using ImageJ software for pixel density, and scored visually (0 to 9 with 0 = no perfusion, 9 = complete perfusion) by 2 independent technicians. The MIXED procedure of SAS was used with day as a repeated measure. LSMeans and pooled SEM are reported. Cows receiving E+ had greater rectal temperatures ( $P = 0.02$ ;  $38.73 \pm 0.08^\circ\text{C}$ ) than those receiving NE ( $38.46 \pm 0.08^\circ\text{C}$ ), providing evidence the cows treated with E+ were influenced by fescue toxicosis. Pulse pressure and mean arterial pressure were decreased ( $P < 0.01$ ) in cows receiving E+ ( $54.08 \pm 3.21$  and  $80.06 \pm 2.71$  mmHg, respectively) than NE ( $67.12 \pm 3.11$  and  $91.37 \pm 2.93$  mmHg, respectively). Concentrations of P4 were similar ( $P = 0.54$ ) between cows receiving E+ ( $6.04 \pm 0.53$  ng/mL) or NE ( $6.36 \pm 0.63$  ng/mL). Pixel densities ( $P = 0.14$ ) and visual perfusion scores were similar ( $P = 0.11$ ) between cows receiving E+ ( $1,477.20 \pm 655.62$  pixels and  $2.23 \pm 0.34$ , respectively) or NE ( $2,934.70 \pm 718.20$  pixels and  $3.00 \pm 0.36$ , respectively). Mean CL volume was similar ( $P = 0.39$ ) between treatments. In conclusion, blood perfusion of CL or peripheral concentrations of P4 were not altered at the onset of fescue toxicosis, indicating that a decrease in blood perfusion of the CL may not be a primary mechanism involved in decreased reproductive efficiency of cattle during fescue toxicosis.

**Key Words:** blood perfusion, corpus luteum, fescue toxicosis

**448 Improved fertility of cows failing to respond to resynchronization of ovulation through presynchronization of ovulation or progesterone supplementation.** Julio O. Giordano\*<sup>1</sup>, Mark J. Thomas<sup>2</sup>, Grace Cuatucumba<sup>2</sup>, Mathew D. Curler<sup>2</sup>, Robert Wijma<sup>1</sup>, and Matias L. Stangaferro<sup>1</sup>, <sup>1</sup>Department of Animal Science, Cornell University, Ithaca, NY, <sup>2</sup>Dairy Health & Management Services, Lowville, NY.

Our objective was to evaluate treatments to increase fertility of timed-AI (TAI) services of lactating dairy cows without a corpus luteum (CL) at the time of the PGF injection of Resynch initiated  $32 \pm 3$  d after AI (RD32; GnRH-7d-PGF-56h-GnRH-16h-TAI). In Exp1, blood collection and ovarian ultrasound was conducted ( $n = 555$ ) at the PGF of RD32 to define a cutoff value for CL size that allows creating groups of cows with low and high fertility after TAI. P/AI were analyzed by logistical regression using PROC GLIMMIX of SAS. Based on P/AI, the CL size selected was 15 mm [ $\text{CL} \geq 15 \text{ mm} = 33.2\%$ ; (165/497) vs. NoCL or  $\text{CL} < 15 \text{ mm} = 10.3\%$  (6/58);  $P < 0.01$ ]. In Exp2, cows without a CL or a  $\text{CL} < 15 \text{ mm}$  (NoCL) were randomly assigned to (1) Ovsynch plus progesterone (P4) [OvP4; GnRH (G1) and CIDR-7d-PGF and CIDR removal-56h-GnRH-16h-TAI;  $n = 212$ ] or (2) presynchronization with GnRH (PreG) 7 d before Ovsynch [PreGOv; GnRH-7d-GnRH(G1)-7d-PGF(P)-56h-GnRH-16h-TAI;  $n = 192$ ]. Cows with a  $\text{CL} > 15 \text{ mm}$  (CLPG) at PGF of RD32 ( $n = 1,797$ ) completed RD32 to receive TAI. Binary data were analyzed by logistical regression using PROC GLIMMIX whereas quantitative data were analyzed by ANOVA using PROC MIXED of SAS. At 39 d after AI, overall P/AI were similar ( $P = 0.13$ ) for cows in OvP4 (34.4%), PreGOv (35.9%), and CLPG (31.1%). Cows inseminated in estrus had similar ( $P = 0.32$ ) P/AI [41.4% ( $n = 29$ ) OvP4, 24.4% ( $n = 41$ ) PreGOv, and 34.1% ( $n = 173$ ) CLPG] but P/AI tended ( $P = 0.06$ ) to differ for TAI cows (33.3% OvP4, 39.1% PreGOv, and 30.8% CLPG). Pregnancy losses were similar ( $P = 0.29$ ) for cows in OvP4 (2.9%), PreGOv (8.7%), and CLPG (11.6%). Ovulatory response to PreG was 86.0%. At G1, cows with a CL (86.6 vs. 16.4%;  $P < 0.01$ ), cows with  $\text{P4} > 1 \text{ ng/mL}$  (82.8 vs. 31.8%;  $P < 0.01$ ), P4 concentrations (3.7 vs. 1.1 ng/mL;  $P < 0.01$ ), and cows with a follicle  $> 10 \text{ mm}$  (94.9 vs. 80.9%;  $P < 0.01$ ) was greater in PreGOv than OvP4 group. Ovulatory response to G1 was 71.9 vs 58.3% for OvP4 and PreGOv ( $P = 0.03$ ). At the PGF before TAI, more cows had a CL in PreGOv than OvP4 (92.1 vs.

77%;  $P < 0.01$ ), cows with  $\text{P4} > 1 \text{ ng/mL}$  was similar (79.1 vs. 82.7%;  $P = 0.50$ ), but P4 concentrations were greater in PreGOv than in OvP4 (4.1 vs. 2.6 ng/mL;  $P < 0.01$ ). We conclude that presynchronization with GnRH 7 d before Ovsynch or Ovsynch plus P4 are 2 feasible alternatives to increase P/AI of cows without a CL at the PGF injection of RD32. Supported by New York Farm Viability Institute project AOR12-012.

**Key Words:** corpus luteum, resynchronization, dairy cow

**449 Interferon-tau and progesterone down-regulate cytochrome P450 1A and 2C in bovine endometrial epithelial cells.** Caleb O. Lemley\* and Christa L. Gilfeather, Mississippi State University, Mississippi State, MS.

The objective of the current study was to examine cytochrome P450 1A (CYP1A) and 2C (CYP2C) activity in bovine endometrial cell cultures following exposure to oxytocin (OT), interferon-tau (IFN), estradiol (E2), and/or progesterone (P4). Bovine endometrial epithelial cells were cultured to 80% confluence in 6-well plates. For experiment 1, cells were treated with 1  $\mu\text{g/mL}$  OT, 10 ng/mL IFN, a combination of OT+IFN, or control (CON) media for 24 h. For the second experiment, cells were treated with 1 ng/mL E2, 15 ng/mL P4, a combination of E2+P4, or CON media for 24 h. Following the hormone treatment, endometrial cells were harvested in lysis buffer containing protease and phosphatase inhibitors and frozen at  $-80^\circ\text{C}$  until further analysis. Treatments were performed in triplicate and the experiment was repeated 4 times ( $n = 12$  per treatment). The activity of CYP1A and CYP2C were determined using specific luminogenic substrates and expressed relative to mg of cellular protein. Data were analyzed using MIXED procedure of SAS and the model statement included hormone and replicate. Treatment with OT alone did not alter activity of CYP1A ( $P = 0.55$ ) or CYP2C ( $P = 0.46$ ) compared with CON cells. Activity of CYP1A was decreased in cells exposed to IFN ( $P < 0.01$ ) or OT+IFN ( $P < 0.01$ ) compared with CON. Similarly, activity of CYP2C was decreased in cells exposed to IFN ( $P < 0.01$ ) or OT+IFN ( $P < 0.01$ ) compared with CON. Treatment with E2 alone did not alter activity of CYP1A ( $P = 0.64$ ) or CYP2C ( $P = 0.06$ ) compared with CON cells. Activity of CYP1A was decreased ( $P < 0.01$ ) in P4 versus CON, while E2+P4 was not different ( $P = 0.38$ ) from CON. Activity of CYP2C was decreased in cells exposed to P4 ( $P < 0.01$ ) or E2+P4 ( $P < 0.01$ ) compared with CON cells. In summary, both interferon-tau and progesterone exposure decreased CYP1A and CYP2C activity. The mixed function monooxygenase enzymes, CYP1A and CYP2C, have been implicated in synthesizing embryotoxic compounds; therefore, down-regulation in the endometrium may be necessary during maternal recognition of pregnancy.

**Key Words:** cytochrome P450, endometrium, interferon-tau

**450 Epidermal growth factor promotes interferon-tau expression in bovine trophectoderm.** Sarah R. McCoski\* and Alan D. Ealy, Virginia Tech, Blacksburg, VA.

Significant pregnancy loss occurs in cattle during the early stages of embryogenesis and placentation. Several critical events occur in the first 3 to 4 weeks of gestation, and aberrations in any one of these events will prompt pregnancy loss. One such event is maternal recognition of pregnancy, when interferon-tau (IFNT) must be produced in sufficient quantities from the conceptus to prevent luteolysis and modify uterine function to favor pregnancy. Epidermal growth factor (EGF) is produced by the uterus and is a recognized embryotrophic factor. Specific activities in bovine embryos include improving rates of bovine embryo development and increasing trophectoderm (TE) proliferation. This

study examined whether EGF affects *IFNT* expression. A stable bovine TE cell line (CT1) was maintained in Dulbecco's Modified Eagle's Medium (DMEM). Cells were serum-starved and then exposed to various concentrations of human recombinant EGF. Total cellular RNA was extracted, and quantitative reverse transcription PCR was completed using *IFNT*-specific primers and ribosomal protein S9 (*RPS9*; internal control). Data were analyzed by ANOVA. In the first study, a 6 h dose response was completed (n = 3 replicate studies). Supplementation with 100 ng/mL EGF increased ( $P < 0.05$ ) *IFNT* transcript abundance when compared with controls ( $2.3 \pm 0.8$  fold effect). Exposure to 1 or 10 ng/mL EGF did not affect *IFNT* transcript abundance. In the second study, a 24 h dose-response was completed (n = 3 replicate studies). A trend in increased *IFNT* mRNA abundance was evident ( $P = 0.07$ ) when CT1 cells were supplemented with 1 ng/mL EGF ( $2.0 \pm 1.5$  fold vs. controls). Incubation with either 10 or 100 ng/mL EGF increased ( $P < 0.05$ ) *IFNT* transcript abundance when compared with controls ( $2.6 \pm 0.8$  and  $3.6 \pm 1.4$  fold effect, respectively). Moreover, the 100 ng/mL effect was more pronounced than the 1 ng/mL effect ( $P = 0.06$ ;  $1.75 \pm 0.7$  fold). These outcomes implicate EGF as a mediator of *IFNT* expression, and provide further evidence of how the maternal system may control *IFNT* production and other developmental events during early pregnancy in ways that dictate pregnancy retention or failure.

**Key Words:** interferon-tau, embryo, trophectoderm

**451 Effects of label-dose permethrin administration on reproductive function in superstimulated beef heifers.** Tyler M. Dohlman<sup>\*1,2</sup>, Marianna M. Jahnke<sup>1</sup>, James K. West<sup>1</sup>, Patrick E. Phillips<sup>1</sup>, and Patrick J. Gunn<sup>2</sup>, <sup>1</sup>*Veterinary and Diagnostic Production Animal Medicine, Iowa State University, Ames, IA*, <sup>2</sup>*Department of Animal Science, Iowa State University, Ames, IA*.

The objective was to study the effects of a commercial pyrethroid-based pour-on product, permethrin, on reproductive performance in superovulated beef heifers by assessing steroid biosynthesis and embryo quality. Nonpregnant, yearling beef heifers (n = 10;  $417 \pm 33$  kg;  $5.5 \pm 0.2$  BCS) were assigned by BW and breed to either 1) saline control (CON) or 2) permethrin pour-on administered at label dose (PYR). Superstimulation was achieved on all heifers utilizing a timed, 17-d, CIDR-based protocol with GnRH and PGF<sub>2a</sub> and decreasing total dosage of 240mg FSH administered twice daily for 4 d. Heifers were AI twice (at onset of estrus and 12 h later) by the same technician with frozen semen from single bull collection. To determine short and long-term effects of permethrin on embryo quality and steroid biosynthesis, superstimulation was initiated twice with collection of embryos occurring at 17 and 51 d post-treatment. Embryos were recovered 6.5 d after first AI via non-surgical flush and were evaluated by International Embryo Transfer Society standards. Blood was collected at standing estrus and d of embryo recovery. Estrogen (E<sub>2</sub>) and progesterone (P<sub>4</sub>) concentrations were analyzed via RIA. MIXED and GLIMMIX procedures of SAS were used to analyze continuous and categorical data, respectively. Heifer per flush was the experimental unit. Total embryos recovered did not differ due to treatment ( $P = 0.30$ ), but did decrease in flush 2 compared with flush 1 ( $P = 0.02$ ). Quality grade, total transferrable quality embryos (TQE), and overall flush success did not differ due to treatment ( $P = 0.16$ ). However, TQE was decreased in flush 2 compared with flush 1 ( $P = 0.05$ ). Total unfertilized oocytes was greater in CON ( $P = 0.04$ ). The PYR heifers tended to have less total P<sub>4</sub> ( $P = 0.15$ ) and P<sub>4</sub> per corpus luteum ( $P = 0.06$ ) at recovery. E<sub>2</sub> per ovulated follicle and E<sub>2</sub> per total ovarian structure was greater in flush 2 ( $P = 0.03$ ) but did not differ due to treatment ( $P = 0.23$ ). In summary, these data indicate

that permethrin administration at label dose in superstimulated beef heifers has a tendency to reduce P<sub>4</sub>, but embryo quality is not affected.

**Key Words:** permethrin, embryo, progesterone

**452 Effects of tamoxifen on pre-pubertal heifer reproductive tissues: Potential for disruption of tract development through alteration of related signaling pathways.** Abdullah Al Naib<sup>\*1</sup>, Ali.Y. Wood<sup>1</sup>, Hannah.L. M. Tucker<sup>1</sup>, Catherine.L. M. Parsons<sup>1</sup>, Victoria.L. McCracken<sup>1</sup>, Abigail.L. Zezeski<sup>1</sup>, Stacie.E. Deaver<sup>2</sup>, Britni.M. Brown<sup>1</sup>, Mike.M. Akers<sup>1</sup>, and Michelle.L. Rhoads<sup>1</sup>, <sup>1</sup>*Virginia Polytechnic Institute and State University, Blacksburg, Virginia*, <sup>2</sup>*Virginia Western Community College, Roanoke, Virginia*.

Pre-pubertal exposure of the developing reproductive tract (RT) to estrogen or xenoestrogens can have long-term consequences that compromise the reproductive performance of dairy cattle. This research examined the effects of the selective estrogen receptor modulator, tamoxifen (TAM), on gene and protein expression in the pre-pubertal RT with particular focus on signaling pathways that affect RT morphology. Tamoxifen was administered to heifer calves (n = 7) daily (0.3 mg/kg subcutaneously) from 28 to 120 d of age. Control calves (CON; n = 7) received an equal volume of excipient. Weight, gross measurements and samples of RT tissues were collected upon sacrifice at  $120.7 \pm 0.3$  d of age. Protein and mRNA were extracted from snap frozen samples of vagina, cervix, uterine body, ovary and oviduct. Data were analyzed using the proc mixed procedure of SAS version 9.1.3 (SAS Institute Inc., Cary, NC) followed by the Tukey-Kramer test as a multiple comparison test. As we previously reported, overall weight of the RT was dramatically decreased following TAM treatment ( $P = 0.01$ ). Both estrogen receptor  $\alpha$  (ER $\alpha$ ) protein and gene expression were dramatically reduced in the uterus, cervix and vagina ( $P < 0.01$ ). In oviduct, ER $\alpha$  protein was reduced in the TAM treated animals ( $P < 0.01$ ) while ER $\alpha$  gene expression was not affected. Similarly, the phosphorylated form of ER $\alpha$  protein was reduced in the cervix and vagina ( $P < 0.01$ ) and tended to be reduced in the uterus ( $P = 0.09$ ) of the TAM group. In contrast to other RT tissues, phosphorylated ER $\alpha$  protein abundance was greater in the ovaries of the TAM animals ( $P < 0.05$ ). Even though insulin-like growth factor-I (IGFI) gene expression was higher in the uterus, oviduct and vagina of the TAM group ( $P < 0.01$ ), IGFI receptor protein and gene expression did not differ. Total mitogen-activated protein kinases (MAPK) protein was higher in the oviduct, vagina and ovary ( $P < 0.01$ ) but was lower in the uterus of TAM treated heifers ( $P < 0.01$ ). The phosphorylated form of MAPK protein was similarly increased in the ovary but was decreased in the cervix of the TAM group ( $P < 0.05$ ). In conclusion, the bovine pre-pubertal RT is affected by TAM treatment. Further research is needed to determine if these effects have long-term consequences for reproductive performance.

**Key Words:** reproductive tract, tamoxifen, estrogen

**453 Effects of label-dose permethrin administration on reproductive function and testicular histopathology in yearling bulls.**

Tyler M. Dohlman<sup>\*1,2</sup>, Darin M. Madson<sup>1</sup>, Patrick E. Phillips<sup>1</sup>, Chris A. Clark<sup>3</sup>, and Patrick J. Gunn<sup>2</sup>, <sup>1</sup>*Veterinary and Diagnostic Production Animal Medicine, Iowa State University, Ames, IA*, <sup>2</sup>*Department of Animal Science, Iowa State University, Ames, IA*, <sup>3</sup>*Armstrong Research and Demonstration Farm, Iowa State University, Lewis, IA*.

The objective of this experiment was to study the effects of a commercial, pyrethroid-based pour-on product, permethrin, on reproductive

parameters and testicular histopathology of yearling beef bulls. Black Angus bulls ( $n = 60$ ;  $369 \pm 17$  d of age;  $509 \pm 33$  kg;  $6.2 \pm 0.5$  BCS) were assigned to either (1) saline control (CON) or (2) permethrin pour-on administered at label dose (PYR). All bulls had blood samples collected and were subjected to an industry standard breeding soundness exam (BSE) via electroejaculation at both 5 d before and 14 d post-treatment. Progressive motility and Eosin-Nigrosin stained morphology were analyzed using high power magnification with phase contrast microscopy. Plasma testosterone concentrations were analyzed via RIA. At 34 d post-treatment, bulls were slaughtered and one testicle per bull was collected for histopathological examination. Categorical and continuous data were analyzed with the GLIMMIX and MIXED procedures of SAS, respectively. Change in motility between BSEs was not different due to treatment ( $P = 0.69$ ). Although morphology improved across treatments between BSEs, PYR bulls had less improvement in percent of head ( $P < 0.001$ ) and tailless sperm ( $P = 0.05$ ) abnormalities compared

with CON, resulting in less improvement of primary abnormalities ( $P = 0.04$ ). Nonetheless, morphological differences did not change the overall outcome for satisfactory breeder status ( $P = 0.82$ ). Change in testosterone concentration did not differ due to treatment ( $P = 0.22$ ). Histopathological examination determined that testicular degeneration and tubule diameter did not differ as a result of treatment ( $P \geq 0.19$ ). It should be noted, however, that degeneration score (higher score having more degeneration) was positively correlated with primary abnormalities ( $P < 0.01$ ;  $r = 0.35$ ) and negatively correlated with normal sperm cells ( $P < 0.001$ ;  $r = -0.43$ ). In summary, these data indicate that a single use of permethrin at label dose in yearling Angus bulls results in minimal detrimental effects on semen morphology, but not to a degree that affects the ability of bulls to pass a standard BSE.

**Key Words:** permethrin, breeding soundness exam (BSE), sperm



## Ruminant Nutrition: Amino acids and metabolism

**454 Energy requirements for pregnant and nonpregnant Nellore cows.** Mateus P. Gionbelli<sup>\*1</sup>, Marcio S. Duarte<sup>2</sup>, Sebastiao C. Valadares Filho<sup>2</sup>, Edenio Detmann<sup>2</sup>, Tathiane R. S. Gionbelli<sup>1</sup>, Diego Zanetti<sup>2</sup>, and Luiz H. P. Silva<sup>2</sup>, <sup>1</sup>University of Lavras, Lavras, Minas Gerais, Brazil, <sup>2</sup>University of Viçosa, Viçosa, Minas Gerais, Brazil.

Forty-nine adult Nellore cows (32 pregnant and 17 nonpregnant) with average initial body weight of 451 ± 10 kg were used in a comparative slaughter study aiming to describe equations and relationships for prediction of net, metabolizable and dietary energy requirements for adult, pregnant and nonpregnant, *Bos indicus* cows. Feeding control was measured individually and cattle were fed either HIGH (ad libitum) or LOW (restricted feeding 1.2 times maintenance according to the NRC). The 32 pregnant cows were separated at random into 4 groups of 8 cows each (4 cows per each feeding level) and harvested at 136 ± 1, 189 ± 1, 239 ± 1 and 269 ± 1 d of pregnancy. The nonpregnant cows were harvested at different times of the experiment (85 to 216 d of feeding control) to keep them in experiment for a similar amount of time as the pregnant cows. The digestible energy and losses of energy as methane and urine were directly measured to establish the relations between GE, DE and ME. Energy content was analyzed in empty body and pregnant compounds. A set of relationships and equations based in the factorial method from ARC was used to estimate the nutrient requirements. The net energy requirement for pregnancy (NE<sub>p</sub>) estimated in this study was about 3/4 of those estimated by NRC. When estimated by a logistic model, the daily requirements for pregnancy showed an exponential increase up to 250 d of gestation and then decreased. However, when an allometric model was used to estimate the daily requirements for pregnancy, the maximum daily requirements were at birth. There were no differences in the dynamics of energy ( $P = 0.388$ ) in the cow's empty body weight pregnant free (EBW<sub>np</sub>) suggesting that the pregnancy does not affect the requirements for accretion of body reserves in cows. The partial efficiencies for use of metabolizable energy for maintenance, weight gain and pregnancy ( $k_m$ ,  $k_g$  and  $k_c$ ) were respectively 70, 53 and 12%. The efficiency of transformation of DE in ME was 0.80.

**Key Words:** *Bos indicus*, conceptus, metabolizable energy

**455 Rumen-protected methyl donors during the transition period: 1. Better postpartal performance in dairy cows supplemented with rumen-protected methionine (Smartamine M) than choline (ReaShure).** Z. Zhou<sup>\*1</sup>, M. Vailati Riboni<sup>1</sup>, E. Trevisi<sup>2</sup>, J. K. Drackley<sup>1</sup>, D. N. Luchini<sup>3</sup>, and J. J. Loo<sup>1</sup>, <sup>1</sup>University of Illinois at Urbana-Champaign, Urbana, IL, <sup>2</sup>Università Cattolica del Sacro Cuore, Piacenza, Italy, <sup>3</sup>Adisseo NA, Alpharetta, GA.

The objective of this study was to evaluate the efficacy of supplemental rumen-protected methionine (MET) and choline (CHO) on performance and health of transition cows. Eighty-one multiparous Holstein cows were used in a randomized complete block design with 2 × 2 factorial arrangement of MET (Smartamine M, Adisseo NA) and CHO (ReaShure, Balchem Inc.) level (with or without). Treatments (20–21 cows each) were control (CON), no MET or CHO; CON+MET (SMA); CON+CHO (REA); and CON+MET+CHO (MIX). From –50 d to –21 d before expected calving, all cows received the same diet (1.24 Mcal/kg DM) with no MET or CHO. From –21 d to calving, cows received the same close up diet (1.54 Mcal/kg DM) and were assigned randomly to treatments (CON, SMA, REA, or MIX). From calving to 30 DIM, cows were on the same postpartal diet (1.69 Mcal/kg DM)

and continued to receive the same treatments through 30 DIM. MET supplementation was adjusted daily at 0.08% DM of diet and REA was supplemented at 60 g/cow/d. No differences ( $P = 0.34$  or greater) were detected for pre- or postpartal body weight and body condition score. However, MET supplementation (SMA, MIX) led to greater ( $P < 0.05$ ) DMI compared with other treatments (CON, REA) in both close-up (14.40 vs. 13.13 kg/d, SEM 0.40) and first 30 d postpartum (19.30 vs. 17.15 kg/d, SEM 0.63). Milk yield in MET-supplemented cows (SMA, MIX) also was greater ( $P < 0.05$ ) compared with other (CON, REA) treatments (44.32 vs. 40.32 kg/d, SEM 1.29). Milk fat % did not differ in response to MET ( $P = 0.91$ ; 3.72% vs. 3.74%, SEM 0.11%) or CHO ( $P = 0.46$ ; 3.78% vs. 3.68%, SEM 0.10%). However, milk protein % was greater in MET-supplemented ( $P < 0.01$ ; 3.32% vs. 3.14%, SEM 0.05%) but not CHO-supplemented ( $P = 0.23$ ; 3.27% vs. 3.19%, SEM 0.05%) cows. CHO led to greater ( $P = 0.02$ ) blood glucose. No MET or CHO effects were detected for blood NEFA ( $P = 0.56$  or greater) or BHBA ( $P = 0.11$  or greater), but a MET × time effect ( $P = 0.10$ ) was observed for NEFA due to lower concentrations on d 4. Results from the present study indicate peripartal supplementation of rumen-protected methionine has positive effects on cow performance.

**Key Words:** methionine, choline, transition cow

**456 Rumen-protected methyl donors during the transition period: 2. Biomarkers of inflammation and oxidative stress reveal better liver and immune function in cows supplemented with rumen-protected methionine (Smartamine M) than choline (ReaShure).** Z. Zhou<sup>\*1</sup>, M. Vailati Riboni<sup>1</sup>, E. Trevisi<sup>2</sup>, F. C. Cardoso<sup>1</sup>, D. N. Luchini<sup>3</sup>, and J. J. Loo<sup>1</sup>, <sup>1</sup>University of Illinois at Urbana-Champaign, Urbana, IL, <sup>2</sup>Università Cattolica del Sacro Cuore, Piacenza, Italy, <sup>3</sup>Adisseo NA, Alpharetta, GA.

The objective of this study was to evaluate the efficacy of supplemental rumen-protected methionine (MET) and choline (CHO) on alleviating inflammation and oxidative stress through their use for synthesis of antioxidants or other methyl-group requiring compounds. Forty-eight multiparous Holstein cows were used in a randomized complete block design with 2 × 2 factorial arrangement of MET (Smartamine M, Adisseo NA) and CHO (ReaShure, Balchem Inc.) level (with or without). Treatments (12 cows each) were control (CON), no MET or CHO; CON+MET (SMA); CON+CHO (REA); and CON+MET+CHO (MIX). From –50 d to –21 d before expected calving, all cows received the same diet (1.24 Mcal/kg DM) with no MET or CHO. From –21 d to calving, cows received the same close-up diet (1.54 Mcal/kg DM) and were assigned randomly to each treatment. From calving to 30 d, cows were on the same postpartal diet (1.69 Mcal/kg DM) and continued to receive the same treatments until 30 d. MET supplementation was adjusted daily at 0.08% DM of diet and CHO was supplemented at 60 g/cow/d. Blood was collected at –10, 4, 8, 20, and 30 d for profiling of 16 biomarkers. Neutrophil and monocyte phagocytosis and oxidative burst were assessed at d 1, 3, 7, 14, and 28 d. MET (SMA, MIX) led to greater ( $P = 0.04$ ; 36.55 vs. 35.53 g/L, SEM 0.35) albumin, lower ( $P = 0.08$ ; 0.35 vs. 0.47 g/L, SEM 0.05) haptoglobin, and greater ( $P = 0.07$ , 93.09 vs. 84.54 U/ml, SEM 3.20) paraoxonase (antioxidant) compared with other treatments. This suggests a more pronounced inflammatory state in cows without supplemental MET. MET-supplemented cows had greater ( $P < 0.01$ ; 60.43 vs. 54.88%, SEM 1.65) neutrophil phagocytosis capacity. Both SMA and REA had greater ( $P < 0.01$ ) monocyte (SMA, 26.93 vs. 16.96%, SEM 2.71; REA, 25.96 vs. 16.96%, SEM 2.52) and

neutrophil (SMA, 59.19 vs. 43.32%, SEM 4.58; REA, 53.89 vs. 43.32%, SEM 4.25) oxidative burst activity compared with CON. Data suggest that supplementing rumen-protected methionine peripartal has positive effects on immune and liver function at least in part by reducing oxidative stress and inflammation.

**Key Words:** methionine, choline, inflammation

**457 Rumen-protected methyl donors during the transition period: 3. Hepatic one-carbon metabolism flux in response to supplemental Smartamine M or ReaShure.** Z. Zhou\*<sup>1</sup>, T. A. Garrow<sup>1</sup>, M. Vailati Riboni<sup>1</sup>, F. C. Cardoso<sup>1</sup>, D. N. Luchini<sup>2</sup>, and J. J. Loor<sup>1</sup>, <sup>1</sup>University of Illinois at Urbana-Champaign, Urbana, IL, <sup>2</sup>Adisseo NA, Alpharetta, GA.

Nutrients such as methionine (MET), choline (CHO), folate, and betaine are vital for provision of methyl groups required by various cellular pathways in tissues such as liver. In the context of dietary methyl-donors, the peripartal dairy cows are characterized by negative MET and CHO balance, both of which can play a role in allowing cows to achieve optimal performance while maintaining good health. Although studies of 1-carbon metabolism and MET cycle in ruminant liver already were performed in sheep, similar data are not available for high-producing Holstein dairy cows. Objectives were to measure activity of enzymes governing S-adenosyl MET formation (methionine adenosyltransferase 1), the transsulfuration pathway (Cystathionine  $\beta$  synthase), and endogenous synthesis of MET via CHO (betaine homocysteine S-methyltransferase, BHMT). Forty multiparous Holstein cows were used in a randomized complete block design with 2  $\times$  2 factorial arrangement of MET and CHO level (with or without). Treatments (10 cows each) were control (CON), no MET or CHO; (SMA), CON+Smartamine M, Adisseo NA; (REA) CON+ReaShure, Balchem Inc.; and (MIX) CON+SMA+REA. From -50 d to -21 d before expected calving, all cows received the same diet (1.24 Mcal/kg DM) with no MET or CHO. From -21 d to calving, cows received the same close-up diet (1.54 Mcal/kg DM) and were assigned randomly to each treatment. From calving to 30 d, cows were on the same postpartal diet (1.69 Mcal/kg DM) and continued to receive the same treatments through 30 d. MET supplementation was adjusted daily at a rate of 0.08% (DM basis) of diet and CHO was supplemented at 60 g/cow/d. Liver samples were harvested at -10, 10, 20 and 30 d relative to calving. Compared with -10 d, liver BHMT activity increased ( $P < 0.01$ ) by 2-fold postpartum regardless of treatment and remained high through at least 30 d. CHO supplementation had no effect ( $P > 0.05$ ). Although no main effect of MET was detected ( $P > 0.05$ ), a significant MET  $\times$  Time interaction ( $P < 0.01$ ) occurred due to higher BHMT activity in MET vs. CON cows on d 20. Data underscore the high demand for endogenous MET synthesis and increased flux through the MET cycle during early lactation.

**Key Words:** methionine, choline, betaine homocysteine S-methyltransferase (BHMT)

**458 Supplemental Smartamine M in high-energy diets during the peripartal period improves production and hepatic biomarkers of oxidative status in Holstein cows.** Mario Vailati Riboni\*<sup>1</sup>, Johan S. Osorio<sup>2</sup>, Erminio Trevisi<sup>3</sup>, James K. Drackley<sup>1</sup>, Daniel Luchini<sup>4</sup>, and Juan J. Loor<sup>1</sup>, <sup>1</sup>University of Illinois at Urbana-Champaign, Urbana, IL, <sup>2</sup>Oregon State University, Corvallis, OR, <sup>3</sup>Università Cattolica del Sacro Cuore, Piacenza, Italy, <sup>4</sup>Adisseo S.A.S., Alpharetta, GA.

Feeding higher-energy prepartum is a common practice in the dairy industry. However, recent data underscore how it could reduce performance, deepen negative energy balance, and augment inflammation and oxidative stress in fresh cows. We tested the effectiveness of rumen-protected methionine in preventing the negative effect of feeding high-energy prepartum. Twenty-one multiparous Holstein cows were fed a controlled-energy diet (CON, n = 7; 1.24 Mcal/kg DM; high-straw) during the whole dry period (~50 d), or switched to a higher-energy (OVE, n = 7; 1.54 Mcal/kg DM), or OVE plus Smartamine M<sup>®</sup> (OVE+SM, n = 7; Adisseo NA) during the last 21 d before calving. Afterward cows received the same lactation diet (1.75 Mcal/kg DM). Smartamine M was top-dressed on the OVE diet (0.07% of DM) from -21 through 30 d in milk (DIM). Liver samples were obtained via percutaneous biopsy at -10, 7 and 21 DIM. Expression of genes associated with energy and lipid metabolism, hepatokines, methionine cycle, antioxidant capacity and inflammation was measured. Although prepartal dry matter intake (DMI) was not affected ( $P = 0.21$ ) by diet, it was lower ( $P < 0.005$ ) in OVE than CON and OVE+SM. Milk yield and ECM were lower ( $P < 0.10$ ) in OVE than CON and OVE+SM. Milk protein and fat percentages were lower ( $P < 0.05$ ) in CON and OVE compared with OVE+SM. Feeding OVE compared with CON led to lower ( $P < 0.05$ ) *PCK1* and *PPARA*. At 7 DIM, *GSR* was greater ( $P < 0.05$ ) in OVE than CON-fed cows, suggesting a greater antioxidant demand. Feeding OVE+SM resulted in similar expression of *GSR* compared with CON. Expression of *SAHH*, *STAT5B* and *MTR* was greater ( $P < 0.05$ ) prepartum in OVE+SM compared with both CON and OVE, and at 7 DIM ( $P < 0.05$ ) for CON and OVE+SM compared with OVE. *FGF21* was lower ( $P < 0.05$ ) prepartum in OVE than CON and OVE+SM, and increased ( $P < 0.05$ ) postpartum only in OVE. Expression of *DMT3A* was greater ( $P < 0.05$ ) in OVE and OVE+SM, while *MTTP* was lower in OVE+SM than CON or OVE. Data suggest, supplemental Smartamine M was able to compensate the negative effect of prepartal energy-overfeeding by increasing production and alleviating the demand for intracellular antioxidants.

**Key Words:** methionine, transition period, nutrigenomics

**459 Effect of strategic ration balancing with use of Prolak, MetaboLys, and Smartamine M on the efficiency of milk protein production and environmental impact in primiparous cows.** Yanting Chen\*<sup>1</sup>, Joe Harrison<sup>1</sup>, Pius Ndegwa<sup>1</sup>, Deb Wilks<sup>2</sup>, Lynn VanWieringen<sup>1</sup>, and John Azzone<sup>3</sup>, <sup>1</sup>Washington State University, Puyallup, WA, <sup>2</sup>EPL Feeds, Lynden, WA, <sup>3</sup>H J Baker, Fayetteville, PA.

The objective of this study was to evaluate the effect of reduced CP on milk production and environmental impact in a commercial dairy herd. Primiparous cows were completely randomized to 2 groups with 84 cows each, and each group had similar DIM (181  $\pm$  7.14 vs. 195  $\pm$  7.85) before initiation of the study. The control diet was the current general herd ration. The reformulated diet was supplemented with Prolak, MetaboLys and Smartamine M, reduced the dietary CP (17.7  $\pm$  0.65 vs. 16.7  $\pm$  0.7% DM), and increased methionine (1.8 vs. 2.2% MP) and lysine (6.6 vs. 6.9% MP) concentrations. Diets were fed in a 6-wk switch back design trial with 2 periods. DMI was recorded daily (1 pen per treatment), and milk yield and composition of individual cows were measured weekly. Urine and feces samples were mixed together and used in a closed chamber incubation to determine NH<sub>3</sub> emission. Feeding the reformulated diet numerically decreased DMI 0.4 kg (19.3  $\pm$  0.49 vs. 18.9  $\pm$  0.58 kg), however milk yield ( $P = 0.91$ ), protein yield ( $P = 0.74$ ), % protein ( $P = 0.49$ ), fat yield ( $P = 0.19$ ), % fat ( $P = 0.09$ ), and lactose yield ( $P = 0.28$ ) did not differ. Lactose % and MUN ( $P < 0.001$ ) decreased when cows were fed the reformulated diet. Cows

fed the reformulated diet had higher Met ( $P = 0.09$ ) and less Tyr ( $P = 0.06$ ) in blood plasma than control cows. Feeding the reformulated diet numerically increased feed efficiency (1.62 vs.  $1.66 \pm 0.01$ ) and milk true protein efficiency (28.4 vs.  $30.8 \pm 0.01\%$ ). Cows fed the reformulated diet consumed 7.2% less N, produced 0.6% more milk total N, excreted 9.9% less predicted urinary N and 18.0% less calculated fecal N than control cows. The difference in income over feed cost for the reformulated diet was \$0.15 and \$0.18 based on the milk price of Washington State in 2013 and 2014, respectively. The emitted  $\text{NH}_3$  flux rates of manure were numerically similar ( $122.6 \pm 10.26$  vs.  $124.1 \pm 11.99 \text{ mg} \cdot \text{h}^{-1} \cdot \text{m}^{-2}$ ) between diets. These results illustrated that feeding low CP with balanced AA diet could improve the efficiency of nitrogen utilization and reduce the environmental impact without compromising the profitability of milk production in primiparous cows.

**Key Words:** bypass lysine, methionine, milk production

**460 Sodium salicylate alters ruminal digestion in vitro and in situ.** Abigail J. Carpenter\*, Claudio F. Vargas-Rodriguez, Jacob A. B. Jantz, and Barry J. Bradford, *Kansas State University, Manhattan, KS.*

Although sodium salicylate (SS) administration after calving increases 305-d milk production, it is associated with hypoglycemia in some circumstances. We hypothesized that this may be in part due to decreased glucogenic substrate supply from fermentation. Six heifers were drenched once daily for 3 d with either 62.5 g of SS in water (SS) or an equal volume of water (CON). A series of batch cultures were performed the day before the start of treatment and 1, 13, and 35 d following. Strained fluid from each heifer was combined in a 2:1 ratio with McDougall's buffer, and 150 mL of the inoculum was added to each flask ( $n = 4/\text{heifer}$ ) with 2.5 g substrate. Gas production was measured with the ANKOM<sup>RF</sup> Gas Production System. Following each rumen fluid collection, Dacron bags containing approximately 1 g of substrate DM were inserted into the rumen of each heifer in duplicate at 2, 8, 16, 24, and 48 h time points to estimate rate of DMD. The 48-h time point was used to estimate rumen-undigested substrate. Measurements from the pre-treatment batch culture and in situ experiments were used as covariates for statistical analysis but were removed from the model if they were not significant ( $P > 0.05$ ). Overall, there was no effect of treatment on batch culture final pH across time ( $P = 0.70$ ), although dry matter disappearance (DMD) was decreased across time due to treatment ( $P < 0.01$ ; treatment  $\times$  day:  $P = 0.01$ ). One day following treatment, SS had no effect on DMD ( $P = 0.70$ ); however, DMD was decreased ( $P < 0.01$ ) in batch culture 13 d after SS treatment ( $45.0$  vs.  $38.9 \pm 0.9\%$  of DM for CON vs. SS, respectively), and it remained lesser for the SS treatment 35 d after the end of treatment ( $P < 0.01$ ,  $44.0\%$  vs.  $40.3 \pm 0.9\%$  of DM for CON vs. SS, respectively). No differences were observed due to treatment for volume, rate, or lag in gas production ( $P \geq 0.60$ ). For the in situ experiment, no differences were detected in any 48-h time points ( $P \geq 0.17$ ). Treatment with SS tended to decrease DMD rate ( $P = 0.10$ ; treatment  $\times$  day:  $P = 0.50$ ). Given the sustained effect of SS drenches on in vitro DMD, it is likely that SS modifies the rumen microbiota to impair ruminal digestion.

**Key Words:** salicylate, fermentation, rumen modification

**461 Effect of rumen acidosis and short-term feed restriction on short-chain fatty acid concentrations and permeability of the bovine gastrointestinal tract.** Rae-Leigh A. Pederzoli\*<sup>1</sup>, Steve Hendrick<sup>2</sup>, John Campbell<sup>1</sup>, Katie M. Wood<sup>1</sup>, and Gregory B. Penner<sup>1</sup>,

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The objective of this study was to identify whether ruminal acidosis (RA) or feed restriction (FR) differentially affect permeability of the gastrointestinal tract (GIT). Twenty-one Holstein steers were randomly assigned to 1 of 3 treatments: control (CON); ruminal acidosis (ACID), and feed restriction (REST). Steers were fed a common diet with a 50:50 F:C ratio once daily at 0800 h for a 5-d baseline period followed by a challenge period. Rumen acidosis was induced by restricting feed to 25% DMI for 1 d and then offering pelleted barley (30% DMI:BW) the next day. Steers on the REST treatment were restricted to 25% DMI for 5 d. Steers were killed and tissues were collected from the rumen, omasum, duodenum, jejunum, ileum, cecum, and proximal and distal colon for measurement of <sup>14</sup>C-mannitol and <sup>3</sup>H-inulin flux in Ussing chambers as markers for gut permeability. Data were analyzed as a randomized complete block design using Proc Mixed. Rumen pH was recorded throughout the study. During baseline, there were no differences for DMI or rumen pH (7.1 kg/d and 6.53, respectively;  $P > 0.1$ ), but DMI and pH during the challenge were 1.7 kg/d and 6.94, and 7.9 kg/d and 5.86 for REST and ACID calves, respectively. The proportion of acetate in the proximal colon from CON (56.8%) was greater (treatment  $\times$  region,  $P = 0.01$ ) than for FR (31.0%) and ACID (31.0%). Butyrate concentration was less in the proximal colon of CON (16.0%; interaction  $P < 0.01$ ) than ACID (31.0%). There were no region  $\times$  treatment interactions or treatment effects ( $P > 0.10$ ) for inulin or mannitol flux. However, inulin flux ( $\text{nmol}/(\text{cm}^2 \times \text{h})$ ) was greater ( $P < 0.01$ ) in rumen (21.1), omasum (26.9), and duodenum (21.3) than ileum (4.2), cecum (7.5), and distal colon (2.4). Mannitol flux ( $\text{nmol}/(\text{cm}^2 \times \text{h})$ ) was greatest ( $P < 0.01$ ) in rumen (36.9), duodenum (59.5), jejunum (48.2), and proximal (41.4) and distal colon (33.0) relative to ileum (13.0). These data indicate that feed restriction and ruminal acidosis do not appear to differentially affect permeability of the GIT. The duodenum and rumen are likely regions with greatest permeability.

**Key Words:** acidosis, feed restriction, permeability

**462 Effect of gluconeogenic precursors on blood metabolites and milk yield in Chilean transition Holstein cattle.** Pedro Melendez\*<sup>1</sup>, Katherine Severino<sup>2</sup>, Maria P. Marin<sup>2</sup>, Patrick Pithua<sup>1</sup>, and Pablo Pinedo<sup>4,5</sup>, <sup>1</sup>*Department of Veterinary Medicine and Surgery, College of Veterinary Medicine, University of Missouri-Columbia, Columbia, MO,* <sup>2</sup>*College of Veterinary Medicine, University Santo Tomas, Viña del Mar, Chile,* <sup>3</sup>*Department of Animal Sciences, College of Veterinary Medicine, University of Chile, Santiago, Chile,* <sup>4</sup>*Texas A&M AgriLife Research, Amarillo, TX,* <sup>5</sup>*Department of Veterinary Pathobiology, College of Veterinary Medicine & Biomedical Sciences, Texas A&M University System, College Station, TX.*

The objective of this study was to determine the effect of feeding a gluconeogenic precursor containing calcium propionate, propylene glycol, and glycerol on serum concentrations of  $\beta$ -hydroxybutyrate (BHBA), nonesterified fatty acids (NEFA), and milk yield in transition Holstein cows under Chilean conditions. The study was conducted in a commercial dairy farm located in central Chile. Cows were housed in a freestall system with headlocks, milked 3 times per day and fed a total mixed ration containing alfalfa hay, corn silage, and concentrate to meet or exceed the requirements proposed by the National Research Council (2001). At 21 d before expected parturition 40 multiparous cows were assigned at random to either a control or a treated group to achieve at least 15 cows per group at 30 DIM. The cows in the control group received the default periparturient TMR diet. Cows in the treatment group received 300 g of a supplement containing 70 g of calcium



propionate, 95 g of propylene glycol and 330 g of glycerol top dressed on the TMR during morning feedings when cows were head-locked. This feeding protocol was continued into the postpartum period until 30 d in milk. Seventeen and 18 cows completed the protocol in the control and the treatment group, respectively. Blood samples were collected at calving for NEFA quantitation and later at 7, 14 and 21 d postpartum for BHBA determination. The average daily milk yield up to 60-d of lactation was significantly higher (2 kg per day) in the treated than the control group ( $P < 0.05$ ). NEFA (mEq/L) at calving were higher in the control ( $0.75 \pm 0.1$ ) than the treated group ( $0.55 \pm 0.1$ ;  $P < 0.05$ ). The concentration of serum BHBA at 14 d postpartum was lower in control ( $0.60 \pm 0.11$  mmol/L) than treated cows ( $0.98 \pm 0.11$  mmol/L;  $P < 0.05$ ). In conclusion, supplementing the default diet with gluconeogenic precursors during the pre- and postpartum period improved milk yield during the first 60 d of lactation and maintained a moderated metabolic energy status in Chilean dairy cattle.

**Key Words:** transition cow, metabolite, dairy cattle

**463 Expression of G-protein coupled fatty acid receptors during the transition period.** Alea Agrawal\*, Johan S. Osorio, and Juan J. Loor, *University of Illinois at Urbana-Champaign, Urbana, IL.*

G-protein coupled receptors (GPR) have been widely studied within human medicine as drug targets for metabolic disorders. They play central roles in many cell signaling processes, and also have application within dairy nutrition as targets for specific dietary components; for example, omega-3 fatty acids. To combat metabolic disorders prevalent in dairy cows during the transition period, which often co-occur with negative energy balance and lipid mobilization, it may be helpful to identify locations, activities, and roles of free fatty acid receptors (FFAR) and other members of the GPR family. To provide insight on tissue-specific differences in gene expression, and therefore, potential downstream pathways of fatty acid-sensing GPR in bovine, quantitative RT-PCR (qPCR) of subcutaneous adipose, liver, and polymorphonuclear leukocyte (PMN) samples during the transition period (-15, +10, and +30 d) were used for expression profiling of medium- (MCFA) and long-chain fatty acid (LCFA) receptors *GPR120* and *GPR40*, MCFA receptor *GPR84*, and niacin receptor *GPR109A*. Results were analyzed in SAS to examine differences in expression over time ( $P < 0.05$ ). In adipose tissue, *GPR120* expression was highest at -15 d, decreased at +10 d, and further decreased at +30 d. *GPR40* expression was highest at +10 d relative to other time points, and *GPR109A* expression was lower prepartum than both postpartum time points. *GPR84* was undetectable. In liver tissue, *GPR84* and *GPR109A* were nearly undetectable and did not differ ( $P > 0.05$ ) across time. *GPR40* had lower expression at +30 d than -15 or +10 d. *GPR120* was undetectable. In PMN, *GPR120* expression was increased between +10 and +30 d but neither were significantly different from -15 d. *GPR84* expression was higher at +10 d relative to other time points. Neither *GPR40* nor *GPR109A* were altered in PMN. The data suggest that there is likely not a direct role for the selected GPR in the liver during the transition period, but they do play variable roles in adipose and PMN. Dietary supplementation or exclusion of LCFA and/or niacin may provide a method of regulating GPR function during the prepartal or early postpartal periods.

**Key Words:** transition cow, fatty acid, G-protein coupled receptor (GPR)

**464 Effects of prepartal dietary intake and calving on blood neutrophil transcriptome in Holstein cows.** Alea Agrawal\*, Muhammad J. Khan, Daniel E. Graugnard, Sandra L. Rodriguez-Zas, and Juan J. Loor, *University of Illinois at Urbana-Champaign, Urbana, IL.*

In the dairy industry, cow health and farmer profits depend on the balance between diet (i.e., nutrient composition, daily intake) and metabolism. This is especially true during the transition period, where dramatic physiological changes foster vulnerability to immunosuppression, negative energy balance, and clinical and subclinical disorders. Using an Agilent microarray platform, the present study examined changes in the transcriptome of bovine PMNL, a representative cell of the immune system, due to time relative to parturition, prepartal dietary intake, or the combination. Sixteen Holstein cows were fed a high-straw, control diet (S; NEL = 1.34 Mcal/kg) or overfed a moderate-energy diet (M; NEL = 1.62 Mcal/kg) during the dry period. Blood for PMNL isolation and metabolite analysis was collected at -14 and +7 d relative to parturition. At an ANOVA false discovery rate (FDR)  $< 0.05$ , time (7 vs. -14 d) significantly influenced expression of 1758 genes, energy intake (M vs. S) influenced 3062 genes, and the interaction had an effect on 1673 genes. Dynamic Impact Approach (DIA) bioinformatics analysis classified effects on KEGG pathways, including: activated carbohydrate metabolism due to time and interactions, and activated amino acid (AA) biosynthesis and ribosome activity with dietary treatment. In contrast, DIA analysis revealed inhibition of riboflavin and fatty acid (FA) metabolism, unsaturated FA synthesis, and calcium reabsorption due to energy intake. These analyses suggest that processes critical for energy metabolism and immune function (e.g., calcium reabsorption, FA and vitamin metabolism, and AA synthesis) were affected by energy overfeeding with mixed results, but overall, strong effects from either main effect were mitigated by the interactions. Ingenuity Pathway Analysis (IPA) of genes significantly affected at an FDR  $< 0.10$  also revealed 50 upstream regulators for each main effect and interaction comparison. The widespread, transcriptome-level changes captured here confirm the importance of dietary energy adjustments around calving on the immune system.

**Key Words:** transition cow, PMNL, intake

**465 A cow mammary epithelial cell-free system based on crude lysosomes and cytosol proteins: Leucine activating mTOR at Ser2448.** Wen-ting Dai<sup>1,2</sup>, Nan Zheng<sup>1,3</sup>, and Jia-qi Wang<sup>\*1,3</sup>, <sup>1</sup>Ministry of Agriculture Laboratory of Quality & Safety Risk Assessment for Dairy Products, Beijing, China, <sup>2</sup>Jilin University, Changchun, China, <sup>3</sup>Ministry of Agriculture-Milk and Dairy Product Inspection Center, Beijing, China.

Essential amino acids, especially leucine, initiated the signaling of the mammalian target of rapamycin complex-1 (mTORC1) and protein synthesis in cow mammary epithelial cells (CMEC). CMEC-H is an immortalized cow mammary epithelial cell line. We established a cell-free system, which was based on crude lysosomes and cytosol proteins from CMEC-H. By trypan blue dye exclusion assay, we found passing through gauge-26 needle was a good cell lysis method of CMEC-H. Crude lysosomes were extracted by lysosome isolation kit and cytosol proteins (supernatant-100) were obtained by ultracentrifugation at  $1 \times 10^5$  g about 1 h. Supernatants 100, which contained all the cytosolic soluble proteins, were identified by SDS-PAGE. Raptor, mTOR and mLST8, the essential components of mTORC1, played a vital role in regulating protein synthesis; Lamp2 represented lysosome. All these proteins were testified by western blots. The constructed cell-free model stimulated by 1× essential amino acids was regarded as positive control,

the cell-free model stimulated by no amino acids was regarded as negative control, and the cell-free model stimulated by 1× leucine was seen as the treatment group. The cell-free models stimulated by 1× leucine readded 20 nmol rapamycin and stimulated by 1× essential amino acids readded 20 nmol rapamycin were both regarded as the corresponding inhibition groups. The results showed that the model can partially duplicate aspects of amino acids signaling mTOR program in vitro; the addition of 1× leucine solely and 1× essential amino acids led mTOR to

be activated, phosphorylated and then moved towards the crude lysosome fractions in the cell-free system. However, combined with 1× leucine or 1× essential amino acids, 20 nmol rapamycin was not able to completely prevent mTOR from being phosphorylated. This study may provide some directions for further construction of cell-free system duplicating amino acid signaling mTOR. Furthermore, the method may be applied to cell-free models of other cell lines.

**Key Words:** cell-free system, crude lysosomes, leucine

## Ruminant Nutrition: Dairy

**466 Effects of including supplemental fat in low and high starch diets on milk production and energy partitioning.** Joshua L. Garver\*, Jonas De Souza, Michael J. VandeHaar, and Adam L. Lock, *Michigan State University, East Lansing, MI.*

Effects of low or high starch diets with or without supplemental fat on the yield of milk and milk components and energy partitioning were evaluated. Thirty-two Holstein cows ( $172 \pm 35$  DIM) were assigned randomly to treatment sequence in replicated  $4 \times 4$  Latin squares with a  $2 \times 2$  factorial arrangement of treatments. Treatment diets contained 16% (LS) or 32% (HS) starch and 0.0% (LF) or 1.5% (HF) palmitic acid-enriched fat supplement (BergaFat F-100). Dietary starch was altered by varying the proportion of ground corn, cottonseed, and soy-hulls, with LS and HS diets containing 17 and 21% forage NDF and 42 and 29% NDF, respectively. Treatment periods were 21 d in length with the final 5 d used for data and sample collection. The statistical model included the random effects of cow and period and the fixed effects of dietary starch level, fat supplementation, and their interaction. Compared with LF, the HF treatment increased DMI in the LS diet (26.8 vs. 25.8 kg/d) and decreased DMI in the HS diet (25.9 vs. 26.5 kg/d; interaction  $P < 0.01$ ). The HF treatment increased milk fat yield in the LS diet (1.57 vs. 1.51 kg/d) but not in the HS diet (1.49 vs. 1.51 kg/d; interaction  $P = 0.06$ ). Compared with LF, the HF treatment also increased milk protein concentration and yield in the LS diet (3.40 vs. 3.35% and 1.31 vs. 1.26 kg/d), but not in the HS diet (interaction  $P = 0.10$  and  $P < 0.05$ , respectively). There was no effect of treatments on milk fat and lactose concentrations or lactose yield. Compared with LF, the HF treatment increased 3.5% FCM (42.2 vs. 41.0 kg/d) in the LS diet but not in the HS diet (40.4 vs. 41.1 kg/d; interaction  $P = 0.07$ ). Similarly, the HF treatment increased ECM (42.3 vs. 41.0 kg/d) in the LS diet but not in the HS diet (40.5 vs. 41.2 kg/d; interaction  $P < 0.05$ ). The milk to feed ratio (ECM/DMI) was not affected by treatments. There was a trend for HS diets to increase change in BCS compared with LS diets ( $P = 0.08$ ). However, fat supplementation did not affect change in BCS. Results demonstrate that under the dietary conditions tested, a palmitic acid-enriched fat supplement fed to mid and late lactation dairy cows maximized yield of milk and milk components when fed in a low starch diet.

**Key Words:** body condition, milk fat, palmitic acid

**467 Effect of breed, energy level of diet, and lactation stage on the evolution of milk lipolysis in dairy cow.** Elise Vanbergue\*<sup>1,2</sup>, Luc Delaby<sup>1</sup>, Ségolène Colette<sup>3</sup>, Yves Gallard<sup>3</sup>, and Catherine Hurtaud<sup>1</sup>, <sup>1</sup>*INRA-Agrocampus Ouest UMR1348 Pegase, Saint-Gilles, France*, <sup>2</sup>*Institut de l'Elevage, Le Rheu, France*, <sup>3</sup>*INRA, Domaine du Pin-au-Haras, Exmes, France.*

Spontaneous lipolysis is the result of the activity of a native milk enzyme, the lipoprotein lipase. Lipolysis leads to a release of free fatty acids (FFA) that cause rancid flavor in milk products. A trial was carried out to study the effects of breed, energy levels, and lactation stage on milk spontaneous lipolysis in dairy cows. Sixty-four cows were used through a year of lactation. Cows were divided into 4 homogenous groups according to their breed [Holstein (HO) or Normande (NO)] and to their feeding system: the intensive system, with a high energy diet (HED) (in winter, corn silage with 30% concentrate; otherwise, pasture with 4 kg/d of concentrate) and the grass system (GS) (in winter, conserved grass with no concentrate; otherwise, pasture with no concentrate). The period of

calving was synchronized between January and March. Individual milk samples were collected every month from both morning and evening milkings. The FFA levels were determined by the difference between the FFA levels after milking and the FFA after 24 h of storage at 4°C. Data were analyzed using SAS mixed procedure. We showed that FFA levels were higher in the evening milks (0.45 vs. 0.25 mEq/100 g of fat,  $P < 0.001$ ) and that evening milks were more susceptible to lipolysis variations. HO cows were more susceptible to lipolysis than NO (0.34 vs. 0.20 mEq/100 g of fat, in morning milks; 0.62 vs. 0.33 mEq/100 g of fat in evening milks,  $P < 0.001$ ). There was no effect of the feeding system on morning milks lipolysis but GS had a tendency to increase milk lipolysis in evening milks (0.41 vs. 0.55 mEq/100 g of fat,  $P = 0.06$ ). Lipolysis was higher in early and late lactation stage, particularly in the GS group. Conserved grass may also affect the lipolysis rate. The energy status in early and late lactation stage is frequently negative so the cows may be more susceptible to a low energy diet. No correlation was found between lipolysis and milk fat content. Poor correlations ( $r < 0.4$ ,  $P < 0.001$ ) were found between lipolysis and milk production, fat globule size, proportion of fatty acid and protein composition, body condition and weight.

**Key Words:** spontaneous milk lipolysis, production factors

**468 Direct and indirect transfer of omega-3 fatty acids to milk fat in dairy cows.** Natalie L. Urrutia\*, Jackie Y. Ying, Samantha R. McKinney, Michael H. Green, and Kevin J. Harvatine, *The Pennsylvania State University, University Park, PA.*

Transfer of dietary fatty acids (FA) to milk has been proposed to fit a 2-pool model with a fast pool representing direct transfer by chylomicrons and a slow pool representing indirect transfer through tissue recycling. The objective of this experiment was to quantify direct and indirect transfer of omega-3 (n-3) FA to milk after an abomasal bolus infusion of n-3 FA. Ten ruminally cannulated multiparous Holstein cows ( $247 \pm 113$  DIM; mean  $\pm$  SD) were used in a crossover design with 7 d periods. Cows were milked 4 times daily (6 h intervals) starting 2 d before initiation of the experiment. Treatments were abomasal infusion of 120 g (infused over 1 h) of a free FA mixture enriched in  $\alpha$ -linolenic acid (18:3 n-3; EALA) or in very long chain n-3 ( $>18C$  n-3; EVLC). The EALA and EVLC treatments provided 80.2 g and 87.6 g of n-3 FA, respectively. Milk was sampled at each milking for determination of milk fat yield and FA profile. The day before bolus infusion was used as a baseline. Total transfer of n-3 FA was analyzed in a model that included random effects of cow nested in sequence, sequence and period and fixed effect of treatment and milk yield (JMP Pro). Time course data was analyzed as repeated measures in SAS and resulting least squares means were fit to a double exponential decay function by nonlinear curve fitting (JMP Pro). Total transfer of n-3 FA to milk differed between treatments ( $P < 0.001$ ) and was 48.2 and 32.7% of the bolus for EALA and EVLC, respectively. Milk n-3 FA concentration and yield peaked at 12 h and returned to baseline at 138 h post infusion in both treatments. Time course of n-3 FA transferred to milk fit a biexponential model ( $R^2 = 0.99$ ). The area (% of total) under the first exponential representing direct transfer was 83.9 and 42.2% and the second exponential representing indirect transfer was 16.1 and 57.8% of the total n-3 FA transferred for EALA and EVLC, respectively. In conclusion, n-3 FA differed greatly in their transfer efficiency mainly due to differences in their direct transfer rates. These differences pre-



sumably occur due to trafficking of very long chain n-3 FA into plasma lipid pools unavailable to the mammary gland.

**Key Words:** omega-3, milk fat

**469 2-Hydroxy-4-(methylthio)butanoate (HMTBa) supplementation increases milk fat and decreases synthesis of alternate biohydrogenation intermediates in diets with risk for milk fat depression.** Michel Baldin\*<sup>1</sup>, Yun Ying<sup>1</sup>, Geoff I. Zanton<sup>2</sup>, Heather A. Tucker<sup>2</sup>, Mercedes Vazquez-Anon<sup>2</sup>, and Kevin J. Harvatine<sup>1</sup>, <sup>1</sup>Penn State University, University Park, PA, <sup>2</sup>Novus International Inc., St. Charles, MO.

We recently reported that supplementation of 2-hydroxy-4-(methylthio)butanoate (HMTBa) reduced the shift to the alternate biohydrogenation pathway and maintained higher milk fat yield in high producing cows fed diets lower in fiber and higher in unsaturated fatty acids (FA). The objective of this experiment was to verify the effect of HMTBa (Alimet, Novus International, Inc., St. Charles, MO) on biohydrogenation intermediates and milk fat synthesis. Twenty-two rumen cannulated high-producing Holstein cows [168 ± 66 DIM; 42 ± 7 kg milk/d (Mean ± SD)] were used in a randomized design performed in 2 blocks (1 = 14 cows, 2 = 8 cows). Treatments were control (corn carrier) and HMTBa (0.1% of diet DM). The experiment included a 7-d covariate period followed by 3 phases that fed diets with increasing risk of milk fat depression (MFD). The diet during the covariate and low-risk phase (7 d) was 32% NDF with no additional oil. The diet during the moderate-risk phase (17 d) was 29% NDF with 0.75% soybean oil. Soybean oil was increased to 1.5% for the last 4 d. Milk yield and DMI were measured daily. Milk was sampled every 7 d and analyzed for fat, protein and FA profile. Data were analyzed using PROC Mixed with repeated measures and the effect of treatment was tested at each time point. There was no effect of block or interaction of block and other fixed effects. There was no overall effect of treatment or treatment by time interaction for DMI ( $P = 0.4$ ), milk yield ( $P = 0.4$ ), and milk protein concentration ( $P = 0.9$ ) and yield ( $P = 0.6$ ). There was an effect of treatment, but no treatment by dietary phase interaction on milk fat with HMTBa increasing milk fat percent (3.2 vs. 3.6%,  $P < 0.01$ ) and yield (1342 vs. 1543 g/d,  $P = 0.02$ ). Additionally, HMTBa decreased the concentration (1.29 vs. 0.81 g/100 g of total FA,  $P = 0.02$ ) and yield (14.7 vs. 10.9 g/d,  $P = 0.01$ ) of *trans*-10 18:1 in milk across the entire feeding period. In conclusion, HMTBa prevented the increase in *trans* FA associated with MFD and maintained milk fat yield when cows were fed a diet with moderate risk of diet-induced MFD.

**Key Words:** 2-hydroxy-4-(methylthio)butanoate (HMTBa), milk fat

**470 Meta-analysis of the effect of plant oils rich in 18:2n-6 on milk fatty acid composition in lactating dairy cows.** Mina Vazirigohar\*, Mehdi Dehghan-Banadaky, Kamran Rezayazdi, and Ardeshir Nejati-Javaremi, *Department of Animal Science, Campus of Agriculture and Natural Resources, University of Tehran, Karaj, Alborz, Iran.*

The objective of this study was to evaluate the effect of dietary plant oils rich in 18:2n-6 (POLA; including safflower, sunflower, corn, soybean and cottonseed oils) on milk fatty acid (FA) profile in lactating dairy cows, using meta-analysis and meta-regression methods. A total of 20 studies were identified with 27 comparisons between treatment and control groups, which met the selection criteria and included in the analysis. Two levels of POLA supplement (less and more than 30 g/kg diet DM) were evaluated in this meta-analysis. Supplementation of cows with

20 g POLA/kg diet DM reduced milk fat 6:0–16:0, enhanced 18:0, *cis* (Δ9–12, and 15) and *trans* 18:1 (Δ6–14), 18:2n-6, *cis*-9,*trans*-11 CLA and *trans*-10,*cis*-12 CLA concentrations. The weighted mean differences for milk fat content of 16:0, *cis*-9 18:1, 18:2n-6 and *cis*-9,*trans*-11 CLA were -5.687 (95% confidence interval (CI) = -7.558 to -3.816), 3.194 (95% CI = 1.139 to 5.249), 0.810 (95% CI = 0.384 to 1.236) and 0.467 g/100 g total FA (95% CI = 0.281 to 0.653), respectively. Inclusion of 20 g/kg DM POLA had no effect on milk 4:0 and 18:3n-3 concentrations. Results of meta-regression showed that dietary 18:2n-6 content explained more than 55% of the variation in 8:0–15:0 results. Dietary FA content, amount of grain in the basal diets and parity were the sources of heterogeneity for milk *cis*-9 18:1. Milk *trans*-11 18:1 increased with increased dietary 18:2n-6 content and forage level of basal diets. Estimated effect size of milk 18:2n-6, *trans*-10,*cis*-12 CLA and 18:0 were greater in safflower > sunflower > corn > soybean and > cottonseed oils. In conclusion, feeding 20 g POLA/kg diet DM lowered milk fat 16:0, increased *cis*-9 18:1, 18:2n-6, *cis*-9,*trans*-18 CLA, without any changes on 4:0 and 18:3n-3.

**Key Words:** meta-analysis, milk fatty acid, plant oil

**471 Prediction of blood nonesterified fatty acid (NEFA) by FTIR analysis of individual cow milk samples.** David M. Barbano\*<sup>1</sup>, Patrick Cree<sup>3</sup>, Tom R. Overton<sup>1</sup>, Heather M. Dann<sup>2</sup>, and Rick J. Grant<sup>2</sup>, <sup>1</sup>Cornell University, Ithaca, NY, <sup>2</sup>William H. Miner Agricultural Institute, Chazy, NY, <sup>3</sup>Delta Instruments, Drachten, the Netherlands.

Our objective was to develop and validate a Fourier transform mid-IR-based milk analysis method to estimate blood NEFA concentrations in lactating dairy cows. High blood NEFA indicates that a cow is mobilizing body fat and increases the risk of metabolic disorders. Milk and blood samples were collected from 60 lactating Holsteins once per week for the first 3 weeks of lactation. Cows were milked 3 times per day. Within + or - one milking of the time of blood collection, a milk sample was analyzed using a Delta Instruments (model FTA) mid-IR milk analyzer. A Wako NEFA HR test kit was used as an in vitro enzymatic colorimetric method for the quantitation of NEFA in blood serum and these values were used as reference values for development of a partial least squares (PLS) regression model to predict blood NEFA from the mid-IR milk spectra. There are no NEFA in milk, so a model to predict blood NEFA from a milk sample uses differences in the milk spectra from sample to sample that are correlated with changes in blood NEFA. The final PLS model had 9 factors, used wavelengths in the following ranges (3000 to 2800, 1800 to 1700, 1585 to 1000  $\text{cm}^{-1}$ ) with a standard error of cross validation of 172  $\mu\text{Eq/L}$ . Validation milk and blood sample pairs ( $n = 53$ ) were collected from Holstein cows from a different herd. The mean value for the blood reference test was 713  $\mu\text{Eq/L}$  of serum and the mean value for the milk based blood NEFA prediction was 703  $\mu\text{Eq/L}$  of serum with a standard deviation of the difference (SDD) of 218  $\mu\text{Eq/L}$  for the 53 validation samples. Blood NEFA measured on blood is a snapshot of the NEFA concentration at an instant in time, while blood NEFA predicted from milk analysis represents a time average for the total time between milkings. The FTIR milk analysis to estimate blood NEFA is rapid (about 10 s), done simultaneously with all other milk component measures, and uses no reagents. This approach could be useful for rapid evaluation of risks of ketosis and reproductive disorders.

**Key Words:** blood NEFA, FTIR, milk analysis

#### **472 Increased NEFA availability promotes plasma ceramide accumulation in Holstein cows.**

J. Eduardo Rico<sup>1</sup>, Luciano S. Caixeta<sup>2</sup>, Yves R. Boisclair<sup>2</sup>, and Joseph W. McFadden<sup>\*1</sup>, <sup>1</sup>West Virginia University, Morgantown, WV, <sup>2</sup>Cornell University, Ithaca, NY.

An increase in plasma NEFA can impair insulin sensitivity in dairy cows by unknown mechanisms. In monogastrics, excess saturated fatty acids can upregulate the hepatic synthesis and secretion of ceramide, a sphingolipid that inhibits insulin action in muscle and adipose tissue. Our objective was to determine whether an increase in plasma NEFA can augment ceramide levels in cows. Six nonpregnant, nonlactating Holstein dairy cows (682 kg ± 22), were used in a crossover design with treatments consisting of i.v. infusion (100 mL/h) of either saline (control) or triacylglycerol (TG) emulsion (Intralipid 20%; Fransenius Kabi) for 16 consecutive hours. The feeding level was set at 120% of estimated energy requirement with meals offered every 2 h. Blood was collected at regular intervals and liver was biopsied at 16 h. LC/MS was used to profile 25 ceramides (Cer), monohexosylceramides (GlcCer), and lactosylceramides (LacCer) in plasma. Data were analyzed using a mixed model with repeated measures (fixed effects of treatment and time). Nonparametric correlations were analyzed. TG infusion increased plasma NEFA by 454% at 3 h relative to control ( $P < 0.01$ ) with no further increase at 16 h. Liver TG were elevated 321% in TG vs control by end of TG infusion ( $P < 0.01$ ). Before infusion, C24:0-Cer, C24:0-GlcCer, and C16:0-LacCer represented 52, 47, and 83%, respectively, of plasma Cer, GlcCer and LacCer. Plasma total Cer increased 149 and 235% by 3 and 16 h of TG infusion, relative to control ( $P < 0.01$ ). Plasma C20:0-, C22:0-, C24:0-, and C26:0-Cer levels increased during TG infusion ( $P < 0.01$ ). Although TG infusion did not modify plasma C16:0-Cer level at 16 h, C16:0-Cer abundance (% of total) was lower at 16 h ( $P < 0.01$ ). TG infusion increased plasma C16:0-, C20:0-, C22:0-, and C24:0-GlcCer levels ( $P < 0.01$ ). Plasma LacCer levels were not modified by TG infusion. Cer levels were positively correlated with NEFA (e.g., C24:0-Cer,  $r = 0.80$ ;  $P < 0.01$ ). Our data indicate that plasma Cer accumulation occurs in dairy cows concomitantly with increases in plasma NEFA and liver TG. Research is needed to determine whether NEFA-induced insulin resistance is mediated by Cer.

**Key Words:** ceramide, dairy cow, insulin resistance

#### **473 An acute increase in circulating NEFA does not lower total plasma sphingomyelin levels in Holstein cows.**

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Insulin resistance in rodents is characterized by inflammation-mediated activation of sphingomyelinase and the transformation of sphingomyelin (SM) into ceramide (Cer), an effect provoked by saturated fatty acids. Our objective was to determine whether an acute increase in circulating NEFA lowers plasma SM levels in cows. Six nonpregnant, nonlactating Holstein dairy cows (682 kg ± 22), were used in a crossover design with treatments consisting of i.v. infusion (100 mL/h) of either saline (control) or triacylglycerol (TG) emulsion (Intralipid 20%; Fransenius Kabi) for 16 consecutive hours. The feeding level was set at 120% of estimated energy requirement with meals offered every 2 h. Blood was collected at regular intervals. LC/MS was used to profile SM in plasma. Data were analyzed using a mixed model with repeated measures (fixed effects of treatment and time). Nonparametric correlations were analyzed. TG infusion increased plasma NEFA by 454% at 3 h relative to control ( $P < 0.01$ ) with no further increase at 16 h. Total plasma Cer concentrations increased in TG-infused cows, relative to control ( $P < 0.01$ ). Before infusion, C16:0- and C18:1-SM represented 30 and 21%,

respectively, of plasma SM. Plasma C16:0-SM levels increased 10% by 16 h of TG infusion, relative to control ( $P < 0.05$ ). Plasma C20:1-SM concentrations were lower in TG-infused cows, relative to control ( $P < 0.05$ ). Plasma C16:1-, C18:0-, C18:1-, C22:0-, C24:0-, and C24:1-SM, as well as total plasma SM were not modified with TG infusion. Before infusion, C20:0-dihydro-SM (DHSM) represented 89% of total plasma DHSM. Plasma C20:0- and C18:0-DHSM increased 79 and 14%, respectively, by 16 h of TG infusion, relative to control ( $P < 0.01$ ). Plasma C18:1- and C20:1-SM were negatively correlated with NEFA (e.g., C20:1-DHSM,  $r = -0.47$ ;  $P < 0.01$ ). In contrast, plasma C18:0- and C20:0-DHSM, as well as total DHSM were positively correlated with NEFA (e.g., C20:0-DHSM,  $r = 0.77$ ;  $P < 0.01$ ). An acute increase in plasma NEFA does not appear to promote sphingomyelin hydrolysis in dairy cows thus suggesting that plasma Cer accumulations occurred due to enhanced de novo Cer synthesis.

**Key Words:** dairy cow, insulin resistance, sphingomyelin

#### **474 Temporal changes in plasma sphingolipids during the transition from pregnancy to lactation in Holstein cows.**

J. Eduardo Rico<sup>\*</sup>, Sina Saed Samii, Alice T. Mathews, and Joseph W. McFadden, West Virginia University, Morgantown, WV.

Ceramide (Cer) and sphingomyelin (SM) are sphingolipids associated with conserved cellular processes; however, their involvement in the homeorhetic adaptation to lactation in cows is unknown. Our objectives were to characterize temporal responses in plasma sphingolipids during the periparturient period and determine whether these changes were related to adiposity and lipolysis. Multiparous Holstein cows were grouped by BCS at d -28 prepartum: lean (BCS  $2.9 \pm 0.13$ ;  $n = 7$ ) or overweight (OVER; BCS  $4.0 \pm 0.21$ ;  $n = 7$ ), fed a balanced diet. Blood samples were collected routinely from d -21 to 21. LC/MS was used to profile 37 Cer, monohexosylceramides (GlcCer), lactosylceramides (LacCer), and SM in plasma. Data were analyzed as repeated measures under a mixed model (fixed effects of BCS and day). Nonparametric correlations were analyzed. Plasma NEFA increased around calving and was higher in OVER from d -5 to 21 ( $P < 0.05$ ). OVER lost more BCS and BW ( $P < 0.01$ ). C16:0-Cer concentration decreased by 40% during transition and was higher in OVER prepartum ( $P < 0.05$ ). C24:0-Cer increased 50% and was 33% higher in OVER at d 21 ( $P < 0.05$ ). C22:0-Cer increased 49 and 66% at d 14 and 21, respectively, in OVER ( $P < 0.05$ ). C24:0-GlcCer increased 52% during transition ( $P < 0.05$ ) and tended to be higher in OVER at d 0 ( $P < 0.1$ ). C16:0-GlcCer increased by 145% at d 4 ( $P < 0.01$ ). C22:0-GlcCer increased over time and was higher in OVER at d 14 ( $P = 0.05$ ). C24:1-GlcCer tended to increase by 100% in OVER during transition ( $P < 0.1$ ). C16:0-LacCer increased postpartum and peaked at d 14 ( $P < 0.01$ ). C18:1- and C24:1-LacCer levels increased during transition and were higher in OVER from d 4 to 21 ( $P < 0.01$ ). Abundant C16:0-, C18:1- and C20:1-SM decreased over time and reached nadir around parturition ( $P < 0.01$ ). C18:1- and C20:1-SM were higher in OVER pre- and postpartum ( $P < 0.05$ ). C20:0-dihydro-SM decreased around calving ( $P < 0.01$ ) and was higher in OVER at d -5 ( $P < 0.05$ ). Cer, GlcCer and LacCer were positively correlated to NEFA and negatively to SM during transition ( $P < 0.001$ ). Our results support a model of NEFA-induced ceramide synthesis during peripartum.

**Key Words:** dairy cow, transition, sphingolipid

**475 Elevations in milk yield from palmitic acid feeding are associated with reduced estimated insulin sensitivity and glucose-stimulated NEFA disappearance.** Alice T. Mathews\*, J. Eduardo Rico, Neil T. Sprenkle, and Joseph W. McFadden, *West Virginia University, Morgantown, WV.*

The ability of saturated fatty acids (SFA) to enhance milk yield in dairy cows may be due to shifts in glucose utilization by reducing insulin sensitivity in adipose and muscle tissues. Our objective was to evaluate the effects of palmitic acid (PA) on milk production and insulin sensitivity in cows. Twenty multiparous mid-lactation Holstein cows were enrolled in a 68-d study consisting of 3 sequential periods: 5-d covariate, 49-d treatment, and 14-d post. All cows received a sorghum silage-based diet pre- and post-treatment, and were randomly assigned to a common diet (control; no fat,  $n = 10$ ;  $138 \pm 45$  DIM) or PA at 4% of ration DM (98% PA; Palmit 98; Global Agri-Trade;  $n = 10$ ;  $136 \pm 44$  DIM). Blood and milk were collected at routine intervals. Intravenous glucose challenges (0.3 g/kg BW; GTT) were performed at d -1, 21, and 49 relative to start of treatments. Data were analyzed as repeated measures using a mixed model (fixed effects of treatment and time), and milk yield served as a covariate. Effects of PA are presented as changes relative to control. PA increased milk yield by wk 7 (30.8 vs. 24.6 kg/d,  $P < 0.05$ ). PA increased milk fat yield (+22 and +18%;  $P < 0.05$ ) and 3.5%-FCM (+19 and +18%;  $P < 0.05$ ) by wk 3 and 7, respectively. PA had no effect on milk protein or lactose content, MUN, SCC, or BW, but increased milk protein yield by wk 7 ( $P = 0.05$ ). PA tended to increase estimated energy balance by wk 7 ( $P = 0.06$ ). PA increased feed efficiency (+10%; 3.5%-FCM/DMI;  $P < 0.05$ ). By d 68, 3.5%-FCM remained elevated in PA cows ( $P < 0.05$ ), while milk yield and components were not different. PA increased plasma NEFA by 48, 93, and 60% by d 4, 6, and 8 ( $P < 0.05$ ), respectively, and had no effect thereafter. PA did not modify plasma insulin or glucose, but reduced estimated insulin sensitivity wk 1 (e.g., -35% on d 8;  $P < 0.01$ ). PA did not modify glucose disposal following GTT; however, PA reduced glucose-stimulated NEFA disappearance by wk 7 ( $P < 0.05$ ). Our data suggest that increased milk yield with PA supplementation may be due in part to alterations in insulin sensitivity.

**Key Words:** insulin sensitivity, milk production, palmitic acid

**476 Effects of timing of chromium propionate supplementation on metabolic and production responses of Holstein cows in early lactation.** Michael S. Allen\* and Richard Longuski, *Michigan State University, East Lansing, MI.*

Forty-eight multiparous Holstein cows were used in a randomized block design experiment with 4 treatments and 12 cows per treatment to determine effects of timing of chromium propionate (CrPr) supplementation on metabolic and production responses in early lactation. Treatments were chromium propionate (C) or control (N), supplemented according to 4 schedules: 1 to 65 d PP (CC); 1–22 d PP (CN); 23–65 d PP (NC); no CrPr supplementation (NN). Supplements were top-dressed at 20 g per cow/d to provide 8 mg of chromium per cow/d for C (KemTRACE Chromium, 0.04% chromium, Kemin Animal Nutrition and Health) or no additional chromium for N (feed-grade limestone). Cows were blocked by calving date, body condition score (BCS) and previous milk yield, and randomly assigned to treatment. Cows were offered a fresh diet (FR) from 1 to 22 d PP and a peak diet (PK) from 23 to  $68 \pm 3$  d PP. CrPr increased daily DMI (2.75 kg/d) and milk yield (4.5 kg/d, both  $P < 0.01$ ), tended to decrease milk fat concentration ( $P = 0.07$ ), and did not affect fat yield compared with control when supplemented during FR only. There were no interactions of the main effects of timing of

CrPr supplementation; effects of CrPr supplementation were sustained through most of the experiment when supplementation ceased at 22 d PP and starting supplementation at 23 d PP did not benefit production. Although CrPr supplementation during FR increased milk yield, the effect diminished by 68 d PP, even when supplementation continued during PK. Plasma metabolites and hormones indicate possible effects of CrPr on insulin sensitivity with a reduction in plasma concentrations of NEFA ( $P = 0.08$ ) and BHBA ( $P = 0.02$ ), lower milk fat concentration ( $P = 0.07$ ), and greater BCS over time (interaction  $P = 0.06$ ) during FR compared with control. Supplementation of CrPr during FR increased DMI and milk yield but effects on production diminished over time with no effects of CrPr by 10 weeks PP, even when supplementation continued during PK. Initiating supplementation at 23 d PP did not benefit production in this experiment.

**Key Words:** postpartum, chromium, insulin sensitivity

**477 The effects of dietary niacin supplementation on FoxO1 and genes involved in hepatic glucose production in dairy cows during the transition period.** Asako Kinoshita\*<sup>1</sup>, Kathrin Hansen<sup>3</sup>, Lena Locher<sup>1</sup>, Ulrich Meyer<sup>2</sup>, Sven Dänicke<sup>2</sup>, Korinna Huber<sup>3</sup>, and Jürgen Rehage<sup>1</sup>, <sup>1</sup>*Clinic for cattle, University of Veterinary Medicine Hannover, Hannover, Lower Saxony, Germany*, <sup>2</sup>*Institute of Animal Nutrition, Friedrich-Loeffler-Institute, Braunschweig, Lower Saxony, Germany*, <sup>3</sup>*Department of Physiology, University of Veterinary Medicine Hannover, Hannover, Lower Saxony, Germany.*

Forkhead box protein O1 (FoxO1) promotes the hepatic glucose production (HGP) by activating the transcription of gluconeogenic enzymes. As a main target of insulin signaling, FoxO1 is phosphorylated by insulin, leading to inhibition of HGP. Dietary niacin supplementation could affect HGP; for example, by modifying the expression of genes or by inhibiting lipolysis. The objective of this study was to investigate the effects of dietary niacin supplements on protein expression of hepatic FoxO1 and mRNA expression of genes involved in HGP in dairy cows during the transition period. Twenty-one pluriparous German Holstein cows were used for  $2 \times 2$  factorial analysis. Cows received diets containing 0 g (C) or 24 g (N) niacin supplementation from -42 d related to calving (d -42) to d 24. Each group was further divided and received diets with 30% (L) or 60% (H) of concentrate on dry matter basis from d-42 to d0. Dietary concentrate proportion was set to 30% at d 0 for all, and then increased up to 50% within 16 d for CL and NL and within 24 d for CH and NH. Liver biopsies were taken at d -42, d 3, and d 21. Protein expression of FoxO1 and phosphorylated FoxO1 at serine 256 (pFoxO1) was measured semiquantitatively by Western blotting. Real-time RT-PCR was performed to measure the mRNA of FoxO1, glucose-6-phosphatase (G6P), pyruvate carboxylase (PC), cytosolic phosphoenolpyruvate carboxylase (PCK1), propionyl CoA carboxylase (PCCA), glucose transporter 2 (GLUT2). Data were evaluated by mixed model for repeated measures to test the effect of time, niacin and prepartal concentrate proportion. The protein and mRNA expression of FoxO1 and the protein expression of pFoxO1 were affected neither by time nor by diet. In cows fed with niacin, the relative quantities of mRNA of G6P, GLUT2, PCCA were higher at d 21 and that of GLUT2 was lower at d 1 ( $P < 0.05$ ). Dietary niacin supplements altered the gene expression, increasing HGP in the transition period of cows. However, the regulation of HGP by FoxO1 seemed to be of less importance on the levels of mRNA, protein and phosphorylation.

**Key Words:** FoxO1, hepatic glucose production, cow



## Teaching/Undergraduate and Graduate Education

### 478 “Pre-Veterinary Academy” course exposes and inspires.

Andrew Fidler\*, *University of Arkansas, Fayetteville, AR.*

In the competitive environment of professional school admissions, and with the massive time, effort, and financial commitment of a professional school program, students benefit from experiences that either reinforce their decision to pursue a professional degree or cause them to change their career path. The “Pre-Veterinary Academy” course exposes students to a veterinary school-like learning experience and delivers a range of subject matter for students to evaluate their interest in and preparation for veterinary school. The course utilizes a condensed 9-d intersession period, providing a unique opportunity to present a large-volume, fast-paced curriculum. Having the full academic attention of the students allows for more intensive lectures, case studies, and hands-on opportunities that are less practical to achieve during a full semester course. The condensed schedule also allows for visiting veterinary college faculty to provide several lectures in a single day, giving the students a more realistic veterinary school-like learning environment. All 10 students enrolled in the course in January 2015 completed a survey following conclusion of the course. All 10 students were either planning on or considering attending veterinary school, with no significant change following completion of the course. All 10 students believed they had at least “some,” with 7 believing they had “most,” and 1 believing (s) he had “all” of the skills, knowledge, and dedication to be successful in veterinary school. All 10 students indicated at least some improvement in their ability to perform and analyze a clinical examination of a veterinary patient as well as their ability to understand and apply research and clinical evidence to a medical case following completion of the course. Seven of 10 students described at least some improvement in their ability to understand and analyze lecture material as well as their ability to work with classmates to achieve learning objectives. The “Pre-Veterinary Academy” course provides students a brief taste of a veterinary curriculum to inspire commitment to and confidence in their pre-veterinary education.

**Key Words:** veterinary, education, teaching

### 479 SafeAssign as a tool for student identification of potential plagiarism. Sarah A. Reed\*, *Department of Animal Science, University of Connecticut, Storrs, CT.*

As writing across the curriculum becomes more common in higher education, the need for instruction about plagiarism is growing. Several plagiarism detection technologies identify matching text in student writing and published work. These technologies have been used to assist instructors in detecting plagiarism and to deter students from committing plagiarism. However, they may also be useful as an instructional tool to assist students to self-identify potential areas of plagiarism in their writing and make appropriate revisions before submission. The hypotheses of this study were that student use of SafeAssign would decrease percent of matching text from first to final submission and improve student awareness of plagiarism. To test these hypotheses, students ( $n = 20$ ) in an upper level, writing intensive Animal Science physiology course were required to submit their first submission of a 15-page literature review to SafeAssign and write a short reflective piece on how they would improve their writing for the final submission based on the results. Students then completed an anonymous online survey to gauge the perceived usefulness of the assignment. Final submissions were also analyzed by SafeAssign and compared with first submissions.

The percent of matching text identified by SafeAssign ranged from 0 to 22% in each submission and was not different between submissions (first:  $8.1 \pm 1.5\%$ , final:  $7.5 \pm 1.6\%$ ,  $P = 0.6$ ). However, much of the text flagged were false positives, including common phrases and citations. Of 5 papers with true plagiarism, 2 were appropriately revised for the final draft and had fewer incidences of plagiarism. Students reported that use of the program was helpful in identifying potential areas of plagiarism ( $n = 69\%$ ) and increased understanding of what is considered plagiarism ( $n = 54\%$ ). Due to inclusion of false positives, the report generated by SafeAssign must be carefully interpreted by both the instructor and student; however, the report provides an additional opportunity for dialog about plagiarism. Use of plagiarism detection programs may be beneficial when used formatively by students during the writing process when supported by other mechanisms of instruction.

**Key Words:** plagiarism, writing, animal science

### 480 Using animal sciences courses to teach general university learning goals. John P. McNamara and Martin Maquivar\*, *Washington State University, Pullman, WA.*

At many universities, there are broad learning goals expected of all undergraduates; including critical and creative thinking, information and scientific literacy, communication, quantitative reasoning, diversity; and depth, breadth and integration of learning. Department often align their learning goals as much as possible, to these goals. To gain an assessment of Animal Science student’s broad abilities, we conducted a survey in 2 semesters to gain a qualitative assessment of student learning related to these broad goals. The classes included Intro. Anim. Sci. (101); Comp. Anim. Nutrition (a university General Biology course open to all majors); AS 285, Rights and Welfare; AS 350, Anim. Reproduction and AS 464 Comp. Anim. Management, a university Capstone course (primarily AS majors with others). The approximately 200 students were more than 80% under 24 years of age, female, Caucasian, with an even distribution of grade levels. For learning information literacy, over 90% of students went to the Internet first to find information, 40 to 55% to textbooks, 20 to 30% to scientific journals, 50% to notes, and only 10% to experts. For information literacy, approximately 55 to 75% self-identified as being comfortable (C) or very comfortable (VC) on using Internet sites from general, university, government agencies, and companies. For scientific and quantitative literacy AS students reported 67, 78, 77, and 65% C and VC on finding and reporting quantitative data, comparing data from graphs and tables, making conclusions from data and preparing graphs and tables. For general communications, on average, AS students reported C and VC at 69, 78, 68 and 56% for communication using verbal conversations, essays, presentations and long papers, with an improvement from start to end of semester of 7, 5, 12 and 13%. For communication on specific AS topics; for writing, oral, using social media, engaging in discussion and explaining complex issues, they reported C and VC at an average of 73, 61, 57, 63 and 56%. Improvement in during the semester was 6, 6, 11, 7 and 17% (a 35% improvement in “explaining complex issues”). Animal science courses can be used effectively to teach broadly applicable skills as well as subject specific skills.

**Key Words:** teaching, undergraduate learning goals, animal science students

**481 Perceptions and effectiveness of teaching methods and technology in animal sciences classes.** John P. McNamara\* and Martin Maquivar, *Washington State University, Pullman, WA.*

New learning styles and careers demand new teaching pedagogies and technologies. Lecture/test, videos, online materials (video, PowerPoints, text, web searches); group projects, and homework may be used to a variety of effects. Different students learn in different ways and a variety of teaching methodologies may lift all students, at the least give them a more diverse skillset. Animal sciences teachers must also adapt to learning styles and technologies, however we should understand their effectiveness. To assess the effectiveness of various teaching technologies, we conducted a survey of animal sciences students over the course of 2 semesters and different grade levels to gain a qualitative assessment of perceptions and effectiveness of methods and technologies on student learning. The classes included Intro. Anim. Sci. (101); Comp. Anim. Nutrition (a university General Biology course open to all majors); AS 285, Rights and Welfare; AS 350, Anim. Reproduction and AS 464 Comp. Anim. Management, a university Capstone course (primarily AS majors with others). The approximately 200 students were more than 80% under 24 years of age, female, Caucasian, with an even distribution of grade levels. The students reported that lectures (with or without slides) and videos, pictures, or other media were their preferred methods of learning at 67 and 50%. In fall semester the percentage choosing lectures as their preferred methodology increased from 60 to 77% and videos decreased from 52 to 42%. When asked about the use of various teaching methods or technology, overwhelmingly 87 to 89% of students responded that in class lectures had the most positive effect on learning, while technologies such as clickers, interactive software, “flipped classroom” (students do work outside class and discuss/work in class) all were fairly neutral to slightly positive (25 to 40% “no effect” and 36 to 53% positive or very positive). Using on line search engines were a positive and very positive for 75% of the students. At this stage it may be concluded that students are most familiar and comfortable with in class lectures and on line searches but other technologies and methods are at least positively affecting learning.

**Key Words:** teaching technology, student perceptions, undergraduate education

**482 Closing the gap between society and university: Effect of teaching animal welfare and animal rights course to animal sciences students at Washington State University.** Martin Maquivar\*, Kristen Koenig, and John McNamara, *Washington State University, Pullman, WA.*

There is increased scrutiny and concern for the welfare of animals used in research, education, food production, and companionship. Animal sciences departments have an opportunity and responsibility to provide education and expertise regarding animal welfare. Our general University learning goals include critical and creative thinking, quantitative reasoning, scientific and information literacy, communication skills, global diversity and integration of learning. The overall objective of the present study was to assess the effect of animal welfare and animal rights course on university learning goals of critical, creative thinking and communication skills related to animal welfare issues. A survey was conducted at the beginning and at the end of the semester. A total of 61 students (84% female and primarily third year) voluntarily answered the survey (questions ranked in a 5-point Likert scale) to assess perceptions and effectiveness of the course to achieve these goals. The course included discussions, assigned readings, term papers and a team project where students prepared a topic related to animal welfare and presented it to community groups (seniors, elementary school students,

and high school students). At the end of the semester students agreed or strongly agreed that “this course increased their ability to define, analyze and solve problems” (90.2%, 55/61); “understand diverse philosophical viewpoints and cultural perspectives” (92%, 56/61); “evaluate arguments based on evidence” (88.5%, 54/61); “tailor messages to audiences” (90.2%, 55/61); “express concepts and beliefs in a coherent, concise and technically correct form” (98.3% 59/61) and “speak with comfort in front of groups” (95%, 58/61). Students felt this course improved and increased their ability to solve problems, engage in discussions with members of the society and also increased their ability to effectively learn.

**Key Words:** animal welfare, undergraduate education, teaching

**483 The relationship between delivery and discovery skills and student performance in animal science courses.** A. Ahmadzadeh\*, J. M. Falk, D. T. Masser, and M. E. Doumit, *University of Idaho, Moscow, ID.*

Different courses require different types of skills by students. The objective was to investigate the relationship between 2 types of students’ personal skills and their performance as measured by course grade. The population consisted of 55 students in 2 animal science courses (Animal Reproduction and Animal Products) at the University of Idaho. A 20-question survey was used to assess an individual’s delivery skills (planning, attention to detail, implementing, and self-discipline) and discovery skills (questioning, observing, networking, and experimenting). Discovery and delivery types were measured (scale of 0 to 50) and based on the score, the skill scores for each type were categorized into 3 levels: high (H; 43–50), moderate (M, 33–42), or low (L;  $\leq 32$ ). Data were analyzed using ANOVA. The model tested the effects of skill level, course, and their interaction. There was no interaction between course type and skill level. There was a moderate association between delivery skills and exam scores ( $r^2 = 0.4$ ,  $P < 0.01$ ). However, overall delivery skill level influenced ( $P < 0.01$ ) the total exams grades. Grades were  $88.1 \pm 3.1$ ,  $79.5 \pm 2.3$ , and  $71.5 \pm 3.6$  for H, M, and L delivery skill, respectively. Discovery skills and exam scores had little association ( $r^2 = 0.1$ ,  $P > 0.5$ ). Discovery skill level did not influence ( $P > 0.2$ ) the total exam scores. Grades were  $87.8 \pm 5.1$ ,  $78.6 \pm 2.1$ , and  $82.1 \pm 3.1$  for H, M, and L discovery skills, respectively. Overall, when exam scores of delivery skills were compared, students with H delivery skill earned the highest grades, whereas discovery skills had little influence on exam scores in these 2 courses. Students with high levels of Delivery skills tend to excel at planning, attention to detail, executing, and self-discipline. If course content is structured for memorization and fact acquisition, then instructors are encouraged to help students develop delivery skills. If course content requires exploration and innovation, then instructors should encourage discovery skills (like questioning, networking and experimenting), and provide feedback on these skills using course grades and assignments.

**Key Words:** discovery and delivery skills, performance, undergraduates

**484 Gender differences in species and career interests among students in first-year seminar courses.** Cody L. Wright, Sara L. Mastellar, Michael G. Gonda\*, and Andie B. Vsetecka, *South Dakota State University, Brookings, SD.*

Changing demographics of students in Animal Science programs have created some uncertainty as to whether traditional curricula adequately address the needs of such a diverse body of students. This study was

designed to identify gender differences in species and career interests among students in first-year seminar courses in both Animal Science (AS 109) and Veterinary and Biomedical Sciences (VET 199). Students were surveyed using an instrument that included demographic questions and a series of 4-point, Likert-type scales for each species and career interest. Eighty-seven students in AS 109 and 45 in VET 199 completed the survey. Animal Science majors comprised 98.5% of the survey respondents, 1.5% had not declared a major. Of the 132 respondents, 68.2% were female and 31.8% were male. The majority of respondents were freshmen (86.4%), 6.8% were sophomores, and 6.8% were upperclassmen. Half of the students (50.8%) either came from a small hobby farm or had no farm or ranch background. Gender differences were analyzed using a Pearson Chi-Square test. Males had greater ( $P = 0.019$ ) interest in feedlot cattle, while females had greater interest in goats ( $P < 0.001$ ), horses ( $P < 0.001$ ), pets ( $P < 0.001$ ), and zoo animals ( $P = 0.021$ ). Interest in cow-calf, dairy, poultry, sheep, swine, and wildlife between genders were not different. Career fields related to agronomy and the feed industry were of more interest to males ( $P = 0.021$  and  $P = 0.016$ , respectively). Females had greater interest in both hands-on and support careers related to horses ( $P = 0.003$  and  $P < 0.001$ , respectively) and in working with zoo animals ( $P = 0.020$ ). Veterinary medicine tended ( $P = 0.059$ ) to be of greater interest to females than males. Interest in careers related to animal breeding, animal reproduction, banking, Extension, farming/ranching, food production, government, meats, teaching, or veterinary technician between genders were not different. These data suggest that more females than males may be pursuing degrees in animal science. Furthermore, females clearly have greater interest in some species that may not be commonly included in traditional animal science curricula.

**Key Words:** animal science, students, gender

**485 Factors affecting first-year academic success in a large animal science program.** Amanda L. Robinson\*, Heather A. Jennings, Jodi A. Sterle, Cheryl L. Morris, Kenneth J. Stalder, David G. Acker, and Howard D. Tyler, *Iowa State University, Ames, IA.*

This study was designed to determine if student's background, interests or non-academic skills affect their grade point average (GPA) during their first year at a large 4-year Animal Science program. All new students ( $n = 344$ ) entering the Department of Animal Science at Iowa State University in the fall of 2013 completed assessments (TalentSmart's Emotional Intelligence (4 categories)) of non-academic skills at the beginning of their first semester. Demographic surveys were used to collect additional data (sex, residency status, entry status (transfer/freshman), species interest, career goals, and home background). Student records were accessed to provide data on previous and current academic performance that included high school rank, ACT scores, first year ISU GPA, and retention data. All data were analyzed using general linear model methods (PROC GLM, SAS Version 9.3). Fixed effects included sex, residency status, entry status, species interest, career goals, and background as well as ACT and overall Emotional Intelligence (EI) scores. For ACT and Emotional Intelligence assessments, students were separated into group quartiles by their scores. Of the higher order fixed effects, only ACT scores significantly affected end-of-first-year GPA. There was no interaction between Emotional Intelligence quartile scores and ACT quartile scores on end-of-first-year GPA. For incoming students in the ISU animal science program, ACT scores are the best predictors of first year grade point average.

**Key Words:** student success, emotional intelligence, demographics

**486 Integration of an immersive experience in public policy and governmental affairs into agricultural sciences PhD training programs.** Catherine W. Ernst\*<sup>1</sup>, Lowell Randel<sup>2</sup>, Mary Malaspina<sup>3</sup>, Kaitlyn R. Perry<sup>1</sup>, Deborah Velez-Irizarry<sup>1</sup>, Camille Scott<sup>1</sup>, and Ronald O. Bates<sup>1</sup>, <sup>1</sup>*Michigan State University, East Lansing, MI*, <sup>2</sup>*The Randel Group LLC, Washington, DC*, <sup>3</sup>*Michigan State University, Washington, DC.*

With the increasing dependency on extramural funding to define the direction of animal sciences research, changes in policy at the federal level heavily influence the discoveries that will improve animal production. Yet few scientists are familiar with this process. Internship opportunities in governmental affairs or science policy are usually summer or full-year programs, and are typically available to advanced graduate students or post-doctoral scientists. Few opportunities exist for students early in their graduate program to explore governmental agencies or interact with policymakers. To address this limitation, we designed an immersive experience for PhD students supported by a USDA NIFA National Needs Fellowship Grant. Three students spent 4 weeks in Washington, DC in June 2014. Students were housed in a local university dormitory, using week 1 to acclimate to the area including familiarization with local transportation systems and locating destinations for subsequent weeks' activities. Weeks 2 to 4 were set up on a rotational basis with each student individually pursuing a different weekly activity. Students spent 1 week on Capitol Hill in the office of a US Congressman (a different congressional office for each student). They also spent 1 week visiting various USDA agencies (a different agency each day). For the third week, students shadowed a professional in either an agriculture-related non-governmental organization or a governmental agency, selected to fit with their individual interests. Through these experiences, students interacted with scientists in federal agencies involved with research and/or policy development, and scientists in trade or commodity organizations, as well as attended congressional hearings, briefings and other meetings on agriculture-related topics. This opportunity not only reinforced the interconnectedness of science, industry, and policy, but also exposed students to many unique scientific careers. This immersive experience in public policy, and its impact on the agricultural industry, greatly influenced the students' professional development.

**Key Words:** science policy, graduate education

**487 Changes in emotional intelligence scores following a peer mentor experience.** Amanda L. Robinson\*, Heather A. Jennings, Jodi A. Sterle, Cheryl L. Morris, and Howard D. Tyler, *Iowa State University.*

This study was designed to determine if participating in a full-year peer mentor experience affects their emotional intelligence (EI) scores. Students selected to the peer mentor program for the Department of Animal Science at Iowa State University ( $n = 34$ ) completed TalentSmart's Emotional Intelligence assessment before starting the program (January, 2014) and then completed the EI reassessment after completing the program (December, 2014). All data were analyzed using paired *t*-tests (PROC TTEST, SAS Version 9.3) to determine potential differences between each student's beginning and ending EI scores (4 distinct categories and 3 composite categories with possible scores ranging from 50 to 100). Scores for self-awareness, self-management, social awareness, and relationship management all significantly increased for peer mentors. The associated composite categories of personal management and social management also were significantly increased, as was the



composite for overall EI. Students that were new to the peer mentor program and started with initial overall EI scores that were below the group average score experienced the largest increase in all scores, with over a 14-point increase in self-awareness scores and over a 12-point increase in social awareness scores. Students showed improvements in

all aspects of their EI as a result of completing the peer mentor program in the animal science program at ISU.

**Key Words:** peer mentor, emotional intelligence

# ADSA Production Division Symposium: The rumen and beyond— Nutritional physiology of the modern dairy cow

**488 Harnessing the physiology of the modern dairy cow to continue improvements in feed efficiency.** Michael VandeHaar\*<sup>1</sup>, Diane Spurlock<sup>2</sup>, and Louis Armentano<sup>3</sup>, <sup>1</sup>*Michigan State University, East Lansing, MI*, <sup>2</sup>*Iowa State University, Ames, IA*, <sup>3</sup>*University of Wisconsin, Madison, WI*.

Feed efficiency, as defined by the fraction of feed energy captured in products, has more than doubled for the US dairy industry in the past 100 years. This increased feed efficiency resulted from increased milk production per cow achieved through genetics, nutrition, and management with the desired goal being greater profitability. With increased milk production per cow, more feed is consumed per cow but a greater portion of the feed is partitioned toward milk instead of maintenance and body growth. The dilution of maintenance has been the overwhelming driver of enhanced feed efficiency in the past, but its impact diminishes with each successive increment in production relative to body size. In the future, we must focus on new ways to enhance digestive and metabolic efficiency. One way to examine variation in efficiency among animals is residual feed intake (RFI), a measure of efficiency that is independent of the dilution of maintenance. Cows that convert feed energy to net energy more efficiently, or have lower maintenance requirements than expected based on BW, use less feed than expected and thus have lower RFI. Cows with low RFI likely digest and metabolize nutrients more efficiently and should have overall greater efficiency and profitability if they are also healthy, fertile, and produce at a high multiple of maintenance. Genomic technologies will help to identify these animals for selection programs. Nutrition and management also will continue to play a major role in farm-level feed efficiency. Helping all farms achieve the efficiency of the best farms would have a major effect on feed efficiency for the industry. Management practices such as TMR-feeding and grouping improve rumen function and efficiency, but they have decreased our attention on individual cows. Perhaps new computer-driven technologies will enable us to optimize efficiency for each individual cow within a herd, or to optimize animal selection to match management environments. In the future, availability of feed resources may shift as competition for land increases. New approaches combining genetic, nutrition, and other management practices will help optimize feed efficiency, environmental sustainability, and profitability.

**Key Words:** efficiency, dairy cow, genetics

**489 Development and physiology of the rumen and the lower gut: Targets for improving production efficiency.** Michael A. Steele\*<sup>1</sup>, Greg B. Penner<sup>2</sup>, Frédérique Chaucheyras-Durand<sup>3</sup>, and Leluo Guan<sup>1</sup>, <sup>1</sup>*Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB, Canada*, <sup>2</sup>*Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, SK, Canada*, <sup>3</sup>*Lallemand Animal Nutrition, Blagnac, France*.

It has been estimated that the gastrointestinal tract accounts for 30% of metabolic and protein synthesis activities of the cow and calf, and thus, plays an important role in animal energetics. Diet and microbiota can positively and negatively affect gut function and the ability to modulate gut function has created an applied interest relevant to improved dairy cow and calf productivity. The mechanisms that govern growth and barrier function of gut tissues have received particular attention, especially with the advancements of molecular based techniques over the past decade. The rumen has been the focal point of dairy cow and calf

nutritional physiology research, yet the intestinal tract has received less attention. Three key areas requiring more discovery-based and applied research: (1) early-life intestinal gut barrier function and growth; (2) how the weaning transition affects gut health of rumen and intestine; and (3) gastrointestinal adaptations during the transition to high energy diets in early lactation. Nutrients are not only seen as substrates, but also as signals that can alter gastrointestinal growth and barrier function. Nutrients can act directly, affecting epithelial cell gene expression, and in concert with somatotrophic axis hormones, insulin-like growth factor (IGF) and growth hormone where they have been shown to play a pivotal role in gut tissue growth. For example, IGF-1 can mediate other hormones involved in cell growth, such as glucagon-like peptide 2 (GLP-2), whereas, intestinal GLP-2 is secreted upon nutrient ingestion causing a stimulation of intestinal growth. The latest research suggests that total-tract barrier function in calves and cows in early life, at weaning and in early lactation is compromised. A major factor for gut health is maintaining proper gastrointestinal barrier function, which is highly influenced by the presence of metabolites (butyrate) and resident microbiota and/or direct fed microbials within the gut. In the first studies that investigated barrier function in cows and calves, it was determined that the expression of genes encoding mucin and tight junction regulating proteins, such as claudins, occludins and desmosomal cadherins, are regulated by diet. Additionally, recent evidence suggests that the upper and lower gut can communicate, but the exact mechanisms of gastrointestinal cross-talk have not been studied in detail. A deeper understanding of how diet and microbiota can affect growth and barrier function of the intestinal tract would provide knowledge of what specific management regimens could effectively impact gut function.

**490 The contribution of the lower gut to altered nutrient partitioning during stress.** Lance H. Baumgard\*<sup>1</sup>, Sara K. Stoakes<sup>1</sup>, Mohannad Abuajamieh<sup>1</sup>, and Robert P. Rhoads<sup>2</sup>, <sup>1</sup>*Iowa State University, Ames, IA*, <sup>2</sup>*Virginia Tech University, Blacksburg, VA*.

Increased animal productivity and production variability are due to changes in nutrient partitioning. The coordination of nutrient trafficking is an incredibly complex system and how tissues/systems are reprioritized or de-emphasized during different physiological states is controlled by homeostatic and homeorhetic adaptations that probably incorporate every tissue and physiological system. There are a variety of situations in an animal's life-cycle that challenges homeostasis. The metabolic adaptations that cows utilize to copiously synthesize milk following parturition are unfortunately often referred to as "metabolic stress." Metabolic maladaptation to lactation results in ketosis and this is in part either caused by inadequate feed intake or causes reduced feed intake. Regardless, although heavily researched for the last 3 decades the specific etiology of periparturient ketosis remains elusive as it unclear why a small percentage of cows are susceptible (or predisposed?) to metabolic imbalances following calving. Heat stress (HS) compromises efficient animal production and jeopardizes animal welfare and HS animals also have a unique metabolic and physiological fingerprint that is uniquely different than their nutritional status predicts. The origin of both ketosis and HS issues may lie at the gastrointestinal tract. Increased intestinal permeability to lipopolysaccharide (LPS) and other luminal contents results in local and systemic inflammatory responses. LPS interferes with hepatic lipid trafficking, stimulates insulin secretion and influences systemic fuel selection. We demonstrated that both

ketotic and HS animals have markedly increased circulating markers of leaky gut. We have also shown that reduced feed intake (a conserved response to stress) compromises intestinal integrity in both thermal-neutral monogastrics and ruminants. Thus, stressors that physically prevent ad libitum feed intake or cause voluntarily reductions in feed take may share a common mechanism(s). Defining the physiology and mechanisms that underlie how intestinal barrier dysfunction jeopardizes animal performance is critical for developing approaches to ameliorate current production issues.

**Key Words:** nutrient partitioning, intestine

#### **491 Nutritional strategies to optimize dairy cattle immunity.**

Lorraine M. Sordillo\*, *Michigan State University, East Lansing, MI.*

Dairy cattle are susceptible to increased incidence and severity of both metabolic and infectious diseases during the periparturient period. Health problems occurring around the time of parturition are especially problematic because they greatly affect the productive efficiency of cows in the ensuing lactation. A major contributing factor to increased health disorders is alterations in bovine immune mechanisms. Indeed, uncontrolled inflammation is a major contributing factor and a common link among several economically important diseases including mastitis, retained placenta, metritis, displaced abomasum, and ketosis. The nutritional status of dairy cows and the metabolism of specific nutrients are critical regulators of immune cell function. There is now a greater appreciation that certain mediators of the immune system also can have a reciprocal impact on the metabolism of nutrients. Thus, any disturbances in nutritional or immunological homeostasis can provide deleterious feedback loops that can further enhance health disorders, increase production losses, and decrease the availability of safe and nutritious foods for a growing global population. This review will discuss the complex interactions between nutrient metabolism and immune functions in periparturient dairy cattle. Details of how either deficiencies or overexposure to macro- and micronutrients can contribute to immune dysfunction and the subsequent development of health disorders will be presented. Specifically, the ways in which altered nutrient metabolism and oxidative stress can interact to initiate and promote uncontrolled inflammatory responses in transition cows will be discussed. Understanding more about the underlying causes of dysfunctional inflammatory responses may facilitate the design of nutritional regimens that will reduce disease susceptibility in early lactation cows. Given the critical role that nutrition plays in supporting

all immune functions, nutritional-based management strategies should have a central position in any disease prevention program.

**Key Words:** immunity, inflammation, oxidative stress

#### **492 Managing complexity: Dealing with systemic cross-talk in bovine physiology.** Barry J. Bradford\*, *Kansas State University, Manhattan, KS.*

Dairy producers rely heavily on advisors with deep expertise in nutrition, reproduction, and health. However, a shift is occurring, driven by both farm size and advances in biology. Larger dairy businesses can investigate management options with a degree of precision never before possible; simultaneously, the lines between the metabolic, immune, and reproductive systems are becoming blurred. For example, new research has revealed a surprising role for immune cells in regulating metabolism and documented the nutrient requirements of the immune system. The gut epithelium has garnered new attention as a tissue that actively manages the commensal microbiome, entrains the responses of the neonatal immune system, and provides a barrier limiting movement of molecules from the gut lumen. New hormone discoveries have added adipose tissue, bone, and muscle to the list of endocrine organs. Finally, nutrients are now seen not only as substrates and cofactors, but also as signals that can alter cellular function. What does all of this mean for the dairy industry? Consultants increasingly need to reach across disciplinary boundaries to best support the physiology of the cow. However, research is needed to move beyond proof-of-principle findings toward applications in dairy cattle. Key unanswered questions include: the degree to which roles of the hindgut in monogastrics translate to ruminants; whether host/microbe crosstalk also occurs in the rumen; whether hormone release by storage organs during a catabolic state affects reproductive function; and the degree to which immunostimulation by dietary signals enhances or disrupts health and productivity. It is critical to address these questions with a 2-pronged approach. Mechanistic studies provide a nuanced understanding of signal interactions, but large-scale commercial studies are also needed to evaluate effects on multiple production outcomes in the environment of interest. Incorporating all aspects of animal health and productivity in management decisions will remain an art for the foreseeable future, but this should not dissuade the industry from pursuing a more holistic approach to management of the cow.

**Key Words:** physiology, endocrinology, dairy cow



# Animal Behavior and Well-Being I

**493 Development of a behavior-based screening tool for disease detection in preweaned group-housed dairy calves.** Mary C. Cramer\* and Amy L. Stanton, *University of Wisconsin- Madison, Madison, WI.*

Producers report bovine respiratory disease (BRD) and diarrhea as the 2 most common diseases in preweaned calves, both of which can affect calf performance and welfare. Group housing has the potential to improve calf welfare, but only if producers are able to detect disease. To reduce time evaluating individual calves, there is a need for a group level health-screening tool to identify potentially sick calves that require further examination. The objective of this study was to develop a screening tool using behavioral measures to detect disease in preweaned group-housed dairy calves. This cross-sectional study included the evaluation of preweaned group-housed calves ( $n = 206$ ) on 4 farms in Wisconsin. Calves were scored for behaviors that were hypothesized to be indicative of illness: abnormal posture when lying or standing, isolation, lethargy, and the willingness to approach a stationary person (2 approach tests were performed and each was scored separately). Each behavior category was scored as normal (0 points) or abnormal (1 point) and then categories were summed to obtain a total behavior score for each calf. Following the behavior scoring, calves underwent an individual health assessment for BRD, diarrhea, and umbilical infections. Total behavior scores ranged from 0 (normal) to 5 (severely abnormal) and reflect the number of abnormal behaviors observed. The highest sensitivity and specificity of the behavior score for disease identification occurred at score cut points of 2 and 3. Therefore, scores of 2 or greater or scores of 3 or greater were considered a positive test result for disease and were further examined. Outcomes of a positive or negative behavior score for cut points of 2 and 3 were analyzed using PROC LOGISTIC in SAS and all models were controlled for calf age and farm. A score of 2 or greater tended to be significant for the detection of diarrhea ( $P = 0.05$ ), but was not significant for BRD ( $P > 0.05$ ). At a cut point of 3 or greater, calves without BRD were 0.24 (95% CI: 0.10 to 0.59) times as likely to have a positive behavior score, compared with calves with BRD ( $P < 0.01$ ). Utilizing a cut point of 3 or greater on this behavior-based screening tool may be useful to identify calves with BRD.

**Key Words:** behavior, BRD, calves

**494 Milk replacer plane of nutrition influences calf nutritive and non-nutritive oral behaviors.** Lindsey E. Hulbert<sup>1</sup>, Sophia C. Trombetta\*<sup>1</sup>, Kate P. Sharon<sup>2</sup>, and Michael A. Ballou<sup>2</sup>, <sup>1</sup>*Department of Animal Sciences and Industry, Kansas State University, Manhattan, KS,* <sup>2</sup>*Department of Animal and Food Science, Texas Tech University, Lubbock, TX.*

The objectives of the study were to (1) determine if milk replacer (MR) plane of nutrition (PN) influences Holstein bull-calf daily duration of non-nutritive sucking (NNS) as well as daily calf starter (CS) and water intake from ages 6–64 d and, (2) determine if automated NNS measures can be used as an additional measure to CS intake to determine if calves are ready for weaning. At age 1 d, calves were randomly assigned to MR treatments of either High (HPN;  $n = 18$ ; 20:28 DM fat:protein; 830g and 1080 g DM/d from ages 1 to 10 and 11 to 51 d, respectively) or Low (LPN;  $n = 15$ ; 20:20 fat:protein; 445 g DM/d ages 1–51 d) PN. All calves were fed milk bottles twice daily at 0700 and 1600 h until step-down weaning. Weaning was initiated at age 51 d by removal of the PM-MR feeding and completed at age 57 d with the removal of the

AM-MR feeding. Water and CS were offered ad libitum and measured once daily. Daily-NNS was measured by event-loggers (1 Hz, HOB0 State Data Logger UX90–001M) that recorded when each calf manipulated a dummy-nipple. Repated data were analyzed using REML PROC MIXED procedure in SAS. Throughout the experiment, LPN-calves drank  $1.5 \times$  more water than HPN-calves ( $P < 0.05$ ). Prior to weaning, LPN-calves consumed more CS than HPN-calves ( $P < 0.01$ ), except for ages 32–41 d. During and after weaning the LPN calves consumed over 18% more CS than HPN ( $P < 0.01$ ). After weaning was completed, HPN-calves doubled CS intake compared with pre-weaning ( $P < 0.05$ ). The LPN-calves performed more daily NNS at ages 13–17 d ( $P < 0.01$ ). Around weaning, HPN-calves tripled NNS from pre-weaning measures ( $P < 0.05$ ), while LPN calves nearly stopped their NNS behaviors ( $P < 0.01$ ). These results indicate that PN influences nutritive and non-nutritive oral behaviors. Calves fed LPN are not motivated to perform NNS once one MR feeding is removed from their diet. In contrast, weaning-initiation increases NNS in HPN-calves, but full-removal of MR stimulates CS intake in HPN-calves. In addition, HPN-calves may not acclimate as easily as LPN-calves to step-down weaning; therefore, alternate weaning strategies for HPN calves may need to be considered and tested utilizing NNS measures.

**Key Words:** behavior, calf, nutrition

**495 Evaluation of pelleting a feed-through larvicide on dairy calf behavior and fly control.** Randi Black\*<sup>1</sup>, Christa Kurman<sup>1</sup>, David Paulsen<sup>2</sup>, Rebecca Trout Fryxell<sup>2</sup>, and Peter Krawczel<sup>1</sup>, <sup>1</sup>*Department of Animal Science, University of Tennessee, Knoxville, TN,* <sup>2</sup>*Department of Entomology and Plant Pathology, University of Tennessee, Knoxville, TN.*

Dipteran pests affect dairy calves through rapid transmission of pathogens and reduction in calf well-being. A feed-through larvicide may effectively manage fly pressure and improve well-being. The objective of this study was to determine the effect of pelleting a feed through larvicide on calf lying behavior. Holstein dairy calves ( $n = 12$ ) were divided into 4 pens and assigned to one of 2 treatment sequences: control (CON)-washout (WAS)-larvicide (LAR) ( $n = 6$ ) or LAR-WAS-CON ( $n = 6$ ). CON and LAR lasted 4 d and WAS lasted 10 d. The control pellet was fed during CON and WAS and the experimental pellet was fed during LAR. Dataloggers recorded lying bouts and posture at 1-min intervals from d 0 to d 24. Fecal samples, collected on d 2, 4, 11, 20, and 14, were seeded with house ( $n = 25$ ), stable ( $n = 25$ ), face ( $n = 25$ ), and horn fly eggs ( $n = 25$ ) to assessed daily emergence of pupae and adults. Data were analyzed using PROC GLM of SAS to determine the effect of treatment period, sequence, and treatment period  $\times$  sequence interaction. No effects on lying bouts were evident ( $P \geq 0.26$ ). Calves laid down longer during CON ( $17.1 \pm 0.2$  h/d) and LAR ( $17.2 \pm 0.2$  h/d) compared with WAS ( $14.4 \pm 0.1$  h/d;  $P < 0.0001$ ). Further, calves laid down less frequently during WAS ( $11.6 \pm 0.2$  bouts/d) compared with CON ( $16.5 \pm 0.4$  bouts/d) and LAR ( $16.0 \pm 0.4$  bouts/d;  $P < 0.0001$ ). Calves on the LAR-WAS-CON sequence had a tendency to stand longer ( $8.0 \pm 0.2$  h/d) than CON-WAS-LAR sequence calves ( $7.5 \pm 0.2$  h/d;  $P = 0.08$ ). Fewer pupae emerged during the WAS period of the LAR-WAS-CON sequence ( $5.8 \pm 1.5$  pupae) compared with CON in CON-WAS-LAR ( $18.0 \pm 1.0$  pupae) and LAR-WAS-CON ( $19.1 \pm 1.0$  pupae), LAR in CON-WAS-LAR ( $18.8 \pm 1.0$  pupae) and LAR-WAS-CON ( $16.3 \pm 1.0$  pupae), and WAS in CON-WAS-LAR ( $13.7 \pm 1.5$  pupae;  $P < 0.0001$ ). This indicates there may be a delayed response of larvicide. Further,

fewer adults emerged during the LAR period ( $1.6 \pm 0.8$  adults) compared with the CON period ( $7.9 \pm 0.8$  adults) of the LAR-WAS-CON sequence ( $P < 0.01$ ). These data suggest a pelleted feed through larvicide may be an effective fly management option for producers without negatively affecting calf behavior.

**Key Words:** dairy calf, larvicide, behavior

**496 An outdoor method of housing dairy calves in groups using individual calf hutches.** Lisa M. Wormsbecher<sup>\*1</sup>, Renée Bergeron<sup>1</sup>, Derek B. Haley<sup>2</sup>, Anne Marie B. de Passillé<sup>3</sup>, Jeff Rushen<sup>3</sup>, and Elsa Vasseur<sup>1</sup>, <sup>1</sup>*Organic Dairy Research Centre, University of Guelph, Alfred Campus, Alfred, ON, Canada*, <sup>2</sup>*Ontario Veterinary College, University of Guelph, Guelph, ON, Canada*, <sup>3</sup>*UBC Dairy Education and Research Centre, University of British Columbia, Agassiz, BC, Canada*.

Housing dairy calves in groups allows natural social interactions between conspecifics. Many farms still house dairy calves individually and feed them milk by hand. Some dairy operations have implemented group-housing using an automated milk feeding system even if the initial set up is expensive. To evaluate the feasibility of using calf hutches in a novel, low input cost group-housing method, we compared individually- and pair-housed calves kept in hutches and examined differences in growth rate, activity level, and competition of paired calves at feeding. Individual calves were given a hutch with an outdoor area (total 6.87 m<sup>2</sup>); 2 hutches were placed side by side with a shared outdoor area for paired calves (6.87 m<sup>2</sup> total per calf). Eighteen Holstein heifer calves were purchased and arrived on farm between the ages of 5–12 d. Calves were assigned to treatments and balanced for age and weight within pairs and across treatments. Over 7 weeks, calves had free-access to milk (up to 16 L/day/calf), calf starter, water and hay. Activity loggers were used to record daily lying time and calves were weighed each week to monitor growth. To evaluate competition in pairs and use of space for all calves, behavioral observations were completed 1 d/wk (7 non-consecutive h/d) and included recording calf location within pen, and for paired calves: displacements at the nipple (indicative of competition between calves), cross sucking (sucking on conspecific), and social grooming (grooming/licking another calf). Over 7 weeks of milk feeding the average feed conversion was  $0.10 \pm 0.005$  kg weight gain/kg milk intake (mean  $\pm$  SE) for paired and individual calves. Social interactions by paired calves did not change across weeks (Wilcoxon sign rank tests;  $P > 0.1$ ):  $5.5 \pm 1.62\%$  social grooming,  $1.4 \pm 0.84\%$  cross sucking, and  $6.5 \pm 3.60\%$  displacements at the nipple. Data for growth, resting time, and space usage are being analyzed while milk and competition data are undergoing further analysis. A 6-week winter replicate of this research is in progress to determine whether the housing method can be utilized in various climatic conditions.

**Key Words:** dairy, calf, housing

**497 The use of single measurements to assess growth of dairy calves and the effect of management practices on calf BW variability.** Guilherme B. Bond<sup>\*1</sup>, Daniel M. Weary<sup>2</sup>, Marina A. G. von Keyserlingk<sup>2</sup>, Lorraine Doepel<sup>1</sup>, Karin Orsel<sup>1</sup>, Herman W. Barkema<sup>1</sup>, and Edmond A. Pajor<sup>1</sup>, <sup>1</sup>*University of Calgary, Calgary, AB, Canada*, <sup>2</sup>*University of British Columbia, Vancouver, BC, Canada*.

Bridging the gap between research methods and application on commercial farm settings is necessary to stimulate producers to use new management practices that promote better calf welfare. Our objectives were to evaluate the association between a single measurement of calf

BW and management practices and identify practices related to higher BW residuals. Farms ( $n = 82$ ) were selected through convenience sampling, had at least 80 Holstein lactating cows and used DHI recording systems. Visits occurred once from June 2010 to August 2012 in AB and BC, Canada. Producers were interviewed about their pre-weaning calf management practices and heart girth was measured for all available calves from 0 to 8 weeks of age. The final database contained data from 865 calves from 57 farms in AB (567 calves) and 25 farms from BC (298 calves). Calf BW was regressed against age and the resulting line equation was  $BW = 40.56 + 4.649 \times \text{Age (wk)}$ . Calf BW residuals were calculated by subtracting the actual BW from the expected BW at each age. Individual animal BW residuals were averaged by farm to represent calf growth on each farm. Average farm BW residuals ranged from  $-16.5$  kg to  $+16.7$  kg. The association between management practices and calf BW residuals was tested at calf-level using mixed models in SAS 9.4. Management variables significant in a univariable model ( $P < 0.20$ ) were included in the final multivariable model ( $P < 0.05$ ). Feeding more than 5 L of milk per day through a teat was associated with higher BW. Conversely, calves fed less than 5 L of milk by bottle had the lowest BW when compared with the other feeding strategies. Feeding milk by bucket, regardless of the amount, was associated with lower BW. Calves introduced to water at an earlier age had higher BW than calves introduced to water later than 2 mo of age. Calves housed in groups had lower BW when compared with calves housed in individual pens; mixed housing (using 2 or more different types of housing) resulted in the lowest BW compared with other systems. The growth measurement used in this study was successful in describing differences of calf growth among farms. These differences were related to practices such as feeding more milk by teat and early introduction to water.

**Key Words:** welfare, feeding, water

**498 Reflective calf hutch covers improve antibody response and decrease panting, but not gain, during hot conditions.** Jade Haberman, Theodore Friend<sup>\*</sup>, and Thomas Hairgrove, *Texas A&M University, College Station, TX*.

Previous research found reflective hutch covers reduced hutch temperature during hot weather, but the biological significance is unknown. This study determined the effect of reflective covers on calves' immune response, body weight, and respiration rate. The study was conducted from June to August, on 2 farms: one near Stanfield, Arizona (10,000 cows) and one near Plainview, Texas (6,000 cows). Agri-Plastic hutches were used in Arizona and Calf-Tel in Texas. Covers were 3.0 mil (aluminized on the external side) white LDPE overlaying the top, sides, and back of the hutches; leaving the front exposed. At each farm, 5 covered and 5 uncovered (control) hutches were also fitted with temperature loggers that recorded internal hutch temperature every 30 min. Calves were given a commercial IBR vaccine at 21-d of age and antibody titer differences at 21 and 42 d determined immune response. Titers were determined using virus neutralization with logarithmic transformation to normalize data. Treatment effects were analyzed with ANOVA and LSD. Calves in covered hutches in AZ ( $n = 50$ ) had a higher ( $P < 0.05$ ) titer responses (25%) than control calves ( $n = 54$ ). Respiration rates taken during sunny conditions at 16:00 at AZ were significantly lower (8 bpm,  $P < 0.01$ ) for covered calves ( $n = 43$ ) than controls ( $n = 50$ ). Body weight was not significantly influenced by treatments at either farm ( $P > 0.05$ ). Covered hutches were  $3.03^\circ\text{C}$  cooler than controls at the hottest time of the day at AZ ( $P < 0.01$ ) and  $0.7^\circ\text{C}$  warmer ( $P < 0.05$ ) during the coolest 2 h period at night. TX was much cooler than average with cloud cover and rain, and daily highs averaged  $7.9^\circ\text{C}$  lower than AZ over the study period, so the cover were not advantageous in

TX. In conclusion, the reflective covers were most effective during sunny and hot conditions, resulting in increased antibody response and decreased respiration rate.

**Key Words:** heat, stress, dairy

**499 Lameness score, pain threshold, temperature and type of lesion of severely lame dairy cows before and after trimming.**

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This study aimed to verify the effect of corrective trimming and the type of lesion on lameness score (LS), nociceptive pain threshold and temperature close to the lesion of lame cows. Six Jersey and 28 Holstein cows were used. The day before trimming (day -1), cows were gait scored using a 1-to-5 numerical rating system where 1 = perfect gait and 5 = severely lame. Cows showing LS of 4 or 5 were selected. On d 0 (trimming), cows were restrained in a tilt chute and before they were hoof trimmed, they were evaluated for pain threshold, temperature close to the lesion and type of hoof lesion, which were classified as dermatitis (digital and interdigital) or ulcers (sole and wall ulcers). Temperature close to the lesion was measured with a thermograph. Pain threshold was measured utilizing an algometer, applying a pressure stimulus at the regions close to the lesion of both rear legs, and observing the foot-lift response. Cows with dermatitis were given an antibiotic and a bandage, those with ulcers received glueing shoes and none received analgesic. After trimming, on d 6, cows were evaluated for LS and on d 7, cows were evaluated for pain threshold and temperature close to the lesion. Data were submitted to variance analysis according to a completely randomized design, considering in the model the effect of trimming, type of lesion and their interaction. Twenty-six cows had dermatitis, 8 cows had sole and wall ulcers. There was no interaction between type of lesion and trimming (Table 1). Trimming decreased LS and increased pain threshold but did not affect temperature close to the lesion. Cows with dermatitis showed lower pain threshold than those with ulcers. Type of lesion did not affect LS nor the temperature close to the affected region. Trimming improved cow's welfare.

**Table 1 (Abstr. 499).** Effect of trimming and type of lesion on lesion pain and temperature

Trait	Corrective trimming (T)			Type of hoof lesion (L)		
	Before	After	P > F	Dermatitis	Ulcers	P > F
Pain threshold (kgf)	7.0 <sup>a</sup>	9.3 <sup>b</sup>	<0.01	6.8 <sup>a</sup>	9.5 <sup>b</sup>	<0.01
Lameness score (1 - 5)	4.5 <sup>a</sup>	2.9 <sup>b</sup>	<0.01	3.5 <sup>a</sup>	3.9 <sup>a</sup>	NS
Temperature of affected region (°C)	29.6 <sup>a</sup>	29.8 <sup>a</sup>	NS	29.7 <sup>a</sup>	29.8 <sup>a</sup>	NS

**Key Words:** lameness, pain sensitivity, trimming

**500 Assessment of calving personnel performance and stillbirth in dairy herds.**

G. M. Schuenemann\*, J. M. Piñeiro, A. A. Barragan, S. Bas, and J. D. Workman, *The Ohio State University, Columbus, OH.*

It is common to observe large between- and within-herd variation in calving personnel performance (CPP) and turnover (TO). The objectives were to assess (1) the association of self-reported conflicts (e.g., lack of communication) on knowledge, skills, and performance of personnel; and (2) the association of CPP and TO on stillbirth and economics. A total of 70 personnel (18 dairy herds and approximately 18,100 cows) who participated in a 2 yr calving training program designed to improve calving management practices and reduce stillbirth were assessed. The educational content was delivered through oral presentation with group discussion and demonstration with supervised practice (skills). Pre- and post-tests of knowledge were used to assess knowledge gain. Participants evaluated the program and provided qualitative feedback about their working environment. Data were analyzed using GLIMMIX. Unresolved conflicts was significantly associated ( $P < 0.05$ ) with personnel performance (determined by proportion of stillbirth) regardless of any differences in knowledge and skills. For the economic simulation, CPP (determined by compliance with calving protocol) was set to either 85% or 95%. Calf loss (stillbirth) was set at 1 preventable death due to late or no intervention at calving, assuming 18% of births need assistance. The overall risk performance percentage (RP) was estimated taking into account the CCP and TO. Costs were set at \$280/head for calf loss, \$1,000 for herd audit, and \$1,000 for training (4 sessions/yr). For a 2,000-cow herd, calf losses were higher (\$43,490 or \$21 per cow/yr) for RP 85% with 30% TO (166 calves lost/yr) compared with RP 95% with 5% TO (19 calves lost/yr). The return on investment (ROI) for high performance teams (RP 95% and TO 5%) was \$21 for every \$1 invested (herd audit and training). The estimated ROI assumes that facilities are adequate, participants are willing to learn and apply newly learned concepts, and the herd audit correctly identifies and addresses the needs with the appropriate training program. Unresolved conflicts (interpersonal issues) within herd are associated with both TO and CPP, which in turns compromise the welfare of cows and calves around parturition.

**Key Words:** dairy personnel, stillbirth, welfare

**501 Do improvements in housing and management voluntarily applied by producers following a cow comfort assessment reduce cow injuries in tie-stall dairies?**

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Cow welfare is an increasing concern for consumers. In Canada new standards on the care and handling of dairy cattle have been established and are currently being enforced. Injuries to hocks, knees, and neck are common in dairy cows and are accepted as being painful and a welfare concern. The objective if this study was to evaluate how voluntary improvements applied by producers in housing and management following a cow comfort assessment with recommendations would lead to improvements in cow welfare in tie-stall farms. The 4 most reported modification made to the tie-stall were: adding new rubber mats, adding or adjusting the brisket board, pushing up and/or out the tie-rail, and/or increasing the chain length. Hock and knee injuries were scored on a scale of 0-3 were scores 2 (swelling 1 - 2.5 cm and/or a lesion) and 3 (swelling >2.5 cm) represent major injuries. Neck injuries were scored on a scale of 0-2 were 2 represents swelling and/or lesions. Forty cows/farm from a total of 19 farms (2 groups: 10 that applied modifications,



9 that did not) visited in the summer were scored during 2 assessments (2011 and 2014). Kruskal-Wallis test was used to compare, between farm groups, the difference in prevalence of injuries between both assessments. Both groups of producers who reported to have made modifications and those that did not had reduced prevalence of major hock injuries by 23.8 and 21.4% (SEM 3.14), major knee injuries by 27.0 and 25.3% (SEM 3.89) and major neck injuries by 32.9 and 34.8% (SEM 4.56) respectively. There were no differences observed in improvement of injury prevalence between both groups of producers (hock:  $X^2 = 0.3$ ,  $P < 0.1$ ; knee:  $X^2 = 0.2$ ,  $P < 0.1$ ; neck:  $X^2 = 0.0$ ,  $P < 0.1$ ). Between the time producers received results of the assessment of their farm, and the new assessment, the level of cow injury decreased indicating that when equipped with knowledge, producers took action to improve cow comfort. Besides the ones reported, producers may have applied other modifications in cow housing or management that could explain the reduction in cow injuries between both assessments.

**502 Social dominance affects body growth, follicle development, and age at puberty in dairy heifers.** Carolina Fiol<sup>\*1,2</sup>, Annie dos Santos<sup>1,2</sup>, Augusto Lacava<sup>1,2</sup>, Ana Maverino<sup>1,2</sup>, Mariana Carriquiry<sup>3</sup>, and Rodolfo Ungerfeld<sup>2</sup>, <sup>1</sup>*Departamento de Bovinos, Facultad de Veterinaria, Montevideo, Uruguay*, <sup>2</sup>*Departamento de Fisiología, Facultad de Veterinaria, Montevideo, Uruguay*, <sup>3</sup>*Departamento de Producción Animal y Pasturas, Facultad de Agronomía, Montevideo, Uruguay*.

The aim of our study was to determine the effects of social dominance in dairy heifers during the prepubertal period on body growth, follicle development and age of onset of cyclic activity. Sixteen Holstein dairy heifers ( $8.2 \pm 0.3$  mo-old,  $208.5 \pm 13.9$  kg BW) were homogeneously allocated according to age and BW to 8 dyads. Each dyad was housed in a different pen separated by electrical fences, had access to only one feeder and received a total mixed ration on a 5% restriction of their potential total dry matter intake to promote competition between heifers. Social dominance (dominant-DH, subordinate-SH) was determined every 30 d in each dyad during the 120 d of the experiment. Heifers were weighed every 20 d, and the number of follicles, the maximum follicle diameter (MFD) and presence of corpus luteum (CL) were determined every 7 d by transrectal ultrasound. Puberty onset was defined as the first day in which a CL—confirmed in the following observation—was recorded. Body weight and follicular parameters were analyzed with an ANOVA for repeated measures, while age at puberty was compared with a paired *t*-test. At the end of the experiment, DH were heavier than SH heifers ( $312.4 \pm 2.5$  vs.  $304.0 \pm 2.5$  kg for DH and SH respectively,  $P = 0.006$ ). In addition, DH reached puberty earlier than SH ( $318.0 \pm 17.5$  vs.  $330.0 \pm 23.3$  d, for DH and SH, respectively;  $P < 0.05$ ), and presented greater MFD from d 53 until the end of the experiment ( $10.0 \pm 0.5$  vs.  $8.3 \pm 0.5$  mm, DH and SH heifers;  $P = 0.04$ ). There was an interaction between social dominance and day of the study for total number of follicles: SH had more follicles than DH heifers on d 70 ( $4.8 \pm 1.1$  vs.  $8.6 \pm 1.1$ , DH

and SH,  $P < 0.01$ ), 79 ( $4.3 \pm 1.1$  vs.  $7.7 \pm 1.1$ , DH and SH,  $P < 0.01$ ) and 86 ( $4.2 \pm 1.1$  vs.  $6.8 \pm 1.1$ , DH and SH,  $P = 0.04$ ). No differences were found on the number of follicles  $>6$  mm between groups ( $1.7 \pm 0.3$  vs.  $2.0 \pm 0.3$ , DH and SH, respectively;  $P > 0.05$ ). In conclusion, with a slight restriction of food availability, dominant heifers presented greater body growth and reproductive development than subordinate ones. Financial support: CSIC, Uruguay.

**Key Words:** dominance, replacement heifer, puberty

**503 Association between social ranking and health of transition dairy cows.** Karen M. Lobeck-Luchterhand\*, Paula R. B. Silva, Ricardo C. Chebel, and Marcia I. Endres, *University of Minnesota, St. Paul, MN*.

The objective of this study was to determine whether social ranking was associated with health of transition dairy cows. A total of 953 Jersey cows were used in the analysis. Cows were examined on DIM 1, 4, 7, 10, and 13 for the diagnosis of retained fetal membranes (RP) and metritis. Metritis was defined as cows with watery, pink or brown, and fetid uterine discharge; acute metritis included cows that had a fever ( $>39.5^\circ\text{C}$ ). Cows were classified with subclinical ketosis when BHBA concentration was  $\geq 1200$   $\mu\text{mol/L}$ . Cows were observed once daily for displacement of abomasum (DA) and thrice daily for mastitis. Displacements from the feedbunk were measured continuously for 3 h after fresh feed delivery 4 d/wk during the 4 wk before calving for determination of social rank. A displacement index (DI) was calculated for each cow as the number of displacements as actor (cow initiated the displacement) divided by total displacements as actor or reactor (cow received the displacement). Displacement index was used on a continuous scale (method 1) and also categorized into social rankings (method 2); cows with a DI  $<0.4$  were considered low-ranking, 0.4 to 0.6 were considered middle-ranking, and  $>0.6$  were considered high-ranking. Additionally, the top and bottom 10th percentile for DI were selected to examine cows on the extreme ends of DI (method 3). Cows were categorized into bottom (DI  $<0.19$ ), middle (0.19–0.71), and top 10th percentile (DI  $>0.71$ ). Proc Logistics in SAS was used to investigate the association between social ranking (defined by each of the 3 methods above) and health. Displacement index was associated with RP (AOR = 1.02, 95% CI 1.00–1.03), metritis (0.99, 0.97–1.00) and incidence of mastitis up to 60 DIM (0.99, 0.97–1.00). High-ranking cows were 2.0 times (1.08–3.70) more likely to have RP than low-ranking cows with no differences between low- and middle-ranking cows. The bottom 10th percentile DI cows were 4.7 times (1.6–13.4) more likely to have a case of metritis than the top 10th percentile ranking cows. A cow's ability to displace another cow from the feedbunk during the prepartum period as a determinant of her social rank was not very consistent in predicting the odds of having a transition health disorder.

**Key Words:** social rank, transition cow, displacement index

## Animal Health: Beef cattle health, lameness and mastitis

**504 Evaluating the metagenome of nasal samples from cattle with bovine respiratory disease complex (BRDC).** Tara G. McDanel<sup>\*</sup>, Larry A. Kuehn, and John W. Keele, *US Meat Animal Research Center, Clay Center, NE.*

Bovine respiratory disease complex (BRDC) is a multi-factor disease, and disease incidence may be associated with an animal's commensal microbiota (metagenome). Therefore, evaluation of the animal's resident microbiota in the upper nasal cavity may help us to understand the effect of the metagenome on incidence of BRDC in cattle. Nasal swabs from approximately 600 calves were collected at various time points including preconditioning, weaning, and when the animal enters the feedlot and is diagnosed with BRDC. Samples from healthy cohorts were also collected for each time point evaluated in the feedlot to compare metagenome profiles of healthy and sick animals. Samples from animals diagnosed with BRDC in the feedlot were pooled in groups of 10 based on when the animal was diagnosed with BRDC (one week, 3 weeks, or 4 weeks after weaning). Samples from these same animals were also evaluated at the time points previous to entering the feedlot (preconditioning and weaning) to evaluate changes in the metagenome across time. Additionally, samples from the preconditioning and weaning time points were pooled in groups of 10 based on location origin of the animals before entering the feedlot, as calves came from 4 pasture locations before being weaned and comingled in the feedlot. To evaluate and compare the metagenome of each pooled sample, the variable region (approximately 1,500 bp) along the 16S ribosomal RNA gene was amplified by PCR to include v1-v8. These amplified products were then sequenced using next-generation sequencing (Pacific Biosciences RSII instrument) and sequence reads were analyzed by WebMGA and GreenGenes to identify subfamilies for the bacterial populations present. Overall, *Mannheimia haemolytica* (34–87%) was the predominant bacterial subfamily present in all pools evaluated at the feedlot, compared with preconditioning and weaning time points. Additionally, metagenome profiles differed across animal location origin before entering the feedlot. These results demonstrate a change in the metagenome of the nasal cavity across different time points of production and confirm the likely role of *Mannheimia haemolytica* in BRDC. USDA is an equal opportunity provider and employer.

**Key Words:** bovine respiratory disease complex, metagenome, 16S sequence

**505 Acute and chronic stress models differentially affect the inflammatory and antibody titer responses to respiratory vaccination in naïve beef steers.** Nathan D. May<sup>\*1</sup>, Jeff A. Carroll<sup>2</sup>, Nicole C. Burdick Sanchez<sup>2</sup>, Shelby L. Roberts<sup>1</sup>, Heather D. Hughes<sup>1</sup>, Paul R. Broadway<sup>2</sup>, Kate P. Sharon<sup>3</sup>, Michael A. Ballou<sup>3</sup>, and John T. Richeson<sup>1</sup>, <sup>1</sup>West Texas A&M University, Department of Agricultural Sciences, Canyon, TX, <sup>2</sup>USDA-ARS, Livestock Issues Research Unit, Lubbock, TX, <sup>3</sup>Texas Tech University, Department of Food and Animal Sciences, Lubbock, TX.

The objective of this research was to determine the effect of an acute vs. chronic stress model on serum antibody titer and acute phase responses. Seronegative beef steers ( $n = 32$ ;  $209 \pm 8$  kg) were stratified by BW and assigned randomly to 1 of 3 treatments (1) Chronic stress (CHR), 0.5 mg/kg BW dexamethasone (DEX) administered i.v. at 1000h on d -3 to 0; (2) Acute stress (ACU), 0.5 mg/kg BW DEX administered i.v. at 1000h on d 0 only; or (3) Control (CON), no DEX. On d -4, steers

were fitted with jugular catheters and placed into individual stanchions in an environmentally controlled facility. At 1200h on d 0, steers were administered a modified-live virus respiratory vaccine containing isolates of infectious bovine rhinotracheitis virus (IBRV), bovine viral diarrhea virus (BVDV), bovine respiratory syncytial virus (BRSV) and parainfluenza-3 virus (PI3V). On d 4, cattle were transported (177 km) to an isolated facility and housed in a single pen. Serum was harvested from d 0, 7, 14, 21, 28, 35, 42, and 56 and subsequently used to determine IBRV-, BVDV-, BRSV-, and PI3V-specific antibody titers. Additionally, serum from d -2, 0, 1, 3, 7, and 14 was used to quantify haptoglobin (Hp) and ceruloplasmin (Cp) concentrations. There was a  $\text{trt} \times \text{d}$  interaction ( $P < 0.01$ ) such that CHR steers had a greater ( $P \leq 0.07$ ) BVDV antibody titer from d 14 to 28; whereas, CHR was greater ( $P = 0.06$ ) than ACU on d 56. Moreover, IBRV antibody titers were increased beginning on d 14 for CHR and d 28 for ACU, and remained elevated through d 56 compared with CON ( $P \leq 0.03$ ). Stress treatment altered Hp such that CON exhibited a greater ( $P < 0.01$ ) Hp concentration than CHR but was not different from ACU ( $P = 0.16$ ). On d 3, Cp was greatest for CON, intermediate for ACU, and least for CHR ( $\text{trt} \times \text{d}$ ;  $P \leq 0.01$ ). Results suggest that immunosuppressive conditions in CHR and ACU may have allowed enhanced viral replication from the vaccine, resulting in a greater antibody titer response. Data further indicate that DEX administration blunted the acute phase response and these alterations were particularly evident in the CHR stress model.

**Key Words:** cattle, stress, vaccination

**506 Effect of injectable trace mineral administration on health, performance and vaccine response of newly received beef cattle.** Shelby L. Roberts<sup>\*1</sup>, Nathan D. May<sup>1</sup>, Casey L. Brauer<sup>2</sup>, Wes W. Gentry<sup>2</sup>, Caleb P. Weiss<sup>2</sup>, Jenny S. Jennings<sup>2</sup>, and John T. Richeson<sup>1</sup>, <sup>1</sup>Department of Agricultural Sciences, West Texas A&M University, Canyon, TX, <sup>2</sup>Texas A&M AgriLife Research, Amarillo, TX.

Previous research has established that trace minerals are necessary for optimal animal health and performance. The objective of this study was to evaluate the effect of an injectable trace mineral supplement containing copper, zinc, selenium, and manganese (Multimin 90) on vaccine response, growth performance and morbidity of beef calves upon entry into a feedlot. A total of 128 crossbred bull ( $n = 40$ ) and steer ( $n = 88$ ) calves were utilized. Cattle were stratified by initial BW ( $276 \pm 3$  kg) and gender, then assigned randomly to treatment pens ( $n = 8/\text{treatment}$ ). Treatment protocols were (1) negative control (CON), or (2) Multimin 90 (MM) administered at 1 mL/45.5 kg BW subcutaneously on d 0. Cattle were also administered a pentavalent modified-live respiratory vaccine, anthelmintic, and metaphylaxis with tilmicosin phosphate on d 0. Individual BW data and blood were collected on d 0, 14, 28, and 42. Harvested serum was used to determine bovine viral diarrhea virus (BVDV) type 1a antibody titer as a proxy for vaccine response. Health was monitored daily by trained personnel blinded to treatment pen assignment. Calves were pulled when assigned a clinical illness score of  $\geq 2$ , and considered morbid and administered antimicrobial treatment if rectal temperature was  $\geq 39.7^\circ\text{C}$  or if lung auscultation score was  $\geq 3$  on a 1 to 5 scale. Overall DMI was not different ( $P = 0.82$ ) between CON and MM. Also, no difference in overall ADG ( $P = 0.21$ ) was detected between CON (1.36 kg/d) and MM (1.25 kg/d) steers. The overall morbidity rate observed for this study was low (14%). There was no statistical difference ( $P = 0.71$ ) in morbidity between treatments,

which averaged 15.6 and 12.5%, for CON and MM, respectively. There was a treatment x d effect ( $P = 0.09$ ) for BVDV-specific antibody titer. On d 14, the MM group had a greater ( $P = 0.02$ ) BVDV antibody titer than the CON group. This data suggests that while administration of an injectable trace mineral did not improve performance or morbidity rate when disease incidence was low, the BVDV-specific antibody response to a respiratory vaccine developed faster for Multimin 90 treated animals.

**Key Words:** cattle, trace mineral, vaccine

**507 Effect of different combination viral-bacterial respiratory vaccines on serum leukotoxin antibody, acute phase response, and performance in beef heifer calves.** Heather D. Hughes<sup>\*1</sup>, Sjoert Zuidhof<sup>2</sup>, Shelby L. Roberts<sup>1</sup>, Joelle L. Pillen<sup>1</sup>, Garrett D. Bigham<sup>1</sup>, and John T. Richeson<sup>1</sup>, <sup>1</sup>Department of Agricultural Sciences, West Texas A&M University, Canyon, TX, <sup>2</sup>Boehringer Ingelheim Vetmedica, St. Joseph, MO.

Vaccination of newly received cattle against respiratory pathogens is a common practice in the stocker or feedlot setting; however, this practice may induce an acute inflammatory or febrile response that could affect clinical presentation or transiently reduce performance. Our objective was to determine if different combination viral-bacterial respiratory vaccines affect the leukotoxin (LKT) antibody concentration, acute phase response, rectal temperature (RT), or gain performance. A total of 30 clinically healthy beef heifer calves (BW = 222.3 ± 27.1 kg) were stratified by pre-trial serum antibody against *Mannheimia haemolytica* (MH) whole cell wall, then assigned randomly to 1 of 3 vaccine treatment regimens consisting of (1) Pyramid 5 + Presponse SQ (P5PS; n = 10), (2) Bovi-shield Gold One Shot (BGOS; n = 10), or (3) unvaccinated control (CON; n = 10). Heifers were housed in a single pen, and blood, BW and RT were collected on d 0, 4, 7, 14, 28, 42 and 56. A treatment x d interaction was observed for MH-specific LKT ( $P < 0.001$ ). Cattle administered either of the vaccines had greater LKT antibody concentrations than CON on d 14 and 28 ( $P \leq 0.04$ ), whereas BGOS was greater than CON on d 7 ( $P = 0.03$ ) but did not differ from P5PS ( $P = 0.49$ ). No differences were detected for RT ( $P = 0.85$ ) or ADG ( $P \geq 0.33$ ), which averaged 1.17, 1.11 and 1.23 kg/d for P5PS, BGOS, and CON, respectively from d 0 to 56. Respiratory vaccination affected serum haptoglobin (Hp) concentration; P5PS exhibited greater Hp concentrations than CON ( $P = 0.01$ ) but was not different from BGOS ( $P = 0.53$ ). No difference in serum ceruloplasmin was observed ( $P = 0.88$ ). Results indicate that either vaccine produced a greater LKT antibody and Hp response compared with CON, but RT and performance were not affected. Respiratory vaccines may have slight inflammatory effects when administered to clinically healthy cattle, yet further research is warranted to elucidate vaccine-induced inflammation in highly stressed cattle.

**Key Words:** acute phase response, cattle, vaccine

**508 Probiotic supplementation improves performance, neutrophil function, and antibody responses of post-weaned Holstein heifers during the commingling phase.** Caleigh E. Payne<sup>\*</sup>, Luis G. D. Mendonça, Lucas D. S. Rocha, Sophia C. Trombetta, Suzy Q. Fowler, Juan C. Gordienko, Sonia J. Moisés, and Lindsey E. Hulbert, Kansas State University, Manhattan, KS.

The risk of disease increases for post-weaned calves when they are transitioned from individual housing to group housing (commingling). Therefore, this study was conducted to determine if probiotic supplementation of mannan-oligosaccharide (MOS) and b-glucan (BG) would

assist calves in the transition from individual hutches to groups of 3. Feed intake, body weight gain, in vivo adaptive immune responses, and ex vivo neutrophil responses were measured from 60, weaned Holstein heifer calves (age 52 ± 4.0 SD d; 83 ± 14.7 SD kg). One week before commingling (-1 wk), calves were randomly assigned to either a daily bolus dose of oral prebiotics (3 g; 10% MOS, 18% BG) dissolved in 15 mL of molasses or control (15 mL molasses only) for 7 weeks. Daily DMI was collected and calves were weighed weekly. Whole blood was collected via jugular venipuncture on wk -1, 1, 2, and 6 relative to commingling. In addition, all calves were administered an innocuous protein injection, ovalbumin (OVA; subQ; 0.5 mg/mL), at commingling and 4 weeks after commingling. All blood samples were measured via flow-cytometry for peripheral neutrophil phagocytosis (PG) and oxidative burst (OB) responses to heat-killed *E. coli* (8739). Plasma OVA-specific IgG and IgA were measured at 2 weeks after each OVA injection. Two weeks after commingling, prebiotic-treated calves had neutrophils with greater OB intensity than control calves ( $P = 0.005$ ). Prebiotic-calves also had greater primary IgA ( $P < 0.01$ ) and IgG ( $P = 0.04$ ) responses to OVA than control calves, as well as greater secondary IgA response ( $P < 0.01$ ) than control calves. One week after commingling, prebiotic-calves had greater ADG (1.07 vs. 0.90 ± 0.059 kg/d;  $P = 0.04$ ) and as well as lower F:G (2.61 vs. 3.60 ± 0.204;  $P = 0.001$ ) than control calves. Probiotic supplements enhanced innate and adaptive immune measures and performance during the commingling phase. This supplementation may help reduce the risk of disease and improve vaccination response during commingling in post-weaned dairy heifers.

**Key Words:** probiotic, immunology, bovine

**509 Shotgun metagenomic analysis of bovine digital dermatitis.** Martin Zinicola<sup>\*</sup>, Hazel Higgins, Svetlana Lima, Vinicius Machado, Charles Guard, and Rodrigo Bicalho, Cornell University, Ithaca, NY.

Metagenomic methods amplifying 16S ribosomal RNA genes have been used to describe the microbial diversity of healthy skin (HS) and lesion stages of bovine digital dermatitis (DD) and to detect critical pathogens involved with disease pathogenesis. In this study, we characterized the microbiome and for the first time, the composition of functional genes of HS, active (ADD) and inactive (IDD) lesion stages using a whole-genome shotgun approach. A total of 16 biopsy samples (HS, n = 8; ADD, n = 4; IDD, n = 4) were collected from Holstein dairy cows housed in one dairy farm. DNA was extracted and the microbiome was determined by shotgun techniques using Illumina MiSeq platform. Metagenomic sequences were annotated using MG-RAST pipeline. Six phyla were identified as the most abundant. *Chordata*, *Firmicutes* and *Actinobacteria* were the predominant phyla in the microbiome of HS, while *Spirochetes*, *Bacteroidetes* and *Proteobacteria* were highly abundant in ADD and IDD. *T. denticola-like*, *T. vincentii-like* and *T. phagedenis-like* constituted the most abundant species in ADD and IDD. Recruitment plots comparing sequences from HS, ADD and IDD samples to the genomes of specific *Treponema* spp., supported the presence of *T. denticola* and *T. vincentii* in ADD and IDD. Comparison of the functional composition of HS to ADD and IDD identified a significant difference ( $P < 0.05$ ) in genes associated with motility/chemotaxis and iron acquisition/metabolism. We also provide evidence that the microbiome of ADD and IDD compared with that of HS had significantly higher ( $P < 0.05$ ) abundance of genes associated with resistance to copper and zinc, which are commonly used in footbaths to prevent and control DD. In conclusion, the results from this study provide new insights into the HS, ADD and IDD microbiomes, improve our understanding of the disease pathogenesis and generate unprecedented knowledge regarding



the functional genetic composition of the DD microbiome. Additionally, an increase in the abundance of copper and zinc resistance genes has been reported, suggesting that further research is necessary to optimize the foot bathing technique for the control and treatment of DD.

**Key Words:** bovine digital dermatitis, dairy cow, metagenomic

### 510 Comparison of milking and lying behavior between lame and sound cows on dairy farms with automated milking systems.

Meagan T. M. King<sup>\*1</sup>, Ed A. Pajor<sup>2</sup>, Stephen J. Leblanc<sup>3</sup>, and Trevor J. DeVries<sup>1</sup>, <sup>1</sup>*Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada*, <sup>2</sup>*Faculty of Veterinary Medicine, University of Calgary, Calgary, AB, Canada*, <sup>3</sup>*Department of Population Medicine, Ontario Veterinary College, University of Guelph, Guelph, ON, Canada*.

To develop better ways to use behavioral data to detect lame cows, comparisons of milking visit behavior, milk yield, and lying behavior were made between lame and sound cows on automated milking system (AMS) farms. Data were collected for 30 cows, over a 6-d period, from each of 26 AMS farms in Eastern Ontario, Canada. Cows were gait scored using a 5-point numerical rating system (1 = sound to 5 = lame). Cows with gait scores < 3 were classified as sound (n = 527) while those with gait scores ≥ 3 were classified as lame (n = 245). Milking visit behavior was extracted from the AMS computer at each farm, while lying behavior was measured continuously using electronic data loggers. Body condition and hygiene were also scored, and were compiled with other cow-level variables such as parity and DIM. Data were summarized across the 6-d observation period/cow and analyzed in multivariable general linear mixed models. When controlling for DIM, milking frequency (#/d; mean ± SE) was lower ( $P < 0.001$ ) for lame cows ( $2.8 \pm 0.1$ ) compared with sound cows ( $3.1 \pm 0.1$ ). Milk yield was lower ( $P = 0.05$ ) for lame cows ( $32.0 \pm 0.9$  kg/d) compared with sound cows ( $33.4 \pm 0.8$  kg/d), while accounting for parity, DIM, and body condition. Compared with sound cows, lame cows had fewer AMS refusals/d (1.0 vs. 1.8; SE = 0.2;  $P < 0.001$ ). Lameness status did not affect the frequency of AMS failures/d ( $0.1 \pm 0.01$ ). Overall, lame cows made fewer visits to the AMS than sound cows (4.0 vs. 5.0 visits/d; SE = 0.3;  $P < 0.001$ ). When accounting for parity and DIM, lying time was greater ( $P = 0.002$ ) for lame cows ( $728.2 \pm 13.7$  min/d) compared with sound cows ( $693.8 \pm 12.0$  min/d). These results demonstrate the differences in behavior and resulting productivity between lame and sound cows, indicating the potential use of behavioral indicators to identify lame cows on AMS farms.

**Key Words:** automatic milking, dairy cow behavior, lameness

### 511 Comparing the prevalence of hoof lesions in dairy cattle classified as high, average or low antibody and cell-mediated immune responders.

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Several studies have shown cattle classified as high immune responders have lower incidence of disease, however the occurrence of hoof lesions has yet to be evaluated in dairy cattle classified for immune response. Therefore the objective of this study was to compare the prevalence of hoof lesions in dairy cattle classified as high, average or low antibody (AMIR) and cell-mediated immune responders (CMIR).

Cattle (n = 190) from a commercial dairy farm in Ontario were evaluated for immune response (IR) using a patented protocol that captures both AMIR and CMIR. They were classified as high, average and low responders based on Estimated Breeding Values for AMIR and CMIR. Hoof health data was collected by the farm's hoof trimmer using Hoof Supervisor software. Only the first trim date for each animal was included in this data set, and multiple lesions per cow were considered. All lesions were analyzed both as individual lesion types and grouped into infectious (digital dermatitis) and horn (sole hemorrhage, sole ulcer, toe ulcer, interdigital hyperplasia and white line). The trimmer scored each lesion for severity as: 1 = least, 2 = middle, 3 = most. Data was analyzed using a SAS mixed model, which included the effects of parity and IR category (high, average and low). Data is presented as number of cases per cows within IR category, and significance is reported at  $P < 0.05$  with trends at  $P < 0.10$ . Results showed that high antibody responders had significantly less digital dermatitis (20%) compared with average (36%) and similarly significantly less severe digital dermatitis (3%) compared with low (16%). Conversely, high antibody responders (50%) had significantly more non-infectious horn lesions compared with average (25%) and low (21%). High cell-mediated responders had a trend toward less interdigital hyperplasia (0%) compared with average (8%). Therefore, not only do cows classified as high immune responders have lower disease incidence, but this study suggests they also have lower prevalence of infectious hoof lesions, a critical problem facing dairy producers.

**Key Words:** hoof health, immune response, dairy cattle

### 512 Calf macrophages exhibit a robust response to LPS which is not affected three weeks after an early life challenge with LPS in vivo.

Filiz T. Korkmaz<sup>\*</sup>, Aimee L. Benjamin, and David E. Kerr, *University of Vermont, Burlington, VT*.

Genetic and epigenetic factors may contribute to animal-to-animal variation in innate immune response to infection with epigenetic effects being imposed by differences in early life environment. To evaluate lasting effects of a severe early-life infection on innate immune response capability we exposed 6 pairs of Holstein bull calves, at 8d of age, to either a 0.5 µg/kg intravenous dose of LPS or saline. Three weeks later we established cultures of monocyte-derived-macrophages (MDMs) from 100 mL blood samples by culturing monocytes for 8 d. The cells were then challenged for 24h with 100 ng/mL of LPS. The LPS treated calves demonstrated clinical signs of endotoxin challenge and markedly elevated ( $P < 0.05$ ) plasma levels of TNF-α ( $3.2 \pm 1.0$  ng/mL) and IL-6 ( $14.4 \pm 2.8$  ng/mL) at 2h post-challenge. Although there was no significant difference ( $P > 0.05$ ) between cells from LPS or saline treated calves, the MDMs produced considerable quantities of IL6 and TNF-α in response to LPS, which averaged  $2.3 \pm 0.4$  ng/mL and  $190 \pm 81$  pg/mL, respectively. Likewise, there was a large LPS-mediated induction of IL8 and IL1-β ( $88.1 \pm 37.6$  and  $1954.9 \pm 1166.5$  fold change, respectively). Readily detectable expression of TLR4 and CD14 was measured with minor induction due to LPS. No differences ( $P > 0.05$ ) were measured between groups in LPS-induced gene expression. Relatively large inter-animal variation was found in most parameters with a significant correlation between expression of IL1-β and IL8 ( $R^2 = 0.68$ ,  $P < 0.01$ ) and TLR4 and CD14 ( $R^2 = 0.57$ ,  $P < 0.01$ ). Dermal fibroblasts were also isolated from 10 calves to determine how their response to LPS compared with that of the MDMs. Fibroblast TLR4 expression was less than MDM TLR4 expression and this was reflected in lower LPS-induced induction of IL8 and IL-6 gene expression. In conclusion, LPS response in MDMs and fibroblasts is variable between animals, lower in fibroblasts, and is not affected in MDMs 3-wks after a single

exposure to LPS at 8d of age. Future studies are needed to determine genetic and epigenetic components which may be influenced by early life environment.

**Key Words:** innate immunity, epigenetic

**513 Dysbiosis of the fecal microbiota in cattle infected with *Mycobacterium avium* ssp. *paratuberculosis*.** Marie-Eve Fecteau, Raymond Sweeney, Sanjay Kumar, Nagaraju Indugu, Bonnie Vecchiarelli, Bhima Bhukya, and Dipti Pitta\*, *Department of Clinical Studies, School of Veterinary Medicine, New Bolton Center, University of Pennsylvania, Kennett Square, PA.*

Johne's disease (JD) is a chronic gastrointestinal infection of cattle caused by *Mycobacterium avium* ssp. *paratuberculosis* (MAP). In the US, more than 68% of dairy herds are infected with MAP, leading to considerable financial losses for producers. We hypothesized that cattle naturally infected with MAP have a reduction in gastrointestinal microbial biodiversity and gastrointestinal dysbiosis may play a role in the pathogenesis of JD. In the present study, fecal samples from 20 naturally infected (positive group); 25 JD-negative herd mates (exposed group); and 25 JD-negative cows from a MAP-free herd (negative group) were obtained from New Bolton Center, JD repository. All fecal samples were processed for genomic DNA and amplified for the V1-V2 regions of the 16S rDNA gene. Sequencing was performed on a 454 Roche Platform and sequences were analyzed using QIIME. Approximately 252,380 reads were analyzed from 70 bacterial communities with an average of 2,843 reads per sample. Approximately 34,606 operational taxonomic units (OTUs) were produced by clustering at 97% sequence similarity. Weighted and unweighted Unifrac distances by principal coordinate analysis showed a substantial difference ( $P < 0.001$ ; Permanova test) in the bacterial community composition of the positive group versus the exposed and negative groups. Taxonomic assignment of the OTUs identified a total of 18 bacterial phyla across all samples. Across negative and exposed groups, *Bacteroidetes* and *Firmicutes* constituted more than 95% of the total bacterial population. However, in the positive group, lineages of *Actinobacteria* and *Proteobacteria* increased and those of *Bacteroidetes* and *Firmicutes* decreased ( $P < 0.001$ ). *Actinobacteria* was highly abundant (30% of the total bacteria) in the positive group compared with <0.1% in exposed and negative groups. Therefore, it is apparent that bacterial communities in exposed and negative groups were homologous whereas significant variation in positive samples was observed. Bacterial diversity in positive group was compromised compared with negative and exposed groups.

**Key Words:** dysbiosis, fecal, Johne's disease

**514 Use of a novel adjuvant to enhance the protective effect of a commercial vaccine against *Staphylococcus aureus* mastitis in dairy heifers.** Charles Hall, Stephen Nickerson\*, David Hurley, Lane Ely, and Felicia Kautz, *University of Georgia, Athens, GA.*

Use of a novel adjuvant (Immunoboost) to enhance antibody titers in response to a commercial vaccine (Lysigin) against *Staphylococcus aureus* mastitis in dairy heifers was evaluated in a 2-phase trial. Heifers 5 to 12 mo of age were used in both study phases and blood samples were collected weekly for processing (ELISA) to determine anti-*S. aureus* antibody levels; baseline titers before each phase did not exceed 1:1600. In phase 1, hyper-immunization with Lysigin to enhance the level and duration of titers did not result in titers that consistently exceeded conventional immunization. In phase 2, anti-*S. aureus* titers

in heifers immunized with Lysigin + 2 mL Immunoboost tended to be elevated ( $P = 0.10$ ) over those of heifers immunized conventionally with Lysigin alone by d 7 after initial immunization, a trend that continued through d 14. By d 21, titers in the Immunoboost group were elevated ( $P = 0.05$ ) over conventional vaccinates and remained significantly elevated through d 35, returning to baseline by d 42. After receiving booster injections on d 42, the Immunoboost group experienced an increase ( $P = 0.05$ ) in titers over conventional vaccinates on d 49 of the trial, and titers remained significantly elevated through d 63. Titers in the Immunoboost group remained elevated over conventional vaccinates through d 84 of the trial, but the difference was not significant, and titers in both groups were approaching baseline values. Findings suggest that Immunoboost is capable of enhancing the antistaphylococcal titer response to the commercial vaccine Lysigin, albeit in the short term. These studies indicate that continued study of using immunization to control *S. aureus* mastitis in dairy heifers is justified.

**Key Words:** antibodies, heifer, mastitis

**515 The efficacy of PlyC endolysin as an alternative therapy for *Streptococcus uberis* mastitis in vitro.** Sara Linden<sup>1</sup>, Parimala Sharma<sup>1,2</sup>, Kasey M. Moyes\*<sup>2</sup>, and Daniel C. Nelson<sup>1,3</sup>, <sup>1</sup>*University of Maryland, College Park, MD*, <sup>2</sup>*Institute for Bioscience and Biotechnology Research, Rockville, MD*, <sup>3</sup>*Department of Veterinary Medicine, College Park, MD.*

The bacteriophage endolysin, PlyC, displays antimicrobial activity against select streptococcal species and may be a promising new therapy for *S. uberis*-associated mastitis. This study investigated the antimicrobial activity of PlyC against a broad spectrum of *S. uberis* isolates, established an in vitro dose response, measured ex vivo binding and antimicrobial efficacy in *S. uberis* infected milk, and assessed safety and response to antibodies. For antimicrobial activity, 2 µg/mL of PlyC was incubated with 7 different strains of *S. uberis* isolated from cows with mastitis as well as ATCC 27958 and BAA-854 (strain 0140J). For dose-response studies, PlyC (0–16 µg/mL) were incubated with 27958. The linear proportion of the initial lytic velocity was measured using a spectrophotometric turbidity reduction assay for both antimicrobial activity and dose-response assays. To demonstrate streptococcal-specific binding in raw cow's milk, 20 µg of AlexaFluor-555-labeled PlyC was incubated with milk containing BAA-854 and binding was visualized via fluorescent microscopy. Finally, polyclonal antibodies against PlyC (pre-immune sera and hyper immune sera [titer ranging from 1:10–1:10,000]) were mixed with 32 µg/mL of PlyC and activity was measured using the spectrophotometric turbidity reduction assay. All data were statistically analyzed using the Student's *t*-test. Our results showed that PlyC possessed potent lytic activity ( $P < 0.01$ ) against all strains of *S. uberis* tested. The dose-response profile demonstrated lytic activity ( $P < 0.01$ ) at concentrations ranging from 1 to 16 µg/mL. Microscopy results indicated that PlyC specifically labeled cell walls of BAA-854 in raw milk ( $P < 0.01$ ). In addition, PlyC retains full lytic activity ( $P < 0.01$ ) against 27958 in the presence of high titer anti-PlyC antibodies, suggesting that even if antibodies are present, they are non-neutralizing. Taken together, the results indicated PlyC has the potential to be used as a novel therapeutic against *S. uberis*-associated bovine mastitis via its specificity against several strains of *S. uberis*, the components in milk do not interfere with activity, and it is not inhibited in the presence of antibodies.

**Key Words:** mastitis, PlyC, treatment

## Beef Cattle Nutrition Symposium: Feeding Holstein steers

**516 Neonatal and young (<205 kg) feeding programs in calf-fed Holsteins.** Luis O. Burciaga-Robles\*, *Feedlot Health Management Services, Okotoks, Alberta, Canada.*

The limited supply of beef cattle is one of the greatest challenges facing the beef industry globally and is a factor reflected in the annual decline in fed cattle harvest. Calf-fed Holsteins (CFH) represent an opportunity to fill the void in beef cattle supply; however, because of the differences that exist compared with beef cattle, a science based approach is required to optimize production and profitability. The CFH often is overlooked in academic research. The research based on dairy heifer calves or beef cattle is often extrapolated to CFH. The objective of this presentation is to discuss the implications of Neonatal Nutrition Programs (NNP) on health and lifetime performance in CFH. The success of NNP (birth to <205 kg) should take into consideration the environment, genetics, immune status, and inherent variation that exist in CFH populations. During NNP, the greatest contributor to the feed-only cost of gain is the milk feeding program. More research is required to optimize growth and production goals without compromising the well-being of the calf. Understanding how NNP affects the immune status/response of the calf and implications on lifetime health and growth is pivotal. Nutrigenomics is an avenue of research that could expand the understanding of how clinical or subclinical disease change nutrient requirements and potentially allowing for development of mitigation strategies. Another area of research required in the CFH encompasses determining the effects of lifetime feeding of CFH through understanding the potential interactions between NNP at the calf ranch and the eventual feeding program at the feedlot. Research regarding grain type, grain processing, by-product use, protein sources and concentrations, and roughage level at the calf ranch is critical. Understanding the biology of the CFH across all growth stages is needed so that economic models can be developed. Understanding the economic contribution of different aspects of biology allows for sensitivity determination of key economic variables that should help identify research priorities. A multidisciplinary research approach to understanding the CFH is required to meet the increasing demand for animal protein without compromising the overall well-being and sustainability of this industry.

**Key Words:** nutrition, calf-fed, feedlot

**517 Morphological, microbiological, and biochemical development of ruminant gastrointestinal tract.** Carl J. Yeoman and Glenn C. Duff\*, *Department of Animal and Range Sciences, Montana State University, Bozeman, MT.*

It is well recognized that morphological development of the rumen is dependent on diet and microbiota. For example, dietary concentrates stimulate the microbial production of VFA, including butyrate. Butyrate is the primary form of energy to the developing rumen wall and promotes development of ruminal papillae. Microbial colonization of the ruminant gut is complex and begins at birth. Microbes found on the teat and in the colostrum rapidly replace early colonizers from the vagina and a dynamic succession takes place until a mature climax community forms between 180 to 360 d of age. By using direct-fed microbials, including live cultures of *Lactobacillus acidophilus*, *L. bulgaricus*, *Streptococcus thermophilus*, and *Aspergillus oryzae* in place of colostrum, we found no loss ( $P > 0.05$ ) in ADG or increase ( $P > 0.05$ ) in intake in neonatal lambs. In Holstein calves supplementation of *L. acidophilus* and *Propionibacterium freudenreichii* resulted in increased ( $P < 0.05$ )

gastrointestinal villus height, crypt depth, and total height (crypt plus villus) before weaning with average ruminal papillae width greater ( $P < 0.01$ ) in calves fed the live cultures versus control calves after weaning. Genus populations of *Bacteroides*, *Roseburia*, and *Eubacterium* were greater ( $P < 0.05$ ) in fecal samples collected in non-scouring calves versus scouring calves. Using serum  $\beta$ -hydroxybutyrate as a marker in serum for ruminal development, milk feeding regimen (bottle versus trough) and housing (group versus individual) did not appear ( $P > 0.10$ ) to affect ruminal development. Alterations of microbiota in the rumen to hasten ruminal development deserves attention to potentially improve animal health and performance.

**Key Words:** ruminant, gastrointestinal, development

**518 Genetic, epigenetic, and management factors contribute to the risk of morbidity and mortality of Holstein feeder calves.** Michael A. Ballou\*<sup>1</sup>, David E. Kerr<sup>2</sup>, Kate P. Sharon<sup>1</sup>, and Aimee L. Benjamin<sup>2</sup>, <sup>1</sup>*Department of Animal and Food Sciences, Texas Tech University, Lubbock, TX,* <sup>2</sup>*Department of Animal Science, The University of Vermont, Burlington, VT.*

Holstein steers contribute significantly to US beef production. Healthy feeder calves are more efficient and produce carcasses with greater value. Holsteins have the genetic capability to produce carcasses with comparable quality to many common beef breeds, but there are unique challenges to feeding Holsteins. Genetic differences between Holsteins and common beef breeds influence health. Additionally, early life management and other environmental factors can have long-term effects on the health and productivity of Holstein feeder calves. Holstein heifers had greater inflammatory responses than Angus heifers when evaluated in both ex vivo fibroblast and in vivo lipopolysaccharide challenge models. In both models, the Holstein heifers produced approximately 3 times more inflammatory mediators than the Angus heifers. The greater inflammatory capacity is likely due to a combination of genetic and epigenetic factors. In fact, pre-weaning nutrition influenced the risk of morbidity and mortality to a combined bovine herpesvirus-1 and *Mannheimia haemolytica* respiratory disease challenge in Holstein calves a month after weaning. Calves fed a restricted quantity of milk replacer, a common management strategy, had greater ( $P \leq 0.05$ ) mortality (4/15) compared with calves fed 2.5 times more milk replacer (0/15). In agreement, the restricted fed calves had greater measures of systemic inflammation, increased peripheral blood neutrophil:lymphocyte ratio and plasma haptoglobin concentrations. In contrast, many management strategies that are common among Holstein calves improve their health in a feed yard. These management strategies include: vaccination (viral and bacterial antigens), weaning, castration, dehorning, and feed bunk and water trough training. Implementing these management strategies to calves will reduce stress and its negative effects on immunity during arrival at the feed yard. Taken together, management decisions of Holsteins during the calf phase are important as they can have either positive or negative effects on future health.

**Key Words:** beef, calf, health

**519 Nutrition and management of calf-fed Holstein steers.** Richard Zinn\*, *University of California, Davis, CA.*

This presentation provides a brief review of feedlot nutrition and management considerations for Holstein steers. Calf-fed Holstein steers



typically enter the commercial feedlot at 100 to 120 d of age, with a full purchase weight of approximately  $137 \pm 16$  kg. On a shrunk-to-shrunk basis (full purchase and final weights reduced 4%), harvest weight, ADG, and days on feed of implanted calf-fed Holstein steers average  $588 \pm 16$  kg,  $1.31 \pm 16$  kg, and  $349 \pm 10$  d, respectively. Rate of gain and gain efficiency are markedly influenced by harvest weight and placement month. Due to high summer temperatures, late summer and fall close-outs are least efficient for feedlots located in the desert southwest. Calf density should be regulated to achieve an average surface moisture level of roughly 20%. Because water consumption is greater (20 to 30%) in Holsteins than conventional beef breeds, they require more pen and shade space. In commercial practice, calf-fed Holsteins are fed a single growing-finishing diet throughout the duration of the feedlot period. A characteristic diet may contain 12–13% crude protein (urea the sole source of supplemental N), 0.70–0.80% calcium, 0.35–0.40% phosphorus, 0.20 to 0.28% magnesium, 0.3 to 0.4% trace mineral salt, and 8% forage NDF. Due to very high growth potential during the initial 112 d on feed, this diet will not meet the calf's metabolizable amino acid requirements, resulting in a predictable decrease (15 to 20%) in energetic efficiency during that phase. The rather high incidence of liver abscess frequently observed among lots of calf-fed Holsteins may warrant continued low-level antibiotic supplementation. Good feedbank management is particularly important with Holsteins. The high energy content of the diet fed over a protracted period of time poses added threat to the possibility of acidosis.

**Key Words:** Holstein, feedlot, nutrition

**520 Feedlot: Liver abscesses.** T. G. Nagaraja\*, *Kansas State University, Manhattan, KS.*

Abscesses in the liver of cattle are focal infections resulting from entry and establishment of pyogenic bacteria. There are several routes by which bacteria reach the liver, but entry via portal blood is by far the most frequent, because it drains the gastrointestinal tract, a source of bacteria. Liver abscesses can occur at all ages and in all types of cattle, but are most common in feedlot cattle fed high-grain finishing diets. The generally accepted pathogenesis of liver abscess includes chronic ruminal acidosis that leads to rumenitis, which then allows bacteria, primarily *Fusobacterium necrophorum*, to cross the epithelial barrier to enter the portal circulation. Bacteria from the portal circulation are filtered by the liver, leading to growth, production of virulence factors, particularly leukotoxin, damage to hepatic parenchyma, and abscess formation. *Trueperella* (formerly *Arcanobacterium*) *pyogenes* is the second most common bacterial species isolated from liver abscesses. Another bacterial species that has been recently isolated from liver abscesses is *Salmonella enterica*. Liver abscesses in cattle are of significant economic concern to the feedlot industry because of liver condemnation, reduction in performance and carcass yield. Liver abscess data collected from several packing plants have indicated a higher incidence of liver abscesses, particularly of the A+ score (most severe), in Holstein steers than the traditional beef cattle. Holstein steers raised for beef production

are different from traditional beef breeds. Among the differences include more digestive and metabolic disorders and death losses in Holstein steers compared with beef breeds. The generally accepted reason for the increased digestive disorders, liver abscesses and death loss is the greater number of days Holsteins are on feed. However, it is not known whether the bacterial flora of liver abscesses of Holstein steers is in any way different from abscesses of the traditional beef cattle. In a recent study, liver abscesses in Holstein steers had a more diverse bacterial flora compared with traditional beef cattle. However, the prevalence of the major bacterial species, *F. necrophorum*, and *T. pyogenes* were similar between the 2 cattle types.

**Key Words:** liver abscess, Holstein steers, etiology

**521 Methods of estimating empty body composition, energy retention, and grading characteristics of calf-fed Holstein steers.**

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A serial harvest trial investigating growth characteristics of calf-fed Holstein steers ( $n = 115$ ; initial BW =  $449.2 \pm 19.9$  kg) was conducted in 28 d intervals starting at 226 d on feed (DOF) and ending at 534 DOF. Five steers were slaughtered on d 226 and utilized as a point for growth modeling. Remaining cattle (110 steers) were randomly allocated to harvest endpoint (10 cattle per slaughter day; 254, 282, 310, 338, 366, 394, 422, 450, 478, 506, and 534 DOF) with one-half of each group receiving zilpaterol hydrochloride (ZH) for 20 d followed by a 3 d withdrawal. Shrunk BW (SBW, kg) and hot carcass weight (HCW, kg) were utilized to predict empty body weight, kg (EBW). Feeding performance {dry matter intake (DMI, kg), net energy for gain, (NEG, Mcal/kg of diet dry matter), and ZH supplementation} variables were utilized to estimate EBW and empty body fat (EBF, %). For prediction of HCW, a 2 variable equation was developed with an  $R^2$  of 0.946 ( $P < 0.01$ ) and root mean square error (RMSE) of 20.13 { $HCW = -41.44 + (0.6637 \times SBW) + (12.974 \times ZH)$ ; 1 = 20-d supplementation, 0 = 0-d supplementation}. For prediction of EBW, an equation with an  $R^2$  of 0.942 ( $P < 0.01$ ) and RMSE of 29.9 was developed { $EBW = 57.6232 + (1.39280 \times HCW) - (11.0994 \times ZH)$ }. Utilizing feeding performance data, a model with an  $R^2$  of 0.919 ( $P < 0.01$ ) and RMSE of 35.2 was developed for prediction of EBW { $EBW = -1547.34 + (1.19 \times DOF) + (21.13 \times DMI) + (1005.73 \times NEG) + (18.87 \times ZH)$ }. To predict EBF using feeding performance variables, a 4 variable equation was developed with an  $R^2$  of 0.621 ( $P < 0.01$ ) and RMSE of 2.9 { $EBF = 0.7769 + (0.0003 \times DOF) + (0.0046 \times DMI) - (0.4346 \times NEG) - (0.0167 \times ZH)$ }. Several variables available to calf-fed Holstein producers may be implicated for use in predictive models related to finishing performance and composition of gain for calf-fed Holstein steers.

**Key Words:** Holstein, body composition, carcass characteristics

## Breeding and Genetics: Feed efficiency and methods

**522 Definition and implementation of a breeding value for feed efficiency.** Jennie E. Pryce<sup>\*1,2</sup>, Oscar Gonzalez-Recio<sup>1</sup>, Gert Nieuwhof<sup>1</sup>, Bill Wales<sup>1</sup>, Michael P. Coffey<sup>3</sup>, Ben J. Hayes<sup>1,2</sup>, and Michael E. Goddard<sup>1,4</sup>, <sup>1</sup>*Department of Economic Development, Jobs, Transport and Resources, Bundoora, VIC, Australia*, <sup>2</sup>*La Trobe University, Bundoora, VIC, Australia*, <sup>3</sup>*SRUC, Edinburgh, Midlothian, UK*, <sup>4</sup>*The University of Melbourne, Melbourne, VIC, Australia*.

The objective was to describe how a new estimated breeding value (EBV) for feed efficiency is calculated. From April 2015, the Australian Dairy Herd Improvement Scheme (ADHIS) has published a new breeding value, called feed saved, that is the amount of feed saved through improved metabolic efficiency and reduced maintenance requirements. The breeding value includes a genomic component for residual feed intake (RFI) combined with either a genomic or pedigree EBV for body weight (BW) predicted using conformation traits. The RFI component of the feed saved EBV has 2 parts: Australian calf RFI and Australian lactating cow RFI. Genomic breeding values for RFI were estimated in a multi-trait analysis (3 traits) that included including Australian growing heifer and lactating cow RFI in addition to overseas (UK and Dutch) lactating cow RFI. The reference population included 50k single nucleotide polymorphism (SNP) genotypes and phenotypes for 234 Australian lactating cows and 843 heifers (approximately 6 mo old) and 958 UK and Dutch lactating cows. In all cases, the RFI phenotypes were trait deviations that were calculated by correcting dry matter intake for bodyweight, growth and yield (in the case of lactating cows). Effects for each SNP were calculated from the output of genomic best linear unbiased prediction and used to predict breeding values of 4,416 sires that were genotyped, but did not have RFI phenotypes themselves. These bulls already had BW breeding values calculated from type traits, from which maintenance requirements in kg of feed per year were inferred. Finally, RFI and the feed required for maintenance (through BW) were combined to construct a feed saved breeding value. Animals with EBVs that are one standard deviation above the mean are predicted to eat 65 kg dry matter less per year at the same level of milk production. The mean reliability associated with the feed saved breeding value was 0.37 in the data set of 4416 genotyped Holstein sires. From April 2015, feed saved has also been included as part of the Australian national selection index published by ADHIS.

**Key Words:** feed efficiency, residual feed intake, genomic

**523 Indices to improve feed efficiency.** Kelli J. Retallick<sup>\*1</sup>, Jennifer M. Bormann<sup>1</sup>, Robert L. Weaber<sup>1</sup>, Michael D. MacNeil<sup>3</sup>, Heather L. Bradford<sup>1</sup>, Harvey C. Freetly<sup>2</sup>, Daniel W. Moser<sup>1</sup>, Warren M. Snelling<sup>2</sup>, Richard M. Thallman<sup>2</sup>, and Larry A. Kuehn<sup>2</sup>, <sup>1</sup>*Kansas State University, Manhattan, KS*, <sup>2</sup>*USDA-ARS Meat Animal Research Center, Clay Center, NE*, <sup>3</sup>*Delta G, Miles City, MT*.

Evaluating feed efficiency of beef cattle has evolved from relying on single trait selection for increased gain to extensive use of multiple trait selection combining measures of gain and intake. Postweaning gain (difference between 365-d and 205-d age adjusted weights) is analyzed as part of National Cattle Evaluation programs and could be used in an index with shorter intake tests (currently >70d to accurately measure gain) to select animals for improved feed efficiency. Under this paradigm, producers could decrease costs per animal and increase genetic change by testing a greater number of animals per year. Objectives of this study were to compare 2 alternative indices for feed efficiency

and to quantify the genetic response to selection for feed efficiency combining an intake test with PWG data. Strong genetic correlation estimates for steers and heifers between average daily gain (ADG) and postweaning gain (PWG) (0.81, 0.65) suggest PWG is a reliable proxy for ADG. On-test ADFI, on-test average daily gain ADG, and postweaning daily gain (PWG) records on 5,606 growing steers and heifers were obtained from the US Meat Animal Research Center in Clay Center, NE. Genetic (co)variances and EBV were estimated using a 6 trait animal mixed model with ADG, ADFI, and PWG as dependent variables for both steers and heifers. Indices combining EBVs for ADFI and ADG and for ADFI and PWG were evaluated. For each index, the weighting of gain was arbitrarily set to 1.0 and the weighting for ADFI was the negative of the average of the intra-contemporary group ratio of mean gain divided by mean ADFI. Values were combined with EBV to compute 2 index values per animal. Pearson correlations for steers (0.96) and heifers (0.45) indicated a strong relationship for steers between the indices. For steers, using the index with PWG rather than with ADG effectively results in the same genetic gain without additional intake records. Because more animals can be measured for intake, using PWG increases relative annual progress of selection for feed efficiency by 15%. These findings support using PWG data in combination with ADFI to determine efficient steers, lessen costs, and increase feed efficiency genetic change per year.

**Key Words:** beef, index, efficiency

**524 Comparison of actual versus predicted feed intake phenotypes for genetic evaluation of feed efficiency in beef cattle.** Kimberly A. Branham<sup>\*1</sup>, Jonathan E. Beever<sup>2</sup>, Dan B. Faulkner<sup>10</sup>, Holly L. Neibergs<sup>3</sup>, Kris A. Johnson<sup>3</sup>, Christopher M. Seabury<sup>4</sup>, Dorian J. Garrick<sup>5</sup>, Daniel D. Loy<sup>5</sup>, Stephanie L. Hansen<sup>5</sup>, Harvey C. Freetly<sup>6</sup>, Matt L. Spangler<sup>7</sup>, Monty S. Kerley<sup>8</sup>, Robert L. Weaber<sup>9</sup>, Daniel W. Shike<sup>2</sup>, Robert D. Schnabel<sup>8</sup>, J. E. Decker<sup>8</sup>, Jerry F. Taylor<sup>8</sup>, and Megan M. Rolf<sup>1</sup>, <sup>1</sup>*Oklahoma State University, Stillwater, OK*, <sup>2</sup>*University of Illinois, Champaign, IL*, <sup>3</sup>*Washington State University, Pullman, WA*, <sup>4</sup>*Texas A&M University, College Station, TX*, <sup>5</sup>*Iowa State University, Ames, IA*, <sup>6</sup>*USDA- Meat Animal Research Center, Clay Center, NE*, <sup>7</sup>*University of Nebraska, Lincoln, NE*, <sup>8</sup>*University of Missouri, Columbia, MO*, <sup>9</sup>*Kansas State University, Manhattan, KS*, <sup>10</sup>*University of Arizona, Tucson, AZ*.

Feed efficiency is expensive to measure in beef cattle because of the technology it requires to measure individual animal dry matter intakes (DMI). One potential solution is to develop methods to effectively utilize pen feed intake data for genetic evaluation. Genetic correlations between predicted DMI (pDMI) and actual DMI reported in the literature indicate that pDMI may be useful as an indicator trait. Therefore, the objective of this study is to evaluate whether quantitative trait loci (QTL) mapping approaches identify the same regions of the genome for pDMI and DMI. Because average daily gain (ADG) is a primary driver of the prediction models, the overlap of pDMI and DMI QTL regions with QTL for ADG will also be evaluated. To achieve these objectives, individual animal feed intake, weight, and carcass data was obtained on 849 Hereford steers and heifers fed within a GrowSafe (GrowSafe Systems Ltd.) feed intake system. The Cattle Value Discovery System (CVDS) growth and carcass data model was utilized to obtain pDMI from DMI pooled within pens and reallocated to individual animals. Phenotypic correlations were 0.64 ( $P < 0.0001$ ) and 0.56 ( $P < 0.001$ ) between pDMI and DMI and pDMI and ADG, respectively. Genotypes

were collected using the Illumina BovineHD Beadchip assay (Illumina Inc., San Diego, CA), and after data filtering, a final data set of 648,625 single nucleotide polymorphisms (SNP) were available for analysis. The SNP effects for ADG, pDMI, and DMI were estimated utilizing a BayesB0 model in GenSel. The 5-SNP windows surrounding the 100 largest SNP effects for each phenotype were compared with determine overlap between QTL regions. Concordance of QTL regions was 50% between pDMI and ADG, 26% between pDMI and DMI, and 19% between DMI and ADG. Seven of the QTL regions in common between pDMI and DMI were independent of ADG QTL regions. These results show that there is concordance between genomic regions for pDMI and DMI independent of the model drivers (ADG), and additional research will be conducted to characterize these regions of interest in genomic prediction for feed efficiency.

**Key Words:** beef cattle, feed efficiency, genomics

**525 Hierarchical Bayesian inference on genetic and non-genetic components of partial efficiencies determining feed efficiency in dairy cattle.** Yongfang Lu\*<sup>1</sup>, Mike Vandehaar<sup>1</sup>, Diane Spurlock<sup>2</sup>, Kent Weigel<sup>3</sup>, Louis Armentano<sup>3</sup>, Charles Staples<sup>4</sup>, Erin Connor<sup>5</sup>, Zhiquan Wang<sup>6</sup>, Mike Coffey<sup>7</sup>, Roel Veerkamp<sup>8</sup>, Yvette Haas<sup>8</sup>, Nora Bello<sup>9</sup>, and Robert Tempelman<sup>1</sup>, <sup>1</sup>Michigan State University, East Lansing, MI, <sup>2</sup>Iowa State University, Ames, IA, <sup>3</sup>University of Wisconsin, Madison, WI, <sup>4</sup>University of Florida, Gainesville, FL, <sup>5</sup>U.S. Department of Agriculture, Beltsville, MD, <sup>6</sup>University of Alberta, Edmonton, AB, Canada, <sup>7</sup>Scottish Agricultural College, Easter Bush, Midlothian, UK, <sup>8</sup>Animal Breeding and Genomics Centre, Wageningen UR Livestock Research, Lelystad, the Netherlands, <sup>9</sup>Kansas State University, Manhattan, KS.

Dairy cattle feed efficiency (FE) can be defined as the ability to convert DMI into milk energy (MILKE) and maintenance or metabolic body weight (MBW). In other words, FE is DMI conditional on MILKE and MBW (i.e., DMI|MILKE,MBW). These partial regressions or partial efficiencies (PE) of DMI on MILKE and MBW can be separately partitioned into genetic or residual PE; furthermore, either PE category might be heterogeneous across various environmental or management factors. We develop a hierarchical Bayesian multivariate mixed model to infer upon such heterogeneity in PE as well as that of variance components (VC) of FE by modeling genetic and residual components of PE and of VC as mixed model functions of various factors such as station (fixed), parity (fixed), days in milk (fixed), and ration within station (random). After validating our proposed model with a simulation study, we applied it to analysis of a dairy consortium data set involving 5,088 Holstein cows from 13 research stations in 4 countries. Although no significant differences were detected across stations for the genetic PE of DMI|MILKE (0.38 kg/Mcal) and of DMI|MBW (0.10 kg/kg<sup>0.75</sup>), as well as the residual PE of DMI|MILKE (0.33 kg/Mcal), the residual PE of DMI|MBW significantly differed across stations ( $P < 0.05$ ), ranging from 0.05 kg/kg<sup>0.75</sup> to 0.18 kg/kg<sup>0.75</sup>. Substantial heterogeneity in genetic and residual VC in FE across stations, rations, and parities was also inferred. Estimated heritabilities of FE ranged from 0.16 to 0.46 across stations, whereas the overall estimated heritability of FE was 0.23. These results suggest that FE is more complex than what is currently considered in most quantitative genetic analyses.

**Key Words:** dairy cattle, feed efficiency, heterogeneity.

**526 Thermal imaging as an indicator of feed efficiency in mid-lactation Holstein cows.** Lydia C. Hardie\* and Diane M. Spurlock, Iowa State University, Ames, IA.

Genetic improvement of feed efficiency in dairy cattle through direct selection is a challenge because of costs associated with measuring feed intake on individual cows. The identification of an easy to measure indicator trait of feed efficiency would help alleviate this problem. The objective of this study was to characterize the genetic variability and determine the relationship between surface body temperature and internal body temperature with feed efficiency in mid-lactation Holstein cows. Feed efficiency was measured as residual feed intake (RFI), defined as the difference between the actual intake and predicted intake based on milk energy, body weight change, and maintenance requirements. Individual daily feed intakes and milk production were recorded for 8 weeks on 124 primiparous cows between 50 and 200 d in milk. Weekly body weight and milk component data were also collected, and average RFI throughout the measurement period was calculated. Surface body temperature was measured weekly by thermal images taken of the lower right rear leg using a Fluke hand-held Thermal Imaging Scanner. The mean temperature in a defined region on the image was averaged across days for each cow. Rectal body temperatures were taken on imaging days and averaged for each cow. The ability of the body temperatures to explain variation in RFI was analyzed with a linear model, which included the fixed effect of replicate along with body temperature (surface or rectal) as a covariate. Heritabilities for body temperatures were estimated in ASReml. Rectal temperature did not significantly explain variation in RFI while surface leg temperature accounted for 2.8% of the total variance in RFI ( $P < 0.10$ ). Average leg surface temperatures for the top and bottom quartiles for RFI were 25.3 and 24.9 C (SE 0.3) for high and low RFI, respectively. Rectal temperatures did not differ between high and low RFI quartiles. Leg surface temperature was moderately heritable ( $0.32 \pm 0.32$ ) while the heritability estimate of rectal temperature was  $0.16 \pm 0.26$ . This research demonstrates surface leg temperature is a heritable trait that explains a small portion of variation in RFI.

**Key Words:** feed efficiency, residual feed intake, thermal imaging

**527 Genetic correlations of lower gastrointestinal tract microflora taxonomic groups with animal intake and gain.** Larry A. Kuehn\*<sup>1</sup>, Warren M. Snelling<sup>1</sup>, Rohita Sinha<sup>2</sup>, James E. Wells<sup>1</sup>, James L. Bono<sup>1</sup>, Harvey C. Freedly<sup>1</sup>, Min Seok Kim<sup>1</sup>, Jennifer Clarke<sup>2</sup>, Stephen D. Kachman<sup>2</sup>, Etsuko Moriyama<sup>2</sup>, Danielle F. Wells<sup>2</sup>, and Andrew K. Benson<sup>2</sup>, <sup>1</sup>USDA, ARS, U.S. Meat Animal Research Center, Clay Center, NE, <sup>2</sup>University of Nebraska-Lincoln, Lincoln, NE.

Diversity and composition of microorganisms in the lower gastrointestinal tract have been shown to affect health and weight in humans and other mammalian species. At least a portion of the microbial diversity is likely under host genetic control. Our objective was to determine whether animal feed intake and growth were genetically correlated with microbial taxonomic groups as measured in fecal samples from the lower gastrointestinal tract through metagenomic analysis. Fall born animals ( $n = 905$ ) of various breed composition (crosses of 18 different breeds) had individual animal feed intake recorded during a 63d period. Steers ( $n = 491$ ) received a high energy concentrate (finishing) ration, and heifers ( $n = 414$ ) received a roughage (growing) ration. During these feeding trials, animals were weighed on 2 consecutive days both when feed intake collection began and ended with additional weights recorded at least every 3 weeks. Fecal samples were collected prior (once), during (at least 3 times), and after (once) the feed intake trial period and pooled as a composite sample by animal. Taxonomic composition of the fecal microbiome was quantified from shotgun metagenomic sequencing of total DNA. Common contigs within and among animals were pooled by



phylogeny yielding a reference metagenome assembly of 87 taxonomic groups. These groups (each present in at least 75% of the animals) were quantified by mapping reads from individual animals onto the reference assembly. Genetic (co)variance components for feed intake, gain, and individual taxonomic groups were estimated using REML. Data were modeled with fixed effects of contemporary group (sex, year, season) and breed composition covariates. Random effects were animal direct genetic effect (pedigree derived relationships) and error. Seven taxonomic groups were estimated to be heritable ( $P \leq 0.10$ ). Genetic correlations were detected ( $P \leq 0.10$ ) for *Blautia* with feed intake ( $r = 0.61$ ) and *Cellulosilyticum* with weight gain ( $r = -0.68$ ). Ration (or sex) specific correlations were not estimable; however, it is likely that ration affects the lower gastrointestinal tract microbial composition. The USDA is an equal opportunity provider and employer.

**Key Words:** beef cattle, feed efficiency, metagenomics

**528 Evaluating residual feed intake in dairy cattle using marker  $\times$  environment interaction genomic selection model.** Chen Yao<sup>\*1</sup>, Gustavo de los Campos<sup>2</sup>, Michael J. Vande Haar<sup>3</sup>, Diane M. Spurlock<sup>4</sup>, Lou Armentano<sup>1</sup>, Yvette de Haas<sup>5</sup>, Roel Veerkamp<sup>5</sup>, Mike Coffey<sup>6</sup>, and Kent A. Weigel<sup>1</sup>, <sup>1</sup>University of Wisconsin, Madison, WI, <sup>2</sup>University of Alabama, Birmingham, AL <sup>3</sup>Michigan State University, East Lansing, MI <sup>4</sup>Iowa State University, Ames, IA <sup>5</sup>Wageningen UR Livestock Research, Lelystad, the Netherlands, <sup>6</sup>Scottish Agricultural College, Easter Bush, Midlothian, UK.

Feed efficiency in dairy cattle has gained much attention recently. To obtain residual feed intake (RFI), a measure of feed efficiency, individual feed intakes must be measured, which is usually cost prohibitive. Combining data from multiple countries could enlarge the reference population, but in this scenario it is essential to model genotype  $\times$  environment ( $G \times E$ ) interactions when making inferences about feed efficiency or selecting efficient cattle. In this study, we constructed an interaction model that allowed marker effects to be peculiar to each environment (marker  $\times$  environment interaction). As a benchmark, a standard genomic selection model that assumes marker effects are constant across environments was also fitted. We applied these models to data from 3,656 cows measured in 3 environments, North America (NAM), the Netherlands (NLD), and Scotland (SAC). The RFI phenotype was calculated as the deviation of an animal's feed intake from its expected intake based on milk energy output and metabolic body weight. Genotypic data included 57,574 SNPs per animal. The proposed model was used to estimate environment-specific genomic heritabilities, marker effects and breeding values. Genomic correlations between environments can also be computed using the variance component estimates from the model. For prediction purposes, the interaction model gave comparable accuracies to the standard model. The interaction model provides a novel way to evaluate traits measured in multiple environments in which  $G \times E$  may exist, and it allows the user to distinguish markers with effects that are stable across environments versus markers for which interactions exist.

**Key Words:** genomic selection, marker  $\times$  environment interaction, residual feed intake

**529 Validating your validation: A consistency check for the R<sup>2</sup> found in a validation to calculate correct reliabilities for genomic EBV in a multi-trait setting.** W. Marianne Stoop\*, H. Eding, and G. de Jong, CRV, Arnhem, the Netherlands.

In 2014, the Netherlands and Flanders implemented a new genomic system, in which direct genomic values (DGV) are used as pseudo-

observations to directly affect the conventional trait. For most trait groups, this pseudo-record system is a multi-trait animal model, where the added R<sup>2</sup> from a validation study is used as correlation between DGV and conventional trait, and a covariance matrix holding partial correlations is derived from it. However, validation of the DGV is done in a single trait setting, where a relatively young –and unreliable– group of bulls is used as validation set, often resulting in unstable added R<sup>2</sup>, depending on which set of bulls you select as validation set. When the added R<sup>2</sup> of traits within a multi-trait system are not consistent to each other, the multi-trait system is unbalanced, even if the defined covariance matrix is positive definite, resulting in incorrect reliabilities. In this study, a consistency check is done on the validation results of a set of fertility traits (nonreturn56, interval-calving-1st-insemination, calving interval, interval-first-last-insemination, conception rate, conception rate heifers, and age at first insemination) used in a multi-trait animal model, to assess the correctness of the added R<sup>2</sup>, the used correlations and the reliability calculated for the GEBV. This consistency check is based on the coefficient of multiple correlations (MCC); where the expectation is that the diagonal of the matrix containing MCC is equal to the corresponding R<sup>2</sup> from the validation. Data of the EuroGenomics consortium was used. The training set varied between 6,000 bulls (for traits with national data only) to 26,000 bulls. Bulls born after 20040630 were considered as validation set. The added R<sup>2</sup> ranged from 0.21 for age-at-first-insemination to 0.64 for interval-first-last-insemination. When the set of added R<sup>2</sup> was consistent, the reliabilities for the validation bulls estimated in the multi-trait model were similar to realized reliabilities. In conclusion, the consistency check is a valuable tool to assess genomic validation.

**Key Words:** dairy cattle, genomics, validation

**530 Genomic heritabilities and SNP associated with lower gastrointestinal tract microflora taxonomic groups and *E. coli* O157:H7 shedding.** Warren M. Snelling\*<sup>1</sup>, Larry A. Kuehn<sup>1</sup>, Rohita Sinha<sup>2</sup>, James E. Wells<sup>1</sup>, James L. Bono<sup>1</sup>, Elaine D. Berry<sup>1</sup>, Min Seok Kim<sup>1</sup>, Jennifer Clarke<sup>2</sup>, Stephen D. Kachman<sup>2</sup>, Etsuko Moriyama<sup>2</sup>, Danielle F. Wells<sup>2</sup>, and Andrew K. Benson<sup>2</sup>, <sup>1</sup>USDA-ARS-US Meat Animal Research Center, Clay Center, NE, <sup>2</sup>University of Nebraska-Lincoln, Lincoln, NE.

Risk of beef contaminated by *E. coli* O157:H7 is affected by prevalence of O157:H7 in cattle feces. Host genetics may influence O157:H7 shedding and overall microbial diversity in the lower gastrointestinal tract. Objectives of this study were to determine influence of animal genotypes on measures of O157:H7 shedding and relative abundance of different taxa in cattle feces, and identify regions of the bovine genome associated with O157:H7 prevalence and microbial diversity. Feces were sampled from fall born animals ( $n = 1,099$ ) of various breed composition (crosses of 18 different breeds) undergoing trials to measure individual animal intake over 63 to 90 d periods. Steers ( $n = 574$ ) received a high-energy finishing ration and heifers ( $n = 525$ ) a high-roughage breeding development diet. Feces were sampled once before, at least thrice during, and once after the feed intake trial period. Individual samples were assessed for enumerable O157:H7. Taxonomic composition of the fecal microbiome was quantified from shotgun metagenome sequencing of total DNA pooled for each animal. Reads were assembled for each animal individually and contigs pooled within and across animals by phylogeny, yielding a reference metagenome assembly of 87 different taxonomic groups that were detected across  $> 75\%$  of the animals. These taxa were quantified by mapping reads from each sample onto the reference assembly. Heritability for relative abundance of each taxa and O157:H7 prevalence measure was estimated with REML, using

genomic relationships described by imputed BovineHD SNP genotypes. Individual SNP effects were solved from animal solutions after REML converged. Heritability estimates ranged from 0.00 for 35 taxa to 0.21 for *Butyrivibrio*, and were greater than 0.05 for 19 taxa. For O157:H7 traits, estimates were 0.10 for the number of times the animal was prevalence positive, and 0.07 for log<sub>10</sub> of the average O157:H7 level. Correlations between SNP effects on O157:H7 and individual taxa were near zero, although a cluster of SNP between *TMEM20* and *PLCE1* on BTA26 was associated with log<sub>10</sub>[O157:H7] and 11 taxa having heritabilities >0.05.

**Key Words:** beef cattle, metagenomics, *E. coli* O157:H7

**531 Utilizing cattle genetic trends to evaluate the long-term use of gene bank collections.** Harvey D. Blackburn\*<sup>1</sup>, Carrie S. Wilson<sup>1</sup>, Samuel Paiva<sup>2</sup>, Scott Spiller<sup>1</sup>, and Phil H. Purdy<sup>1</sup>, <sup>1</sup>ARS/USDA, Fort Collins, CO, <sup>2</sup>EMBRAPA, Brasilia, Brazil.

The National Animal Germplasm Program has developed substantial germplasm collections (>800,000 samples) for more than 300 unique livestock populations or breeds. Gene bank utilization is relatively new to the livestock sector and the long-term genetic relevance of such collections has not been documented in terms of how far into the future the captured diversity might be commercially useful. Comparing the annual average PTA or EPD for various traits computed by the respective breed associations with the collection's PTA/EPD statistics (mean, standard deviation, and extremes) may be a comparison that provides insight as to how well the collection mirrors the in situ population. Six breeds selected for comparison were Holstein (n = 5393), Jersey (n = 873), Angus (n = 703), Hereford (n = 252), Charolais (n = 86), and Brangus (n = 70). Birth years for the bulls in each breed spanned: 50, 60, 55, 60, 45, and 35 years, respectively. Trends for PTA or EPD for each trait computed by the respective associations were developed. Breed average (in situ population) vs. collection: average, standard deviation and minimum and maximum values for the respective breeds were compared. The collection average across breeds and traits closely mirrored the in situ populations' PTA or EPD estimates over time. When comparing the repository PTA or EPD outliers of any breed to the mean of its in situ counterpart across traits it was determined that it would take the in situ average PTA/EBV estimates, for a given birth year, 25 to 35 years to reach the collection's maximum or minimum value. This finding was consistent across all breeds regardless of collection size and age. These results demonstrate that animals with samples in gene bank collections could produce progeny that are commercially competitive for at least a quarter century (post collection) and demonstrates that gene bank collections are sufficient to provide protection for the various breeds in the collection. These results demonstrate that continued targeted sampling by gene banks over time will further ensure collection utility. In addition to the issue of genetic security, these findings suggest genetic progress has not advanced as quickly in cattle populations as might have been assumed.

**Key Words:** genetic resource, cattle, gene bank

**532 Estimating the heritability of gene expression profiles using RNAseq data.** Deborah Velez-Irizarry\*<sup>1</sup>, Catherine W. Ernst<sup>1</sup>, Ronald O. Bates<sup>1</sup>, Pablo Reeb<sup>1</sup>, Yeni Bernal Rubio<sup>2</sup>, Nancy E. Raney<sup>1</sup>, and Juan P. Steibel<sup>1</sup>, <sup>1</sup>Michigan State University, East Lansing, MI, <sup>2</sup>University of Buenos Aires, Buenos Aires, Argentina.

Estimation of heritability is crucial for breeding purposes and for understanding the genetic basis of phenotypic traits. For the specific case of expression traits, heritability estimates can be used to prioritize

expression quantitative trait loci (eQTL) genes. Estimates of heritability are usually obtained by linking phenotypic records with the estimated relationship matrix. The relationship matrices can be derived from pedigree, marker information or both. We propose an approach to estimate heritability of gene expression that takes full advantage of next generation sequencing platforms. A GBLUP-based animal model was used to fit all genetic markers simultaneously to each gene expression profile individually. A preliminary study using longissimus muscle (LM) transcriptome sequence data (RNaseq) for 24 animals from the Michigan State University pig resource population (MSUPRP) showed that more power was needed to identify significant genetic effects when using gene expression profiles as a trait phenotype. A subsequent study involving LM RNaseq for 144 MSUPRP animals was conducted using a similar GBLUP-based model. Results showed great improvement in the detection of significantly heritable expression traits (HET). A total of 226 statistically significant HET were discovered at FDR = 1%. The range of heritability estimates for these significant expression traits was between 0.27 and 1.00. Furthermore, 3 gene expression profiles showed extremely high heritability ( $h^2 > 0.99$ , with GBV q-values  $< 1 \times 10^{-8}$ ). Pathway Analyses of the significant HET revealed multiple genes involved in organismal development and transcriptional regulation emphasizing cellular growth and proliferation. The top genes involved in these molecular processes include *TGFB3*, *BMPRIA* and *MCM8*, the former 2 associated with weight gain. This research shows that genomic prediction models can be effectively used to elucidate the molecular mechanisms driving variations in heritable expression traits and to identify important regulatory gene networks.

**Key Words:** heritability, RNAseq, quantitative genetics

**533 Exploitation of population-wide whole-genome genotyping to identify the founder of a deleterious mutation in cattle.** Andreas Kromik<sup>1</sup>, Phillip Widmann<sup>1</sup>, Frieder Hadlich<sup>1</sup>, Dierck Segelke<sup>2</sup>, Rosemarie Weikard<sup>1</sup>, and Christa Kühn\*<sup>1,3</sup>, <sup>1</sup>Leibniz Institute for Farm Animal Biology (FBN), Institute for Genome Biology, Dummerstorf, Germany, <sup>2</sup>Vereinigte Informationssysteme Tierhaltung w.V. (vit), Verden/Aller, Germany, <sup>3</sup>University Rostock, Faculty of Agricultural and Environmental Sciences, Rostock, Germany.

Genomic selection programs generate deep pedigrees with whole genome genotyping data, which can also be used for the analysis of genetic defects. Recently, studies highlighted the relevance of novel mutations for human health. The use of non-progeny tested very young sires in genomic selection programs increases the danger of an unnoticed, rapid spread of novel mutations, if no respective precautions are taken. Recently, we identified a novel defect (spinal and vertebral malformation, VSD) and its causal mutation in Holstein cattle. Based on 50K SNP data, in our project 73 offspring of a carrier sire for VSD (41 VSD affected, 31 VSD unaffected) were haplotyped using CRIMAP options, and the paternal haplotype carrying the VSD mutation was identified. To further trace the origin of the haplotype associated with VSD, we subsequently haplotyped 50K SNP genotypes of a total of 55,384 individuals from the German Holstein population including all available relatives of the carrier sire using BEAGLE. Genotypes were provided by VIT Verden, the central database for genomic evaluation in German Holstein cattle. Tracking of haplotypes and haplotype breakpoints through the pedigree excluded the father of the carrier sire as potential source of the mutation and indicated that the mutation was located on the paternally inherited haplotype of the dam. Sequencing of the bovine *brachyury* gene in the maternal grandsire, the dam and the carrier sire itself revealed that the mutation developed de novo in the dam, because the haplotype of the maternal grandsire that was transmitted to the dam

contained the wild type allele, whereas the respective haplotype in the dam showed the mutated allele. Additional genotyping of 94 offspring of the maternal grandsire revealed that none of those offspring showed the mutated allele. In conclusion, population-wide SNP genotyping and haplotype tracking enabled exclusion of a major Holstein blood line as source for VSD and confirmed the origin of the defect. Due to the recent

de novo mutation and the confirmed knowledge about the founder, the defect can easily be eradicated from the Holstein population before any further outspread.

**Key Words:** genetic defect, cattle, vertebro-spinal dysplasia



## Breeding and Genetics: Genomic methods

**534 Use of genomic recursions in single-step genomic BLUP with a large number of genotypes.** Breno D. Fragomeni<sup>\*1</sup>, Daniela A. L. Lourenco<sup>1</sup>, Shogo Tsuruta<sup>1</sup>, Yutaka Masuda<sup>1</sup>, Ignacio Aguilar<sup>2</sup>, Andres Legarra<sup>3</sup>, Thomas J. Lawlor<sup>4</sup>, and Ignacy Misztal<sup>1</sup>, <sup>1</sup>*Department of Animal and Dairy Science, University of Georgia, Athens, GA*, <sup>2</sup>*Instituto Nacional de Investigacion Agropecuaria, Las Brujas, Canelones, Uruguay*, <sup>3</sup>*INRA, UMR1388 GenePhySE, Castanet Tolosan, France*, <sup>4</sup>*Holstein Association USA Inc., Brattleboro, VT*.

The purpose of this study was to evaluate accuracy of genomic selection in single-step genomic BLUP (ssGBLUP) when the inverse of the genomic relationship matrix (G) is derived by the APY (algorithm for proven and young animals). This algorithm implements genomic recursions on a subset of “proven” animals. Only a matrix due to the subset needs to be inverted and extra costs of adding “young” animals are linear. Analyses involved 10,102,702 Holsteins with final scores on 6,930,618 cows. A total of 100k animals with genotypes included 23k sires (16k with more than 5 progenies), 27k cows, and 50k young animals. Genomic EBV (GEBV) were calculated with a regular inverse of G, and with the G inverse approximated by APY. Initially, animals in the “proven” subset included only sires or cows. Later, animals in the “proven” subset were randomly sampled from all genotyped animals in sets of 5k, 10k, and 20k; each sample was replicated 4 times. Genomic EBV with APY were accurate when the number of animals in the “proven” subset was  $\geq 10k$ , with little difference between the ways of creating the subset. Numerical properties as shown by the number of rounds to convergence were best with random subsets. The ssGBLUP with APY can accommodate a large number of genotypes at low cost and with high accuracy.

**Table 1 (Abstr. 534).** Correlations (or range of correlations) between genomic EBV with regular and APY ssGBLUP for young genotyped animals and rounds to convergence for different subset of animals used in recursions

Definition of subset	Animals in subset	Correlation	Rounds to convergence
All	100,000	1.00	567
Sires	23,174	0.99	432
Cows	27,215	0.99	797
2k random	2,000	0.94	356
5k random	5,000	0.97	360
10k random	10,000	0.99	396
20k random	20,000	0.99	420

**Key Words:** single-step method, genomic selection, genomic recursion

**535 Genomic predictions with approximated G-inverse for a large number of genotyped animals.** Yutaka Masuda<sup>\*1</sup>, Ignacy Misztal<sup>1</sup>, Shogo Tsuruta<sup>1</sup>, Daniela A. L. Lourenco<sup>1</sup>, Breno Fragomeni<sup>1</sup>, Andres Legarra<sup>2</sup>, Ignacio Aguilar<sup>3</sup>, and Tom J. Lawlor<sup>4</sup>, <sup>1</sup>*University of Georgia, Athens, GA*, <sup>2</sup>*INRA, Castanet-Tolosan Cedex, France*, <sup>3</sup>*Instituto Nacional de Investigación Agropecuaria, Canelones, Uruguay*, <sup>4</sup>*Holstein Association USA Inc., Brattleboro, VT*.

The objective of this study was to compare the accuracy of genomic predictions in final score for young Holstein bulls calculated from single-step GBLUP models with the regular  $G^{-1}$  and approximated  $G^{-1}$  ( $G^{-1}_{ap}$ ) matrices. The  $G^{-1}_{ap}$  was calculated with recursions on a small subset of

animals. The regular  $G^{-1}$  has a quadratic memory cost and cubic computational cost as the number of genotyped animals increases, whereas  $G^{-1}_{ap}$  has a linear cost for animals outside the subset. The predictor data set consisted of 77,066 genotyped animals, 9,009,998 pedigree animals, and 6,384,859 classified cows born in 2009 or earlier. For calculation of  $G^{-1}_{ap}$ , 9,406 high accuracy bulls or 16,828 high accuracy bulls and cows were used as the small subset. Genomic predictions (GEBV2009) were calculated for predicted bulls that had no classified daughters in 2009 but did in 2014. The validation data set contained phenotypes and pedigree recorded up to March 2014. Daughter yield deviations (DYD2014) were calculated for the predicted bulls with at least 30 daughters in 2014 ( $n = 2,948$ ). Coefficient of determination ( $R^2$ ), calculated from a linear regression of DYD2014 on GEBV2009, was 0.44 with the regular  $G^{-1}$  and 0.45 with  $G^{-1}_{ap}$  for either subset. Genomic predictions using all available 569,404 (570K) genotypes were also calculated with  $G^{-1}_{ap}$  on 9,406 bulls. The computation was performed using 16 CPU cores and 61 G bytes of working memory. Setting up  $G^{-1}_{ap}$  took 1.8 h, setting up matrices associated with  $A_{22}^{-1}$  took 7 min, and iterations took 4.6 h, resulting in 6.4 total hours. In contrast, BLUP computations without genotypes took 1.6 h in total. The  $R^2$  value from 570K genotypes was similar to the result from GEBV2009. Genetic predictions can be obtained with substantially less computational cost but without loss of reliability using  $G^{-1}_{ap}$ . The single-step GBLUP with  $G^{-1}_{ap}$  is applicable to very large genotyped populations.

**Key Words:** computing, Holstein, single-step genomic BLUP (ssGBLUP)

**536 Theoretical aspects of the APY algorithm for inverting a large genomic relationship matrix.** Ignacy Misztal<sup>\*</sup>, *University of Georgia, Athens, GA*.

The algorithm for proven and young animals (APY) implements the inverse of the genomic relationship matrix by recursion on a subset of animals. If the subset is small, storage and computations are approximately linear with the number of genotyped individuals, allowing for processing of practically an unlimited number of animals. The APY algorithm was tested with many subsets, including proven bulls, bulls and cows, cows only and random subsets. GEBVs calculated with APY were accurate when the number of animals in the subset was  $\geq 10k$ , with little difference between different subsets. Best convergence rates when solving equations with APY were obtained with subsets composed of randomly chosen animals. The properties of the APY algorithm can be explained using the concept of a finite number of independent chromosome segments. Assume that each segment has a fixed value, that a fraction of each segment has a value proportional to the length of that segment (infinitesimal model), and that a genome of an individual is composed of a fraction of each segment. Subsequently, in the absence of confounding,  $n$  animals allow for identification of a population with  $n$  segments. Assuming some errors, lowest estimation errors are with heterogeneous animals. For traits where genes are distributed unequally across the genome, a conceptual division of segments into smaller segments with quasi-equal distribution would result in a larger subset required for the same accuracy of GEBV. If genome sequencing allows for identification of all  $m$  QTN, each QTN may be treated as one segment and, assuming a purely additive model, a recursion on  $m$  sufficiently heterogeneous animals will capture all variability in the genome. Computation in APY assume ability to compute parts of such a genomic relationship matrix that reflects the genetic architecture for

each trait. The APY algorithm may allow routine genomic evaluation for any number of genotyped animals with any model at a cost not much above BLUP.

**Key Words:** genome selection, genomic recursion, genomic relationship matrix

**537 Effect of increasing the number of single nucleotide polymorphisms from 60,000 to 85,000 in genomic evaluation of Holsteins.** George R. Wiggans\*, Tabatha A. Cooper, Paul M. VanRaden, Curt P. Van Tassel, Derek M. Bickhart, and Tad S. Sonstegard, *Animal Genomics and Improvement Laboratory, Agricultural Research Service, USDA, Beltsville, MD.*

The periodic need to restock reagent pools for genotyping chips provides an opportunity to increase the number of single nucleotide polymorphisms (SNP) on a chip. As an improved replacement for the GeneSeek Genomic Profiler HD for dairy cattle, a set of >140,000 SNP was selected that included all SNP on the current chip, all SNP used in genomic evaluations, SNP that are possible functional mutations, and other informative SNP. Most added SNP were selected from the Illumina Bovine HD Genotyping BeadChip based on the magnitude of effects on evaluated traits. Some SNP with lower minor allele frequency were considered because of their potential for better tracking of causative variants. Genotypes already available from other chips were used to impute and evaluate the SNP set. Effects for 134,511 usable SNP were estimated for all breed-trait combinations; SNP with the largest absolute values for effects were selected (5,000 for Holsteins, 1,000 for Jerseys, and 500 each for Brown Swiss and Ayrshires for each trait), which resulted in 78,032 SNP after removing duplicates. An additional 9,130 SNP with many parent-progeny conflicts after imputation were removed, which resulted in 72,843 SNP. Of those, 38,515 were among the 60,671 SNP currently used in genomic evaluation. To minimize possible accuracy loss, 12,094 of the SNP currently used but not already selected and with the largest effects were added for a total of 84,937 SNP. Three cutoff studies were conducted with 60,671, 84,937, and 134,511 SNP to determine gain in reliability over parent average when evaluations based on data from August 2011 were used to predict genetic merit from December 2014. Across all traits, mean gains were 32.5, 33.4, and 32.0 percentage points, respectively. Previous experience indicates that gains from the highest number of SNP will increase as the number of genotypes from the new SNP set increases. The gain of 0.9 percentage points from adding nearly 25,000 SNP justifies the extra computation time needed. However, the gain may be overestimated because data used to select the most informative SNP were also the data used to determine gain.

**Key Words:** dairy cattle, genomic evaluation, single nucleotide polymorphisms

**538 Genome-wide association study of fertility traits in dairy cattle using high-density single nucleotide polymorphism marker panels.** Kristen L. Parker Gaddis<sup>1</sup> and John B. Cole\*<sup>2</sup>, <sup>1</sup>*Department of Animal Sciences, University of Florida, Gainesville, FL,* <sup>2</sup>*Animal Genomics and Improvement Laboratory, ARS, USDA, Beltsville, MD.*

Unfavorable genetic correlations between production and fertility traits are well documented. Genetic selection for fertility traits is slow, however, due to low heritabilities. Identification of single nucleotide polymorphisms (SNP) involved in reproduction could improve reliability of genomic estimates for these low heritability traits. Additionally, high-density marker panels can increase the power of resultant GWAS by providing increased coverage and stronger linkage disequilibrium

between markers and causal variants. The objective of this study was to identify SNP associated with 3 fertility traits in dairy cattle, daughter pregnancy rate (DPR), heifer conception rate (HCR), and cow conception rate (CCR), using high-density marker panels. Deregressed predicted transmitting abilities were available for 10,000 bulls sampled from the National Dairy Database that had high-density genotypes. Of those, 725 had been genotyped with the Illumina BovineHD Genotyping BeadChip. The remaining bulls had genotypes from various chip densities that were imputed up to the same level. After editing, 312,614 markers were included in the analyses. Univariate analyses were performed for DPR, HCR, and CCR using REMLF90 (version 1.79) with genomic options. postGSf90 (version 1.170) was used to calculate SNP effects and 10-SNP window variances. The largest proportion of variance explained for DPR (0.126%) was located on chromosome 6. Peaks were also identified on chromosomes 5, 18, and 28 associated with DPR. For HCR, the region explaining the largest proportion of variance (0.155%) was located on chromosome 1. Large peaks were also identified for HCR on chromosomes 6, 8, 14, and 17. The largest proportion of variance explained for CCR (0.181%) was located on chromosome 18. Large peaks associated with CCR were also identified on chromosomes 6, 15, and 19. Numerous markers and regions aligned with those previously identified. Significant SNPs could be used in genomic selection programs as well as in identification of genes and networks involved with fertility.

**Key Words:** fertility, genomic evaluation, high-density genotype

**539 Segment-based methods to calculate weights for weighted single-step GBLUP.** Xinyue Zhang\*, Daniela A. L. Lourenco, and Ignacy Misztal, *University of Georgia, Athens, GA.*

The purpose of this study was to explore additional options for calculating weights in weighted single-step GBLUP (WssGBLUP). In GWAS by ssGBLUP, GEBV are converted to marker (SNP) effects. Unequal variances for markers are then derived from SNP solutions and subsequently incorporated into a weighted genomic relationship matrix. Improvements on the SNP weights were obtained iteratively by recomputing both the SNP effects and the GEBV. Six options were used to calculate the weights: (1) proportional to  $u_i^2$  where  $u_i$  is the effect of the  $i$ -th SNP; (2) proportional to  $u_i^2 + \text{constant}$ ; (3) weights as  $v^{s-2}$ , where  $v$  is a scale standing for the departure from normality, and  $s$  is number of standard deviation from mean for each  $u_i^2$  where  $p_i$  is frequency of the second allele; (4) as the largest effect ( $u_i^2$ ) among every 20 SNP; (5) as the mean effect of every 20 SNP; (6) as the summation of effects of every 20 SNP. A simulated data set was used that included 15,600 animals in 5 generations, of which 1,540 were genotyped for 50k SNP. The simulation involved phenotypes for a trait with heritability of 0.5 and affected by 5, 100, and 500 QTL. Accuracy between TBV and GEBV for genotyped animals in the last generation was used for evaluation. Comparisons also involved BayesB and BayesC with deregressed proofs or EBV from BLUP, and  $\pi = 0.99, 0.9$  or  $0.5$ . In single-step, SNP effects were tracked along 10 iterations and weights were equal to 1.0 in the first iteration. Option 5 was the best in identifying simulated QTL without background noise and with precision in most of the regions. Option 2 kept accuracy of GEBV at the plateau after 2 iterations and was 0.81 as opposed to 0.70 for BayesC and 0.48 for BayesB under 500 QTL scenario. All methods reached better accuracies than BayesB and BayesC when number of QTL approached or exceeded 100 (0.2% of all SNP) due to automatically including PA in GEBV. Weights based on a sum of SNPs may be superior to those based on individual SNPs.

**Key Words:** weighted SNP, single-step genomic BLUP (ssGBLUP), BayesB

**540 Multi-allelic haplotype model based on genetic partition for genomic prediction and variance component estimation.** Yang Da\*, *Department of Animal Science, University of Minnesota, Saint Paul, MN.*

Functional genomic information has been growing rapidly but remains largely unused in genomic selection. Genomic prediction and estimation using haplotypes in genome regions with functional elements such as all genes of the genome can be an approach to integrate functional genomic information with genomic selection. Toward this goal, a multi-allelic haplotype model treating each haplotype as an ‘allele’ was developed for genomic prediction and estimation based on the partition of a multi-allelic genotypic value into additive and dominance values. Each additive value is expressed as a function of  $h-1$  additive effects, where  $h$  = number of alleles or haplotypes, and each dominance value is expressed as a function of  $h(h-1)/2$  dominance effects. For a sample of  $q$  individuals, the limit number of effects is  $2q-1$  for additive effects and is the number of heterozygous genotypes for dominance effects. Additive values are factorized as a product between the additive model matrix and the  $h-1$  additive effects, and dominance values are factorized as a product between the dominance model matrix and the  $h(h-1)/2$  dominance effects. Genomic additive relationship matrix is defined as a function of the haplotype model matrix for additive effects, and genomic dominance relationship matrix is defined as a function of the haplotype model matrix for dominance effects. Based on these results, a mixed model implementation for genomic prediction and variance component estimation that jointly use haplotypes and single SNPs is established, including 2 computing strategies for genomic BLUP (GBLUP) and genomic REML (GREML) with identical results. The multi-allelic genetic partition fills a theoretical gap in genetic partition by providing general formulations for partitioning multi-allelic genotypic values and provides a haplotype method based on the quantitative genetics model toward the utilization of functional genomic information for genomic selection.

**Key Words:** haplotype, GBLUP, GREML

**541 Revisiting allelic frequencies estimation: A decision theory approach to derive Bayes, minimax, and admissible estimators.** Carlos A. Martinez\*<sup>1,2</sup>, Kshitij Khare<sup>2</sup>, and Mauricio A. Elzo<sup>1</sup>, <sup>1</sup>*Department of Animal Sciences, University of Florida, Gainesville, FL,* <sup>2</sup>*Department of Statistics, University of Florida, Gainesville, FL.*

Decision theory was used to derive point estimators of allelic frequencies with optimal statistical properties. Uniparameter (2 alleles) and multiparameter (multiple alleles) estimation problems were addressed for an arbitrary locus, then results were extended to multiple loci. First, estimators satisfying the Bayes principle of average risk optimality were obtained using a multinomial sampling model, a Beta (biallelic loci) and a Dirichlet (multiallelic loci) prior and 3 different loss functions: Squared Error Loss (SEL), Kullback-Leibler Loss (KLL) and a Quadratic Error Loss (QEL). Second, these Bayes estimators were used to obtain minimax estimators by finding values of the hyperparameters such that the frequentist risk functions were constant, a condition that implies minimaxity. Finally, the admissibility of the estimators was checked using standard theorems from decision theory. The frequentist risk function of the Bayes estimator derived from KLL involved a finite sum without a closed form, hence this risk function could not be written as a simple algebraic expression. However, this does not prevent its computation. Under SEL and QEL it was possible to find Bayes-minimax-admissible estimators (BMAE). Sufficient conditions for the usual maximum likelihood estimator to be BMAE and for the risk functions tending to infinite or converging to zero were also found. In addition to optimal statistical

properties, these estimators have the appealing feature of taking into account random variation in allelic frequencies. The impact of using these estimators in different areas of quantitative and population genetics needs be assessed either empirically or theoretically and this poses a problem for further research.

**Key Words:** admissibility, allele frequency, minimaxity

**542 Strategies for estimating hyperparameters based on single-step Bayesian models.** Lei Zhou\* and Robert J. Tempelman, *Michigan State University, East Lansing, MI.*

Single-step BLUP (SS-BLUP) genomic prediction has the advantage of combining phenotypic information on both genotyped and nongenotyped animals. Recently developed single-step Bayesian regression models (SSBR) provide potentially even greater flexibility (e.g., heavy-tailed or variable selection alternatives) for modeling the prior distribution of SNP markers in tandem with polygenic effects. We discuss and present strategies for inferring upon these hyperparameters, particularly when data from either category (i.e., genotyped or nongenotyped) of animals is limiting. For example, when most animals in a genetic evaluation are not genotyped, inferences on key hyperparameters (i.e., marker variance, scale parameters) are compromised if these inferences are primarily based on marker data from genotyped animals only (strategy 1) whereas information on these hyperparameters might be readily borrowed from polygenic inferences involving nongenotyped animals as well (strategy 2). To compare these 2 strategies, a simulation study with 10 replicates was conducted. Five generations and a total of 2000 animals were simulated in each replicate. The heritability of the trait was 0.5 based on 1500 SNPs. The proportion of animals genotyped ranged from 10 to 90%. Results showed that strategy 2 estimated hyperparameters with greater accuracy than strategy 1 (Table 1), particularly when the proportion of animals that were genotyped was low (i.e., 10%). Nevertheless both strategies lead to similar accuracies of estimated breeding values for both genotyped and non-genotyped animals under the various genotyping rate scenarios. We also present methodology for both strategies using REML to infer upon these hyperparameters based on Gaussian specifications as well as how to infer upon hyperparameters in heavy-tailed (BayesA) and/or variable selection (BayesB) specifications. In conclusion, our proposed strategy had some advantage upon inferring hyperparameters, and further research is necessary for SSBR models.

**Table 1 (Abstr. 542).** Genetic variance by scenario and strategy

Scenario	Strategy	Total genetic variance (mean ± SD)
		24.60 ± 4.48 (True)
10% genotyped	1	24.92 ± 5.64
	2	25.70 ± 4.33
50% genotyped	1	22.35 ± 4.70
	2	24.05 ± 2.87
90% genotyped	1	20.73 ± 7.81
	2	24.26 ± 3.22

**Key Words:** single-step, Bayesian, hyperparameter

**543 Reassessing hierarchical Bayesian genome-wide association analyses.** C. Chen\*, J. P. Steibel, and R. J. Tempelman, *Michigan State University, East Lansing, MI.*

Genomic best linear unbiased prediction (GBLUP) analyses have been increasingly adapted for genome-wide association (GWA) analyses. A



currently popular modification of GBLUP for GWA, which we label as classic GBLUP, is to treat all genetic markers as random, except for the marker being tested; conversely, shrinkage GBLUP treats all markers as random in a whole genome prediction (WGP) analysis. The classic GBLUP modification has been demonstrated to preserve Type I error rates whereas shrinkage GBLUP leads to a very conservative GWA test. Nevertheless, shrinkage estimation has recently been shown to have GWA properties under alternative prior specifications. Some popular WGP model specifications are heavy-tailed (i.e., BayesA) or involve variable selection (i.e., BayesSSVS) that do not shrink large marker effects as much as shrinkage GBLUP. Given that MCMC implementations of these models are computationally onerous, we propose inferences under these alternative priors based on the EM algorithm (i.e., EMBayesA and EMBayesSSVS). In a simulation involving 10 replicated data sets, each involving about 2000 individuals and 5000 SNP markers with average pairwise LD  $r^2 = 0.30$  across 5 chromosomes, we discovered that EMBayesA and EMBayesSSVS shrink the majority of the posterior  $z$ -score based  $P$ -values to be larger relative to classic GBLUP, whereas markers in QTL regions tend to have substantially smaller  $P$ -values in EMBayesA and EMBayesSSVS compared with classic GBLUP. In an application involving backfat data from a Duroc-Pietrain F2 cross, we determined that EMBayesSSVS inferred SNP effects in albeit fewer putative QTL regions compared with classical GBLUP, although GWA using EMBayesA did not detect any such association. We suggest that EMBayesSSVS or other hierarchical variable selection models represent promising alternatives for GWA analyses of complex traits for which null marker effects might not appropriately represent a global null hypothesis; however, we also demonstrate that recently developed regularization techniques are vitally important in helping avoid posterior multimodality concerns in large dimensional EM-based inferences as well.

**Key Words:** expectation maximization, genome-wide association (GWA)

#### **544 Approximating realized additive relationships in absence of genomic information.** Romdhane Rekaya\*, Sajjad Toghiani, and L. Y. Chang, *The University of Georgia, Athens, GA.*

Superiority of genomic selection is due to the use of the realized relationships. Unfortunately, only a small portion of animals included in the evaluation are genotyped. Consequently, the majority of animals will still be evaluated based on their expected relationships. With the availability of genomic information, it is possible to assess the variation of expected co-ancestry coefficients for different orders (degrees) of relationships. Thus, this available information represents a good prior and could be used to better assess the Mendelian sampling in absence of genomic data. Furthermore, a sizeable portion of non-genotyped animals are already phenotyped for several traits with moderate to high heritabilities. Phenotypic records contain information about the true genetic relationship between animals. Using these 2 sources of information, we can improve the pedigree based relationship without using genomic data. A simulation was carried out to investigate the ability to infer realized additive relationships in absence of genomic information. A single trait with heritability of either 0.5 or 0.8 was simulated. A pedigree with large number of full (FS) and half (HS) sibs reflective of a chicken population was also simulated. All animals in the pedigree were assigned 60K SNP genotypes. The average (A) and the realized (G) matrices were computed using the pedigree and the genomic informa-

tion, respectively. Using our procedure a realized relationship matrix ( $G^*$ ) was computed without use of the simulated genomic information. The results showed that the similarity between G and  $G^*$ , measured by the absolute value of difference between corresponding elements of both matrices, was greater than between G and A. The average difference between G and A was 0.07 compared with 0.03 between G and  $G^*$ . The realized relationships for FS and HS estimated using our proposed method overlap largely with those obtained using genomic information. Additionally, the similarity between G and  $G^*$  increased, as expected, with the increase of heritability. The fact that the difference between G and  $G^*$  is smaller than between G and A indicate that the matrix  $G^*$  is a better approximation of G than A.

**Key Words:** realized, relationship, genomic

#### **545 Imputation using whole-genome sequence data in Brown Swiss and Original Braunvieh.** Christine F. Baes<sup>\*1,2</sup>, Beat Bapst<sup>2</sup>, Franz R. Seefried<sup>2</sup>, Heidi Signer-Hasler<sup>1</sup>, Christine Flury<sup>1</sup>, Dorian Garrick<sup>3</sup>, Christian Stricker<sup>4</sup>, and Birgit Gredler<sup>2</sup>, <sup>1</sup>*Bern University of Applied Sciences, Zollikofen, Bern, Switzerland*, <sup>2</sup>*Qualitas AG, Zug, Zug, Switzerland*, <sup>3</sup>*Iowa State University, Ames, IA*, <sup>4</sup>*agn Genetics, Davos, Grison, Switzerland.*

The distinct half-sib population structure common in dairy cattle populations permits imputation of array-based genotypes up to sequence level using sequence information of key ancestors. This approach provides a cost-effective alternative to sequencing all animals. The process works well in single breeds (e.g., Holstein), however it is less accurate when validation and reference animals are not of the same breed or population (e.g., Brown Swiss or Original Braunvieh). The objective of this study was therefore to investigate imputation strategies for composing reference and validation sets using Brown Swiss (BS) and Original Braunvieh (OB) animals. Whole genome sequence information (WGS) of 70 BS, 17 BS  $\times$  OB and 8 OB animals was available for analysis. WGS was masked to mimic medium (50K; 54,609 SNP), and high-density (HD; 777,962 SNP) SNP arrays and then imputed back up to sequence level. Imputation was conducted using fimpote, which employs an overlapping sliding window approach to exploit haplotype similarities between validation and reference animals. Principal component analysis was used to determine which animals constituted reference and validation sets; scenarios involving various breed compositions were compared. The accuracy of imputation from 50K and HD to WGS was evaluated for each scenario by calculating the number of correctly imputed genotypes and concordance between true and imputed genotypes. Furthermore, imputation accuracy when the reference population was solely comprised of the breed to be imputed was compared with results obtained when the reference population was comprised of multiple breeds. Validation of OB genotypes resulted in 64.6–69.6% (50K to WGS) and 79.5–83.8% (HD to WGS) correctly imputed genotypes. BS validation resulted in 78.0–78.9% (50K to WGS) and 85.5–86.1% (HD to WGS) correctly imputed genotypes. Validation of intermediary animals using both BS and OB as reference resulted in the highest percentage of correctly imputed genotypes (79.1–79.6% for 50K to WGS and 86.5–86.7% for HD to WGS). Results show that breed composition of reference and validation sets has a considerable effect on imputation accuracy, and that imputation quality is improved if reference animals from similar populations are included.

**Key Words:** imputation, whole-genome sequence, accuracy

## Companion Animal Symposium: Bioenergetics of pet food

**546 Pet obesity and bioenergetics of pet food.** Kelly Swanson\*, *University of Illinois at Urbana-Champaign, Urbana, IL.*

It is estimated that 30 to 40% of dogs and cats in the United States are overweight or obese. Pet obesity increases the risk of several diseases and disorders, including insulin resistance, diabetes mellitus, hyperlipidemia, cancers, and many others, and results in a shorter life span. Although genetics, a sedentary lifestyle, and neutering contribute to weight gain, the over-consumption of energy-dense foods is a major driver. There are many challenges to preventing pet obesity, including owner beliefs and feeding tendencies, estimation of energy density, and development of appropriate feeding guidelines. Owner education, especially as it pertains to body condition scoring and feeding guidelines, is a critical need. Feeding guidelines are required on all pet foods, but many owners either do not read them carefully and/or do not understand the importance of adjusting intake based on body condition score. Ad libitum feeding and excessive feeding of table scraps and treats contribute to overfeeding. Estimating caloric content of ingredients or pet foods may also be difficult and quite different depending on the methodology used. Numerous equations are used to estimate the caloric density of diets based on proximate analysis, but they are all based on assumptions pertaining to the digestibility and metabolism of nutrients. The use of crude fiber, a highly inaccurate method of fiber estimation, also contributes to inaccuracies in determining digestible carbohydrate content and consequent caloric density estimates. In vitro assays that estimate nutrient digestibility have been developed, but in vivo tests are by far the most accurate method by which metabolizable energy content of diets may be determined. Even when an accurate caloric density has been determined, developing accurate feeding instructions are difficult because a large variation among pet animals exists, including differences in physical activity level, age, life stage, and metabolism. Fortunately, AAFCO regulations will be requiring a calorie statement on all pet foods in the near future. Given the issues listed above, however, it is questionable whether that requirement will aid in obesity prevention or provide false hope to veterinarians and pet food professionals.

**Key Words:** pet obesity

**547 Bioenergetics of pet food.** Ellen Kienzle\* and Britta Dobenecker, *Chair of Animal Nutrition and Dietetics, Ludwig-Maximilians-Universität München, Oberschleissheim Germany.*

Energy of pet food is evaluated as metabolizable energy (ME). To determine ME heat of combustion in food (gross energy, GE), feces and urine is analyzed. Losses by gases are negligible. GE of the food can be calculated by standard values for heat of combustion of nutrients. Given a typical pet food potential errors in the standard values for heat of combustion of nutrients can add up to a deviation of a maximum of about 8% between GE as analyzed and GE as calculated. In practice there are often deviations of up to 30% between analyzed and calculated GE, mostly due to pitfalls of bomb calorimetry. Five repetitions with an intra-analysis variation coefficient of < 0.4% are necessary to ensure quality. To predict digestibility a fixed value for the digestibility of each nutrient can be used. This works well in a group of rather homogenous foods. Examples are unmodified Atwater factors for homemade western diet type foods for humans and pets. If digestibility is variable within a group of foods it is either necessary to make subgroups of foods with similar digestibilities or to use equations which adjust digestibility such as the prediction of digestibility by fiber in dry matter. For this crude

fiber is the analysis of choice because in pet foods it detects mostly unfermentable fiber, which has a stronger impact on energy digestibility than fermentable fiber. Even so subgrouping the foods may present an advantage. The depressive effect of fiber on digestibility is stronger for carbohydrates than for fat, thus subgrouping according to carbohydrate or fat content may increase accuracy of prediction. Subgrouping of foods could even be done by in vitro tests such as an HCl-pepsin test to identify reducing diets with low digestibility of protein. There is little data on renal energy excretion in pets. So far the prediction of renal losses is done by a factor linked to protein intake, which is lower for cats than for dogs in most equations. Presumably the species difference reflects a difference in renal excretion of high energy nitrogen compounds such as hippuric acid, which is derived from phenolic food compounds. Both the content of such compounds in the diet and the ability to degrade these compounds to hippuric acid may contribute to the difference.

**Key Words:** pet food, bomb calorimetry, prediction of digestibility

**548 Animal and in vitro digestion models for estimates of value of energy and energy-yielding nutrients.** R. T. Zijlstra\* and L. F. Wang, *University of Alberta, Edmonton, AB, Canada.*

In vitro digestion (IVD) techniques can evaluate digestibility of energy and energy-yielding nutrients in feedstuffs or complete diets for pigs and other monogastric species. These techniques can mimic the conditions of the gastro-intestinal tract and may have advantages compared with in vivo models of digestion. For example, IVD analyses require less sample material and less time, and are cheaper than in vivo digestibility analyses. Therefore, IVD provides the possibility to screen many samples and support the development of feedstuff databases and rapid feed quality evaluation systems. However, one critical step for IVD analyses is rigorous validation of in vitro digestibility data using the target animal model. For pigs, considerable validation efforts have been made. Using purified enzymes, buffers, and controlled pH, a 3-step IVD model, mimicking digestion in the stomach, small intestine, and large intestine, respectively can estimate the apparent total-tract digestibility (ATTD) of energy. The impact of evaluating the ATTD of energy is clear instantly, because it relates strongly to the DE value of feedstuffs or diets. Similarly, kinetics of glucose release during in vitro digestion of starch is an excellent indicator of net portal appearance of glucose in pigs. Kinetics of fiber degradation can be characterized by in vitro fermentation models, but these models not been validated quantitatively in the pig model. Nevertheless, kinetics of both starch digestion and fiber fermentation are related to important aspects of metabolism of nutrients, gut physiology, and health. In summary, IVD techniques can be a valuable tool to describe the ATTD of energy and kinetics of starch digestion and fiber fermentation of feedstuffs in swine.

**Key Words:** in vitro digestion, energy, pig

**549 Nature's pet food: Energy of raw meat-based and whole prey diets.** Katherine Kerr\*<sup>1</sup> and Cheryl Morris<sup>2</sup>, <sup>1</sup>*University of Florida, Gainesville, FL,* <sup>2</sup>*Iowa State University, Ames, IA.*

Raw and cooked meat-based and whole prey diets for pets have seen increased popularity in past decades. This rise has been attributed to owner perceptions of increased control of dietary ingredients and processing along with feeding more biologically appropriate foods that contributes to the human animal bond. Nutritional therapy with these diet

types can play an integral role in maintaining pet health and, for some pets, in mitigation of disease, such as allergy, gastroenteric diseases, kidney disease and obesity. Meat-based and whole prey diet formulations can mimic the natural history of pets with positive implications for pet health; that is, distribution of energy among macronutrients, dilution of energy and nutrients with moisture – while providing opportunities for modulations that optimize health (i.e., addition of animal or plant fibers). Felines have evolutionarily lacked the need for rapid adaptation to a variety of diet types, and are metabolically prepared for protein and fat energy, with less emphasis on carbohydrate utilization. Similar to the wild type diet of the cat, meat-based and whole prey diets provide > 90% of metabolizable energy (ME) from protein and fat, with little contribution to ME from digestible carbohydrates. Meat-based and whole prey diets also mimic the high moisture of wild type diets (~20 to 30% DM), diluting energy concentration compared with extruded diets (~90% DM). Isocalorically (DM basis), an owner may need to feed 4 times more whole prey on an as-fed basis to provide the same amount of ME as the extruded diet. In wild type diets, fermentable and non-fermentable animal (hide, hair, etc.) and plant fibers play an important role in energy metabolism and gut health: addition of plant and animal fibers, dilutes ME concentrations, decreases inflammation, and beneficially modulates gut bacterial populations. Meat-based and whole prey diets have been underutilized, undervalued, and under-researched as pet foods or as nutritional therapy options. Research has predominately focused on microbial contamination and public health concerns and health implication research are lacking. The flexibility and ingredient control of meat-based diets paired with proper formulations can provide palatable, highly digestible energy sources and targeted nutrient concentrations for pets.

**550 Bioenergetics of pet obesity.** Dennis Jewell\*, *Hill's Pet Nutrition Inc., Topeka, KS.*

Most pet owners in the United States fulfill their pet's nutritional needs through feeding commercially prepared pet foods. These are supplied

with suggested feeding amounts which individual pet-parents adjust to maintain body weight and influence body composition. Significant research efforts have been expended to develop methods for estimating energy concentration of pet foods; however, even the poorest estimates of pet food energy are less variable than the variation of energy need between pets. For example, the estimate for canned cat food has an average difference of 9% and including 2 standard deviations, still had an estimate within 20% of the measured value. The variation of dogs and cats, which are living in the same environment, shows that dogs on average maintain weight consuming 121 kcal/kg<sup>3/4</sup> daily, while the cat uses 73. There is a greater than 2-fold difference between the dog or cat using the least or most amount of energy. Therefore, although the relationship between dietary energy and the bioenergetics of pet food starts with the energy concentration of pet food it is most significantly concerned with the energy use associated with life. Obese dogs and cats use less energy to maintain weight than do their lean counterparts. However, the amount of energy needed to maintain weight in obese pets is subject to change and may be influenced to equal or even exceed the energy needed by normal pets to maintain weight. It has been shown that when dogs were fed a food with increased amounts of fiber there was spontaneous reduction in ad libitum energy intake and repartitioning of energy away from fat so that the energy deficit was completely balanced by energy mobilization from fat. This is also the normal response to food restriction. However, in both dogs and cats a feeding program with controlled weight loss, followed by weight maintenance, when consuming a nutritionally enhanced food, resulted in a significant change in body composition and a significant change in energy needed to maintain weight. This suggests that the bioenergetics of pet obesity is established by the food being eaten, the individual pet, and the feeding regimen.

**Key Words:** bioenergetics, pet, obesity



# Contemporary and Emerging Issues and International Animal Agriculture Symposium: Ahead to 2050—Global livestock production challenges: Current status, future needs, production obstacles

**551 Global livestock production challenges: Current status, future needs, and production obstacles—North America.** Michael Galyean\*, *Texas Tech University, Lubbock, TX.*

Feeding a rapidly growing global population is an enormous challenge. Livestock ownership and production is common in all economies, and demand for animal products grows as economic standards rise, suggesting that livestock will remain an integral component of worldwide food production systems. To achieve global food security, resources used for livestock production must be managed efficiently and sustainably. Current North American (NA) livestock numbers are decreasing or stable, reflecting greater market weight and production efficiency of individual animals and stable or decreased demand for products in NA. Exports of NA livestock products, largely to middle- or upper-income countries, have increased steadily in recent years. Rebuilding animal numbers in some NA livestock segments (e.g., beef cows) will be challenging, which could diminish growth potential. Consolidation in the NA livestock industry has been extensive, which could limit or redirect investments in production infrastructure. Urbanization of agricultural land, increasing regulation, consumer concerns for food safety and environmental effects of livestock production, and a general backlash against technology-based approaches to increase animal growth and efficiency are obstacles to increased production in NA. Challenges associated with climate (e.g., increased drought, decreasing water for irrigation, and shifts in crop production and availability of traditional feedstuffs) are likely but difficult to predict. With these obstacles and challenges, what role will NA livestock production systems play in meeting global food needs? Exporting livestock products should remain an important feature of the NA role, but expansion could be limited by trade issues and will likely continue to focus on developed countries. A potentially significant role for NA to play in meeting worldwide food production needs is to provide leadership in research and development related to sustainable livestock production systems that fit economies and cultures in developing nations. This will require renewed national commitments to support agricultural research and a recognition of the historical and cultural importance of livestock production.

**Key Words:** livestock production, research and development, sustainability

**552 Opportunities and challenges in animal nutrition.** Leo A. den Hartog\*<sup>1,2</sup>, <sup>1</sup>*Nutreco, Boxmeer, the Netherlands*, <sup>2</sup>*Wageningen University, Animal Nutrition Group, Wageningen, the Netherlands.*

The global livestock sector is characterized by differences in dynamics. In developing economies, the livestock sector is evolving in response to rapidly increasing demand for livestock products. In developed economies, demand for animal products is stagnating. Livestock production will increasingly be affected by external factors. These include surging demands for animal products and struggling supplies of feed raw materials, resulting from the competition for natural resources and trade barriers. Simultaneously, there is growing concern about food and its impact on health, and the effect of production systems on animal welfare and the environment. Optimization of productivity and efficiency within such constraints are important objectives, as well as maximization of the profit for all stakeholders. Animal feed and nutrition are the essential

link in the livestock production chain, i.e., between crop cultivation and animal protein production and processing. It is usually the biggest cost factor in livestock production. Several indicators demonstrate that further optimization of animal feed and nutrition is potentially possible. The genetic potential is only partially utilized. The utilization of most nutrients appears to be low and there is a huge variation in performance among farms and within farms, among animals. In addition, environmental performance can be improved significantly. New science and technologies seem to offer many opportunities for innovation in animal feed and nutrition. Key drivers for future innovation are basically (gen) omics, microsystem- and nanotechnology and information and communication technology (ICT). These mainstream technologies are the foundation of many application technologies of relevance for animal feed and nutrition. Acceptance by consumers and society is a critical success factor. In conclusion, animal feed and nutrition are crucial in livestock production. Innovations have the potential to meet the challenges and to result in resource efficiency, healthy livestock and people, responsible production systems and optimal profit throughout the value chain.

**Key Words:** animal nutrition, livestock production, innovation

**553 The future of feed business and live animal production in Asian and Chinese markets to year 2050.** Genfeng (Ian) Yi\*, *Aquaculture Science & Technology Group of Beijing Dabeinong (DBN) Technology Group Co. Ltd., Beijing, China.*

By 2050, the world's population will be 9 to 10 billion people, with at least 40% from Asia. Currently, the feed production volume of Asia contributes about 40%, with Chinese production of 190 million metric tonnes (MMT). The major live animal production countries include China, Indonesia, Thailand, Vietnam, India, Malaysia, and Japan. China is the dominant animal producer, with live pork production about 53 MMT, aquaculture 46 MMT, egg 27 MMT, white and yellow poultry 6 MMT, and ruminant (lamb, sheep, and beef) 5 MMT. With fast economic growth and increasing revenue, urbanization, a growing middle-class population, food safety concerns, environmental pressures, and progress of science and technology, the Asian market, especially the Chinese market, faces numerous development opportunities and challenges. The major challenges for Asian feed and live animal production are as follows: (1) how to supply sufficient safe animal protein and milk to the growing demands of consumers; (2) how to run large-scale and high-input farms efficiently; (3) how to train and attract professionals for live animal production; (4) how to find financial resources for modern agribusiness; (5) how to ensure biosecurity and disease control; (6) how to deal with the conflict of the environment and increased live animal production; (7) how to control soaring feed ingredient costs and reduce overall live animal production cost; (8) how to solve international trading interests and conflicts; (9) how to upgrade and use Internet and "web of things" technology for modern agribusiness; (10) how to integrate and consolidate industry chains and face international competition; (11) molecular and disease-resistance breeding technology; (12) how to innovate and apply new science and technology; (13) organic animal production; (14) antibiotic residue issues; (15) impact of climatic change on animal and aquaculture production (Asia accounts for 90% of world aquaculture production); (16) limited water and land resources; (17) GMO and transgenic issues of plants and their effect on animal produc-

tion; (18) nanotechnology; (19) bioactive substances and biotechnology application; and (20) local governmental policies and regulations. The future of live animal production must include high production efficiency, low carbon production, organic foods, and minimal production cost and maximal production profitability. Asian live animal production needs to be more environmentally friendly, health-oriented, nutritious, safe, sustainable, and affordable to consumers.

**554 Ahead to 2050—Latin American livestock production challenges.** Fernando Rutz\*, *Universidade Federal de Pelotas, RS, Brazil.*

According to FAO, the world's population will be over 9 billion people and food production will need to be increased by 70% by the year 2050. Latin America (LA) has the potential to be an important player in providing food to fulfill this requirement, having the largest global fresh water reservoir, and one third of the world's available land for sustainable livestock production. With a wide range of climate zones, LA has diversified agriculture capabilities, ranging from tropical- to temperate-zone products. However, there is a lot of productivity variation among countries and even among different regions within a country. From the ruminant side, LA has suitable production conditions for increased beef, dairy and sheep production. From the monogastric point of view, the swine, poultry and aqua industries are well developed and constantly growing. This capability is exemplified by the fact that Brazil is now the largest beef and chicken meat exporter in the world. The dairy industry has also been experiencing significant growth and development during the last 2 decades. In almost all LA regions, milk (M) and milk-derivatives (MD) are increasing and diversifying into more complex products. In addition, M and MD export have been increasing in some LA countries. In spite of that, within some countries in LA and Caribe the average consumption of milk is below 25% of requirement. This fact shows the need to stimulate production and consumption of milk in some regions. To cope with the demands of food production, LA needs to increase the efficiency of production. To do this it can use technologies to supplement on pasture, increase grazing-tillage integration, and use more intensive livestock systems to shorten the time to slaughter. Use of tools like predicting feed efficiency with in

vitro fermentation, development of DNA technology, exploring gene expression (nutrigenomics), genetic improvement and crossbreeding, artificial insemination, embryo transfer and sexed semen, can improve production capabilities too. Improvements in livestock health, product safety and quality, and control global warming gas emissions are also essential. Increased LA livestock production efficiency will result in decreased land degradation, and natural resource conservation.

**Key Words:** ruminant, monogastric, climate

**555 Global livestock production challenges: Current status, future needs, production obstacles in Africa—The Uganda**

**example.** L. Okedi\*<sup>1</sup>, Y. Baguma<sup>2</sup>, J. M. Kabirizi<sup>1</sup>, and J. Kungu<sup>1</sup>,  
<sup>1</sup>*National Livestock Resources Research Institute, Tororo, Uganda,*  
<sup>2</sup>*National Agricultural Research Organization Secretariat, Entebbe, Uganda.*

The livestock sector in Uganda contributes 5.2% to the national gross domestic product and 18% to the overall agricultural GDP. National livestock numbers consist of 12.8 million cattle, 12.5 million goats, 4 million sheep, 3.6 million pigs and 42 million chickens. A total of 5 million households in Uganda own livestock (UBOS, 2008). Farming households that include livestock in their enterprise mix tend to have higher incomes than those involved only in crop farming. This is a result of livestock ownership and also improved crop productivity in that livestock manure improves nutrient availability to plants and increases soil organic matter. Economic benefits of including livestock production within farming enterprises are the greater accumulation of assets and the ability to secure credit. Uganda leads Africa in pork consumption, but cattle, goats, poultry and fish still provide significant sources of dietary animal protein. The expansion of meat and milk production in Africa has been disappointing despite efforts to stimulate increased production. Constraints cut across socioeconomic, technological, institutional and financial sectors. As well, little attention has been given to the development of the livestock value chain due to uncertainty in feed availability, reproductive inefficiency, the lack of livestock genetic improvement, public and animal health issues, and the prevailing policy environment.

**Key Words:** Africa

## Dairy Foods: Cheese and chemistry

**556 Release of bioactive peptides and essential amino acids as affected by sodium chloride reduction and substitution in Akawi cheese.** Akanksha Gandhi\* and Nagendra P. Shah, *The University of Hong Kong, Hong Kong.*

The aim of this study was to evaluate the effects of sodium chloride reduction and its substitution with potassium chloride on selected probiotic bacteria and their functionality in Akawi cheese during storage for 30 d at 4°C. The survival of selected probiotic bacteria (*Lactobacillus acidophilus*, *Lactobacillus casei* and *Bifidobacterium longum*) and starter bacteria (*Streptococcus thermophilus* and *Lactobacillus delbrueckii* ssp. *bulgaricus*), the angiotensin-converting enzyme-inhibitory and antioxidant activities, and the concentrations of essential amino acids as affected by storage in different brine solutions [10% NaCl, 7.5% NaCl, 7.5% NaCl+KCl (1:1), 5% NaCl and 5% NaCl+KCl (1:1)] were investigated. Peptide profiles of cheese extracts, from different salt concentrations, were observed by RP-HPLC and in vitro bioactivities of the peptide fractions were evaluated using Caco-2 cell line. Survival of probiotic bacteria increased significantly in cheeses with low salt after 30 d, and after 20 d for starter bacteria. No significant difference in texture profile of cheeses during 30-d storage was observed except for fracturability and gumminess, which was found to be lesser in cheeses brined with only NaCl solutions. No significant changes were observed in ACE-inhibitory activity and antioxidant activity of the cheeses during storage. Interestingly, concentrations of 4 essential amino acids (phenylalanine, tryptophan, valine and leucine) increased significantly during storage in brine solutions containing 7.5% (w/v) total salt. The study revealed that at total salt concentration of 7.5% (w/v), 50% substitution of NaCl with KCl in Akawi cheese would not affect the activities of potential bioactive peptides, and would lead to increased release of essential amino acids.

**Key Words:** salt reduction, cheese, essential amino acids

**557 Generation of highly antioxidative peptides from purified bovine  $\alpha_{s2}$ -casein.** Zahur Z. Haque\* and Xue Zhang, *Department of Food Science, Nutrition and Health Promotion, Mississippi State University, Mississippi State, MS.*

Alpha<sub>s2</sub>-casein is particularly rich in  $\pi$  electron rich aromatic amino acids that, in the absence of structural constraints, partake in  $\pi$ - $\pi$  stacking and interact with positively charged residues (cation- $\pi$ ) resulting in electronic continuity required for rapid quenching of lone-pairs of oxidative radicals. This study investigated isolation of  $\alpha_{s2}$ -casein from bovine milk, standardization of its chymotryptic hydrolysis, visualization of resulting peptide profiles by tricine-SDS-polyacrylamide gel electrophoresis (TSDS-PAGE) and size-exclusion high performance liquid chromatography (SE-HPLC), and determination of the antioxidative efficacy of the fractions. Alpha<sub>s2</sub>-casein was isolated from whole casein by a 2-step 1-proponal-precipitation method and hydrolyzed using chymotrypsin (EC 3.4.21.1) because it selectively cleaves peptide bonds adjacent to aromatic residues, increasing the chances for inter-peptide aromatic-stacking. Five mg/mL of  $\alpha_{s2}$ -casein was hydrolyzed with chymotrypsin (50  $\mu$ g/mL) for 5–60 min at 37°C. A novel fixing method with sodium hypochlorite was used to fix the highly amphipathic peptides after TSDS-PAGE and visualization was by silver staining. The original protein was completely hydrolyzed to smaller peptides within 5 min. Peptide profiles determined by SE-HPLC, using a Superdex Peptide 10/300 GL column and 30% acetonitrile (v/v) as the eluent, substantiated the effectiveness

of the peptide fixation following TSDS-PAGE peptide profiling. Average molecular weight of each fraction was determined by comparing its retention time with those of molecular weight standards. Data depicted an inverse relationship between molecular weight and the duration of enzymatic hydrolysis. Average molecular weight of the smallest fraction at 5 min of hydrolysis was 981 Da, and it was gradually reduced to 130 Da after 60 min of hydrolysis. As expected all hydrolyzates had oxygen radical absorbance capacity values (2000–3000  $\mu$ M TE) that were significantly ( $P < 0.05$ ) higher than that of the original protein (580  $\mu$ M TE). Correlation of the antioxidative efficacy with residual secondary structural constraints and intensity of association tendency of the peptide fractions is presently being investigated.

**Key Words:** milk protein, reactive oxygen species, oxygen radical absorbance capacity

**558 Efficacy of Cheddar whey combined with chitosan in edible coatings to reduce protein-fat oxidation and accumulation of reactive oxygen species in retail-cut catfish fillet.** Zahur Z. Haque\* and Dipaloke Mukherjee, *Department of Food Science, Nutrition and Health Promotion, Mississippi State University, Mississippi State, MS.*

Retail-cut muscle foods are especially susceptible to oxidative degradation and formation of harmful reactive oxygen species (ROS) owing to exposure to oxygen, bright lights and contact with metal counters. Due to extended cooking at temperatures near the pH optima of most of starter culture proteases during its Cheddar cheese manufacture, the resulting whey (CW) is naturally rich in antioxidative peptides and Maillard reaction products. In the present study, we report the antioxidative efficacy of CW when used in an edible coating, with and without chitosan (Ch), to reduce protein and fat degradation in stored farm-raised retail-cut catfish and the accumulation of ROS in the muscle. Fresh catfish fillet of uniform thickness were cut into cubes of unvarying weight (5g) and geometry, and immersed in dispersions of CW (2% w/v), Ch (1%, w/v) or mixture of both (CW+Ch) in 0.2 M Mcllvaine's iso-ionic buffer (pH 7) at 22°C for 2 min. Protein and lipid oxidation [evident from carbonyl contents (CC) and peroxide values (PV)] of the samples were determined after storage at 4°C for nil, 1, 3, 5 and 7 d. Accumulation of ROS at cellular level was studied using a fluorogenic molecular probe (CellROX Deep Red Reagent, Life Technologies). Results showed significantly ( $P < 0.05$ ) superior efficacy of the coatings in reducing overall oxidative degradation of samples for most of the storage periods compared with control (immersed only in the buffer). The CW+Ch mixture showed dramatic effects in alleviating lipid oxidation and ROS accumulation. It also reduced CC by ~60% compared with control after 3 d of storage, thus indicating a considerable extension in shelf life. The PV was reduced to ~22 and 16% in the samples immersed in CW+Ch and CW, respectively, after 5 d of storage compared with control. The same samples also exhibited 28 and 21% reduction in ROS accumulation relative to control after the same number of days of storage. The results indicated a vivid potential of this combined approach for combating oxidative degradation of foods and prevention of the formation/accumulation of harmful ROS in foods.

**Key Words:** reactive oxygen species, muscle food, sweet whey.



**559 Influence of fish oil alone or in combination with hydrogenated palm oil on sensory characteristics and fatty acid composition of bovine cheese.** Einar Vargas-Bello-Pérez<sup>\*1</sup>, Gonzalo Ñiguez-González<sup>1</sup>, Karen Fehrmann-Cartes<sup>1</sup>, Paula Toro-Mujica<sup>1</sup>, and Philip C. Garnsworthy<sup>2</sup>, <sup>1</sup>*Pontificia Universidad Católica de Chile, Santiago, Chile*, <sup>2</sup>*The University of Nottingham, Loughborough, UK*.

The objective of the present study was to evaluate the effect of dietary supplementation of fish oil (FO) alone or in combination with hydrogenated palm oil (FOPO) on the fatty acid (FA) profile of milk and cheese from dairy cows and the sensory characteristics of cheese. Nine Holstein cows (173 ± 21 DIM) were used in a replicated 3 × 3 Latin square design with 21-d periods. Dietary treatments consisted of a basal diet (Control; no fat supplement), and fat-supplemented diets containing fish oil (FO; salmon oil; 500 g/d/cow) and FO (250 g/d/cow) + hydrogenated palm oil (PO; palm oil; 250 g/d/cow). Milk collected on d 21 was pooled from 3 cows within the same treatment and period and made into cheese. Three cheeses per treatment per period at 14 d of aging were used for sensory evaluation. Cheese color measurements were determined according to the CIE L\*, a\*, and b\* LAB color system. Sensory evaluation considered the following attributes: color homogeneity, holes, overall odor, ripe cheese odor, cow milk odor, salty, acid, bitter, overall flavor, ripe cheese flavor, sharpness, toughness, graininess, screeching, moisture and greasiness. Except for milk lactose yield, milk and cheese components were not affected by dietary treatments. Milk and cheese contents of C6:0, C8:0, C10:0 and C14:0 and atherogenicity index were lower ( $P < 0.05$ ) with FO and FOPO diets than control diet. Compared with control and FOPO, FO increased ( $P < 0.05$ ) C18:1 trans 11 and C22:6n3 content in milk and cheese. The color obtained from cheeses elaborated from cows given FO resulted in a more ( $P < 0.05$ ) intense yellowness than control and FOPO. Compared with control and FOPO, FO resulted in higher ( $P < 0.05$ ) notes for color homogeneity, whereas in comparison with control and FO, FOPO resulted in higher ( $P < 0.05$ ) tough texture. In conclusion, supplementation of dairy cow diets with FO alone or in combination with hydrogenated palm oil can enhance the FA profile of milk and cheese without deleterious effects on sensory characteristics of cheese. This study was sponsored by a research grant from FONDECYT 11121142 (Fondo Nacional de Desarrollo Científico y Tecnológico, Chile).

**Key Words:** cheese, fatty acids, fish oil

**560 Comparison between whey dilution during cheese-making and standardization of milk lactose by ultrafiltration on the properties of low and reduced fat Gouda cheese.** Rodrigo A. Ibáñez<sup>\*1,2</sup>, Selvarani Govindasamy-Lucey<sup>3</sup>, John J. Jaeggi<sup>3</sup>, Mark E. Johnson<sup>3</sup>, Paul L. H. McSweeney<sup>1</sup>, and John A. Lucey<sup>2,3</sup>, <sup>1</sup>*University College Cork, Cork, Ireland*, <sup>2</sup>*University of Wisconsin-Madison, Madison, WI*, <sup>3</sup>*Wisconsin Center for Dairy Research, Madison, WI*.

In recent years, the consumption of cheeses with reduced fat content has experienced growth due to health concerns. However, these products are associated with a hard and rubbery texture, poor melting properties and excessive development of acidity. In the manufacture of Gouda cheese, some whey is removed and replaced with water to decrease the residual lactose content and hence to control pH. This step is also known as whey dilution (WD) and may be a cause of high variability depending on the amount of water added, temperature, holding times and stirring rates. As an alternative, the standardization of lactose content in cheesemilk by ultrafiltration (UF) prior cheese manufacture could potentially reduce the residual lactose content in cheese and thus control the final pH. This study aimed to compare the effect of WD during cheese manufacture,

with the alternative approach of adjustment of the lactose content of cheesemilk using UF, on the composition, texture, functionality and sensory properties of reduced-fat (RF) and low-fat (LF) Gouda-type cheeses during 6 mo of ripening. A stirred curd direct-salted cheese manufacture was used, differing in the levels of WD at 30, 15 and 0% (WD<sub>30</sub>, WD<sub>15</sub> and WD<sub>0</sub>, respectively). The RF and LF milks used in WD<sub>30</sub> and WD<sub>15</sub> had a lactose-to-casein (L:C) ratio of ~1.8, which is the typical ratio found in milk. The WD<sub>0</sub> treatments were made with UF standardized milks to L:C ratio of ~1.1. Similar trends between treatments were observed in both RF and LF treatments. WD<sub>0</sub> exhibited lower residual lactose and lactic acid contents than WD<sub>30</sub> and WD<sub>15</sub>, leading to higher pH values ( $P < 0.05$ ). WD<sub>0</sub> had softer texture and were more meltable ( $P < 0.05$ ), probably due to a lower proportion of insoluble Ca caused by the addition of water required to achieve the lower L:C ratio in UF milks. Sensory analysis also indicated that WD<sub>0</sub> cheese had lower acidity and softer texture. These results suggest that UF standardization of the L:C ratio of cheesemilk could be a useful alternative to WD to reduce the acidity, improve texture and functionality of reduced- and low-fat Gouda cheese.

**Key Words:** Gouda cheese, lactose standardization, whey dilution

**561 Growth and gas formation by a novel obligatory heterofermentative nonstarter lactic acid bacterium in cheese made using a *Streptococcus thermophilus* starter.** Fatih Ortakci<sup>\*1</sup>, Jeffery Broadbent<sup>1</sup>, Craig Oberg<sup>2,1</sup>, and Donald McMahon<sup>1</sup>, <sup>1</sup>*Utah State University, Logan, UT*, <sup>2</sup>*Weber State University, Ogden, UT*.

A novel slow-growing obligatory heterofermentative nonstarter lactic acid bacterium, *Lactobacillus wasatchii* sp. nov., was studied for growth and gas production in Cheddar cheese made using a *Streptococcus thermophilus* starter. Cheesemaking trials were conducted using starter *St. thermophilus* alone or in combination with *Lb. wasatchii* deliberately added to cheese milk at a level of ~10<sup>4</sup> cfu/ml. Then cheeses were ripened at 6 or 12°C. At d 1, starter streptococcal numbers were similar in both cheeses (~10<sup>9</sup> cfu/g) and nonstarter lactic acid bacteria (NSLAB) counts were below detectable levels (<10<sup>2</sup> cfu/g). As expected, *Lactobacillus wasatchii* counts were 3 × 10<sup>5</sup> cfu/g in cheeses inoculated with this bacterium. Starter streptococci decreased over time at both ripening temperatures but fell more rapidly at 12°C, especially in cheese with *Lb. wasatchii* ( $P < 0.05$ ). Populations of NSLAB and *Lb. wasatchii* reached 5 × 10<sup>7</sup> and 2 × 10<sup>8</sup> cfu/g, respectively after 16 wk of ripening at 12°C, and their emergence was correlated with ~0.5% reductions in galactose concentrations. Levels of galactose at 6°C had also similar decrease after 16 wk storage. Gas formation and textural defects were only observed in cheese with added *Lb. wasatchii* ripened at 12°C. Results demonstrate that, *Lb. wasatchii* can contribute to late gas blowing in Cheddar cheese made with *St. thermophilus*, especially when the cheese is ripened at elevated temperature.

**Key Words:** cheese late blowing, nonstarter lactic acid bacteria, *Streptococcus thermophilus*

**562 Late blowing of Cheddar cheese induced by accelerated ripening and ribose and galactose supplementation in presence of a novel obligatory heterofermentative nonstarter lactobacilli species.** Fatih Ortakci<sup>\*1</sup>, Jeffery Broadbent<sup>1</sup>, Craig Oberg<sup>2,1</sup>, and Donald McMahon<sup>1</sup>, <sup>1</sup>*Utah State University, Logan, UT*, <sup>2</sup>*Weber State University, Ogden, UT*.

A novel nonstarter lactic acid bacterium, *Lactobacillus wasatchii* sp. nov. has been studied for growth and gas formation in a control Cheddar

cheese and in cheese supplemented with 0.5% ribose, 0.5% galactose, or 0.25% ribose plus 0.25% galactose using regular and accelerated cheese ripening temperatures of 6 and 12°C. Cheese milk along with starter lactococci was inoculated with *Lb. wasatchii* at a level of 10<sup>4</sup> cfu/mL, whereas a control vat was inoculated with starter lactococci only. Starter numbers in both cheeses decreased from 10<sup>7</sup> to ~10<sup>3</sup> cfu/g at 23 wk of ripening at both temperatures, except the control cheese at 6°C which had one log higher final cell counts. Unlike starter bacteria, nonstarter lactic acid bacteria started at <10<sup>2</sup> cfu/g in the cheese and reached 10<sup>6</sup> to 10<sup>7</sup> cfu/g with higher numbers observed at 12°C. *Lactobacillus wasatchii* grew to ≥10<sup>8</sup> cfu/g in cheese supplemented with ribose (alone or with galactose) at elevated temperature which was ~1-log higher compared with the control and galactose-supplemented cheeses. In all cheeses with adjunct *Lb. wasatchii*, highest growth and gas formation was observed at 12°C although most gas production occurred at ≥16 wk. Adding both ribose and galactose provided substantially higher growth and gas formation because of the ability of *Lb. wasatchii* to co-utilize both sugars; producing gas from galactose as a result of the obligatory heterofermentative nature of the bacterium. Even without sugar supplementation, gas was observed in the presence of adjunct *Lb. wasatchii* after 16 wk. We have observed that *Lb. wasatchii* can grow to high cell densities when grown in carbohydrate-restricted broth containing lactococcal cell lysate. During cheese ripening, lysis of starter bacteria would provide sufficient substrate (such as ribose) to allow growth of *Lb. wasatchii* during cheese ripening and the presence of any hexoses in cheese would allow *Lb. wasatchii* to produce gas. We conclude that *Lb. wasatchii* is a previously undetected contributor to late gas formation in Cheddar cheese and the defect is more pronounced when elevated ripening temperatures are used.

**Key Words:** nonstarter lactic acid bacteria, gas, cheese

**563 Demonstration of pH micro-heterogeneity in cheese matrices by fluorescence microscopy.** Zuzana Burdikova<sup>1</sup>, Zdenek Svindrych<sup>2</sup>, Jan Pala<sup>3</sup>, Cian D. Hickey<sup>1,4</sup>, Martin G. Wilkinson<sup>4</sup>, Jiri Panek<sup>5</sup>, Mark A. E. Auty<sup>1</sup>, Ammasi Periasamy<sup>2</sup>, and Jeremiah J. Sheehan<sup>\*1</sup>, <sup>1</sup>*Teagasc Food Research Centre Moorepark, Fermoy, Co. Cork, Ireland*, <sup>2</sup>*Department of Biology, University of Virginia, Charlottesville, VA*, <sup>3</sup>*Third faculty of Medicine, Charles University, Prague, Czech Republic*, <sup>4</sup>*Dept of Life Sciences, University of Limerick, Ireland*, <sup>5</sup>*Institute of Macromolecular Chemistry, Academy of Sciences of the Czech Republic, Prague, Czech Republic*.

Cheese, a product of microbial fermentation, may be defined as a protein matrix entrapping fat, moisture, minerals and solutes as well as dispersed bacterial colonies. The growth and physiology of bacterial cells in these colonies may be influenced by the microenvironment around the colony, or alternatively the cells within the colony may modify the microenvironment (e.g., pH, redox potential) due to their metabolic activity. To date there remains a significant knowledge gap relating to the degree of micro-heterogeneity in compositional and biochemical parameters such as pH within the cheese matrix and its relationship with microbial, enzymatic and physicochemical parameters and ultimately with cheese quality, consistency and ripening patterns. The objective of this study was to investigate whether pH micro-heterogeneity exists in cheese matrices. For the first time and arising from the development of a method using fluorescent lifetime imaging (FLIM) microscopy with Oregon green 488 dye, it is now possible to examine cheese for localized differences in pH. A ripened, semi-hard, dry salted cheese manufactured with thermophilic cultures was analyzed. Our study showed micro-heterogeneity in pH within the matrix of that cheese with pH ranging between 4.0 and 5.5. This is particularly interesting as it shows,

contrary to previous assumptions, that the pH of a cheese matrix is not homogenous at micro-scale but contains localized variation. This may be due to localized differences in the aqueous phase or concentrations of constituents of the aqueous phase including lactose, lactate, minerals or salt. It may also be influenced by variations in buffering capacity of the surrounding cheese matrix. It is envisaged that future work using this method will focus on determining whether manufacture processes influence pH at local level within different cheese matrices and whether different cheese types may have different patterns of micro heterogeneity. It is also envisaged that this methodology will be employed to examine a broad range of food matrices in differing food products.

**Key Words:** pH, micro-heterogeneity, cheese

**564 Evaluation of X-ray fluorescence spectroscopy for determination of minerals in process cheese.** Catherine Shawl<sup>\*1</sup>, Jordan S. Rose<sup>2</sup>, and David R. McCoy<sup>3</sup>, <sup>1</sup>*Kraft Foods Group, Glenview, IL*, <sup>2</sup>*Oxford Instruments, Concord, MA*, <sup>3</sup>*Dairy Management Inc., Rosemont, IL*.

X-ray fluorescence spectroscopy (XRF) has been demonstrated to have the ability to rapidly and directly measure sodium in a variety of cheeses by Stankey et al. Their rapid and simple method used a disk of cheese as the sample. However, inaccuracies may result from the formation of a thin layer of butter oil forming between the cheese and the surface of the holder and from using cheese samples that do not have a uniform surface such as shredded or ground cheeses. It is also important in process cheese manufacture to control the concentration of chloride, phosphorus and potassium as well as sodium. This study investigated various modifications to the sample preparation procedures for XRF technology that would remove the mentioned limitations, expand the minerals assayed, and evaluate the feasibility of using XRF in a manufacturing situation. The optimum sample preparation was created by homogenizing process cheese into 0.1 N nitric acid at 50°C. The homogenate was then centrifuged for 2 min in a microcentrifuge to separate the sample into a solids, an aqueous, and an oil layer. Two to 3 mL of the aqueous layer was removed from the centrifuge tube and assayed in an Oxford Instruments X-Supreme8000 Energy Dispersive X-Ray Fluorescence (EDXRF) spectrometer. Instrument assay time was 3 min, which allowed simultaneous measurement of the minerals of interest. Total time from sample submission to result was 15 min, making it rapid enough for in-plant quality control use. Relative standard deviation for multiple trials with the same block of process cheese product was 3.0% for sodium, 6.5% for phosphorus, 0.9% for chlorine, and 1.4% for potassium in preliminary studies. The instrument calibration to a reference ICP method has an R<sup>2</sup> of 0.915 for sodium using a straight line fit. The modified XRF method extends the range of minerals assayed and addresses the causes of the inaccuracies that had limited the desirability for industrial use.

**Key Words:** sodium, cheese, assay

**565 Novel sample preparation for smear ripened cheese rinds evaluated by powder X-ray diffractometry.** Gil F. Tansman<sup>\*1</sup>, Paul S. Kindstedt<sup>1</sup>, and John M. Hughes<sup>2</sup>, <sup>1</sup>*Department of Nutrition and Food Sciences, University of Vermont, Burlington, VT*, <sup>2</sup>*Department of Geology, University of Vermont, Burlington, VT*.

Recently we developed methods to evaluate surface crystals in Cheddar and hard Italian-style cheeses, and internal crystals in Cheddar, Gouda, and Parmigiano Reggiano cheeses by powder X-ray diffraction (PXRD). However, new sample preparation techniques are needed to analyze

surface crystals of washed rind cheeses because of the complex nature of the surface smear. Our objectives were to develop repeatable sample preparation protocols for smear evaluation by PXRD and to validate the protocols by single crystal X-ray diffraction (SCXRD). Initial efforts to evaluate crystalline inclusions in smears involved scraping the surfaces of 3 washed rind cheeses with a spatula and loading them onto PXRD slides. Fresh samples were used deliberately because ikaite ( $\text{CaCO}_3 \cdot 6\text{H}_2\text{O}$ ), which in cheese has thus far only been observed in surface smears, decomposes in the presence of acetone. Diffraction patterns showed limited repeatability between repeated measures when data collection was performed in succession at roughly 25 min intervals. The lack of repeatability was found to be an artifact of sample desiccation during data collection. To prevent drying during data collection, 2 modifications were investigated; sample slides were coated in a thin layer of immersion oil, and alternatively allowed to air-dry before pul-

verization in a mortar. PXRD data collected using both PXRD method modifications revealed that all 3 smears contained ikaite and that one of the smears also contained struvite ( $\text{NH}_4\text{MgPO}_4 \cdot 6\text{H}_2\text{O}$ ). The presence of ikaite and struvite was confirmed by single crystal x-ray diffraction (SCXRD) in the corresponding cheeses. It appears that smear material used in PXRD requires additional prep due to its extensive loss of volume upon drying, which causes diffraction artifacts. As confirmed by SCXRD, both modified PXRD methods provide valid diffraction data on the identities of crystals present in smear samples. Although SCXRD is a superior method for identifying the single crystals from surface smears, a reliable PXRD method is useful because powder diffractometers are typically less expensive, more widely available, and require less training to operate.

**Key Words:** cheese, crystals, PXRD



# Dairy Foods Symposium: Recent developments in manufacturing and applications of lactose and lactose derivatives

**566 Overview of technological advances in manufacture and applications of lactose and lactose derivatives: Present and future.** Shantanu Agarwal\*, *Dairy Management Inc., Rosemont, IL.*

Lactose is a major component of milk and has been extensively studied over the years. Much work has focused on the manufacture and application of lactose. Lactose and its derivatives have wide physico-chemical and bioactive properties making it appropriate for a wide range of applications food, animal feed, and pharmaceuticals. Primary uses of lactose in the food industry are largely based on its relatively low sweetness, protein stabilizing ability, great crystallization attributes, ability to accentuate flavor/ color and significant nutritional value. Extensive research continues on various fronts as to how to bring added value to lactose and its derivatives in various food and nutraceutical applications. In this presentation, technological advancements and challenges in manufacture of lactose from various product streams (e.g., permeate, acid whey) will be reviewed. The presentation will also review latest research in understanding drivers and inhibitors for lactose crystallization. The role of lactose and its derivatives in various food applications will be discussed with regards to attributes such as product quality and sensory attributes (color, texture, mouthfeel, and sweetness). Nutritional significance of lactose and its derivatives will be discussed in relation to glycemic index, dietary fiber, and mineral absorption. Last, this presentation will focus on the future growth opportunities for lactose and its derivatives.

**Key Words:** lactose, permeate, crystallization

**567 Lactose and lactose derivatives: More than prebiotics?** Michael Gänzle\*, *University of Alberta, Edmonton, AB, Canada.*

Commercially available lactose derivatives include lactulose ( $\beta$ -4' galactosylfructose), lactosucrose ( $\beta$ -4' galactosylsucrose), and galacto-oligosaccharides. 3-, 4-, and 6-Galactobiose, allolactose, and 6-, 3-, and 4-galactosylsucrose are the major components of commercial galacto-oligosaccharide preparations. Commercial processes for lactose conversion generally employ enzymatic transglycosylation with fructose, sucrose, and lactose, respectively, as galactosyl acceptors. Because human  $\beta$ -galactosidase is specific for lactose, enzymatic transgalactosylation converts lactose, a disaccharide that is indigestible in about 75% of human adults, to oligosaccharides that are indigestible in all human infants and adults. Galacto-oligosaccharides and other lactose derivatives are fermented by colonic microbiota. Galacto-oligosaccharides increase the abundance of bifidobacteria in intestinal microbiota and are thus recognized as prebiotic compounds. However, the increase of bifidobacteria in human microbiota depends on the individual composition of the microbiota and is observed in most but not in all individuals. Health benefits of consumption of galacto-oligosaccharides are predominantly derived from intestinal fermentation to short chain fatty acids and may be independent of the stimulation of specific taxa in the human gut. In analogy to lactose intolerance, over-consumption of galacto-oligosaccharides results in intestinal discomfort and osmotic diarrhea. In addition to their prebiotic activity, galacto-oligosaccharides have been shown to prevent the adhesion of enteric pathogens to mucosal surfaces. Their activity is likely mediated by specific interaction with bacterial glycan recognition proteins that are involved in early steps of pathogenesis. The prevention of pathogen adhesion is a complementary mechanism of galacto-oligosaccharides to specifically benefit host

health. The concept of preventing pathogen adhesion was validated in vivo with swine models; anti-adhesive glycans may be used as functional food ingredient but also in animal production to reduce the use of antimicrobial growth promoters.

**Key Words:** lactose  $\beta$ -galactosidase galacto-oligosaccharide, pathogen adhesion

**568 Role of lactose in dairy and food products: State of understanding.** Tonya C. Schoenfuss\*, *University of Minnesota, St. Paul, MN.*

Lactose is the most abundant nonfat solid in milk and its properties present both opportunities and challenges for its presence in food and dairy products. Important properties that affect dairy ingredient use due to lactose are crystallization, stickiness, hygroscopicity, and reactions it participates in. Positive attributes of lactose include its low sweetness, low cost, and the ability to participate in non-enzymatic (Maillard) browning reactions because it is a reducing sugar. It is also important as a solids replacer to balance mouth feel and the solution properties in beverages, ice cream mixes, and the liquid phase during baking. The effect of lactose on the viscosity of the solution phase can alter the ability to hold gas during baking, and maintain ice crystal stability during the storage of frozen products. Thermodynamic parameters that are measured such as the glass transition ( $T_g$ ), melting temperature ( $T_m$ ) and temperature of crystallization ( $T_{cr}$ ) are useful to evaluate the effects of processing and other ingredients on lactose state and stability. But in formulated food systems, it can be complicated to attribute the functional attributes entirely to these measured properties. This objective of this symposium talk is to focus on the challenges of using lactose in formulated foods, and what is understood about the important chemical, physical and thermodynamic properties involved in caking and crystallization. Challenges with using lactose in beverages, chocolate, frozen desserts and bars will be highlighted, as will methods to avoid undesirable changes to texture, viscosity and handling characteristics.

**Key Words:** lactose, food

**569 Industry perspective on managing quality and yield of lactose: From food to pharmaceutical grade.** A. Kent Keller\*, *Keller Technologies Inc., Mantorville, MN.*

Lactose has been commercially produced since the early part of the twentieth century. Worldwide production increased phenomenally in the 1940s when it was found that lactose was the best substrate for penicillin production. Further increases in lactose production resulted when mothers started to use infant formula, which contains lactose as the major constituent. Late in the twentieth century, confectioners found many reasons to use crystalline lactose in chocolates. Today chocolates are second only to infant formulae as the major use of lactose. In this presentation, the author shares some of his 40 years of experience operating and designing commercial-scale lactose production systems. Each unit operation for lactose production is discussed with a focus of how each unit operation can affect lactose quality and yield. The following unit operations are covered: raw material supply, concentration (RO and evaporation), crystallization, refining, drying and packaging. Particular attention will be given to the current state of the art, which is not typi-

cally covered in various textbooks. Finally, a review of traditional and new methods for producing pharmaceutical grade lactose are discussed.

**Key Words:** lactose, lactose production

**570 Processing and stability of high lactose powders .** Thom Huppertz\*<sup>1,2</sup>, Inge Gazi<sup>1</sup>, and Hasmukh Patel<sup>2</sup>, <sup>1</sup>*NIZO food research, Ede, the Netherlands*, <sup>2</sup>*South Dakota State University, Brookings, SD*.

Lactose strongly affects the stability of dairy products on drying and storage. In skim milk powder, WPC35 and infant formula, lactose represents >50% of dry matter and forms an amorphous matrix in which other constituents are dispersed. When temperature and water activity exceed critical values, the amorphous lactose matrix changes to a rubber-like state, where the powder can become sticky. Lactose crystallization and Maillard reactions can also occur as self-propagating reactions due to water release from amorphous lactose on crystallization and the reaction between the carbonyl and the amino group, thus further increasing water activity. Hence strict control of heat-load and drying conditions and prevention of moisture gradients (e.g., in dry-blending ingredients) are needed to maintain powder properties. Otherwise, lumping, caking and other handling properties can occur. Moisture sorption isotherms

and glass transition temperatures can be predicted based on composition and can be used as input parameters for optimization of processing conditions. For powders with even higher lactose content, e.g., permeate or whey powder, pre-crystallization of lactose in the concentrate before drying is required to avoid development of excessive stickiness during drying. Pre-crystallization is achieved by (flash) cooling and addition of seeding crystals. Optimization of pre-crystallization conditions (temperature and solids content) in relation to crystallization rate and viscosity is needed to attain sufficient crystallization within an acceptable time. Typically, up to 75% of lactose can be crystallized by pre-crystallization. Post-crystallization after drying can further increase crystallization levels, up to 95%. Failure to attain sufficiently lactose crystallization (i.e., >75%) can lead to caking, lumping and browning when powders are stored. Other constituents also influence the behavior of lactose during processing and storage. Lactose-phosphate, formed during fermentation, is a potent inhibitor of lactose crystallization, whereas lactate salts present can add considerable hygroscopicity to powders. Overall, careful control of composition, processing and storage conditions is paramount to attain and maintain high-lactose powders of desirable properties.

**Key Words:** lactose, powder, processing

## Nonruminant Nutrition: Feed ingredients

**571 Nutrient profile and digestibility of agro-industrial coproducts as determined using an in vitro model of swine.** Utsav P. Tiwari\*, Halina M. Zaleski, and Rajesh Jha, *University of Hawaii at Manoa, Honolulu, HI.*

Market availability and price of some conventional feedstuffs, such as corn, wheat, and soybean meal, are highly variable because of high demand for food, feed and fuel. Exploring alternative feedstuffs that can either completely or partially replace these feedstuffs is very important to ensure the sustainability of the swine industry. Use of these alternative feedstuffs in pig diets can be optimized by characterizing their nutrient profile and digestibility. To explore the nutritional value, 3 agro-industrial coproducts, barley brewers grain (BBG), okara and wheat millrun (WMR) were analyzed for basic nutrients, fiber and GE content. Digestibility of DM and GE was determined using an in vitro 3-step enzymatic assay using pepsin, pancreatin and viscozyme (which mimics the digestion occurring in the gastrointestinal tract of swine) with 4 replicates of each sample digested over 2 batches. All replicates were used to determine DM digestibility while 2 replicates from each batch of each feedstuff were used to determine GE digestibility. On a DM basis, nutrient profile were as follows. GE: 4073 (BBG), 4736 (WMR) and 4824 kcal/kg (okara); CP: 11.7 (BBG), 15.9 (WMR) and 22.7% (okara); NDF: 31.0 (okara), 35.0 and 42.0% (BBG); ADF: 19.7 (okara), 24.2 and 34.0% (BBG), and hemicellulose: 8.0 (BBG), 10.8 and 11.2% (okara). The DM digestibility of okara (74.1%) was higher ( $P < 0.05$ ) than BBG (61.3%), WMR was in between (69.9%). Similarly, GE digestibility of okara (66.2%) was higher ( $P < 0.05$ ) than BBG (43.0%), WMR was in between (53.1%). Okara is not merely a good source of protein and energy, but also had fairly high in vitro DM and GE digestibility in swine. In conclusion, some agro-industrial coproducts can be potential substitute for common ingredients used as energy sources in pig diets, especially for small farms where these coproducts are widely available.

**Key Words:** coproduct, in vitro digestibility, swine

**572 Feeding liquid dairy derivatives (whey) to nursery pigs.**

Laura Eastwood, Mike R. Deibert, Dakota L. Wightman, and Denise Beaulieu\*, *Prairie Swine Centre Inc., Saskatoon, SK, Canada.*

Two experiments were conducted to examine the feeding value of liquid whey for newly weaned pigs. The experiments used pigs weaned at  $26 \pm 2$  d of age, housed 2 per pen. A water/whey mix was provided in buckets suspended above each pen allowing accurate measurement of intake. No additional water source was provided. Pig body weights, and liquid and feed intakes were calculated weekly. In Expt. 1, 72 pigs received 0, 8 or 16% sweet whey (vol/vol) in their drinking water (providing 0, 16.4 or 24.6 g solids/L). Pigs were acclimated for 7 d, followed by 14 d data collection, and fed a standard pre-grower diet. In Expt. 2, 160 pigs were assigned to 1 of 7 treatments ( $3 \times 2$  factorial + control). Treatments consisted of 0% whey (control;  $n = 20$ ), and sweet (pH 5.8, 20.5% DM) or acid (pH 2.9, 29.9% DM) whey at levels providing 16.4, 24.6 and 32.8 g solids/L (8, 12 or 16% inclusion (vol/vol) in drinking water for sweet whey, and 5.5, 8.2 and 10.9% for acid whey;  $n = 10$ /whey level). Diets were adjusted to account for expected nutrient intakes from the whey, based on intakes from Expt. 1. Data for both experiments were analyzed as a randomized complete block with treatment as a fixed effect, and block and pen as random effects. Effects of whey level were determined using linear and quadratic polynomial contrasts and

an orthogonal contrast was used in Expt. Two to test the effect of whey type.  $P < 0.05$  was considered significant. In Expt. 1, liquid intake and the intake of DM and calories from the whey increased with increasing whey concentration ( $P < 0.01$ ), Total caloric intake (2.64 Mcal DE/d), and piglet ADG (0.32 kg/d) were not affected by treatment. In Expt. 2, whey type (sweet vs. acid) had no effect on growth or nutrient intake. Liquid, nutrient and energy intake increased, and ADG and G:F ratios were improved with increasing whey regardless of whey type (linear  $P < 0.05$ ). Diet ADFI was unaffected by type or amount of whey (0.50 kg/d). Pigs compensated for the nutrients in the whey by decreasing feed intake (Expt. 1) or consuming similar amounts of a less nutrient dense diet (Expt. 2). Depending on the price of the whey, cost savings could therefore be achieved.

**Key Words:** swine, nursery, whey

**573 Growth performance, serum lipids, and intestinal volatile fatty acids contents in growing pigs fed flaxseed meal- and oat hulls-containing diets.** Saymore P. Ndou\*, Elijah Kiarie<sup>1,2</sup>, and Charles M. Nyachoti<sup>1</sup>, <sup>1</sup>*Department of Animal Science, University of Manitoba, Winnipeg, MB, Canada,* <sup>2</sup>*DuPont Industrial Biosciences-Danisco Animal Nutrition, Marlborough, UK.*

The objective of the study was to determine growth performance, serum lipids, and intestinal volatile fatty acids (VFA) concentrations in pigs fed flaxseed meal (FM)- and oat hulls (OH)-containing diets. Forty-eight Genesus [(Yorkshire-Landrace)  $\times$  Duroc] barrows with initial BW of  $25.0 \pm 0.32$  kg were penned in pairs. Pigs were allotted to 3 experimental diets: (corn-soybean meal-based diet (Control), 12% FM- and 10% OH-containing diets, in a completely randomized design, to give 8 replicates per treatment. Diets were formulated to be iso-energetic. However, incorporation of FM and OH increased the soluble fiber (sDF) and insoluble fiber (iDF) content of the diets, respectively. Feed intake and BW of pigs were measured weekly for 28 d. On d 28, blood samples were collected via jugular vein puncture for measuring concentration of serum lipids. Pigs were subsequently slaughtered for ileal, cecal and colonic digesta collection for VFA analysis. Pigs fed the control and OH-containing diet had greater final BW ( $P < 0.01$ ), feed intake ( $P = 0.01$ ) and weight gain ( $P < 0.01$ ) than FM-containing diets-fed pigs. Feed efficiency was reduced by 15.6% in pigs fed FM-based diets compared with the control ( $P = 0.01$ ). Total cholesterol was reduced ( $P < 0.01$ ) more in pigs fed OH-containing diets (1.99 mmol/L) than in FM-containing diets (2.25 mmol/L), compared with the control (2.36 mmol/L). Pigs fed FM- and OH-containing diets had greater ( $P < 0.01$ ) cecal pooled VFA concentration (135 and 126 mmol/L) than those fed the control diet (87.5 mmol/L). Dietary inclusion of FM and OH increased the cecal ( $P < 0.01$ ) and colonic ( $P < 0.01$ ) concentration of acetate by more than 100% and 20%, respectively. A similar trend was observed in the colon in which case pigs fed the FM- and OH-containing diets tended to have highest concentrations of valerate ( $P = 0.09$ ) and pooled VFA ( $P = 0.10$ ). In conclusion, iDF reduced serum cholesterol more than sDF and the former depressed growth performance by reducing feed intake. However, both sDF and iDF induced variable effects on VFA concentrations depending on intestinal segment.

**Key Words:** dietary fiber, growth performance, hindgut fermentation



**574 Effects of feeding increasing inclusion of canola press-cake on diet nutrient digestibility and growth performance of weaned pigs.** X. Zhou<sup>\*1</sup>, E. Beltranena<sup>1,2</sup>, and R. T. Zijlstra<sup>1</sup>, <sup>1</sup>University of Alberta, Edmonton, AB, Canada, <sup>2</sup>Alberta Agriculture and Rural Development, Edmonton, AB, Canada.

Canola press-cake (CPC) is a co-product from biodiesel production in small-scale processing plants that mechanically press oil from cleaned canola seed without seed conditioning or solvent extraction. The CPC contains 37% CP and 20% remaining oil; thus, CPC could be a source of AA and energy in pig diets. However, growth responses to increasing inclusion of CPC have not been evaluated in pigs. To evaluate the feeding value of CPC, 240 pigs (7.5 kg) starting 1 wk after weaning at 19 d of age were fed 5 wheat-based diets containing 0, 5, 10, 15, or 20% CPC to replace soybean meal in 2 phases (Phase 1 and 2). Diets were formulated to contain 2.45 and 2.41 Mcal NE/kg and 5.02 and 4.20 g standardized ileal digestible (SID) Lys/Mcal NE, respectively, and were fed for 2 wk for phase 1 (d 0–14) and 3 wk for phase 2 (d 15–35). Feed added and left and pig BW were measured weekly to calculate pen ADFI, ADG, and G:F. Freshly-voided feces were collected on d 12–13 and d 33–34 for phase 1 and 2, respectively, to determine diet apparent total-tract digestibility (ATTD) of DM, GE, and CP and diet DE using the index method. The diet NE value was predicted by Eq. 1–8 in NRC (2012). Data were analyzed using the MIXED procedure in SAS by orthogonal contrasts tested the linear or quadratic effects of CPC inclusion. Increasing dietary inclusion of CPC linearly reduced ( $P < 0.05$ ) the ATTD of GE by 1.3% and CP by 2.3% and diet DE and NE values in phase 1 and 2. Increasing dietary inclusion of CPC did not affect overall (d 0–35) ADFI and ADG of pigs but linearly reduced ( $P < 0.01$ ) ADFI for d 29–35, linearly increased ( $P < 0.05$ ) ADG for d 15–21, and linearly reduced ( $P < 0.05$ ) ADG for d 29–35. Increasing CPC inclusion linearly increased ( $P < 0.05$ ) feed efficiency for d 15–21 and overall. In conclusion, feeding up to 20% of CPC reduced ATTD of nutrients but did not reduce overall growth performance of weaned pigs when diets were balanced for NE and SID Lys/NE ratio. The CPC may be used as an alternative feedstuff to soybean meal to reduce feed cost while maintaining performance.

**Key Words:** canola press-cake, performance, weaned pig

**575 Apparent and true ileal and total-tract digestibility of fat in diets including canola press-cake or canola oil and endogenous fat loss in growing pigs.** X. Zhou<sup>\*1</sup>, E. Beltranena<sup>1,2</sup>, and R. T. Zijlstra<sup>1</sup>, <sup>1</sup>University of Alberta, Edmonton, AB, Canada, <sup>2</sup>Alberta Agriculture and Rural Development, Edmonton, AB, Canada.

Residual oil in canola press-cake (CPC) and extracted oil (CO) are both sources of dietary fat for pigs. However, fat digestibility may be lower in cake as oil remains trapped in the crushed seed matrix. Dietary fat source may affect endogenous fat losses that must be determined to calculate true fat digestibility. To test these hypotheses, 9 ileal-cannulated pigs (25.4 kg BW) were fed 9 diets in a  $9 \times 8$  Youden square. A basal diet of wheat and barley grains and canola meal was formulated. Eight test diets were prepared by mixing 10, 20, 30, and 40% CPC or 1.5, 3.0, 4.5, and 6.0% CO with basal diet, respectively, to match dietary fat content. Apparent total-tract digestibility (ATTD) and apparent ileal digestibility (AID) of acid-hydrolyzed ether extract (EE) were calculated for each diet using TiO<sub>2</sub> as marker. True total-tract digestibility (TTTD) and true ileal digestibility (TID) of EE in CPC and CO and endogenous loss of EE were estimated by regressing apparent digested EE (g/kg of DM intake) against dietary EE intake (g/kg of DM). The ATTD and AID of EE in CPC diets were 61.5 and 78.9% and were lower ( $P < 0.01$ ) than the 63.4 to 81.9% in CO diets, respectively. Apparent total-tract and

ileal digested EE content in CPC and CO diets increased linearly ( $P < 0.01$ ) with increasing EE intake. Endogenous loss of EE was greater ( $P < 0.05$ ) for the total-tract (–23.4 g/kg of DM intake) than by the ileum (–9.4 g/kg of DM intake). Dietary fat source did not affect ( $P > 0.05$ ) total-tract or ileal endogenous EE loss. The TTTD and TID of EE in CPC were 94.5 and 92.3% and were lower ( $P < 0.01$ ) than 100 and 96.5% in CO, respectively. In conclusion, CPC had reduced ATTD, AID, TTTD, and TID of EE compared with CO. Dietary fat source did not affect endogenous losses of EE. The reduced fat digestibility of CPC compared with CO indicates that fat digestibility of CPC should be considered to predict its nutritional value accurately before diet formulation.

**Key Words:** canola oil, fat digestibility, pig

**576 Nutrient profile and digestibility of macadamia nut cake as determined using an in vitro model of swine.** Utsav P. Tiwari and Rajesh Jha<sup>\*</sup>, University of Hawaii at Manoa, Honolulu, HI.

Highly variable price and market availability of some conventional feedstuffs, such as corn, wheat, soybean meal (SBM) make imperative to explore alternative feedstuffs to be used in swine diets. Macadamia nut cake (MNC), a coproduct from macadamia nut oil extraction process, is available and can serve as a potential feedstuff. However no or limited information of its nutrient profile and digestibility value limit its use in swine diets. To explore the nutritional value, MNC was analyzed for its basic nutrients, fibers, GE, AA and fatty acids content using standard methods. Digestibility of DM and GE of MNC was determined using an in vitro 3-step enzymatic assay using pepsin, pancreatin and viscozyme (which mimics the digestion occurring in the gastrointestinal tract of swine) with 4 replicates of sample digested over 2 batches. All replicates were used to determine DM digestibility while 2 replicates from each batch were used to determine GE digestibility. The MNC sample contained 93.1% DM. On DM basis, Ash, CP, crude fat, NDF, ADF, lignin and GE were found to be 3.7%, 25.5%, 11.9%, 35.8%, 28.0%, 16.0% and 5581 kcal/kg, respectively. In vitro DM and GE digestibility were found to be 76.7 and 71.4%, respectively. The concentration of Lys, the first limiting AA in swine, was found to be 0.7%. Linoleic and linolenic acid content were 2.5 and 0.2%, respectively. Gross energy content in MNC is comparable with that of corn and higher than SBM, due to high residual oil content, while protein content is twice as high as corn but lower than SBM. However, high fiber and presence of potential antinutritional factors like tannin and phytic acid need to be considered while using MNC in swine diets. In conclusion, MNC is not merely a good source of protein and energy but also had fairly high DM and GE digestibility when studied using an in vitro model of swine. Hence, it may serve as a potential alternative of some conventional sources of protein and energy in swine diets.

**Key Words:** in vitro digestibility, macadamia nut cake, swine

**577 Effect of duck grease on growth performance, carcass characteristics, and meat quality in growing-finishing pigs.** Jie Yu<sup>\*</sup>, Li Zhu, Bing Yu, Jun He, Ping Zheng, Xiangbing Mao, Quyan Wang, Zhiqing Huang, Junqiu Luo, and Daiwen Chen, Animal Nutrition Institute, Sichuan Agricultural University, Chengdu, Sichuan, China.

Duck grease (DG), a by-product of the duck industry, is abundant and inexpensive as a potential energy feed source because more than 60% of global ducks are produced in China. However, the nutritive value of DG in swine diet has not been assessed. In our previous study, the apparent digestible energy and metabolizable energy have been evaluated. The

present study addressed the effects of DG compared with soybean oil (SO) on growth performance, carcass characteristics and meat quality in a 82 d feeding program. A total of 72 growing barrows (31.62 ± 2.31 kg initial body weight) were randomly assigned into 2 dietary treatment groups in 6 replicates/pens utilizing a completely randomized design. The pigs were fed a corn-soybean meal based diet containing same amount (2% in growing diet, 1% in finishing diet) of SO or DG. One pig at average body weight in each pen was electrically stunned and slaughtered at the end of the feeding experiment, and carcass characteristics and pork quality were measured. Unpaired *t*-test was applied to evaluate the comparisons between 2 groups. Growth performance and carcass characteristics were not different ( $P > 0.05$ ). Pigs fed DG improved the apparent total-tract digestibility of dry matter (DM; 82.19 vs. 86.46%,  $P < 0.05$ ) and gross energy (GE; 81.22 vs. 84.17%,  $P < 0.05$ ). Visual color, ultimate pH, marbling score, and driploss were not different between DG and SO treatment. The intramuscular fat (IMF) content (2.77 vs. 3.67%,  $P < 0.05$ ) and muscle fiber density (1112.82 vs. 1389.97 fiber number /mm<sup>2</sup>,  $P < 0.05$ ) in longissimus dorsi (LM) were increased, while slice shear force was decreased in DG fed pigs (5.94 vs. 4.54 kg,  $P < 0.05$ ). In conclusion, dietary DG improved pork quality without negatively influencing growth performance and carcass characteristics in growing-finishing pigs.

**Key Words:** duck grease, pork quality, growing-finishing pigs

**578 Feeding layer hens with a new type of defatted green microalgae produced dose-dependent enrichments of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) in their egg yolk and tissues.** Theodore Derksen, Meghan Manor, and Xin Gen Lei\*, *Cornell University, Ithaca, NY.*

There is little EPA or DHA present in the yolks of eggs produced by hens fed corn-soybean meal-based diets. The purpose of this experiment was to enrich these n-3 fatty acids in the egg yolk by feeding hens with defatted green microalgae. A total of 50 White Leghorn hens (46-wk old, 1.70 ± 0.27 kg BW) were housed individually in cages, divided into 5 groups (n = 10), and fed a corn-soybean meal-based diet containing 0, 2.9, 5.8, 11.5, or 23% of a new type of defatted green microalgae (*Nannochloropsis oceanica*, Cellana, Kailua-Kona, HI) for 6 wk. Data were analyzed by one-way ANOVA with or without time-repeated measurements (SAS Version 9.1, SAS Institute, Inc., Cary, NC). The *P*-values for multiple regression analyses were adjusted using a Bonferroni correction procedure (significance at  $P \leq 0.01$ ). The microalgae supplementation produced dose-dependent linear ( $P < 0.01$ ) enrichments of EPA, DHA, and total n-3 fatty acids consistently in egg yolk ( $R^2 > 0.9$ ) assayed biweekly and of DHA in Liver ( $R^2 = 0.78$ ), breast ( $R^2 = 0.82$ ), and thigh ( $R^2 = 0.66$ ) assayed at wk 6. Concentrations of EPA + DHA in egg yolk, liver, breast muscle, and thigh muscle of hens fed 11.5 and 23% microalgae were 1.4–2.1, 0.6–1, 3.3–5.3, and 6–7-fold greater ( $P < 0.001$ ) than those in the controls (0% microalgae), respectively. Supplemental microalgae showed no effect on body weight, egg production rate, or tibia dimensions, but produced linear increases ( $P < 0.05$ ) in tibia bone strength ( $R^2 = 0.71$ ), egg shell weight ( $R^2 = 0.58$ ), and egg yolk redness ( $R^2 = 0.71$ ) and linear decreases ( $P < 0.05$ ) in ADFI ( $R^2 = 0.88$ ), egg yolk lightness ( $R^2 = 0.97$ ), and egg yolk yellowness ( $R^2 = 0.80$ ). In conclusion, this new type of defatted green microalgae was very effective in elevating EPA and DHA in the egg yolk, liver, and muscle tissues of layer hens. Eggs produced by hens fed the 23% defatted green microalgae diet may serve as a major food source of EPA and DHA to meet human needs. Supported in part by USDA/DOE Biomass

R&D Initiative Grant, a Hatch Grant of Cornell University, and a Hatch Grant Supplement from Cornell University for Undergraduate Research.

**Key Words:** egg, EPA/DHA, microalgae

**579 Moderate levels of supplemental defatted green microalgae in broiler diets had no adverse effect on bone properties.**

Eleanore O'Neil, Stephanie Gattrell, Meghan Manor, and Xin Gen Lei\*, *Cornell University, Ithaca, NY.*

Two experiments were conducted to determine if supplementing broiler diets with defatted green microalgae (*Nannochloropsis oceanica*, 0.69% phosphorus, Cellana, Kailua-Kona, HI) affected bone properties and phosphorous nutrition. Data were analyzed using one-way ANOVA and(or) linear regression procedure (SAS Version 9.1, SAS Institute, Inc., Cary, NC). In Experiment 1, day-old male Ross broiler chicks (total = 180) were fed a corn-soybean meal basal diet (BD) containing 0 (Control), 2, 4, 8, or 16% of the defatted microalgae for 6 wk. Chicks were housed in cages (6/cage) and each treatment consisted of 6 cages. Supplemental microalgae had no effect on tibial maximum load, maximum slope, extension at maximum load, energy to maximum load, or weight. However, tibial length of birds fed the 16% microalgae diet was 6.9% shorter (113 ± 0.3 mm vs. 105 ± 4.6 mm,  $P < 0.05$ ) than that of those fed the control diet. This decrease was consistent with a 10.2% lower BW of birds fed the 16% microalgae compared with the control (2817 ± 70.6 vs. 2528 ± 75.8 g,  $P < 0.01$ ). There were dose-dependent increases ( $P < 0.01$ ) in soluble inorganic phosphorous retention, and decreases ( $P < 0.05$ ) in soluble inorganic phosphorous excretion with elevated microalgal inclusions. Plasma inorganic phosphorous concentrations were not affected by the diet treatments. In Experiment 2, 3-wk old male Cobb chicks (total = 120) were fed the control, BD + 10% microalgae, BD + 10% microalgae + high vitamin E (DL- $\alpha$ -tocopheryl acetate, 150 IU/kg), or BD + 10% microalgae + high selenium (selenium-enriched yeast, 0.5 mg Se/kg) for 3 wk. Chicks were housed in cages (5/cage) and each diet treatment consisted of 6 cages. Compared with the control (BD), the other 3 diets produced no differences in all the tibial measures (including length) as described in Experiment 1. In conclusion, feeding broiler chicks with the defatted green microalgal biomass up to 10% in their diets did not show any adverse effect on bone health, but seemed to improve dietary phosphorus retention. Supported in part by a USDA/DOE Biomass R&D Initiative Grant, a Hatch Grant of Cornell University, and a Morley Student Research Fund Award of Cornell University.

**Key Words:** broiler, microalgae, tibial strength

**580 Low concentrations of supplemental defatted microalgae affect egg and tissue fatty acid composition differently in layers fed diets containing corn and flaxseed oils.** Jonggun Kim and Xin Gen Lei\*, *Cornell University, Ithaca, NY.*

Our laboratory previously showed that high concentrations of supplemental defatted microalgae (DMA, 10–15%) or flaxseed oil (FO, 5%) altered fatty acid profiles of egg yolk and tissues of layers. This experiment was to determine if and how lower concentrations of these supplementations affected fatty acid composition of egg yolk, liver, adipose tissue, and plasma of layers. A total of 60 Shaver leghorn layer hens (individually caged, 20 wk old) were allotted into 6 groups (n = 10) and fed a corn soybean meal-basal diet containing 0, 3, or 5% DMA (*Nannochloropsis oceanica*, Cellana, Kailua-Kona, HI) and 0 (1.5% corn oil) or 1.5% FO (Dyets, Bethlehem, PA) for 6 wk. Data were analyzed by 2-way

(2 by 3 factorial) ANOVA with or without time-repeated measurements using PC-SAS (Version 9.1, SAS Institute, Inc., Cary, NC). Although BW, ADFI, egg production rate, egg weight, or egg albumen, yolk, and shell were not affected by any of the dietary treatments at various time points, egg yolk color was changed ( $P < 0.05$ ) from 7 to 13 (Roche color fan) with increasing concentrations of DMA. There was no diet effect on plasma concentrations of triglyceride, cholesterol, or uric acid. While concentrations of n-3 fatty acids in egg yolk and plasma were elevated by FO up to 9-fold ( $P < 0.05$ ) starting at wk 1 or 2, such improvements by DMA were only 39 to 83% ( $P < 0.05$ ). Although DMA also improved ( $P < 0.05$ ) n-6/n-3 fatty acid ratios in egg yolk and plasma from 13 to 23 to 7–13, this effect was seen only in hens fed diets without FO. In the presence of 1.5% FO, only 5% DMA produced 8–9% increase ( $P < 0.05$ ) in n-3 fatty acid concentration of egg yolk at wk 2 and 3. Fatty acid profiles of liver and adipose tissue (collected at wk 6) displayed responses to supplemental DMA and FO similar to those of egg yolk or plasma. In conclusion, supplemental 2.5 or 5% DMA caused moderate enrichments of n-3 fatty acids and decreases of n6/n3 fatty acid ratios of egg yolk and tissues in hens fed diets containing corn oil. The DMA effects on these measures were very limited in hens fed diets containing FO. Supported in part by a USDA/DOE Biomass R&D Initiative Grant and a Hatch Grant of Cornell University.

**Key Words:** eggs, microalgae, n-3 fatty acid

**581 Supplemental defatted green microalgae and phytase improve hemoglobin repletion in weanling pigs.** Meghan L. Manor, Theodore J. Derksen, Rebecca L. Schwartz, Carol A. Roneker, and Xin Gen Lei\*, *Cornell University, Ithaca, NY.*

Our previous studies demonstrated that supplementing 7.5% defatted microalgae in diets for weanling pigs improved their blood hemoglobin

concentrations over the controls ( $11.0 \pm 1.5$  vs.  $13.0 \pm 1.4$  g/dL). This experiment was to determine if a lower concentration of microalgae alone or in combination with phytase produced similar improvements. A total of 30 weanling pigs (Hampshire  $\times$  Yorkshire  $\times$  Landrace cross-bred,  $7.5 \pm 0.4$  kg BW, hematocrit:  $27.5 \pm 1.7\%$ , and hemoglobin:  $9.4 \pm 0.6$  g/dL) were allotted to 5 dietary treatment groups ( $n = 6$ /group). The 5 diets included (1) a corn-soybean meal basal diet (BD) without supplemental inorganic iron (negative control), (2) BD + 50 mg Fe/kg (as  $\text{FeSO}_4$ , positive control), (3) BD + 0.5% defatted green microalgae (*Nannochloropsis oceanica*, Cellana, Kailua-Kona, HI), (4) BD + 500 units/kg of phytase (OptiPhos, Huvepharma, Peachtree City, GA), and (5) BD + defatted microalgae + phytase. The trial lasted for 6 wk and growth performance and hematology of pigs were measured biweekly. Individually penned pigs were considered the experimental unit. All data were analyzed by one-way ANOVA with or without time-repeated measurements using PC-SAS (Version 9.1, SAS Institute, Inc., Cary, NC). Blood hemoglobin concentrations and hematocrit of pigs fed the negative control diet were lower ( $P < 0.05$ ) than those fed the positive control diet at wk 4 and/or 6, whereas pigs fed the 3 diets containing microalgae and/or phytase had similar values to the positive control group for both measures at both time points. The experimental diets exerted effects on BW, ADG, ADFI, and gain/feed ratio similar to those on hemoglobin and hematocrit. However, the diets had no effect on plasma lipid or glucose profiles of pigs. In conclusion, supplementing 0.5% defatted green microalgae alone or with 500 units of phytase/kg was very effective in improving iron utilization for hemoglobin repletion in the weanling pigs. Supported in part by USDA/DOE Biomass R&D Initiative Grant, a Hatch Grant of Cornell University, and a Morley Student Research Fund Award of Cornell University.

**Key Words:** iron, microalgae, phytase



## Physiology and Endocrinology: Gametes and stress

**582 The effects of coenzyme Q10 supplementation on in vitro fertilization in porcine oocytes.** Caitlin A. Streacker\* and Brian D. Whitaker, *The University of Findlay, Findlay, OH.*

Polyspermic penetration as a result of in vitro fertilization (IVF) has shown to be a major obstacle in the production of porcine embryos. The objective of this study was to reduce the incidence of polyspermic penetration by supplementing coenzyme Q10 during oocyte maturation. Oocytes (n = 50/well) were matured in tissue culture media 199 supplemented with 0, 10, 50 or 100 mM of coenzyme Q10 during the last 24 h of maturation. Frozen thawed semen from a single boar was used for fertilizing groups of approximately 30 oocytes in a concentration of 200 sperm/oocyte. Approximately 12 h after IVF, oocytes were evaluated for fertilization kinetics and subsequent embryonic development. A total of 600 oocytes over 3 replications were used in this study with one well/treatment/replicate. Data were analyzed using ANOVA with the main effects including treatment well and replicate. Chi-squared analysis was used to determine percentages of embryos reaching the different developmental states for each treatment. Oocytes supplemented with 50 mM coenzyme Q10 had significantly higher ( $P < 0.05$ ) penetration rates ( $100 \pm 14.97\%$ ) and pronuclear formation (MPN;  $58.33 \pm 19.54\%$ ) compared with 100 mM coenzyme Q10 supplementation. There was no difference between not supplementing coenzyme Q10 and 10 mM supplementation with respect to penetration and MPN formation rates. There was no difference between the treatments groups when considering the incidence of on polyspermic penetration. Cleavage at 48 h after IVF and blastocyst formation at 144 h after IVF were evaluated. Supplementing 100 mM coenzyme Q10 during oocyte maturation significantly decreased ( $P < 0.05$ ) the percentage of embryos cleaved by 48h after IVF ( $3.33 \pm 19.06\%$ ) and the percent of embryos at the blastocyst stage of development by 144 h after IVF ( $0.00 \pm 15.76\%$ ) compared with all the other treatment groups. There was no difference between the other treatment groups with respect to cleavage and blastocyst formation rates. These results indicate that 50 mM coenzyme Q10 supplementation during oocyte maturation will not be detrimental to IVF and embryo culture in pig oocytes.

**Key Words:** coenzyme Q10, IVF, swine

**583 Melatonin supplementation during oocyte maturation improves embryonic development in pigs.** Rachel L. Lane\* and Brian D. Whitaker, *University of Findlay, Findlay, OH.*

High levels of reactive oxygen species (ROS) in and around maturing oocytes lead to oxidative stress, which decrease fertilization success rates. Melatonin reduces levels of ROS during in vitro fertilization in swine. The objective was to determine the effects of melatonin during oocyte maturation on: IVF kinetics and embryonic development. Oocytes (n = 50/well) were supplemented during the last 24 h of maturation with 0, 75, 100, or 150 nM melatonin and then subjected to IVF and embryo culture. Frozen-thawed semen from a single boar was used (approximately 30 oocytes/well, 200 sperm/oocyte). A total of 600 oocytes over 3 replicates were used in this study with one well/ treatment/ replicate. Post IVF, a percentage of the embryos were evaluated for penetration, polyspermy, and male pronuclear formation rates. The remaining embryos were evaluated 48 h after IVF for cleavage and 144 h for blastocyst formation. Data were analyzed using ANOVA with the main effects including treatment, well and replicate. Chi-squared analysis was used to determine percentages of embryos

reaching the different developmental stages for each treatment. There were no differences between treatment groups with respect to penetration and polyspermy. Supplementation of 150 nM melatonin produced a significant ( $P < 0.05$ ) percent of embryos with MPN ( $25.93 \pm 21.17\%$ ) compared with those supplemented with 75 nM ( $66.67 \pm 21.17\%$ ) or 100 nM ( $79.17 \pm 21.17\%$ ). There was no difference between the 75 nM and 100 nM treatment groups. Supplementation of 150 nM ( $22.00 \pm 21.37\%$ ) produced significantly ( $P < 0.05$ ) less of cleavage between all of the other treatment groups 48 h after IVF. Embryo supplementation of 75 nM melatonin during maturation had significantly higher ( $P < 0.05$ ) percentage of blastocyst formation by 144 h after IVF compared with those supplemented with 150 nM ( $10.00 \pm 17.32\%$ ) of melatonin. There was no difference in the percent of embryos reaching the blastocyst stage of development by 144 h after IVF between no supplementation, 100 nM, and 75 nM of melatonin. These results indicate that the supplementation of 75 nM melatonin during the later stages of maturation improves embryo development by increasing cleavage and blastocyst rates 48 and 144 h after IVF, respectively.

**Key Words:** melatonin, IVF, swine

**584 Cumulus-oocyte complex gene expression in bovine preovulatory follicles after administration of porcine luteinizing hormone.** Amir Behrouzi\*<sup>1</sup>, Marcos G. Colazo<sup>1</sup>, Ana Ruiz-Sanchez<sup>2</sup>, and Divakar J. Ambrose<sup>1,2</sup>, <sup>1</sup>*Alberta Agriculture and Rural Development, Livestock Research Branch, Edmonton, AB, Canada,* <sup>2</sup>*Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB, Canada.*

In previous work, the use of porcine luteinizing hormone (pLH) in lieu of gonadotropin releasing hormone (GnRH) for synchronizing ovulation improved pregnancy rates in lactating dairy cows. Later, we found that pLH treatment not only altered the LH profile, but also influenced expression of the BMP-15, GDF-9 and TGF- $\beta$ 1 proteins known to promote cumulus expansion and oocyte competence in nonlactating cows. We currently hypothesized that pLH would alter the expression of target genes in the cumulus-oocyte complexes (COC), which regulate oocyte competence. Cyclic nonlactating Holstein cows (n = 8) were subjected to ovarian stimulation with 200 mg FSH. Follicles = 10 mm were aspirated at  $21 \pm 1$  h after giving either 100  $\mu$ g GnRH or 25 mg pLH im, and COC recovered and frozen until qRT-PCR analysis. Serial blood samples were collected from before giving GnRH/pLH until 20 h after, for plasma LH analysis using an anti-bovine LH monoclonal antibody that cross-reacts equally with both bovine and porcine LH. Plasma LH concentrations and qRT-PCR data were analyzed using MIXED and GENMOD procedures of SAS, respectively. When LH concentrations (ng/mL) were compared in GnRH- and pLH-treated cows, effects of time and time-by-treatment interactions were detected ( $P < 0.01$ ) with mean LH being higher in GnRH than pLH cows ( $2.8 \pm 0.2$  vs.  $2.0 \pm 0.2$ ) from 30 min until 4 h post-treatment. However, mean plasma LH from 5 to 20 h post-treatment was greater ( $P < 0.01$ ) in pLH- than in GnRH-treated cows ( $1.1 \pm 0.2$  vs.  $0.4 \pm 0.1$ ). Treatment with pLH altered *HAS-2* and *GREM-1* mRNA abundance ( $P < 0.01$ ) by about 26- and 7-fold, in COC of pLH- and GnRH-treated cows, respectively. Whereas the relative expression of *AREG* abundance tended ( $P = 0.06$ ) to increase in GnRH-treated cows, *EREG*, *BTC*, *PGr*, *COX-2* and *PTX-3* mRNA expression were not affected by treatment. This study provides evidence that a prolonged, higher-than-basal LH profile in pLH-treated cows increased the expression of *HAS-2* and *GREM-1*, genes known to

improve oocyte competence, which may explain the higher pregnancy rates previously reported when pLH was used to induce ovulation.

**Key Words:** cumulus-oocyte complex, porcine LH, GnRH

**585  $\alpha$ -Lipoic acid improves the post-thaw quality and survival of Nili-Ravi buffalo bull sperm.** Muhammad Hammad Fayyaz<sup>1</sup>, Sajid Iqbal<sup>1,2</sup>, Muhammad Binyameen<sup>3</sup>, Nasim Ahmad\*<sup>1</sup>, and Mush-taq Ahmad<sup>1</sup>, <sup>1</sup>Department of Theriogenology, University of Veterinary and Animal Sciences, Lahore, Pakistan, <sup>2</sup>Semen Production Unit, Qadirabad, Sahiwal, Pakistan, <sup>3</sup>Buffalo Research Institute, Kasur, Pakistan.

Cryopreservation exerts damages to sperm including crystal formation, oxidative stress, osmotic shock and lipid peroxidation.  $\alpha$ -Lipoic acid (ALA); an antioxidant that plays a role in ATP production and breakdown of free radicals thus reduces oxidative stress. The aim of this study was to optimize the concentration and effect of ALA on post-thaw quality and survival at 37°C of buffalo (*Bubalus bubalis*) bull sperm. Semen of 3 mature Nili Ravi breeding bulls was collected twice a week using artificial vagina. The ejaculates of 3 bulls were pooled bull wise (replicates = 6), divided into 6 aliquots, diluted with the Tris-citrate-egg yolk extender supplemented with different concentrations of ALA (0, 0.5, 1, 1.5, 2 and 2.5mM) and cryopreserved in 0.5-mL French straws using standard procedure. Two straws per replicate were thawed and pooled and incubated in modified synthetic fluid (mSOF) at 37°C in a humidified CO<sub>2</sub> incubator. Semen was evaluated for motility, plasma membrane integrity (PMI; hypo-osmotic swelling test), viability and acrosomal integrity simultaneously (FITC-PNA/PI assay) and DNA integrity using acridine orange assay; at 0, 1.5, 3 and 4.5h of incubation. The data were presented as Mean  $\pm$  SEM and analyzed 6  $\times$  4 factorial repeated measure ANOVA for 6 concentrations and 4 times using MIXED procedure in SAS Enterprise Guide (version 4.2) taking time as repeated measure. The results revealed that extender supplemented 0.5 mM and 1 mM ALA resulted in higher ( $P < 0.05$ ) sperm motility ( $55 \pm 1.3\%$  and  $55.5 \pm 1.3\%$  respectively than control;  $48.5 \pm 1.37\%$ , 1.5 mM;  $49 \pm 1.37\%$  and 2 mM;  $43.5 \pm 1.37\%$ ), PMI ( $59.8 \pm 1.8\%$  and  $60 \pm 1.8\%$  respectively), viability ( $67.3 \pm 1.5\%$  and  $68.5 \pm 1.5\%$  respectively) and DNA integrity ( $99.29 \pm 0.48\%$  and  $99.39 \pm 0.48\%$ ) than control and other groups after thawing. The survival of sperm was also recorded higher ( $P < 0.05$ ) due to 0.5 and 1 mM ALA resulting in motility ( $23.5 \pm 1.3\%$  and  $25.5 \pm 1.3\%$ ), PMI ( $34.3 \pm 1.8\%$  and  $37.8 \pm 1.8\%$ ), viability ( $38.6 \pm 1.5\%$  and  $42.6 \pm 1.5\%$ ) and DNA integrity ( $97.34 \pm 0.48\%$  and  $98.0 \pm 0.48\%$ ) than control and other groups at 4.5 h of incubation. In conclusion,  $\alpha$ -lipoic acid enhances the post-thaw quality and survival of buffalo bull sperm.

**Key Words:**  $\alpha$ -lipoic acid, buffalo sperm, in vitro incubation

**586 Comparison of fertility of liquid and frozen semen when varying the interval from CIDR removal to insemination.** Brit-tany N. Richardson\*<sup>1</sup>, Erin L. Larimore<sup>1</sup>, Julie A. Walker<sup>1</sup>, Matthew D. Utt<sup>2</sup>, J. Mel DeJarnette<sup>2</sup>, and George A. Perry<sup>1</sup>, <sup>1</sup>Department of Animal Sciences, South Dakota State University, Brookings, SD, <sup>2</sup>Select Sires Inc., Plains City, OH.

Cryopreservation allows for long-term storage of semen. However, cryopreservation and thawing imposes stress on spermatozoa, and prematurely initiates the process of capacitation; possibly decreasing sperm lifespan. Liquid semen is not exposed to these stressors, leading to a potential longer lifespan in the female reproductive tract and thus increasing the window for successful insemination. The objective of

this study was to compare fertility of liquid and frozen semen when varying the interval from CIDR removal to insemination using the 7-d CO-Synch + CIDR protocol. Within age group cows ( $n = 389$ ) were randomly assigned to insemination at 36 or 60 h after CIDR removal with either liquid (36L and 60L) or frozen semen (36F and 60F). Cows were monitored for estrus activity from CIDR removal to 60 h after CIDR removal. Cows that failed to exhibit estrus received an injection of GnRH (100  $\mu$ g) and a blood sample was collected for analysis of estradiol concentration. Data were analyzed using the GLIMMIX procedure of SAS with sire and herd as random variables. There was no difference in pregnancy rates between liquid or frozen semen (53% and 52%), but cows inseminated at 60 h had greater ( $P < 0.01$ ) pregnancy rates than cows inseminated at 36 h (72% and 31%). There was no interval by semen type interaction ( $P = 0.57$ ). Estrus was detected in 63%, 61%, 56%, and 62% of 36F, 36L, 60F, and 60L, respectively (only 5% and 1% of 36F and 36L were detected in estrus before insemination). Overall cows that exhibited estrus had greater pregnancy rates compared with cows that did not ( $P < 0.01$ ; 79% and 24%). Among cows that did not exhibit estrus, those inseminated with liquid semen tended to have greater pregnancy rates than those inseminated with frozen semen ( $P = 0.06$ ), and ones that became pregnant had elevated ( $P < 0.01$ ) concentrations of estradiol at 60 h ( $10.7 \pm 1.9$  vs  $7.9 \pm 2.9$  pg/mL). In summary, there was no difference in pregnancy success between liquid and frozen semen. However, cows that exhibited estrus and inseminated at 60 h after CIDR removal had greater pregnancy success compared with cows that did not exhibit estrus.

**Key Words:** pregnancy, liquid semen, estrus

**587 The effects of seasonal heat stress on sperm nuclear shape in boars.** Teyanna M. Loether, Ricky L. Monson, Cathy Miller-Gaudette, and John J. Parrish\*, University of Wisconsin-Madison, Madison, WI.

Negative effects on germ cell development have been correlated with elevated scrotal temperatures in domestic livestock species, including the boar. To fully comprehend the effect of environmental heat stress on male fertility, a total of 1,181 boar ejaculates, with an average of  $49.2 \pm 1.7$  per week, were collected during June–November 2012 from a commercial boar stud. Boars were housed in facilities equipped with cool cell technology and facility temperature was measured every 30 min in 2012. Environmental temperature data were obtained from the National Oceanic and Atmospheric Administration for 2012 and 2014, and expressed as a weekly average of the maximum daily temperature. Sperm were assessed using Fourier Harmonic Amplitude (FHA) analysis to determine sperm nuclear shape and harmonic values were generated (HA0–5). Using the first collection week as the control, a GLM procedure with Dunnett's mean separation test was performed to measure changes occurring in HA0-HA5. Changes were found in HA0 and HA2 for wk 8–21, or the weeks of July 25–October 24 ( $P < 0.05$ ). The HA0 at week one was  $2.80 \pm 0.01$   $\mu$ m and peaked during wk 14 at  $2.87 \pm 0.01$   $\mu$ m. Effects on HA0 appeared 3 weeks after external temperatures rose above 90°F (facility temperature = 82°F); this corresponds to when affected round spermatids first appear in the ejaculate as mature sperm. Fluctuations in HA0 followed the oscillations in temperature until the average maximum external and internal temperature fell below 68°F. To confirm these results, samples from the same facility were collected in 2014 3 weeks after external temperatures rose above 90°F. A period of heat stress was effectively captured as changes in HA0 and HA4 were again found 3 weeks post-heat event ( $P < 0.05$ ). Increases seen in HA0, which is a reflection of the overall size of the sperm head, indicate that boar sperm nuclei enlarge during seasonal heat stress. Fourier Harmonic

Amplitudes, particularly HA0, 2, and 4, can be utilized as an effective tool in predicting specific periods where nuclear shape may be affected by heat stress. Furthermore, FHA analysis could be utilized in the future to correlate changes in nuclear shape as a result of heat stress to fertility.

**Key Words:** boar reproduction, seasonal heat stress, sperm nuclear shape

**588 Expressional regulation of chemerin and its receptors in the liver and adipose tissues of young cattle by weaning and nutrition.** Yutaka Suzuki<sup>\*1</sup>, Daichi Kato<sup>1</sup>, Mitsuhiko Kondo<sup>1</sup>, Hizuru Hatanaka<sup>1</sup>, Satoshi Haga<sup>1,2</sup>, Takafumi Gotoh<sup>3</sup>, and Sanggun Roh<sup>1</sup>, <sup>1</sup>Graduate School of Agricultural Science, Tohoku University, Sendai, Miyagi, Japan, <sup>2</sup>Grassland Management Research Division, NARO Institute of Livestock and Grassland Science, Nasushiobara, Tochigi, Japan, <sup>3</sup>Kuju Agricultural Research Center, Kyushu University, Takeda, Oita, Japan.

Chemerin is highly expressed and secreted from liver and adipose tissues, and acts through its receptors in an endocrine, paracrine, autocrine manner in ruminant. We previously reported the administration of chemerin analog peptide induced insulin secretion in vivo. However, there are currently no data on the expressional regulation of chemerin and its receptors depending on weaning and nutrition. In the present study we investigated the expressional levels of chemerin and its receptors (CMKLR1 and CCRL2) gene in the liver and adipose tissue of weaning calves and young cattle fed with different diets. Japanese Black calves were divided into (1) pre-weaning and post-weaning group (euthanized before and after weaning; 1.5 or 3.5 mo of age, respectively), or (2) concentrate-fed group received concentrate (CP 12%, TDN 73%; 2.5% of BW daily) plus Italian ryegrass (CP 9.4%, TDN 55.3%; ad libitum) and hay-fed group received Italian ryegrass ad libitum from 3 to 10 mo of age. The liver and/or adipose tissue (subcutaneous, mesenteric, perirenal and epididymal) samples were collected from cattle to quantitate mRNA expressional levels of chemerin, CMKLR1 and CCRL2 by qRT-PCR. Statistical analysis was performed by Mann-Whitney U test (significant difference;  $P < 0.05$ ). Chemerin mRNA level was higher in mesenteric adipose tissue of post-weaning group and CCRL2 mRNA level was higher in the liver of post-weaning group, compared with pre-weaning group. Chemerin mRNA level in liver was higher in the concentrate-fed group than in hay-fed group at 10 months of age. This study revealed the effects of weaning and dietary energy source on gene expression of chemerin and CCRL2, suggesting the role of chemerin in the regulation of insulin secretion in cattle around weaning and in different nutritional states.

**Key Words:** chemerin, weaning, nutrition

**589 Modulation of the metabolic response to vaccination in naïve beef steers using an acute versus chronic stress model.**

Nicole C. Burdick Sanchez<sup>\*1</sup>, Jeffery A. Carroll<sup>1</sup>, Nathan D. May<sup>2</sup>, Shelby L. Roberts<sup>2</sup>, Heather D. Hughes<sup>2</sup>, Paul R. Broadway<sup>1</sup>, Kate P. Sharon<sup>1,3</sup>, Michael A. Ballou<sup>3</sup>, and John T. Richeson<sup>2</sup>, <sup>1</sup>USDA-ARS, Livestock Issues Research Unit, Lubbock, TX, <sup>2</sup>West Texas A&M University, Department of Agricultural Sciences, Canyon, TX, <sup>3</sup>Texas Tech University, Department of Animal and Food Sciences, Lubbock, TX.

Available energy plays a critical role in the initiation and maintenance of an immune response to a pathogen a process that is further altered by activation of stress system. This study was designed to determine the effect of an acute versus chronic stress model on the metabolic

response to vaccination in naïve beef steers. Steers ( $n = 32$ ;  $209 \pm 8$  kg) were blocked by BW and assigned to 1 of 3 treatments: (1) Chronic stress (CHR), 0.5 mg/kg BW dexamethasone (DEX) administered i.v. at 1000h on d 3 to d 6; (2) Acute stress (ACU), 0.5 mg/kg BW DEX administered i.v. at 1000h on d 6; or (3) Control (CON), no DEX. On d 2, steers were fitted with jugular vein catheters and moved into individual stanchions in an environmentally controlled facility. Blood samples were collected at -74, -50, and -26 h, at 0.5-h intervals from -4 h to 6 h, and at 12, 24, 36, 48, and 72 h relative to vaccination with Pyramid 5 + Prespense SQ at 1200h on d 6. Data were analyzed by the MIXED procedure of SAS specific for repeated measures. Feed intake was not different ( $P = 0.44$ ) between CON ( $4.9 \pm 0.2$  kg), ACU ( $4.9 \pm 0.2$  kg) and CHR steers ( $5.1 \pm 0.2$  kg). There was a treatment  $\times$  time interaction ( $P < 0.001$ ) for serum glucose concentrations. Specifically, glucose concentrations increased at -50 h in CHR steers and at 12 h in ACU steers, and remained elevated through 72-h post-vaccination period compared with CON steers. The change in NEFA concentrations was affected by treatment ( $P < 0.001$ ) and time ( $P < 0.001$ ) such that the change in NEFA was greater in CHR ( $0.06 \pm 0.01$  mmol/L), followed by CON ( $-0.01 \pm 0.01$  mmol/L) and ACU steers ( $-0.04 \pm 0.01$  mmol/L). Blood urea nitrogen (BUN) was affected by treatment ( $P < 0.001$ ) and time ( $P < 0.001$ ) such that BUN concentrations were greatest in CHR ( $12.0 \pm 0.1$  mg/dL) followed by ACU ( $10.4 \pm 0.1$  mg/dL) and CON steers ( $9.6 \pm 0.1$  mg/dL). These data demonstrate that activation of the stress and immune axes using an acute or chronic stress model can increase energy mobilization before and following vaccination in naïve steers, potentially affecting energy availability needed to mount an adequate antibody response to vaccination.

**Key Words:** cattle, metabolism, vaccination

**590 Mimicking acute and chronic stress exposure in naïve beef steers alters the acute phase response (APR) associated with vaccination.**

Jeffery A. Carroll<sup>\*1</sup>, Nicole C. Burdick Sanchez<sup>1</sup>, Nathan D. May<sup>2</sup>, Shelby L. Roberts<sup>2</sup>, Heather D. Hughes<sup>2</sup>, Paul R. Broadway<sup>1</sup>, Kate P. Sharon<sup>1,3</sup>, Michael A. Ballou<sup>3</sup>, and John T. Richeson<sup>2</sup>, <sup>1</sup>USDA-ARS, Livestock Issues Research Unit, Lubbock, TX, <sup>2</sup>West Texas A&M University, Department of Agricultural Sciences, Canyon, TX, <sup>3</sup>Texas Tech University, Department of Animal and Food Sciences, Lubbock, TX.

This study was designed to determine the effect of an acute versus chronic stress model on the APR associated with vaccination in naïve beef steers. Steers ( $n = 32$ ;  $209 \pm 8$  kg) were blocked by BW and assigned to 1 of 3 treatments: (1) Chronic stress (CHR), 0.5 mg/kg BW dexamethasone (DEX) administered i.v. at 1000h on d 3 to d 6; (2) Acute stress (ACU), 0.5 mg/kg BW DEX administered i.v. at 1000h on d 6 only; or (3) Control (CON), no DEX. On d 2, steers were fitted with indwelling rectal temperature (RT) devices and jugular catheters, and then moved into individual stanchions in an environmentally controlled barn. Blood samples were collected and serum isolated at -74, -50, and -26 h, at 0.5-h intervals from -4 h to 6 h, and at 12, 24, 36, 48, and 72 h relative to vaccination with Pyramid 5 + Prespense SQ at 1200 h on d 6. A second blood sample was used to measure complete blood cell counts. Data were analyzed using the Mixed procedure of SAS specific for repeated measures. There was a treatment  $\times$  time interaction ( $P < 0.01$ ) for RT such that DEX treatment in CHR and ACU steers decreased RT on d3 and d6, respectively, compared with CON steers. Vaccination on d 6 increased RT in CON but not in CHR or ACU steers with RT remaining elevated in CON for the remainder of the study. There was a treatment  $\times$  time interaction ( $P < 0.01$ ) for total white blood cells (WBC), neutrophils, lymphocytes, and monocytes. Specifically, DEX



increased WBC and neutrophils in CHR and ACU steers, yet decreased lymphocytes in CHR steers. Monocytes initially increased in response to DEX treatment in CHR and ACU steers. Also, eosinophils were greater ( $P < 0.01$ ) in CON than ACU and CHR steers. There was a treatment  $\times$  time interaction ( $P < 0.01$ ) for interferon- $\gamma$  (IFN- $\gamma$ ) such that IFN- $\gamma$  was greater in CON and CHR than ACU steers. Concentrations of tumor necrosis factor- $\alpha$  were elevated ( $P < 0.01$ ) in CHR compared with ACU and CON steers. These data suggest that ACU stress may prime, while

CHR stress may cause hyper-activation, of the APR associated with vaccination in naïve beef calves, thus potentially negatively affecting the overall immunological response to vaccination.

**Key Words:** cattle, immunity, vaccination

## Production, Management, and the Environment II

**591 Global impact of improving feed efficiency and technology transfer efficacy.** Robin R. White<sup>\*1,3</sup>, Todd J. Applegate<sup>2</sup>, Gary L. Cromwell<sup>3</sup>, Donald C. Beitz<sup>4</sup>, Michael L. Galyean<sup>5</sup>, Mary Beth Hall<sup>6</sup>, Phillip S. Miller<sup>7</sup>, Jack Odle<sup>8</sup>, William P. Weiss<sup>9</sup>, and Mark D. Hanigan<sup>1</sup>, <sup>1</sup>National Animal Nutrition Program, Virginia Tech, Blacksburg, VA, <sup>2</sup>Purdue University, West Lafayette, IN, <sup>3</sup>University of Kentucky, Lexington, KY, <sup>4</sup>Iowa State University, Ames, IA, <sup>5</sup>Texas Tech University, Lubbock, TX, <sup>6</sup>USDA/ARS, Madison, WI, <sup>7</sup>University of Nebraska, Lincoln, NE, <sup>8</sup>North Carolina State University, Raleigh, NC, <sup>9</sup>The Ohio State University, Columbus, OH.

Providing protein for the expanding population is an imperative challenge to which animal nutrition research is vital. The objective of this work was to model how improving livestock feed efficiency and adoption of agricultural technologies would affect the number of people's annual protein requirements that could be met from a constant feed base. Global production data from FAOStat and occupation statistics from World Bank were collected to estimate the meat, milk and eggs (food) produced by an average farmer. Increasing feed efficiency was modeled as a % reduction in the feed required to produce 1 kg of food. Technology transfer efficiency was also accounted for. Net improvement in food produced was modeled as the sum of food produced from the proportion of farmers adopting the technology and food produced from the proportion of farmers not adopting the technology. Conversion of meat live weight to retail weight was based on dressing percentages (sheep/goat, 50%; beef, 63%; pork 70%) and a 50% conversion of dressed to retail weight. Retail milk and eggs were assumed 90% of produced weights. Protein production was calculated based on protein content. Meat protein (125 g/kg to 183 g/kg) and milk or egg protein (33 g/kg to 126 g/kg) varied by species. Protein produced by the system was divided by an average human's protein requirement (45 g/d) multiplied by 365 d. Assuming a 30% efficiency in technology transfer, a 34% improvement in feed efficiency would be required to meet the protein needs of an additional  $0.5 \times 10^9$  people. A myriad of combinations of improved feed efficiency and improved technology transfer could result in sufficient protein to feed  $2 \times 10^9$  people. In general, feed efficiency improvements of over 50% were required and technology transfer efficiencies between 50 and 70% greatly improved practicality of meeting the protein requirements of  $2 \times 10^9$  billion people. Both biological research aimed at improving feed efficiency, and social research aimed at understanding factors influencing technology adoption rates will be required to improve global food security. Supported by USDA NIFA and State Agricultural Experiment Stations.

**Key Words:** food security, nutrition, feed efficiency

**592 Nutrient cycling on dairy farms: Status and opportunities.** Andrew Henderson<sup>1</sup>, Ying Wang<sup>\*2</sup>, Karin Veltmank<sup>3</sup>, and Olivier Jolliet<sup>3</sup>, <sup>1</sup>University of Texas, Houston, TX, <sup>2</sup>Innovation Center for US Dairy, Rosemont, IL, <sup>3</sup>University of Michigan, Ann Arbor, MI.

Nutrient management represents both a challenge and opportunity to dairy and other livestock industries. Current practices may lead to lost nutrients from feed, manure, and commercial fertilizers, possibly impairing water bodies or affecting air quality. These losses may have direct economic implications for dairy farms, via possible phosphorus supply shortages [Van Vuuren et al., 2010. *Glob. Environ. Change* 20:428–439] or future nutrient regulation (for eutrophication or greenhouse gases). Losses also present an opportunity for improvement, if farmers can take

advantage of nutrient cycling. Using a combination of literature meta-analysis and case study modeling, this research outlines and estimates the potential scale of improved nutrient cycling on dairy farm. For the meta-analysis, we compiled ~300 research articles for nutrient flows on dairy farms. We cataloged the variety of nitrogen, phosphorus, and potassium flows crossing the farm boundary (e.g., purchased feed) and internal to the farm (e.g., manure application to crops). Some flows had large coefficients of variation (CV), such as total excretion of nitrogen (CV = 2.3, n = 43) and total intake of nitrogen (CV = 2.7, n = 94). To complement this analysis, we compared modeled nutrient flows on a commercial dairy farm, using the process-based models Integrated Farm System Model [IFSM; Rotz et al., 2014; The Integrated Farm System Model (IFSM): Reference Manual Version 4.1, USDA] and ManureDNDC [Manure De-Nitrification De-Composition; Li et al., 2012. *Nutr. Cycl. Agroecosys.* 93:163-200]. Field N<sub>2</sub>O emissions differed between the models (3.8–11.3 t of N<sub>2</sub>O/yr), but other emissions predicted P and N losses to (ground)water through leaching, run-off and erosion are comparable across models. Whole-farm ammonia emissions are comparable across models (87.6 – 122.1 t NH<sub>3</sub>/yr). The variation among farms, in the literature and modeled, represents a potential for improvement. This study compares and contrasts these variations, identifying the magnitude of potential nutrient cycling efficiencies on farm.

**Key Words:** dairy, nutrient cycling, environmental impact

**593 Comparing climate impacts of grass-finished beef production strategies in the upper Midwest using a partial life cycle analysis.** Jason E. Rowntree<sup>\*1</sup>, Rebecca Ryals<sup>2</sup>, Marcia S. DeLonge<sup>3</sup>, Marilia B. Chiavegato<sup>4</sup>, W. Richard Teague<sup>5</sup>, and Peter Byck<sup>6</sup>, <sup>1</sup>Michigan State University, East Lansing, MI, <sup>2</sup>University of Hawaii, Honolulu, HI, <sup>3</sup>Union of Concerned Scientists, Washington, DC, <sup>4</sup>University of São Paulo, Piracicaba, São Paulo, Brazil, <sup>5</sup>Texas A&M Agrilife Research, Vernon, TX, <sup>6</sup>Arizona State University, Tempe, AZ.

Cattle grazing management strategies can vary widely and have important effects on ecosystem carbon storage, greenhouse gas (GHG) emissions and land requirements. Existing GHG life cycle assessments of beef production often lack refined details about grazing strategies and their associated impacts on ecosystem carbon and greenhouse gas dynamics. A partial life cycle analysis was conducted on an upper Midwest grass finishing beef production system that compared 2 different grazing management strategies. The approaches were: 1) a non-irrigated, high-density grazing system stocked at 1.0 AU/ha (100,000 kg LW/ha; SysA) and 2) an irrigated, low-density grazing system stocked at 2.5 AU/ha (30,000 kg LW/ha; SysB). Our life cycle boundary only included the grass-finishing portion of beef production. Steers were born in April, weaned in November, backgrounded for 6 mo and grazed until slaughter in November, the following year, with an average age at slaughter of 19 mo and a 295 kg HCW. We included GHG associated with enteric methane (CH<sub>4</sub>), soil nitrous oxide and CH<sub>4</sub> emissions, alfalfa and mineral supplementation and farm energy use. We used 2 years of on-farm corrected data for soil and enteric emissions and animal performance from Lake City Research Center, Lake City, MI. The assumed boundaries for potential soil C loss or gain ranged from an emission of 3 Mg C/ha/yr to a positive sink of 3 Mg C/ha/yr. Enteric CH<sub>4</sub> emissions had the largest effect on overall GHG flux and this varied by year and grazing system. In years 2012 and 2013, enteric CH<sub>4</sub> was 89 and 49% of the overall flux for SysA compared with 72 and 65% for SysB, respectively. Both systems are net GHG sources when soil

C sequestration is excluded. When soil C sequestration is considered, each grazing strategy has potential to be an overall sink. These results indicate SysA and SysB would have to sequester up to 1.0 and 2.0 Mg C/ha/yr to have a net zero GHG footprint, respectively.

**Key Words:** grass-finished, methane, carbon

**594 Predicting methane emission of dairy cows using fatty acids and volatile and non-volatile metabolites in milk.** Sanne van Gastelen<sup>\*1,2</sup>, Elsa C. Antunes-Fernandes<sup>1,3</sup>, Kasper A. Hettinga<sup>3</sup>, and Jan Dijkstra<sup>2</sup>, <sup>1</sup>Top Institute Food and Nutrition, Wageningen, the Netherlands, <sup>2</sup>Animal Nutrition Group, Wageningen University, Wageningen, the Netherlands, <sup>3</sup>Food Quality Design Group, Wageningen University, Wageningen, the Netherlands.

The objective of this study was to develop prediction equations for methane (CH<sub>4</sub>) emission of dairy cows using fatty acids (FA), volatile metabolites, and non-volatile metabolites in milk. Data from a randomized block design experiment with 32 multiparous Holstein Friesian cows and 4 diets was used. All diets had a roughage:concentrate ratio of 80:20 based on dry matter. Roughage consisted of grass silage only, corn silage only, or mixtures of both silages. Methane emission was measured in climate respiration chambers, and expressed per unit dry matter intake (DMI) and per unit fat- and protein-corrected milk (FPCM). Milk samples were analyzed for FA by gas chromatography, volatile metabolites by gas chromatography-mass spectrometry, and non-volatile metabolites by nuclear magnetic resonance. A multivariate model was developed using a stepwise procedure with selection of FA, volatile, and non-volatile metabolites. Only variables with  $P < 0.05$  entered the model, and variables with  $P < 0.10$  were retained in the final model. Multivariate analysis, using only FA (g/100g total FA), resulted in equations: CH<sub>4</sub> (g/kg DMI) = 29.5 (±1.33) - 2.13 (±0.98) × C18:2n-6 - 5.37 (±1.31) × total CLA (adjusted R<sup>2</sup> = 0.53), and CH<sub>4</sub> (g/kg FPCM) = 11.7 (±2.13) + 42.7 (±7.55) × C15:0iso - 9.88 (±3.23) × C17:0 (adjusted R<sup>2</sup> = 0.51). Multivariate analysis, using FA, volatile metabolites (peak area) and non-volatile metabolites (area change), resulted in equations: CH<sub>4</sub> (g/kg DMI) = 28.6 (±0.97) - 6.33 (±1.00) × total CLA - 6.21 (±1.65) × N-acetyl sugar + 2.46 (±0.83) × choline (adjusted R<sup>2</sup> = 0.69), and CH<sub>4</sub> (g/kg FPCM) = 7.2 (±2.32) + 1.23 × 10<sup>-6</sup> (5.26 × 10<sup>-7</sup>) × 3-nonanone + 22.1 (±5.69) × C15:0iso - 205.1 (±66.21) × uridine diphosphate hexose + 62.7 (±28.17) × Unknown (adjusted R<sup>2</sup> = 0.68). The potential of milk FA only to predict CH<sub>4</sub> emissions was moderate. Including volatile and non-volatile metabolites enhanced the predictive power, suggesting that these metabolites, in combination with milk FA, hold potential to predict CH<sub>4</sub> emission of dairy cows.

**Key Words:** methane emission, milk fatty acids, milk volatile and non-volatile metabolites

**595 Methane prediction equations for beef cattle fed low forage diet.** Paul Escobar-Bahamondes<sup>\*1,2</sup>, Masahito Oba<sup>1</sup>, and Karen A. Beauchemin<sup>2</sup>, <sup>1</sup>University of Alberta, Edmonton, AB, Canada, <sup>2</sup>Agriculture and Agri-Food Canada, Lethbridge, AB, Canada.

The goal of this study was to develop equations to predict CH<sub>4</sub> emissions from beef cattle fed low forage diets (≤20% DM basis) and compare their predictions with IPCC 2006. A dietary forage ≤20% DM database was constructed with treatment means from 17 beef studies published between 2000 and 2014. Criteria for inclusion in the database were:

intake, diet composition and enteric CH<sub>4</sub> production. To overcome the limited size of the database (n = 34) each study was resampled 1,000 times using a Monte Carlo technique to create a new virtual data set. Outliers were excluded by Mahalanobis procedure. Three steps were used to obtain new equations: 1) variables associated with CH<sub>4</sub> production were detected by Principal Component Analysis, 2) stepwise forward multiple regression was used with the original database to obtain prediction equations with study included as a random effect using the Mixed procedure, and 3) stepwise forward regression and K-fold cross validation (n = 5) was used with the Monte Carlo data set (n = 34,000). Observed-predicted values were used to evaluate model performance using the concordance correlation (r<sub>c</sub>) and root mean square prediction error (RMSPE, g/d). Statistical analysis was performed using JMP v.11. The best-fit equation using the original database was: CH<sub>4</sub> (g/d) = -26.4(±20.17) + 0.21(±0.04) × BW(kg) + 38.1(±11.83) × CP(kg/d) - 70.5(±25.48) × fat<sup>2</sup>(kg/d) + 10.1(±5.12) × (NDF-ADF)<sup>3</sup>(kg/d) with  $P < 0.05$ , r<sub>c</sub>: 0.91 and RMSPE: 13.74; where BW = body weight, CP = crude protein, NDF = neutral detergent fiber, and ADF = acid detergent fiber. The best-fit Monte Carlo equation was: CH<sub>4</sub> (g/d) = -10.1(±0.62) + 0.21(±0.001) × BW(kg) + 0.36(±0.003) × DMI<sup>2</sup>(kg/d) - 69.2(±1.65) × fat<sup>3</sup>(kg/d) + 13.0(±0.45) × (CP/NDF) - 4.9(±0.07) × (starch/NDF) with  $P < 0.001$ , r<sub>c</sub>: 0.92 and RMSPE: 12.6 where DMI = dry matter intake. These new prediction equations had greater r<sub>c</sub> and lower RMSPE than IPCC 2006 (r<sub>c</sub>:0.29; RMSPE:43.2), indicating greater prediction accuracy. Using the Monte Carlo equation data set improved accuracy of prediction compared with the original equation database. Both equations specifically developed for cattle fed low forage diets increase the accuracy of predicting CH<sub>4</sub> emissions compared with IPCC (2006).

**Key Words:** methane, beef cattle, equation

**596 Evaluating extant empirical models for predicting enteric methane emissions from lactating dairy cows.** J. A. D. R. N. Appuhamy\* and E. Kebreab, Department of Animal Science, University of California, Davis, CA.

Empirical models are widely used to estimate enteric methane (CH<sub>4</sub>) emissions from dairy cows worldwide. The objective of this study was to evaluate extant models for predicting CH<sub>4</sub> emissions from dairy cows using literature data. Thirty-nine extant models developed based on dairy cow data were evaluated using measurements from 47 studies published after 2000. The data containing dietary, production and animal information included 50, 83, and 41 enteric CH<sub>4</sub> measurements of lactating dairy cows from North America (NA), Europe (EU), and Australia and New Zealand (AUNZ), respectively. The models were evaluated using root mean square prediction error (RMSPE), concordance correlation coefficient, and Nash-Sutcliffe efficiency statistics. An index including equally weighted statistics was used to rank the models within each region. For NA, the 5 top ranked models were those by Hristov et al. (2013), Moe and Tyrrell (1979), Ellis et al. (2007), Moraes et al. (2014), and Moate et al. (2011) [Ref 1–5, respectively, in Table 1] and had RMSPE 15–17% of the average observed value. The majority of the best performing models in NA were developed on data from NA cows. A completely different Set of models performed best on both EU (RMSPE = 11–13%) and AUNZ (RMSPE = 11–15%) data. The best performing models were those by FAO (2010), IPCC (1997), Storlien et al. (2014), Yan et al. (2000), and Nielsen et al. (2014) [Ref I–V respectively in Table 1]. Regional origin and perhaps diet type of the data on which models have been developed need to be considered when selecting a model to predict CH<sub>4</sub> emissions successfully.



**Table 1 (Abstr. 596).** Top ranked models for predicting enteric CH<sub>4</sub> (MJ/cow/d) emissions<sup>1</sup>

NA	Ref	EU and AUNZ	Ref
= [2.54 + 19.14 × DMI] × 0.05565	1	(0.0975 – 0.0005 × DMd) × GEI	I
= 3.14 + 0.51 × NSC + 1.74 × HC + 2.65 × C	2	0.065 × GEI	II
= 4.08 + 0.068 × MEI	3	1.47 + 1.28 × DMI	III
= –9.311 + 0.042 × GEI + 0.094 × NDF% – 0.381 × FA% + 0.008 × BW + 1.621 × MilkF	4	3.234 + 0.0547 × GEI	IV
= [exp(3.15 – 0.0035 × FA)] × DMI × 0.05565	5	1.26 × DMI	V

<sup>1</sup>DMI, NSC, HC = hemicellulose, C = cellulose (all in kg/d), FA and FA% = dietary fat (g/kg of DM and % of DM), DMd = diet DM digestibility (%), GEI and MEI = gross and metabolizable energy intake (MJ/d), NDF% (% of DM), BW (kg), MilkF = milk fat %.

**597 Comparison between the GreenFeed system and the sulfur hexafluoride tracer technique for measuring enteric methane emissions from dairy cows.** Joonpyo Oh<sup>\*1</sup>, Fabio Giallongo<sup>1</sup>, Tyler Frederick<sup>1</sup>, Mike T. Harper<sup>1</sup>, Holley Weeks<sup>1</sup>, Antonio F. Branco<sup>2</sup>, Alexander N. Hristov<sup>1</sup>, William J. Price<sup>3</sup>, Peter J. Moate<sup>4</sup>, Matthew H. Deighton<sup>4</sup>, S. Richard O. Williams<sup>4</sup>, Maik Kindermann<sup>5</sup>, and Stephanie Duval<sup>6</sup>, <sup>1</sup>The Pennsylvania State University, University Park, PA, <sup>2</sup>Universidade Estadual de Maringá, Maringá, Paraná, Brazil, <sup>3</sup>University of Idaho, Moscow, ID, <sup>4</sup>Agriculture Research Division, Ellinbank Centre, Ellinbank, Victoria, Australia, <sup>5</sup>DSM Nutritional Products, Base, Switzerland, <sup>6</sup>DSM Nutritional Products France, Saint Louis Cedex, France.

The objective of this study was to compare 2 commonly used techniques for measuring CH<sub>4</sub> emissions from ruminant animals, the GreenFeed (GF) system and the sulfur hexafluoride (SF<sub>6</sub>) technique. The study was part of a larger experiment, in which a CH<sub>4</sub> inhibitor, 3-nitrooxypropanol (3NOP), fed at 4 application rates (0, 40, 60, and 80 mg/kg feed DM) decreased enteric CH<sub>4</sub> emission by 25 to 32% in a 12-wk experiment with 48 lactating Holstein cows. The larger experiment used a randomized block design and was conducted in 2 phases (Feb-May, phase 1 and Jun-Aug, phase 2), with 24 cows in each phase. Methane emissions using GF were measured during experimental wk 2, 6, 9, and 12. During each GF measurement, 8 spot samples of gas emissions were collected from each cow, staggered over a 3-d period (a total of 0.67 h/cow). Emission data using the SF<sub>6</sub> technique were collected for 3, 24 h periods (a total of 77 h/cow) during wk 2, 6 or 9, and 12. An outlier analysis removed 1 observation from the GF data set (1,271 observations) and 6 observations from the SF<sub>6</sub> data set (451 observations). Methane yield data (g/kg DMI) were averaged per cow for the statistical analysis. The mean CH<sub>4</sub> yield, SD, lower and upper 95% CL, CV, and min and max values for the GF data set were (g CH<sub>4</sub>/kg DMI or as indicated): 12.8, 3.63, 12.8 and 13.9, 27.2% (18.1 and 21.2%; control and 3NOP cows, respectively), and 6.7 and 26.4. For the SF<sub>6</sub> data set these values were: 14.7, 5.60, 14.7 and 17.0, 35.3% (30.4 and 29.9%, control and 3NOP cows), and 7.2 and 36.5. Data were analyzed within experimental phase, sampling week, and treatment to compare CH<sub>4</sub> yield between GF and SF<sub>6</sub>. The difference between the 2 methods (SF<sub>6</sub> – GF) within treatment was 1.9 to 4.1 g CH<sub>4</sub>/kg DMI ( $P < 0.001$  to 0.06) for phase 1 and 1.1 to 2.4 g/kg DMI ( $P = 0.06$  to 0.38) for phase 2. In the conditions of this experiment, the SF<sub>6</sub> technique produced larger variability in CH<sub>4</sub> yield than the GF method. The difference between the 2 methods was not consistent over time, perhaps influenced by barn ventilation and background CH<sub>4</sub> and SF<sub>6</sub> concentrations.

**Key Words:** methane, GreenFeed, sulfur hexafluoride

**598 Breed and lactation stage affect rumen methanogens in co-housed primiparous dairy cattle.** L. M. Cersosimo<sup>\*1</sup>, M. Bainbridge<sup>1</sup>, J. Kraft<sup>1</sup>, and A.-D. G. Wright<sup>2</sup>, <sup>1</sup>University of Vermont, Burlington, VT, <sup>2</sup>University of Arizona, Tucson, AZ.

Rumen methanogens are anaerobic, methane-producing archaea that decrease feed efficiency in dairy cattle and contribute to global warming. The objective of this study was to determine if breed or lactation stage affect the rumen methanogen diversity and density. Seven Holstein (H), 8 Jersey (J), and 7 Holstein-Jersey crossbreeds (X) were co-housed in a free stall on a total mixed ration, and whole rumen digesta samples were collected at 3, 93, 183, and 273 d in milk (DIM). To determine the methanogen diversity, the V1-V3 region of the 16S rRNA gene was amplified. Sequences were generated using Illumina MiSeq v.3 and all bioinformatics analyses were performed using Mothur. Real-time PCR amplified the methanogen-specific mcrA gene and the mcrA copy numbers per mL rumen digesta were calculated. Data were analyzed using PROC MIXED in SAS. The model included the effects of breed (B), lactation stage (LS), and B × LS. A total of 1,683,569 16S sequence reads were classified to species-level taxa by B and LS. The *Methanobrevibacter (Mbr.) smithii-gottschalkii-millerae-thaueri* and *Mbr. ruminantium-olleyae* clades did not differ. Notably, the mean % abundance of the species *Mbr. thaueri* differed ( $P < 0.01$ ) by LS. At 93 DIM, *Mbr. thaueri* abundance was higher in J (35.0%) compared with both H (24.5%,  $P < 0.05$ ) and X (19.4%,  $P < 0.01$ ), while no significance was observed at 183 and 273 DIM, respectively. Differences by LS were seen in less abundant species (<5%), *Mbr. woesei* ( $P < 0.001$ ), *gottschalkii* ( $P < 0.05$ ), *millerae* ( $P < 0.05$ ), and *Methanosphaera stadtmanae* ( $P < 0.01$ ). The top operational taxonomic units (OTU) from a 2% cutoff were related to *Mbr. smithii*, *Mbr. thaueri*, *Mbr. ruminantium*, and *Mbr. millerae*. There were no differences in methanogen density, the number of OTUs (18–37 per breed), or Shannon and Inverse Simpson indices. Chao values differed by LS ( $P < 0.01$ ). Our data suggest that LS may have had a greater effect on the methanogen diversity than B and that core rumen methanogens persist over a lactation period.

**Key Words:** archaea, diversity, *Mbr. thaueri*

**599 Aerodynamic design of a TMR feed bin to measure gaseous emissions from cattle.** Scott Zimmerman<sup>\*1</sup>, John Roche<sup>2</sup>, and Pat Zimmerman<sup>1</sup>, <sup>1</sup>C-Lock Inc., Rapid City, SD, <sup>2</sup>Dairy NZ, Hamilton, New Zealand.

The need to measure methane (CH<sub>4</sub>) and carbon dioxide (CO<sub>2</sub>) emissions from cattle in production environments is increasing. Previously, CH<sub>4</sub> and CO<sub>2</sub> emissions have been accurately measured for short-term repeated periods using a supplemental feeding station (GreenFeed, C-Lock Inc.). Considering the interest in residual feed intake, there is an opportunity to combine the measurement of feed intake with the

measurement of CH<sub>4</sub>, by incorporating this technology into a TMR-feeder. To do this effectively, it is necessary to design a feed bin that has desirable airflow characteristics, that ensure a high capture of emitted gasses and produces a fast response time to emitted gasses. The purpose of this study was to design a TMR feeder with optimal airflow characteristics. A TMR bin, 92 × 92 × 81 cm was constructed (dead-volume = 710 L). An opening for the cow's head of 30 × 6 cm was made and the lid was sealed. Bin dimensions enabled adequate TMR to be offered, while allowing the cow to reach all corners. Airflow was extracted from the bin at a rate of 45 L/s using a fan. From the outlet gas, CO<sub>2</sub> concentrations were measured. Artificial smoke was used to visualize flow patterns inside the TMR bin and results were recorded on video. Separately, CO<sub>2</sub> gas was injected into the TMR bin at 7 g/min until the system reached equilibrium, at which point the CO<sub>2</sub> was removed. The T-90 equilibrium time of CO<sub>2</sub> disappearance was analyzed. Three gas extraction methods were tested: (m1) a 10-cm-diameter circular outlet, (m2) a manifold with 36 evenly spaced 1.27-cm-diameter holes in only the upper half of the bin, (m3) a manifold with 72 evenly spaced 1.27-cm-diameter holes throughout the bin. There was no observed sample loss from the head opening with any outlet configuration. However, m2 and m3 produced a significantly longer T-90 equilibrium time compared with m1 (24, 33, and 66 s, respectively). The multiple air outlets in m2 and m3 produced improved airflow characteristics. Although m3 produced the fastest equilibrium time, the air holes at the bottom of the bin could become plugged with TMR; therefore, manifold 2 is likely the optimal option. In conclusion, a well-designed TMR bin can produce acceptable airflow characteristics for the measurement of gaseous emissions from cattle while they are feeding.

**Key Words:** methane, emissions, cattle

#### 600 Predicting nitrogen excretion from lactating dairy cattle.

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A genetic algorithm was implemented to select models to predict fecal, urinary, and total manure nitrogen (N) excretion by lactating dairy cows. Akaike's Information Criterion was used as the criteria for model selection. Data for model development consisted of 1,047 indirect calorimetry observations on lactating cows collected at the USDA's Energy and Metabolism laboratory from 1963 to 1995. Two tiers of model classes based on model input requirements were developed resulting in 6 models. Tier 1 selects a single covariate model to estimate N excretion and the Tier 2 selects models from many potential dietary and animal covariates including BW, DIM, CP, NDF, ash content, and proportion of concentrate in the diet. Animal and study were designated as cross-classified random effects and the final selected mixed models were fit using restricted maximum likelihood in R statistical software with the lme4 package. The root mean square prediction error (RMSPE) was used to evaluate the models in 3 ways: (1) K-fold cross validation based on all data, (2) evaluation with the most recent 6 years of data, and (3) evaluation with N balance data collected from literature published from 1996 to 2014. The number of published studies reporting covariates required for tier 2 models was not sufficient for model evaluation. Results listed in Table 1 for tier 1 models show better prediction for total manure N and fecal N compared with urinary N excretion. Tier 2 models had lower RMSPE than Tier 1 models across all forms of excretion.

**Table 1 (Abstr. 600).** Model selection and evaluation reported in the RMSPE (% mean N excretion); parameter estimates with standard errors in parentheses

Model estimates	USDA cross-validation	USDA 1990-1995	Literature data 1995-2014
UN = 12.0(5.80) + 0.333(0.0106) × NI	23.7	12.7	23.8
FN = -18.5(3.59) + 10.1(0.169) × DMI	12.6	18.2	16.2
TN = 20.3(4.72) + 0.654(0.00926) × NI	9.80	8.14	10.8

UN = urine nitrogen, FN = fecal nitrogen, TN = total manure nitrogen, NI = nitrogen intake (g/d).

**Key Words:** dairy, nitrogen, prediction

#### 601 Checks and balances: Evaluating reliability of dairy nutrient management data to better protect groundwater resources.

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To protect groundwater from further nitrate contamination, California regulations require dairy farmers to follow a strict nutrient management plan. The regulations require copious annual reporting of crop field management, farm infrastructure, and animal population. The data collected in these annual reports could be integral to evaluating and improving both farm practices and the regulations themselves. The objectives of this project were to (1) assess the reliability and accuracy of annual report data to allow the information to be used responsibly and (2) to suggest promising methods of improving data quality. Annual reports from 18 dairies were obtained to assess data reliability. Mass balance calculations were performed to check the self-consistency of data within a facility. The results of mass balance calculations show that the annual reports do not account for a remarkably large percentage of the nutrients being produced on the farms. Literature suggests that over 60% of nitrogen (N) and 90% of phosphorus (P) should be recovered; however, a median of only 25% of both N and P in cattle manure was recovered based on annual reports. This could be due to many different causes including inaccurate nutrient sampling and analysis techniques or fraudulent reporting. It is likely that the sampling and analysis methods are a significant source of error because the accuracy of the majority of the protocols has not been assessed. The results of this study have led to projects that should improve data collection protocols in both the short and long-term. In the short term, online decision trees are being developed in collaboration with extension agents and consultants to help farmers self-assess their current data collection practices, and provide personalized suggestions for improvement. Suggestions are currently based on the recommendations of experts, but will be improved in the long term using a statistical modeling approach paired with field experiments to examine the uncertainty in each of the recommended protocols. With more reliable data, we can identify regulations and management practices that are more protective of groundwater quality.

**Key Words:** dairy waste management, nitrate leaching

#### 602 Economic and environmental implications of wheat crop rotations on organic dairy farms.

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Our objective was to determine the sustainability for 8 crop rotation sequences of 3-yr rotations in a long-term (25-yr) well-managed organic

dairy farm in Maine. A medium-sized organic dairy farm was simulated with the Integrated Farm System Model (version 3.6) to evaluate crop rotation (management) effects on crop performance, environmental impacts and profitability. The 9 cropping strategies included continuous ryegrass/red clover (continuous grass), corn harvested early followed by winter wheat (corn-wwheat-grass), corn followed by spring wheat (corn-swheat-grass), ryegrass/red clover rotated with winter wheat (grass-wwheat-grass), ryegrass/red clover in rotation with spring wheat (grass-swheat-grass), soybean rotated with both winter wheat (soybean-wwheat-grass) and spring wheat (soybean-swheat-grass), corn double cropped (corn-corn-grass) and soybean followed by corn (soybean-corn-grass). Wheat was harvested as a cash crop in all simulated years and sold at an organic premium price. All rotations were in long rotation with perennial ryegrass/red clover over the 3 yr period. When the continuous grass-based system was simulated, farm net return was higher (\$742.15/cow). Under crop rotation, the option of growing and selling winter wheat instead of spring wheat improved farm profitability when

the results were compared with other wheat cropping rotations. The higher winter wheat yields for soybean-wwheat-grass resulted in 7% more income from feed sales (\$1,027) and 1% more in total revenue (\$1,065). Also, winter wheat should be encouraged to reduce soil nutrient accumulation, particularly soybean replaced with cash crop wheat (soybean-wwheat-grass). Here the major reductions in nutrient flows were a 0.8 kg/ha/yr decrease in both P runoff and leachate losses and K accumulation (-17%); there was also a 4% reduction in water footprint (kg/kg FPCM). Use of corn harvested as silage in place of grass or/and soybean in the first year or winter/spring wheat in the second year provided relatively less profitability to the farm. In general, there were lower economic and more favorable environmental benefits to shifting land from continuous grass-based production to specified small grain cropping rotations.

**Key Words:** dairy, wheat, integrated farm system model



## Ruminant Nutrition: Manipulating rumen function

**603 The effects of Megalac and a fatty acid prill containing high levels of palmitic acid supplementation on milk fatty acid composition with early lactation dairy cows.** Guiling Ma<sup>1</sup>, Elliot Block<sup>2</sup>, Limin Kung<sup>3</sup>, Joe Harrison\*<sup>1</sup>, and C. Merrill<sup>2</sup>, <sup>1</sup>Washington State University, Puyallup, WA, <sup>2</sup>Arm & Hammer Animal Nutrition, Princeton, NJ, <sup>3</sup>University of Delaware, Newark, DE.

The objective of this study was to compare 2 fat sources, Megalac (Church & Dwight Co. Inc., Princeton, NJ) and a high palmitic acid prill (Guarantee-palmitate (min) 80%) on composition of milk fatty acids of early lactation cows in a feeding trial lasting 12 wks. Thirty multiparous Holstein cows were randomly assigned to 1 of 2 diets. Except for the fat products, other feeding ingredients were identical in the diets. Fat products were supplemented at 1.2% (DMI). Milk components were assessed from AM and PM milkings. Milk fatty acids were analyzed with Proc Mixed. The interaction between time and treatment was also analyzed. There were no differences in the composition of short chain fatty acids (C6:0, 8:0, 10:0, 11:0, 12:0, 14:1T, 14:1, 15:0 and 15:1T) between treatments ( $P > 0.05$ ). Palmitic supplementation increased palmitic acid in milk ( $37.2 \pm 0.5$  vs.  $40.7 \pm 0.6$ ,  $P < 0.05$ ), while there was a fat source x week interaction ( $P < 0.05$ ). Megalac supplementation increased the concentration of stearic acid ( $10.1 \pm 0.3$  vs.  $8.8 \pm 0.3$ ,  $P < 0.05$ ), and no interaction between fat source and time ( $P > 0.05$ ). MEGALAC<sup>TM</sup> supplementation also increased ( $P < 0.05$ ) unsaturated fatty acids C18:1-8T ( $0.081 \pm 0.004$  vs.  $0.067 \pm 0.004$ ), C18:1-9T ( $0.31 \pm 0.03$  vs.  $0.21 \pm 0.03$ ), C18:2 ( $2.5 \pm 0.1$  vs.  $2.2 \pm 0.1$ ), C20:1-8-eicosenoate ( $0.054 \pm 0.003$  vs.  $0.042 \pm 0.004$ ), and there was no time interaction ( $P > 0.05$ ). Megalac supplementation increased ( $P < 0.05$ ) C18:1-11T ( $1.13 \pm 0.08$  vs.  $0.89 \pm 0.08$ ,  $P < 0.05$ ), tended to increase C18:1-12C ( $0.34 \pm 0.01$  vs.  $0.31 \pm 0.01$ ,  $P < 0.07$ ), increased C18:2-*cis*-9,*trans*-11 ( $0.45 \pm 0.02$  vs.  $0.38 \pm 0.03$ ,  $P < 0.06$ ), and there was a time interaction. There was no difference ( $P > 0.05$ ) between fat sources for C16:1T, C16:1, C17:0, C17:1-10-heptadecenoate, C18:1-10-T, C18:1, C18:1-11-C, C18:3- $\gamma$ , C18:3- $\alpha$ , C18:2CLA, and C18:2-*trans*-10,*cis*-12, in milk. Except for C18:1-12C, there was no difference between AM and PM sampling times ( $P > 0.05$ ). Our findings suggest that in the future, it is not necessary to separately analyze the AM, PM samples for milk fatty acid analysis. Feeding of Megalac appears to promote the T 11 vs. T 10 pathway of biohydrogenation.

**Key Words:** milk, fatty acid, dairy

**604 Effects of different levels of supplementation of a molasses and crude glycerol mixture on ruminal fermentation parameters of beef steers.** Francine M. Ciriaco\*, Darren D. Henry, Vitor R. G. Mercadante, Tessa M. Schulmeister, Martin Ruiz-Moreno, G. Cliff Lamb, and Nicolas DiLorenzo, *North Florida Research and Education Center, University of Florida, Marianna, FL.*

We determined the effects of feeding different levels of a 50:50 molasses:crude glycerol supplement on ruminal fermentation and blood parameters. Eight ruminally cannulated Angus crossbred steers (323  $\pm$  42 kg BW) were used in a 4  $\times$  4 duplicated Latin square design. In each of the 4 28-d periods, animals were housed in individual pens at the University of Florida Feed Efficiency Facility, had ad libitum access to Tifton 85 bermudagrass hay, and were randomly assigned to one of 4 treatments: 0, 0.45, 1.36, and 2.27 kg/d (as fed) of a 50:50 liquid mixture of molasses:crude glycerol. Ruminal fluid and blood samples were collected before supplement feeding (0 h) and every 3 h postfeeding

for 24 h. Immediately after each collection, ruminal pH was measured. Plasma was analyzed for blood urea nitrogen (BUN) and ruminal fluid for VFA and NH<sub>3</sub>-N concentrations. Data were analyzed as repeated measures and orthogonal polynomial contrasts were used to determine the effects of supplementation level on ruminal fermentation and blood parameters. As the level of supplementation increased, mean ruminal pH (6.73, 6.83, 6.74, 6.65 for 0, 0.45, 1.36, and 2.27 kg/d, respectively;  $P = 0.03$ ) and concentrations of BUN and NH<sub>3</sub>-N ( $P < 0.001$ ) decreased linearly. Molar proportions of acetate decreased ( $P < 0.001$ ) whereas molar proportions of propionate ( $P < 0.001$ ) and butyrate ( $P = 0.007$ ) increased linearly as the level of supplementation increased. Total VFA concentrations were affected cubically ( $P = 0.005$ ) by liquid supplementation. Feeding up to 2.27 kg/d of the liquid supplement to steers consuming bermudagrass hay caused a decrease in ruminal pH; however, values were not below 6.0, which is the threshold known to affect fiber digestion. Therefore, we concluded that the inclusion of up to 2.27 kg/d of a 50:50 mixture molasses:crude glycerol in forage based diets fed to growing steers, positively affected ruminal fermentation, increasing propionate concentrations at the expense of acetate, which should improve animal performance.

**Key Words:** crude glycerol, forage, molasses

**605 Total-tract pdNDF digestibility in heifers fed with TMR or pelleted ration.** Elena Bonfante\*, Mattia Fustini, Nicola Negri, Alberto Palmonari, Giorgia Canestrari, and Andrea Formigoni, *DIMEVET, University of Bologna, Ozzano Emilia, Italy.*

The aim of this study was to evaluate the total-tract pdNDF digestibility (TTdpdNDF) in heifers fed with the same ration as TMR or pellet. Eight tie-stall heifers (age 336  $\pm$  30d, BW 346  $\pm$  35kg) were used in a 12 weeks study (4 periods of 3 weeks: 2 adaptive and 1 for data collection). Diets had the same ingredients (hay 41.8%, barley straw 27.4%, sunflower 13.7%, grain 16.4%, NaCl 0.7%) but fed in 2 physical forms: TMR and PELLET ( $\emptyset = 8$ mm), thus differing in structure, evaluated through the physical effective factor (pef = 38.73% in pellet, 66.12% in TMR). Heifers, divided in 2 groups, were fed ad libitum, individually, with the 2 diets in alternate periods, and dry matter intake (DMI), DMI/BW, water intake, rumination time, rumen temperature and pH were evaluated daily. Fecal samples for TTdpdNDF determination were collected and average daily gain (ADG) was calculated at the end of each period. Data of the third week of each period were statistically analyzed with ANOVA for repeated measures, while ADG and TTdpdNDF by *t*-test (Statistica v10). The DMI and DMI/BW was higher ( $P < 0.001$ ) in pelleted diet (11.49  $\pm$  1.72 vs. 9.03  $\pm$  1.33 kg) (3.05  $\pm$  0.33 vs. 2.40  $\pm$  0.24%) and water intake increased ( $P < 0.01$ ) during pellet administration (52.0  $\pm$  13.0 vs. 41.0  $\pm$  9.0 L/d). ADG at the end of the study was 1.0  $\pm$  0.45 kg/d. The rate for the pellet diet was greater but not significantly different (1.07  $\pm$  0.32 vs. 0.90  $\pm$  0.54 kg/d). Rumination time was considerably lower ( $P < 0.001$ ) with pellet than TMR (256  $\pm$  58 vs. 521  $\pm$  66 min/d). Diet had no effect on rumen temperature or pH. TTdpdNDF, evaluated using uNDF<sub>240 h</sub> as a marker, was statistically different between pellet and TMR (87.88  $\pm$  3.72 vs. 91.45  $\pm$  1.92%). The results of this study suggest that a complete pelleted diet was well accepted by animals, as showed by higher DMI. Moreover, even if rumination time dropped with the pelleted diet, ruminal pH was maintained similar to those with TMR. TTdpdNDF, despite the difference between 2 diets was significant, maintained high values in both diets. In conclusion, a complete pelleted diet, designed

to provide sufficient amount of physically effective fiber, could be fed to growing ruminants without generating digestive disorders.

**Key Words:** TTdpdNDF, physical effective fiber, heifer nutrition

**606 Rumen degradability of wheat straw is related to changes in lignin properties after fungal treatment.** Sandra J. A. van Kuijk\*<sup>1</sup>, Anton S. M. Sonnenberg<sup>2</sup>, Johan J. P. Baars<sup>2</sup>, Wouter H. Hendriks<sup>1</sup>, and John W. Cone<sup>1</sup>, <sup>1</sup>*Animal Nutrition Group, Wageningen University, Wageningen, the Netherlands*, <sup>2</sup>*Plant Breeding, Wageningen University, Wageningen, the Netherlands*.

The aim was to improve the rumen degradability of wheat straw (WS), which has relatively high cell wall content. Plant cell walls consist of hemicellulose and cellulose that are bound to lignin. These carbohydrates can be an important source of energy for rumen microbes. However, rumen microbes cannot degrade lignin, which blocks the availability of the carbohydrates. The availability of carbohydrates can be increased when lignin is removed in a pre-treatment. In nature, dead plants can be degraded by fungi. Some fungal species degrade lignin without consuming cellulose during vegetative growth. One of the selective lignin degrading fungi, *Lentinula edodes* was used to test the improvement in rumen degradability of WS. Two conditions were tested in triplicate: autoclaved WS inoculated with *L. edodes* and autoclaved WS as control. After 12 weeks of incubation at 24°C, rumen degradability was determined with the in vitro gas production (IVGP) technique (Cone et al., 1996). Lignin, hemicellulose and cellulose content were determined according to the methods described by Van Soest et al. (1991). Changes in chemical composition and IVGP upon fungal treatment were compared with the control, using the generalized linear model method in SAS (v9.3). To test the effect of changes in lignin structure and properties, pyrolysis gas chromatography-mass spectrometry (py-GC/MS) was done on fungal treated WS and the control. *L. edodes* treatment for 12 weeks increased ( $P < 0.05$ ) IVGP of WS compared with untreated WS. Cellulose content was unchanged, while hemicellulose and lignin content decreased ( $P < 0.05$ ). In addition to a decrease in total lignin, py-GC/MS showed an increasing amount of lignin degradation products. Upon *L. edodes* treatment not only a total degradation of lignin occurred, but the composition of lignin also changed. Lignin in WS consists of syringyl (S) and guaiacyl (G) units in a 1:1 ratio. *L. edodes* degraded more S than G units, since the S/G ratio decreased. This decrease in S/G ratio was correlated to an increase in IVGP. We conclude that the *L. edodes* treatment increased the IVGP of WS, which was correlated to both lignin content and composition.

**Key Words:** rumen degradation, plant cell wall, fungal pretreatment

**607 Inoculant effects on silage fermentation and aerobic stability of sorghum wet ethanol co-product/roughage blends.** Pedro R. B. Campanili\*, Jhones O. Sarturi, Michael L. Galyean, Sara Trojan, Lauren A. Ovinge, Barbara J. M. Lemos, Alex Thompsom, David Klein, Mendu Venugopal, and Bradley Johnson, *Texas Tech University, Lubbock, TX*.

Anaerobic storage of blends containing sorghum wet distillers grains plus solubles (WDGS) and low-quality roughages with or without microbial inoculants (DeLaval Manufacturing) on fermentation, losses, and aerobic stability were evaluated. Experimental silos (n = 90; 18.9-L plastic containers) were assigned randomly to 1 of the 15 treatments using a 3 × 3 + 6 treatment arrangement as follows: 65:35 blend of WDGS and one of the following roughage sources: wheat straw (WS), corn stalks (CS), and alfalfa hay:cottonseed hull (CSH) blend, with or

without inoculant (n = 5; inoculant A and B, all roughages; C, D, and E only for CSH); roughages with no inoculants (cotton burs (CB), and corn dry DGS:WDGS blend (52:48; DGS-blend); and pure WDGS. Data were analyzed using GLIMMIX procedures of SAS with d 0 as a covariate. Regardless of inoculation, pure WDGS showed greater ( $P < 0.01$ ) total DM losses (8 vs. 2%), and less acetate (0.44 vs. 0.80%, DM basis) than silage blends. Low acetate concentration (0.13% of DM) was observed on d 0, regardless of treatment. Disappearance of fiber fractions was greater ( $P < 0.01$ ) for blends than for pure WDGS. Inoculation tended ( $P = 0.07$ ) to increase ADF digestibility for the CSH blend, and increased ( $P < 0.01$ ) fiber disappearance for WS blends. Greater ( $P < 0.01$ ) disappearance of NDF and ADF was observed for the WS blend plus inoculants A or B than for the average of other roughage treatments (16.80 vs. 4.57%, and 12.27 vs. 0% for NDF and ADF, respectively). Blends of CSH and CS had less ( $P \leq 0.04$ ) fat loss when inoculant A or B was applied vs. those without inoculant. The CSH blends took more time ( $P = 0.04$ ; 6 h) to lose aerobic stability than other treatments. Independent of inoculation, pH and losses increased ( $P < 0.01$ ) during the aerobic stability for WS, but was less for CSH, with CS blends being intermediate. Inoculation positively affected fermentation profile of ensiled WDGS/low-quality roughage blends, although this depended on type of roughage. Silo post-opening management of blends must be considered once it can account as an important source of loss.

**Key Words:** distillers, ensiling, stability

**609 Effects of urea and fibrolytic enzymes on chemical composition, in vitro digestibility, in vitro degradability, and gas production of cotton gin trash.** Alexandro Pereira Andrade<sup>1,2</sup>, Mauro Pereira de Figueiredo<sup>2</sup>, Danilo Gusmao de Quadros\*<sup>1</sup>, Joel Queiroga Ferreira<sup>2</sup>, and Yann Santos Luz<sup>2</sup>, <sup>1</sup>*Bahia State University, Barreiras, Bahia, Brazil*, <sup>2</sup>*Southwest Bahia University, Vitoria da Conquista, Bahia, Brazil*.

The objective of this study was to evaluate the effects of treatment with urea (0 and 6%) and fibrolytic enzymes (0, 2, 4 and 6%; 75% cellulase and 25% hemicellulase) on chemical composition, IVDMD, dry matter in vitro degradability (IVDEG), and gas production of cotton gin trash. A completely randomized design in a 2 × 4 factorial design was used, with 4 replications. Two kilograms of residue were treated by 60 d with urea dissolved in water, applied to raise the moisture to 25%. The enzymes were sprayed, acting for 24 h. There was significant interaction between urea and fibrolytic enzymes for DM, EE, CP, and ADF ( $P < 0.05$ ). Urea treatment increased CP in 157% ( $P < 0.05$ ). Enzymatic hydrolysis affected quadratically CP in the untreated residue, but positively linear when it was ammoniated. Urea and enzymes were an effective way in reducing cell wall constituents ( $P < 0.05$ ). The IVDMD was greater with ammoniation and when the enzymes doses were increased ( $P < 0.05$ ). There was no significant interaction between urea and fibrolytic enzymes on IVDEG ( $P < 0.05$ ). Although the values of the fraction *a* decreased from 19.1% to 13.6%, ammoniation increased *b* and *c* fractions from 34.2% to 40.4%, and from 0.038 to 0.045, respectively ( $P < 0.05$ ). Using 5% of passage rate, the addition of urea reduced the effective degradability from 33.9% to 32.6%. No effect of urea was observed for potential degradability ( $P > 0.05$ ). Fibrolytic enzymes did not affect the soluble fraction ( $P > 0.05$ ); however, they increased linearly all other IVDEG parameters. Urea did not affect the rate of digestion of  $K_{d1}$  and  $K_{d2}$  ( $P > 0.05$ ), while the enzymes just decreased linearly  $K_{d2}$ . The  $Vf_1$  and  $Vf_2$  were greater with ammoniation ( $P < 0.05$ ), increasing from 16.2 to 51.2 and 51.2 to 62.4 mL/g, respectively, and they were increased linearly with the enzymes doses. The lag time was affected by the interaction between urea and enzymes ( $P < 0.05$ ), being

different in the ammoniated material without or with 2% of enzymes. Urea and fibrolytic enzymes associated treatment enhanced nutritional value of cotton gin trash.

**Key Words:** ammoniation, cellulase, hemicellulase

**610 Effects of *Saccharomyces cerevisiae boulardii* supplementation during the receiving period on growth efficiency, and behavioral and health responses in newly weaned beef heifers.** Monica L. Jenks<sup>\*1</sup>, Gordon E. Carstens<sup>1</sup>, Abbey G. Cupples<sup>1</sup>, Jason E. Sawyer<sup>1</sup>, William E. Pinchak<sup>2</sup>, Kerry S. Barling<sup>3</sup>, and E. Chevaux<sup>3</sup>, <sup>1</sup>Department of Animal Science, Texas A&M University, College Station, TX, <sup>2</sup>Texas A&M AgriLife, Vernon, TX, <sup>3</sup>Lallemand Animal Nutrition, Milwaukee, WI.

Objectives of this study were to evaluate the effects of live yeast (LY; *Saccharomyces cerevisiae boulardii* strain I-1079;  $0.35 \times 10^9$  cfu/g ProTernative) supplementation during the receiving period on growth efficiency, feeding behavior, activity and vaginal temperature in 72 newly weaned beef heifers (initial BW of  $203 \pm 22$  kg). Immediately upon weaning, heifers were vaccinated (Pyramid 5) and ship stressed (800 km) before being returning to the research center. Upon arrival, heifers were allotted to 1 of 4 pens each equipped with 3 GrowSafe feed bunks, and pens to 1 of 2 treatments ( $n = 36$ ) consisting of standard receiving diet (ME 2.36 Mcal/kg, CP 16.5% DM) without LY, and control diet containing LY (5 g ProTernative/kg diet; Lallemand Animal Nutrition). Temperature sensors (iButton) were placed intra-vaginally (CIDR) to record temperature, and HOBO devices attached (hind leg) to measure physical activity for the first 14 d ( $n = 18$ ). LY treatment did not affect morbidity rate (10.4%), vaginal temperature ( $39.2 \pm 0.2^\circ\text{C}$ ), or frequency ( $16.6 \pm 2.2$  events/d) and duration ( $46 \pm 5$  min/event) of standing bouts. ADG tended ( $P < 0.1$ ) to be greater for LY heifers during the first 28 d ( $0.625$  vs.  $0.432 \pm 0.08$  kg/d), but was not affected by LY treatment during the 56-d study. LY treatment did not affect DMI, but DMI increased as the study progressed from  $2.06 \pm 0.22$  (first 14 d) to  $2.91 \pm 0.19\%$  of BW during the 56-d study. LY heifers consumed more ( $P < 0.05$ ) meals ( $16.8$  vs.  $14.6$  vs.  $\pm 1.1$  events/d) that were shorter ( $P = 0.08$ ) in length ( $12.8$  vs.  $14.9 \pm 1.2$  min/event) and smaller ( $P < 0.05$ ) in size ( $0.48$  vs.  $0.55 \pm 0.04$  kg/event) and at a slower ( $P < 0.05$ ) meal-eating rate ( $4.61$  vs.  $5.54 \pm 0.39$  g DM/min) compared with control heifers. Moreover, heterogeneities of DMI (SD =  $0.59$  vs.  $0.92$  kg/d) and RFI (SD =  $0.48$  vs.  $0.73$  kg/d) were less ( $P < 0.05$ ) in LY than control heifers. While the LY treatment did not affect growth efficiency or health status, supplementation with live-yeast may have favorably affected meal patterns of newly weaned beef heifers.

**Key Words:** live yeast, morbidity, feeding behavior

**611 Direct addition or pre-incubation of exogenous xylanase affects in vitro gas production kinetics, degradability and ruminal fermentation activities of three fibrous feeds.** M. M. Y. Elghandour<sup>1</sup>, A. E. Kholif<sup>2</sup>, S. Lopez<sup>3</sup>, A. Z. M. Salem<sup>\*1</sup>, and T. A. Morsy<sup>2</sup>, <sup>1</sup>Facultad de Medicina Veterinaria y Zootecnia, Universidad Autónoma del Estado de México, Toluca, Estado De México, Mexico, <sup>2</sup>Dairy Science Department, National Research Centre, Giza, Egypt, <sup>3</sup>Instituto de Ganadería de Montaña (IGM) CSIC-Universidad de León, Departamento de Producción Animal, Universidad de León, León, Spain.

The use of exogenous fibrolytic enzymes (EFE) technology to improve the utilization of fibrous feeds is a novel approach for enhancing feed utilization ruminants performance. The effects of EFE xylanase on in

vitro fermentation characteristics of maize stover (MS), oat straw (OS) and sugarcane bagasse (SCB) were examined using an in vitro gas production (GP) technique. Three doses of xylanase plus EFE (Dyadic PLUS, Dyadic International Inc., Jupiter, FL) in liquid form were used at 0 (control), 60 (low), 120 (medium) and 240 (high)  $\mu\text{g/g}$  DM of substrate. Ruminant GP,  $\text{CH}_4$  and  $\text{CO}_2$  concentrations were recorded at 2, 4, 6, 8, 10, 12, 14, 24, and 48 h of incubation at 2 methods of application (direct or 72-h pre-incubation). Separate ANOVA was performed for each feedstuff (maize stover, oat straw and sugarcane). Each experiment was laid down in a 2 (mode of EZ application either by pre-treatment or directly)  $\times$  3 (rates of application of 60, 120, and 240  $\mu\text{g}$  xylanase/g DM) factorial plus one control (no enzyme additive) completely random design, for a total of 7 experimental treatments. The analysis was performed using the PROC MIXED of SAS for factorial arrangements of treatments plus one control. Increased ( $P < 0.05$ ) GP at different incubation times was observed with EFE doses addition for the incubated fibrous feeds versus control. The pre-incubation versus direct addition of EZ increased GP of the 3 feeds. However, the direct addition of EFE improved ( $P < 0.05$ ) DM, neutral detergent fiber (NDF), and acid detergent fiber (ADF) degradabilities. Substrates incubated with higher doses of EFE increased ( $P < 0.05$ ) GP, DM, NDF, and ADF degradabilities versus low and medium doses. Enzyme decreased ( $P < 0.05$ ) rumen pH of MS and SCB compared with control. Ammonia-N and total volatile fatty acids (VFA) was unaffected ( $P > 0.05$ ) by EFE application, doses, and application methods in MS and OS. However, total and individual VFA increased ( $P < 0.05$ ) when SCB was incubated with xylanase and this was EZ dose dependent ( $P < 0.05$ ). Methane and  $\text{CO}_2$  concentrations were not affected ( $P > 0.05$ ) with EZ application methods or its doses in MS and OS. However, increasing EFE doses with SCB increased  $\text{CH}_4$  ( $P = 0.013$ ) and  $\text{CO}_2$  concentrations ( $P = 0.006$ ). It could be concluded that application of xylanase enzyme improved GP and rumen fermentation activities. However, the effects were substrate, application methods, and EFE doses dependent, and increasing EZ doses had more effects than low doses.

**Key Words:** enzyme, fibrous feed, gas production

**612 Effects of essential oils and exogenous enzymes for finishing Nellore cattle in feedlot.** Murillo Alves Porto Meschiatti<sup>1</sup>, Lucas Agostinho Pellarin<sup>1</sup>, João Ricardo Rebouças Dórea<sup>2</sup>, Tiago Sabella Acedo<sup>2</sup>, Luis Fernando Tamassia<sup>2</sup>, Cristina Simões Cortinhas<sup>2</sup>, and Flávio Augusto Portela Santos<sup>\*1</sup>, <sup>1</sup>University of São Paulo, Piracicaba, SP, Brazil, <sup>2</sup>DSM Produtos Nutricionais Brasil SA, São Paulo, SP, Brazil.

The objective with this experiment was to evaluate the combination of essential oils and exogenous enzymes on performance of Nellore bulls finished in feedlot. Three hundred Nellore bulls (initial BW =  $330 \pm 33$  kg) were fed during a total period of 90 d with diets containing 82.5% corn, 8.5% sugarcane bagasse, 5% soybean meal, 3% mineral, 1% urea and were randomly allocated to 50 pens. Animals were blocked based on initial BW. The treatments were MON (Monensin, Tortuga – 26 mg/kg DM), CRINA (Essential Oils: Crina Ruminants, DSM – 90 mg/kg DM), CRINA+MON (90 and 26 mg/kg DM, respectively), CRINA+RUM (CRINA +  $\alpha$ -amylase: Ronozyme RumiStar, DSM – 90 and 560 mg/kg DM, respectively) and CRINA+RUM+P (CRINA+RUM+Protease: Ronozyme Proact, DSM – 90; 560 and 840 mg/kg DM, respectively). Response variables included: dry matter intake (DMI), average daily gain (ADG), feed efficiency (ADG/DMI), final body weight (final BW), hot carcass weight (HCW) and dressing. The data were analyzed using PROC MIXED of SAS and means were compared by Tukey test considering the block as random effect and treatments as fixed effects.



The DMI, ADG, final BW, HCW were greater for CRINA+RUM in comparison with MON, CRINA+MON and CRINA+RUM+P (Table 1). Cattle fed CRINA+RUM had improvements 9.4% in DMI, 12.2% in ADG, 3.7% in final BW and 4.6% HCW compared with cattle fed MON diets. In conclusion, Nellore bulls fed diets containing amylase and essential oils were slaughtered heavier than bulls fed monensin diets.

**Table 1 (Abstr. 612).** Combination of essential oils and exogenous enzymes on feedlot performance of finishing Nellore bulls

Item	MON	CRINA	CRINA+ MON	CRINA+ RUM	CRINA+ RUM+P	P-value	SEM
Initial BW, kg	330.8	330.8	330.9	330.6	330.7	0.5422	10.9
Final BW, kg	476.4 <sup>b</sup>	486.5 <sup>ab</sup>	474.1 <sup>b</sup>	494.1 <sup>a</sup>	463.1 <sup>c</sup>	0.0001	12.6
DMI, kg/d	8.64 <sup>bc</sup>	9.24 <sup>ab</sup>	8.50 <sup>c</sup>	9.45 <sup>a</sup>	8.44 <sup>c</sup>	0.0001	0.27
ADG, kg/d	1.615 <sup>b</sup>	1.722 <sup>ab</sup>	1.584 <sup>b</sup>	1.812 <sup>a</sup>	1.465 <sup>c</sup>	0.0001	0.06
FE, G:F	0.187 <sup>ab</sup>	0.187 <sup>ab</sup>	0.188 <sup>ab</sup>	0.193 <sup>a</sup>	0.175 <sup>b</sup>	0.0001	0.005
HCW, kg	264.8 <sup>b</sup>	272.5 <sup>ab</sup>	262.3 <sup>b</sup>	277.0 <sup>a</sup>	257.4 <sup>c</sup>	0.0002	8.01
Dressing, %	55.5	56.0	55.5	56.1	55.8	0.2652	0.25

**Key Words:** additive, beef, feedlot

**613 Effect of supplementing grazing cattle with *Saccharomyces cerevisiae* on fiber digestibility and rumen cellulolytic bacteria population.** D. O. Sousa<sup>\*1</sup>, M. A. Arcari<sup>1</sup>, M. V. Biehl<sup>1</sup>, A. V. Pires<sup>1</sup>, E. Chevaux<sup>2</sup>, L. J. Mari<sup>2</sup>, and L. F. P. Silva<sup>1</sup>, <sup>1</sup>University of São Paulo, Pirassununga, São Paulo, Brazil, <sup>2</sup>Lallemand Animal Nutrition, Aparecida de Goiânia, Goiás, Brazil.

The aim of this study was to evaluate the effect of live yeast (*Saccharomyces cerevisiae* CNCM I-1077) supplementation on fiber digestibility and rumen population of cellulolytic bacteria of grazing Nellore cattle, throughout the 4 seasons of the year. Eight rumen cannulated steers were used in a completely randomized design in a 2 × 4 factorial arrangement of treatments: with or without yeast and seasons of the year (spring, summer, fall, and winter). The live yeast product (Levucell SC Farm, Lallemand, Brazil) was given daily, in capsules, via the rumen cannula, to achieve 8 × 10<sup>9</sup> cfu per animal. Animals were kept on an intensive rotational grazing system throughout the year, receiving similar mineral supplementation. Every 45 d, in situ rumen NDF degradability (NDFD) of 5 reference forages was determined after 24 and 48 h incubation: (1) corn silage, (2) bermudagrass hay, (3) sugarcane silage, (4) marandu-grass, and (5) guinea grass (mombaça-grass). In addition, a composite sample representing the liquid and solid phases of the ruminal content was collected for DNA extraction and real-time PCR quantification of 4 rumen cellulolytic bacteria species (*Butyrivibrio fibrisolvens*, *Fibrobacter succinogenes*, *Ruminococcus flavefaciens* and *Ruminococcus albus*). On average, live yeast supplementation increased 24 h-NDFD by 6.8% throughout the year (40.7 vs. 38.1%,  $P < 0.01$ ), without a yeast × season interaction ( $P = 0.45$ ), or yeast × roughage interaction ( $P = 0.39$ ). When analyzed after 48h of rumen incubation, there was a yeast × season interaction ( $P = 0.08$ ). Live yeast supplementation increased

48h-NDFD only during the autumn months ( $P = 0.01$ ). Among the 4 species evaluated, *R. flavefaciens* was the most prevalent cellulolytic bacteria, and yeast supplementation increased by 78% ( $P < 0.01$ ) the relative population of *R. flavefaciens* in the rumen, and this effect was more pronounced during the summer and spring months (yeast × season interaction,  $P = 0.10$ ). Supplementation with live *Saccharomyces cerevisiae* increased fiber digestibility of roughages in steers grazing tropical grasses, and this effect was related to the greater population of *R. flavefaciens* in the rumen.

**Key Words:** cellulolytic bacteria, fiber digestibility, yeast

**614 Effects of enzymatically hydrolyzed yeast supplementation and supplementation frequency on immune parameters among periparturient beef cows and calves.** Janine E. Swartz<sup>\*</sup>, Derek W. Brake, Elaine E. Grings, Eric A. Nelson, Cody L. Wright, Julie A. Walker, Ethan J. Blom, and George A. Perry, South Dakota State University, Brookings, SD.

We evaluated effects of enzymatically hydrolyzed yeast (EHY) and supplementation frequency (FREQ) on immune parameters among beef cows and calves. Eighty multiparous (parity = 4.2 ± 0.3) cows were fed a common brome hay-based diet (CP = 8.0 ± 0.1%). Cows were blocked by expected calving date and stratified by BCS within block before random assignment to treatment. Beginning 88 ± 5 d prior and up to parturition, cows were provided 1 kg daily or 3 kg every 3 d of a soybean hull-based supplement that contained 0 or 3 g/kg EHY. The daily supplement was designed to meet ruminal N requirements. Cows were vaccinated against rotavirus at 62 and 48 ± 5 d before parturition. Jugular blood was collected from cows at 62, 48, 40, 24 and 14 d before parturition. At parturition, colostrum was milked from cows before feeding to calves and jugular blood was collected from cows and calves. Subsequently, calf plasma was collected at 2 and 14 d after parturition. Calf plasma IgG concentration increased (*Quadratic* < 0.01) as age increased and the passive transfer status among calves was 'excellent' (i.e., calf 2 d plasma IgG = 37 ± 1.9 g/L; APHIS, 2010). Nonetheless, plasma IgG was greater ( $P = 0.03$ ) among calves born to cows supplemented EHY; FREQ had no effect on plasma IgG in calves. Despite differences among calf plasma IgG concentrations, there was no effect of EHY or FREQ on colostrum yield, colostrum concentration of IgG or calf intake of colostrum. Similarly, apparent efficiency of IgG absorption and sera rotavirus neutralization titers among calves aged 14 d was not affected by treatment ( $P \geq 0.36$ ). Cow plasma IgG decreased (*Quadratic* = 0.02) as cows neared parturition and was not affected by EHY ( $P = 0.56$ ) or FREQ ( $P = 0.14$ ). We observed a quadratic increase in rotavirus neutralization titers in cow sera in response to vaccination, as expected. Sera rotavirus neutralization titers were not affected by EHY ( $P = 0.70$ ) nor FREQ ( $P = 0.42$ ). These data suggest that EHY but not FREQ may affect passive transfer of IgG.

**Key Words:** immunity, cattle, supplementation

## Ruminant Nutrition: Protein metabolism

**615 Water partitioning in lactating Holstein cows fed two levels of dietary forage and crude protein contents.** J. A. D. R. N. Appuhamy\*<sup>1</sup>, M. Niu<sup>1</sup>, A. Leytem<sup>2</sup>, R. Dungan<sup>2</sup>, and E. Kebreab<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of California, Davis, CA, <sup>2</sup>USDA-ARS, Northwest Irrigation Research Lab, Kimberly, ID.

Knowledge on relationships between diet composition and water kinetics of dairy cows will assist in understanding water utilization and determining water footprint. The objective of the study was to investigate the effects of dietary forage and CP contents on water partitioning in lactating dairy cows. Twelve Holstein cows were randomly assigned to a 2 × 2 factorial arrangement of 2 forage levels [38 (LF) vs. 53% (HF)] and 2 CP levels [15.2 (LP) vs. 18.5% (HP)] in a 4 × 4 Latin square design with 4 periods. Cows were kept in metabolic cages indoors, where drinking water intake (dWI), DMI, urine weight, feces weight, and milk yield were measured over 3 d. The dietary treatment effects were analyzed in mixed-effect models including random cow effect. Average DMI, dWI, water via feed (fWI), milk yield, and ambient temperature (T) were 20.0 ± 2.2, 100.6 ± 14.3, 2.5 ± 0.2 and 31.4 ± 5.8 kg/cow/d, and 25.9 ± 1.4°C, respectively. Drinking water intake was positively related to DMI ( $P < 0.01$ ). Independent of DMI, dWI increased respectively by 5.0 ± 2.1 and 5.9 ± 2.2 kg/cow/d ( $P < 0.05$ ) for the increases in dietary fiber and CP contents. Irrespective to DMI and the diet composition, dWI increased by 2.2 ± 0.8 kg/cow/d ( $P = 0.02$ ) for unit increase in T. Fecal water output (fWO) increased as DMI increased ( $P < 0.01$ ). Regardless of DMI, fWO increased (FL = 27.2 vs. FH = 30.4 kg/cow/d) as dietary fiber content increased ( $P < 0.01$ ). Urinary water output (uWO) was positively related to dWI ( $P < 0.01$ ). When adjusted for dWI, HP and HF diets were associated with greater uWO (by 3.4 ± 1.1 and 2.0 ± 1.1 kg/cow/d, respectively) than LP and LF diets ( $P < 0.08$ ). Each dietary nutrient modification independently and equally increased total manure water output (fWO+uWO) by 4.7 ± 1.2 kg/cow/d ( $P < 0.01$ ). Water in milk (mWO) and BW were not affected by the dietary treatments. Overall, 26.8 ± 5.4, 28.1 ± 5.7, and 23.5 ± 6.8% of total water intake (fWI+dWI) were partitioned to mWO, fWO, and uWO, respectively. The majority of the rest (21.6 ± 10.7%) may be respiratory-cutaneous water losses. Forage content, dietary composition and temperature should be included for a better estimate of water utilization and partitioning in dairy cows.

**Key Words:** diet composition, lactating cow, water balance

**616 Milk yield and composition responses to changes in supply of net energy and metabolizable protein: A meta-analysis.** Jean-Baptiste Daniel\*<sup>1,2</sup>, Nicolas C. Friggens<sup>1</sup>, Patrick Chapoutot<sup>1</sup>, Harmen Van Laar<sup>2</sup>, and Daniel Sauvant<sup>1</sup>, <sup>1</sup>INRA-AgroParisTech UMR791, Paris, Îles-de-France, France, <sup>2</sup>Nutreco R&D, Boxmeer, North Brabant, the Netherlands.

This study aimed to quantify, through a meta-analysis of literature data, average milk yield, milk composition and energy balance responses to changes in supply of: net energy (NE<sub>L</sub>), metabolizable protein (MP), and their interaction, for dairy cows. Experiments with designs focused on the effects of protein and/or energy levels on milk performance were selected. The dietary NE<sub>L</sub> and MP supplies were estimated for all treatments using the updated digestive model from INRA (Sauvant and Nozière, 2013). From all studies included in the database, 308 experiments (917 treatments) with experimentally induced variations in either NE<sub>L</sub> or MP supplies were kept for this analysis. These treatments

covered a wide range of diet characteristics and therefore a large part of the plausible NE<sub>L</sub> and MP supplies that can be expected in practical situations. The average predicted MP supply was (mean ± SD) 2080 ± 467 g/d and average NE<sub>L</sub> supply was 143 ± 25 MJ/d. The majority of those studies used multiparous Holstein-Friesian cows between 50 and 200 d in milk. Between and within-experiment variations were split to estimated milk responses. Linear and quadratic effects of NE<sub>L</sub> and MP supplies and their interaction on milk responses were systematically tested, and removed from the model when non-significant ( $P > 0.05$ ). Increasing either NE<sub>L</sub> or MP supplies resulted in positive responses for milk, lactose, protein and fat yields. Interactions between NE<sub>L</sub> and MP supplies were significant ( $P < 0.01$ ) for milk yield, milk protein yield and milk lactose yield. Greater responses were found with increasing NE<sub>L</sub> at higher MP supply, and vice versa. Energy balance was related, positively with NE<sub>L</sub> supply, and negatively with MP supply. The negative interaction found between NE<sub>L</sub> and MP supply diminished the effect of MP on energy balance at high NE<sub>L</sub> supply. The current meta-analysis provides accurate estimation of milk yield (RMSE = 1.04 kg) and milk composition responses to change in supply of NE<sub>L</sub> and MP. This study also found significant interactions between NE<sub>L</sub> and MP supply for milk, protein and lactose yields.

**Key Words:** dairy cow, milk composition, meta-analysis

**617 Updating protein requirements and responses of metabolizable protein efficiency in dairy cows and goats.** Daniel Sauvant\*<sup>1</sup>, Gonzalo Cantalapiedra-Hijar<sup>2</sup>, and Pierre Nozière<sup>2</sup>, <sup>1</sup>AgroParisTech-INRA, Paris, France, <sup>2</sup>INRA-VetagroSup, Theix, France.

One of the major target of renewing the INRA feed evaluation system for ruminants was to update their metabolizable protein (MP) requirements as well as their responses to changes in protein supply. These updates have been made from meta-analyses of large databases of INRA and of literature. For the requirements, the nitrogen losses observed in practical situations were quantified. The 3 main routes of losses, which result in non-productive protein requirements, are, in decreasing order of importance (1) metabolic fecal protein (MFP) which was calculated from non-digestible organic matter intake, (2) urinary endogenous nitrogen losses (NUE) which was the NU losses corresponding to digestible N intake = 0 and corrected for purine-N derivative from rumen microorganisms, (3) loss of protein from phaneres (PPH). After, the values of “non-productive” MP requirements were applied to 873 treatments of 296 experiments performed on lactating cows or goats with the aim to study the milk response to MP supply. MP supply was calculated using the renewed INRA model (Sauvant and Nozière, 2013). The efficiency of available MP utilization (EffMP%) was determined with an iterative calculation, assuming the same common EffMP value not only for milk protein synthesis but also for MFP, for PPH as well as for the accretion or mobilization of body proteins which was related to the calculated energy balance. This led to the most accurate prediction of EffMP in response to the dietary concentration of MP (g/kg DM). EffMP presented the same decreasing function to the MP concentration for cows and goats. Moreover, for a given MP content, inter-experiments variations were significantly explained, positively by actual or potential milk protein yield (MPY, g/d) and negatively by the level of dry matter intake (DMI, kg/d). Therefore, for dairy cows, the following regression was calculated: EffMP = 68.3 - 0.54 (MP-100) + 0.029 (MPY-1000) - 1.04 (DMI - 21) [n = 873, R<sup>2</sup> = 0.83, RMSE = 2.9]. In conclusion, the major

novelties of these updates were the fecal and urinary nitrogen losses and the common value of EffMP for all the functions of proteosynthesis.

**Key Words:** protein requirement, efficiency, dairy

**618 The effect of concentration allocation strategy on the performance, health and fertility of high-genetic-merit dairy cows offered a grass silage-based diet.** Mark W. Little\*<sup>1,2</sup>, Niamh O'Connell<sup>2</sup>, and Conrad P. Ferris<sup>1</sup>, <sup>1</sup>Agri-Food and Biosciences Institute, Hillsborough, UK, <sup>2</sup>School of Biological Sciences, Queens University Belfast, Belfast, UK.

Few studies have examined the effect of concentrate allocation strategy on the performance high yielding cows (approximately 40 kg/day). This 140-d study (commencing at calving) examined the effect of concentration allocation strategy: group fed (GF) or individual cow fed (ICF) on feed intake, milk production and composition, tissue changes, energy balance, health and fertility of Holstein Friesian dairy cows (n = 72). With GF, cows were offered a total mixed ration comprising grass silage and concentrates (50:50 DM ratio) plus 0.35 kg chopped straw/cow/day throughout the study. With ICF, cows were offered a basal ration consisting of grass silage, concentrates (6 kg/cow/day) and chopped straw (0.35 kg/cow/day), with this diet designed to meet the cows energy requirements for maintenance plus 24 kg milk/cow/day. With this treatment additional concentrates were offered 'feed-to-yield' via an out-of-parlor feeding system (0.45 kg concentrate/kg milk) based on each individual cow's milk yield during the previous week. Production data were analyzed using a Residual Maximum Likelihood (REML) mixed model analysis; fertility and health data were analyzed using generalized linear model regression analysis with the logit link function, using GenStat Version 16.2. Total concentrate intakes were 1616 kg/cow and 1635 kg/cow for GC and ICF, respectively. Neither silage DM intake ( $P = 0.059$ ) nor total DM intake (22.4(GF), 22.2(ICF) kgDM/day:  $P = 0.243$ ) was affected by allocation strategy. Similarly, concentrate allocation strategy had no effect on milk yield (39.3 (GF), 38.0 (ICF) kg DM/day:  $P = 0.113$ ), milk fat content ( $P = 0.819$ ), milk protein content ( $P = 0.308$ ), somatic cell score ( $P = 0.125$ ), BCS loss to nadir ( $P = 0.521$ ) or mean daily energy balance ( $P = 0.356$ ). Concentrate allocation strategy has no effect on the numbers of cows with mastitis ( $P = 0.617$ ), lameness ( $P = 0.144$ ) or pneumonia ( $P = 0.451$ ). Cows on the ICF treatment tended to have more abomasal disorders ( $P = 0.096$ ), and an improved pregnancy rate to first and second service ( $P = 0.014$ ). Offering concentrates according to milk yield had no effect on cow performance or health, while improving fertility performance.

**Key Words:** concentrate allocation, performance, fertility

**619 How the efficiency of utilization of histidine varies with supply in dairy cows.** Helene Lapiere\* and Daniel R. Ouellet, *Agri-culture and Agri-Food Canada, Sherbrooke, QC, Canada.*

To determine the effect of a limited supply of His, 5 multiparous Holstein cows fitted with catheters were used in a 4 × 4 Latin square plus one cow, with 14-d periods. Cows were fed a diet balanced to supply 103% of NE<sub>L</sub> and 72% of MP requirements (1610 g/d; NRC, 2001). Treatments were abomasal infusion of His at 0, 7.6, 15.2 or 22.8 g/d plus a mixture of AA (595 g/d, CN profile; from Ajinomoto). At each period, 6 sets of blood samples were collected from arterial, portal, hepatic and mammary sources. Splanchnic plasma flow was calculated by downstream dilution of deacetylated para-amino-hippurate and mammary plasma flow, using the Fick principle. Results are given in Table 1. Mammary plasma flow was not affected ( $P = 0.75$ ) by His deletion,

averaging 690, 644, 682, and 690 ± 34 L/h, for incremental doses of His. Portal absorption accounted for 108% of the infused His at the highest dose suggesting very little catabolism of His across the gut. The mammary uptake:output ratio did not differ from unity across infusion rates. However, the splanchnic release was not sufficient to account for MG uptake at the 0 dose, in line with decreased muscle carnosine (a labile pool of His dipeptide) concentration at low His supply (JDS 97 E-Suppl.1:757). The increased hepatic removal observed as His supply increases was the sole responsible of the decreased His efficiency and was directly related to liver inflow of His.

**Table 1 (Abstr. 619).**

His flow (mmol/h)	His infused (g/d)				SEM	P-value	
	0	7.6	15.2	22.8		Linear	Quadratic
Digestive flow <sup>1</sup>	8.9	8.9	9.1	9.1	0.12	0.08	0.92
Total supply (Tsupply)	8.9	10.9	13.2	15.2	0.12	<0.001	0.92
Portal absorption	7.2	8.7	11.2	14.2	0.88	<0.001	0.39
Hepatic removal	-1.2	-1.4	-3.8	-5.0	0.83	0.005	0.53
Splanchnic release	5.9	7.5	7.5	9.8	1.48	0.11	0.82
Mammary (MG) uptake (U)	-6.7	-7.7	-7.5	-9.0	0.49	0.01	0.64
Milk output (O) <sup>2</sup>	6.3	7.5	7.8	8.3	0.24	<0.001	0.21
Liver fractional removal	0.04	0.02	0.04	0.04	0.016	0.72	0.55
MG fractional removal	0.65	0.40	0.22	0.23	0.033	<0.001	0.004
U:O	1.06	1.02	0.97	1.08	0.054	0.92	0.21
Splanchnic/MG	0.92	0.98	0.99	1.09	0.174	0.50	0.91
Maintenance net rqt <sup>3</sup>	1.9	1.9	2.0	2.0	0.02	0.08	0.91
Efficiency <sup>4</sup>	0.95	0.90	0.76	0.70	0.022	<0.001	0.93

<sup>1</sup>NRC (2001).

<sup>2</sup>In protein synthesized within the MG, 28.9 mg His/g true protein.

<sup>3</sup>Maintenance requirement = scurf + endogenous urinary + metabolic fecal (Florida Symposium, 2014:166) excluding efficiency of utilization.

<sup>4</sup>(Milk output + maintenance)/Total supply.

**Key Words:** histidine, metabolism, dairy cow

**620 Effects of rumen-protected methionine, lysine, and histidine on lactation performance of dairy cows.** F. Giallongo\*<sup>1</sup>, J. Oh<sup>1</sup>, M. Harper<sup>1</sup>, J. Lopes<sup>1</sup>, A. N. Hristov<sup>1</sup>, H. Lapiere<sup>2</sup>, R. A. Patton<sup>3</sup>, I. Shinzato<sup>4</sup>, J. Tekippe<sup>4</sup>, and C. Parys<sup>5</sup>, <sup>1</sup>Department of Animal Science, The Pennsylvania State University, University Park, PA, <sup>2</sup>Dairy and Swine Research and Development Centre, Agriculture and Agri-Food Canada, Sherbrooke, QC, Canada, <sup>3</sup>Nittany Dairy Nutrition Inc., Mifflinburg, PA, <sup>4</sup>Ajinomoto Co. Inc., Tokyo, Japan, <sup>5</sup>Evonik Industries AG, Hanau, Germany.

The objective of this study was to evaluate the effects of rumen-protected (RP) Met, Lys and His supplementation to a metabolizable protein (MP)-deficient diet on performance of dairy cows. The experiment was a 9-wk randomized complete block design with 36 Holstein cows (DIM, 132 ± 30 d; BW, 611 ± 81 kg). After a 2-wk covariate period, cows were blocked by DIM, milk yield, and parity, and randomly assigned to 1 of the following 6 treatments: control [AMP; +245 g/d of MP over NRC (2001) requirements]; MP-deficient diet (DMP; -118 g/d of MP); DMP supplemented with RPMet (30 g/d of Mepron; Evonik Industries AG; DMPM); DMP supplemented with RPLys (130 g/d of AjiPro-L; Ajinomoto Co., Inc.; DMPL); DMP supplemented with RPHis (120 g/d of an



experimental product; DMPH); and DMP supplemented with RPMet, RPLys and RPHis (DMPMLH). The AMP and DMP diets consisted of (DM basis): 42% corn and 21% alfalfa silages and 37% concentrates and contained 16.5 and 14.5% CP, respectively. DMI tended to be decreased ( $P = 0.07$ ) by DMP compared with AMP (28.0 vs. 29.4 kg/d). Milk and energy-corrected milk yields were decreased ( $P < 0.03$ ) by DMP (40.5 and 36.1 kg/d) vs. AMP (44.1 and 42.1 kg/d). Milk protein content was increased ( $P \leq 0.03$ ) by DMPH and DMPL (3.17 and 3.20%) compared with DMP and AMP (3.01%), and tended ( $P = 0.06$ ) or was numerically higher ( $P \leq 0.15$ ) for DMPMLH and DMPM (3.15 and 3.12%) vs. DMP and AMP. Milk fat content was decreased by DMP vs. AMP (3.33 and 3.90%;  $P = 0.04$ ) and was increased by DMPH and DMPMLH (3.93 and 4.01%;  $P \leq 0.03$ ) compared with DMP. Yields of milk protein and milk fat were decreased ( $P = 0.01$ ) by DMP vs. AMP (by 10 and 20%, respectively). Cows fed AMP had higher MUN (11.7 mg/dL;  $P < 0.01$ ) compared with cows fed the DMP diets (on average 8.09 mg/dL). Overall, feeding an MP-deficient diet decreased DMI and yields of milk, protein, and fat. Addition of RPAA to the DMP diet generally increased milk protein content but did not affect protein yield. Supplementation of RPHis alone or in combinations with RPMet and RPLys also increased milk fat content.

**Key Words:** methionine, lysine, histidine

### 621 Evaluation of a rumen-protected methionine product for lactating dairy cows at 2 concentrations of dietary crude protein.

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This study evaluated the effect of a supplemental rumen-protected Met product (Timet; VETAGRO S.p.A.; Reggio Emilia, Italy) on lactation performance and rumen measures of dairy cows fed 2 concentrations of dietary CP. Sixteen multiparous Holstein cows (8 ruminally-cannulated) were used in a replicated 4 × 4 Latin square design with 28-d periods. Cows were fed 1 of 4 diets: (1) a control diet containing (DM basis) 40.8% corn silage, 9.5% hay crop silage, and 49.7% grain mix (CON); (2) the CON diet plus Timet (TMT); (3) the CON diet plus Smartamine (Adisseo USA Inc.; Alpharetta, GA; SMT); and (4) a reduced-CP diet plus Timet (TMT-RCP). The TMT, SMT, and TMT-RCP were balanced to contain the same supply of metabolizable Met and Lys, and contained (DM basis) 16.2, 16.2, 15.9, and 13.7% CP, respectively. All diets contained 30.9% NDF and 26.8% starch. Dry matter intake and milk yield were measured on d 22–28. Milk samples were collected d 26–27 to determine composition. Rumen NH<sub>3</sub>-N concentrations were measured on d 26–27 at 4-h intervals. Data were analyzed by ANOVA using the MIXED procedure of SAS. Dry matter intake was unaffected by diet, averaging 26.8 kg/d ( $P > 0.10$ ). Milk yield was unaffected by diet and was 45.6, 46.8, 44.4, and 44.9 kg/d for the CON, TMT, SMT, and TMT-RCP diets, respectively ( $P > 0.10$ ). Milk protein percentage was increased for cows fed SMT (3.33%) compared with the other 3 diets (3.24%;  $P = 0.01$ ), but protein yield was unaffected by diet and was 1.44, 1.49, 1.45, and 1.46 kg/d for the CON, TMT, SMT, and TMT-RCP diets, respectively ( $P > 0.10$ ). Milk lactose percentage was decreased for cows fed SMT (4.55%) compared with the other 3 diets (4.61%;  $P = 0.01$ ). Consistent with reduced CP, MUN (mg/dL) was decreased for cows fed TMT-RCP (11.3) compared with the other 3 diets (13.5;  $P \leq 0.01$ ), and ruminal NH<sub>3</sub>-N (mg/dL) was decreased for cows

fed TMT-RCP (3.4) compared with the other 3 diets (6.9;  $P = 0.001$ ). In conclusion, milk and protein yields were unaffected by sources of rumen-protected Met products. Supplementation of Timet maintained the lactation performance of cows fed reduced CP diets.

**Key Words:** amino acid, reduced CP, rumen measure

### 622 Milk protein yield and plasma concentration of methionine increase in dairy cows fed field peas supplemented with rumen-protected amino acids.

Andre B. D. Pereira<sup>\*</sup>, Andre F. Brito, Kayla M. Aragona, Simone F. Reis, and Kelsey A. Juntwait, *University of New Hampshire, Durham, NH.*

Previous studies showed that diets with more than 15% field peas (FP; DM basis) resulted in reduced milk and milk protein yield in dairy cows. We hypothesize that decreased yields were caused by limited supplies of MP-Lys and MP-Met due to extensive degradation of FP RDP in the rumen. The objective of this study was to evaluate diets with 25% of FP supplemented with rumen-protected (RP) Lys (AjiPro-L) and Met (Smartamine-M) as a substitute for corn meal (CM) and soybean meal (SBM). Twelve multiparous and 4 primiparous lactating Holstein cows were blocked by DIM, milk yield and parity, and randomly assigned to 1 of 4 diets in a replicated 4 × 4 Latin square design. Diets were 35.5% corn silage, 15.5% grass-legume haylage, 5.9% roasted soybean, and: (1) 36% CM and 1.3% urea (3.59:1 MPLys:MPMet ratio; negative control: NC), (2) 29.7% CM, 9.8% SBM, and RPLys-Met (3.07:1 MPLys:MPMet ratio; positive control: PC), (3) 25% FP, 12.3% CM, and 2.4% SBM (3.88:1 MPLys:MPMet ratio; PEAS), and (4) 25% FP, 12.2% CM, 2.4% SBM, and RPLys-Met (3.13:1 MPLys:MPMet ratio; PEASAA). Data were analyzed using the MIXED procedure of SAS and Tukey adjustment for means separation ( $\alpha = 0.05$ ). All dietary treatments had 15.1% CP and 1.58 Mcal/kg. As expected, cows fed NC had decreased DMI, milk protein yield and total concentration of ruminal VFA and increased excretion of MUN. Cows fed NC and PEAS had decreased plasma concentration of Met. Feeding cows PC, and PEASAA mitigated these negative responses. Results showed that plasma Lys was higher for diets fed FP, suggesting that Met is the first limiting AA in FP-based diets.

**Table 1 (Abstr. 622).** Feeding field peas to dairy cows

Item	Diet				SEM	P-value
	NC	PC	PEAS	PEASAA		
DMI, kg/d	23.30 <sup>b</sup>	24.96 <sup>a</sup>	24.63 <sup>a</sup>	25.00 <sup>a</sup>	0.39	<0.01
Milk, kg/d	41.79	42.93	42.69	42.71	1.04	0.53
Milk fat, kg/d	1.40	1.41	1.45	1.49	0.04	0.09
Milk protein, kg/d	1.15 <sup>b</sup>	1.25 <sup>a</sup>	1.21 <sup>ab</sup>	1.24 <sup>a</sup>	0.03	<0.01
MUN, mg/dL	9.85 <sup>c</sup>	7.93 <sup>a</sup>	9.09 <sup>b</sup>	8.77 <sup>b</sup>	0.27	<0.01
Ruminal total VFA, mM	103.08 <sup>c</sup>	113.23 <sup>b</sup>	111.92 <sup>b</sup>	120.92 <sup>a</sup>	1.84	<0.01
Plasma Lys, mM	67.20 <sup>b</sup>	69.94 <sup>b</sup>	77.21 <sup>a</sup>	76.97 <sup>a</sup>	3.96	0.03
Plasma Met, mM	19.92 <sup>c</sup>	27.71 <sup>b</sup>	19.63 <sup>c</sup>	31.89 <sup>a</sup>	1.06	<0.01

**Key Words:** field peas, dairy cow, rumen-protected amino acid

**623 Increased pre-wilting increases metabolizable protein concentration in grass-clover silage.** Marianne Johansen\* and Martin R. Weisbjerg, *Department of Animal Science, AU Foulum, Aarhus University, Tjele, Denmark.*

In organic dairy farming supply with metabolizable protein (MP) is often limited. Therefore, the potential for increased MP concentration by increased pre-wilting of grass-clover before ensiling was investigated. Spring growth and first regrowth grown by 2 Danish organic farmers were cut and pre-wilted to a planned dry matter (DM) content of 35% and 70%, respectively, giving in total 8 silages with DM contents ranging from 28.3 to 72.5%. Four rumen, duodenal and ileal fistulated Holstein dairy cows (551 ± 33 kg body weight, 216 ± 23 d in milk, mean ± SD) were fed ad libitum with the silages in a crossover design, with 5 periods of 3 weeks. No concentrate was offered, but minerals and vitamins were offered daily. Three markers (Cr<sub>2</sub>O<sub>3</sub>, TiO<sub>2</sub> and YbCl<sub>3</sub>·6H<sub>2</sub>O) were dosed in the rumen twice a day for measurement of flow at duodenum and ileum. In the last week of each period daily feed intake was registered and 12 subsamples from duodenum and ileum, respectively, were collected over 94 h to cover the diurnal variation, pooled, and subsequently analyzed. Microbes were isolated from the rumen and analyzed for amino acids (AA) and purines to estimate the flow of microbial protein at the duodenum. Statistical analyses were done in R using a linear random regression model with DM content as fixed effect, cow and growth x farmer as random intercepts and with a growth x farmer random slope. Results given below are per 10% units increase in DM content of grass-clover silage. The absorption of AA in the small intestine was increased ( $P = 0.03$ ) with 5.59 g/kg DM intake. The increased absorption was caused by a higher small intestinal digestibility of AA (1.27 g/100g AA,  $P = 0.03$ ) and a higher flow of AA in the duodenum (5.63 g/kg DM intake,  $P = 0.08$ ). The higher flow of AA in the duodenum derived from both a higher flow of microbial AA (2.12 g/kg DM intake,  $P = 0.06$ ), and a lower true rumen degradation of AA from the silage (-4.04 g/100g AA,  $P = 0.04$ ). In conclusion, the MP concentration in grass-clover silage can be improved by pre-wilting to a higher DM content before ensiling.

**Key Words:** pre-wilting, metabolizable protein, grass-clover silage

**624 Precision-feeding dairy heifers different levels of dietary fiber and high rumen undegradable protein.** Louisa Bowen\*, Ashley Bowyer<sup>1</sup>, Sonya Weeks<sup>1</sup>, Gustavo Lascano<sup>1</sup>, and N. A. Gomez<sup>2</sup>, <sup>1</sup>Clemson University, Clemson, SC, <sup>2</sup>The California Polytechnic State University, San Luis Obispo, CA.

The objective of this experiment was to determine the effects of feeding a high rumen undegradable protein (RUP) diet when dietary fiber level is manipulated within differing forage to concentrate ratio (F:C) on nutrient utilization of precision-fed dairy heifers. Six rumen cannulated Holstein heifers (555.4 ± 31.4 kg BW) were randomly assigned to 2 levels of concentrate, HC [55%] and LC [40%] and to a fiber level sequence [100% oat hay and silage OA, 0% wheat straw WS (low fiber); 83% OA, 17% WS (medium fiber); and 67% OA, 33% WS (high fiber)] administered according to a split-plot 3 × 3 Latin square design (21-d periods). Similar levels of N intake (1.70 g N/kg BW<sup>0.75</sup>) and RUP (55% of CP) were provided. All dependent variables were analyzed as a 3 × 3 Latin square using the PROC MIXED procedure of SAS. No differences were observed for dry matter (DM), neutral detergent fiber (NDF), and acid detergent fiber (ADF) apparent digestibility

(AD) between HC and LC-fed heifers. Heifers receiving HC diets had greater starch AD ( $P < 0.01$ ) than LC heifers. Increasing the fiber level through WS addition resulted in a linear reduction of organic matter AD ( $P = 0.05$ ). There was a linear interaction for DM AD ( $P = 0.05$ ) with a concurrent linear interaction in NDF AD. Nitrogen intake, AD, and retention were not different among treatments, however, urine N excretion increased linearly ( $P = 0.03$ ) with added fiber. Uric acid excretion differed among F:C treatments ( $P < 0.01$ ) but total purine derivatives (PD), microbial CP, and protozoa did not differ between F:C treatments. Microbial CP flow and PD quadratically increased ( $P < 0.05$ ) with WS inclusion. Mean rumen fluid pH quadratically increased ( $P = 0.04$ ) with WS addition, although no F:C effect was detected. Ruminal ammonia concentration did not differ among treatments. Apparent digestibility, purine excretion, and ruminal pH were affected differently as dietary fiber was added through the addition of WS in the diet, suggesting that high RUP can have differential effects on nutrient utilization as dietary fiber is manipulated in precision-fed dairy heifers.

**Key Words:** fiber level, protein degradability, precision-feeding

**625 Dietary fiber and crude protein contents can be modified to minimize enteric methane emissions and nitrogen excretions from dairy cows simultaneously.** M. Niu\*, J. A. D. R.N. Appuhamy<sup>1</sup>, A. Leytem<sup>2</sup>, R. Dungan<sup>2</sup>, and E. Kebreab<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of California, Davis, Davis, CA, <sup>2</sup>USDA-ARS, Northwest Irrigation and Soils Research Lab, Kimberly, ID.

Simulation of enteric methane (CH<sub>4</sub>) emissions and nitrogen (N) excretion from cows fed grass silage-based diet indicated that there may be a trade-off between reduced N excretions and increased CH<sub>4</sub> emissions when certain dietary manipulations were considered. The study aimed to examine effects of changing dietary forage and CP contents on CH<sub>4</sub> emissions and N excretions from lactating dairy cows. Twelve post-peak lactating Holstein cows were randomly assigned to a 2 × 2 factorial arrangement of 2 forage (alfalfa hay) levels [37 (LF) vs. 53% (HF)] and 2 CP levels [15.2 (LP) vs. 18.5% (HP)] in a 4 × 4 Latin square design with 4 periods. Cows were fed and milked twice daily. During the first 14 d, cows were housed in a free-stall barn, where enteric CH<sub>4</sub> emissions were measured using the GreenFeed system from d 8 to 14 in each period. Cows were then moved to metabolic cages, where total feces and urine output (kg/cow/d) were measured using total collection approach for 3 d. Treatment effects were analyzed by a mixed model with the random effect of cow. Dietary forage content was negatively associated with DMI ( $P = 0.09$ ) and milk yield (MY,  $P = 0.02$ ) and positively related ( $P < 0.05$ ) to CH<sub>4</sub> emission per unit of DMI (HF = 20.4 (0.83) vs. LF = 17.8 (0.84) g/kg) and MY (HF = 13.7 (0.80) vs. LF = 11.6 (0.80) g/kg). Dietary CP content did not affect CH<sub>4</sub> emissions but was positively related to milk urea nitrogen content, urine, urine N (UN), and total manure N outputs (MN) per kg of DMI or MY ( $P < 0.01$ ). Dietary forage content was positively associated with only UN (HF = 170 (8.5) vs. LF = 152 (8.5) g/cow/d,  $P < 0.01$ ). Fecal N output was not affected by dietary CP levels. Dietary forage content was positively associated with only UN (HF = 170 (8.5) vs. LF = 152 (8.5) g/cow/d,  $P < 0.01$ ). The enteric CH<sub>4</sub> emissions and MN per unit of milk yield were positively correlated ( $r = 0.67$ ,  $P < 0.01$ ). There were no interactions between dietary CP and forage contents on CH<sub>4</sub> emissions or N excretion. Overall, increasing dietary forage and CP contents independently

increased CH<sub>4</sub> emissions (g/kg of MY) and total manure N excretions (g/kg of MY), respectively.

**Key Words:** dairy cow, methane emissions, nitrogen excretions

**626 Effects of substituting true protein with non-protein nitrogen in Holstein dairy heifers precision-fed different forage to concentrate ratios.**

Prabha G. Ranasinghe\*<sup>1</sup>, Noe A. Gomez<sup>2</sup>, Krysztina Rowland<sup>1</sup>, Ashley Caprio<sup>1</sup>, and Gustavo J. Lascano<sup>1</sup>, <sup>1</sup>*Clemson University, Clemson, SC*, <sup>2</sup>*California Polytechnic State University, San Luis Obispo, CA*.

The objective of this experiment was to determine the effects of feeding different combinations of true protein (TP) and non-protein nitrogen (NPN) when forage to concentrate (F:C) is manipulated on nutrient utilization in precision-fed dairy heifers. Eight ruminally fistulated Holstein heifers (386 ± 9.1 kg BW) were randomly assigned to 2 concentrate levels: HC (25% forage) and LC (75% forage) and to a combination of TP and NPN sequence [U:C; 0% from urea U, 100% casein C (0% U, 100% C; 33% U, 67% C; 67% U, 33% C; 100% U, 0% C)] within concentrate level administered according to a split-plot, 4 × 4 Latin square design (21 d periods). Data were analyzed using the PROC Mixed

procedure from SAS. Intake of N was similar and use U and C to provide 1.80 g N/kg BW 0.75. Neutral detergent fiber (NDF) and acid detergent fiber (ADF) apparent digestibility (AD) were positively affected in HC diets ( $P \leq 0.05$ ). ADF AD linearly increased ( $P < 0.01$ ) in HC, and the opposite occurred in the LC group. Fecal N, urine N, and total excreted N were reduced ( $P \leq 0.05$ ) in the HC treatment (TRT) which resulted in an increased retained N. No U:C effects were observed in AD of DM, OM, and NDF, but NAD, N (% of intake), N (% of digested) decreased linearly as U level increased ( $P < 0.05$ ). A quadratic interaction ( $P < 0.01$ ) was observed; decreasing total excreted N and retained N (g/d) in the HC and the opposite in LC. Total purine derivatives (PD) were positively affected by the HC group ( $P = 0.04$ ) but did not affect microbial protein flow. There was a positive quadratic ( $P < 0.01$ ) response in total PD as U increased in both F:C TRT. Protozoa numbers were higher in the HC diets ( $3.3 \times 10^5$  vs.  $2.8 \times 10^3$ ;  $P < 0.01$ ) and increase quadratically ( $P < 0.01$ ) with U inclusion. No effects for ruminal NH<sub>3</sub>N between F:C TRT was observed, but there was a linear increase ( $P < 0.01$ ) as U level increased. Ruminal pH was lower for HC diets (6.3 vs. 6.6) and there was a quadratic interaction as U increased. Nutrient utilization was significantly affected by manipulation of F:C and U:C.

**Key Words:** heifer, true protein, urea



## Small Ruminant: General

**627 Use of a staphylococcal vaccine to reduce prevalence of mastitis and lower somatic cell counts in a registered Saanen dairy goat herd.** Felicia Kautz, Stephen Nickerson\*, and Lane Ely, *University of Georgia*.

The purpose of this investigation was to evaluate the efficacy of a staphylococcal bacterin in reducing the prevalence of staphylococcal mastitis and somatic cell counts (SCC) in a commercial dairy goat herd. Does were vaccinated or left as controls, and the levels of mastitis and SCC monitored over 18 mo. *Staphylococcus caprae* (42.5%), *S. xylosum* (15.1%), and *S. simulans* (10.0%) were the predominant causes of intramammary infections (IMI). The new infection rate was 1.64 IMI/doe among vaccinates, which tended to be lower ( $P < 0.12$ ) than controls (2.67 IMI/doe). The spontaneous cure rate of existing IMI after immunization was 1.28 cures/doe in vaccinates, which was higher than that observed in controls (0.6 cures/doe;  $P < 0.043$ ). Average SCC of milk samples from vaccinates showed a tendency to be lower than that of controls ( $1274 \times 10^3/\text{mL}$  vs.  $1529 \times 10^3/\text{mL}$ , respectively;  $P < 0.10$ ). Results support the continued study of mastitis vaccines for use in managing staphylococcal mastitis and SCC in dairy goats.

**Key Words:** dairy goat, mastitis, vaccination

**628 Keeping trends and practices for various exotic, cross-bred, and indigenous sheep breeds in sub-tropical highlands of Pakistan.** Muhammad Abdullah\*<sup>1</sup>, Muhammad Mudassir<sup>1</sup>, Jalees Ahmed Bhatti<sup>1</sup>, Abu Saeed Hashmi<sup>2</sup>, Nisar Ahmad<sup>1</sup>, and Umair Younas<sup>1</sup>, <sup>1</sup>*Department of Livestock Production, University of Veterinary & Animal Sciences, Lahore, Punjab, Pakistan*, <sup>2</sup>*Department of Biochemistry, Institute of Biochemistry and Biotechnology, Lahore, Punjab, Pakistan*.

Northern areas of Pakistan are rich in natural resources including livestock as a major source of income for local residents and landless herders. The study objective was to fulfill the knowledge gap about herder's perception toward the relative performance and ecological adaptability of different sheep breeds and their crosses. The herds of 150 pastoralists were considered as case study to investigate their herds. The study was conducted at Kaghan valley of Mansehra district and Haripur according to availability of landless farmers during summer and winter season, respectively. Three pastoral units were selected each from Naran upland and Haripur/Attock lowlands. Herders ( $n = 20$ ) were interviewed using questionnaire from each unit. The collected information were entered in to Epi-info software program (version 6.04b) for descriptive statistics. Various sheep breeds found in northern areas were Afghani, Kaghani, Crossbreed and Rambouillet. Average number of sheep belonging to Kaghani, Rambouillet (exotic), Ramghani (crossbreed) and Afghani sheep per herd were found to be 41%, 6%, 29% and 29%, respectively. Trends for keeping Kaghani, Afghani, Crossbreed and Rambouillet ram as an animal with beauty was among 28%, 40%, 26% and 16% herders. Kaghani sheep was found to be preferred by majority of population for its mutton quality. Highest replacement preference (44.3%) for ram was given to Kaghani breed as compared with Rambouillet which was given least preference (16.78%) due to its low disease resistance against our local conditions. Herders (92%) suggested that high disease resistance was noted in Kaghani breed. In conclusion, such survey studies helped in better understanding of landless farmer's preferences about keeping herd among indigenous, cross and exotic sheep breeds.

**Key Words:** Kaghani, Rambouillet, sheep

**629 Motility of Boer buck spermatozoa stored fresh for 72 hours.** Olumide A. Ajao\*, Daniel M. Barry, and Kow K. Benyi, *University of Venda, Thohoyandou, Limpopo Province, South Africa*.

This study was aimed at evaluating the effects of Biladyl and Triladyl extenders, ambient and refrigerated temperatures (5°C, 12°C and 17°C) on the motility of Boer buck spermatozoa stored for 72 h period of storage. Four ( $n = 4$ ) healthy Boer bucks aged  $3.12 \pm 0.55$  years were ejaculated using artificial vagina (AV) once every 4 d for 6 replicates. The semen samples were pooled and divided into 2 equal halves. One half was extended in a 2-step Biladyl extender (semen sample plus Biladyl Fraction A made up half of the final volume) and the other with Triladyl, both at ratio 1: 5 v/v (semen to extender). Each of the extended semen divisions was further divided into 4 equal parts; one of the 4 was stored at ambient temperature and the others at 5°C, 12°C and 17°C. Data were analyzed by ANOVA for a  $2 \times 4 \times 6$  factorial in a completely randomized design, using the GLM procedure of Minitab (Minitab 2013). Spermatozoa motility was evaluated using Sperm class Analyzer (SCA) after every 12 h for 72 h. Extender type (E) and storage temperature (T) had highly significant effects ( $P < 0.01$ ) on the total motility (TM) of Boer buck spermatozoa kept at the refrigerated temperatures after 72 h of storage compared with the TM of spermatozoa stored at ambient temperature which were the lowest in both extenders. Biladyl had 68% TM on spermatozoa kept at ambient temperature, 85% TM at 5°C, 89% TM at 12°C and 90% TM at 17°C; and Triladyl had 64% TM at ambient temperature, 88% TM at 5°C, 89% TM at 12°C and 90% TM at 17°C. The highest progression obtained after the 72 h was 34% as observed from the semen kept at 17°C in Biladyl and 45% in the semen kept at 17°C Triladyl. With suitable protocol, both extenders can maintain goat sperm motility at specific refrigerated temperatures when kept fresh for up to 72 h.

**Key Words:** Boer buck, ambient, artificial vaginal

**630 *Salix babylonica* as a phytogetic anthelmintic alternative on sheep and goat farms in México.** A. Z. M. Salem\*<sup>1</sup>, M. M. Y. Elghandour<sup>1</sup>, A. E. Kholif<sup>2</sup>, J. C. Vázquez-Chagoyán<sup>1</sup>, R. M. de Oca-Jiménez<sup>1</sup>, A. B. Pliego<sup>1</sup>, and T. A. Morsy<sup>2</sup>, <sup>1</sup>*Facultad de Medicina Veterinaria y Zootecnia, Universidad Autónoma del Estado de México, Toluca, Estado De México, Mexico*, <sup>2</sup>*Dairy Science Department, National Research Centre, Giza, Egypt*.

There is an increased interest in screening phytogetic extracts and medicinal plants as alternatives to the traditional drugs. The efficacy of *Salix babylonica* (SB) extract as a alternative control of gastrointestinal and pulmonary parasites in sheep and goats at the commercial scale was tested. A sample of 20% of the population of 8 sheep and 7 goat farms was included. About 20 mL of SB extract was orally administered weekly at 0700–0900 h before morning feeding for 60 d. A fecal egg or oocyst count technique was performed after 0 (pre-extract administration), 1, 20, 40 and 60 d after the extract administration. Fecal samples were evaluated for the presence of coccidian oocysts, cestode and nematode eggs by a salt flotation technique. Afterward, oocysts or eggs were counted using the McMaster method. Significant differences ( $P < 0.01$ ) in the fecal oocyst and egg output of *Eimeria*, *Dictyocaulus*, and *Moniezia* were observed between sheep and goats. The treatment influenced ( $P < 0.05$ ) egg outputs of *Cooperia*, *Dictyocaulus* and *Trichuris*. The fecal egg or oocyst counts of *Haemonchus contortus*, *Eimeria*, *Cooperia*, *Chabertia*, *Dictyocaulus*, *Moniezia* and *Ostertagia* were

time-dependent ( $P < 0.05$ ). For sheep, administration of SB decreased ( $P < 0.05$ ) the fecal eggs count of *H. contortus*, *Cooperia*, *Chabertia*, *Dictyocaulus*, *Moniezia* and *Trichostrongylus*. After 20 d of treatment, no *H. contortus*, *Cooperia* or *Moniezia* were detected. For goats, only a few species were affected ( $P < 0.05$ ) after the SB administration. The SB reduced ( $P < 0.05$ ) the fecal eggs count of *H. contortus*, *Cooperia*, *Chabertia* and *Moniezia*. Quadratic decreases were observed ( $P < 0.05$ ) for *Chabertia*, *Trichostrongylus* and *Ostertagia*. Eggs of *H. contortus* and *Moniezia* were not present in the feces of goats after one day of treatment; in contrast to the egg output of *Trichostrongylus* and *Ostertagia*. It is therefore concluded that weekly administration of SB at 20 mL weekly can be a promising alternative to the use of synthetic anthelmintics to treat gastrointestinal and lung nematodes of small ruminants in organic and conventional production systems.

**Key Words:** anthelmintic, *Salix babylonica*, small ruminants

**631 A comparison of ewe colostrum and a colostrum alternative (Volostrum) as a colostrum source for artificially reared triplet lambs.** Tommy M. Boland<sup>\*1</sup>, Fiona M. McGovern<sup>1</sup>, Francis P. Campion<sup>1</sup>, and Jessica Cooke<sup>2</sup>, <sup>1</sup>*School of Agriculture and Food Science, University College Dublin, Dublin, Ireland*, <sup>2</sup>*Volac International Ltd., Orwell, Royston, UK*.

Prolificacy is a key driver of output and profitability in sheep-meat production systems. An increase in multiple births is associated with increasing prolificacy. Litter sizes of 3 or more lambs can potentially increase sheep meat production if managed appropriately however such litters are often associated with increased mortality and low post natal growth. Potential mechanisms to reduce these performance bottlenecks include artificially rearing one lamb of each triplet set. The objective of this study was to compare the pre-weaning, post-weaning and slaughter performance of triplet lambs, removed from their dams at birth. Animals were subjected to one of 2 treatments ( $n = 15$  per treatment) for the first 24 h of life. Lambs received either ewe colostrum at one, 10 and 18 h postpartum (C) or they received a commercial colostrum alternative (Volostrum; V). From 24 h postpartum until weaning (at 6 weeks of age) all lambs received artificial milk replacer ad libitum (Lamlac). Concentrate feed was introduced at 10 d of age and lambs were weaned to an all concentrate diet, once daily concentrate intakes reached 250 g FW. Pre-weaning lamb growth rate averaged 384 g/d and did not differ with treatment. Post weaning lamb growth rate tended to be higher for lambs offered ewe colostrum in the first 24 h of life (412 vs. 325 g/d;  $P = 0.06$ ). The C lambs had higher live weights at 10, 11 and 12 weeks of age ( $P < 0.05$ ). There was a tendency for higher lifetime growth rates for the lambs offered ewe colostrum (392 vs. 324 g/d;  $P = 0.07$ ). Lambs offered ewe colostrum during the first 24 h of life reached target slaughter weight (44 kg) 24 d earlier (109 d) than lambs offered Volostrum ( $P < 0.01$ ) however other slaughter and carcass parameters did not differ with treatment. In conclusion, a colostrum alternative supports high pre-weaning and post-weaning lamb growth rates, however performance advantages in terms of post weaning growth rate and reduced days to slaughter are conferred when lambs consume ewe colostrum during the first day of life.

**Key Words:** colostrum, colostrum alternative, lamb

**632 The relationship of body linear measurements and body weight with real-time ultrasound body composition measurements in Boer x Spanish yearling goats.** Flavio R. B. Ribeiro\*, Louis C. Nuti, Shaye K. Lewis, William B. Foxworth, Yoonsung Jung, Bianca Garza, Brandi Owens, Rosemarie Somers, and Gary

R. Newton, *Cooperative Agricultural Research Center, Prairie View A&M University, Prairie View, TX*.

The objective of this study was to determine the relationship of body linear measurements such as hip height (HH), girth circumference (GC), point of the shoulder to hip length (HipShL), forearm circumference (FntLC) and BW with real-time ultrasound (RTU) measurements of body composition, in Boer  $\times$  Spanish cross yearling goats ( $n = 90$ ;  $n = 48$  wethers and  $n = 42$  doelings). The body composition traits measured by RTU were 12–13th rib longissimus lumborum muscle area (uLMA), 12–13th rib fat thickness (uBF), and rump fat thickness (uRUMP). Ultrasound measurements were taken using an Aloka 500 with a 12 cm 3.5 MHz transducer, hair was clipped and vegetable oil was used as a coupling agent to enhance image quality. Data were analyzed by gender and also pooled using the Proc CORR and Proc REG procedures of SAS. BW was highly correlated ( $P < 0.0001$ ) to HH, GC, HipShL, FntLC, uLEA, uBF, and uRUMP (0.65, 0.85, 0.62, 0.70, 0.89, 0.75, and 0.70, respectively) with the pooled data. Similar results were observed when data were analyzed by gender. However, a lower correlation was observed between BW and HipShL and FntLC with the doelings (0.46 and 0.48, respectively) and between BW and HH, uBF and uRUMP for the wethers (0.35, 0.38, and 0.36, respectively). Prediction equations were also developed to predict uLEA, and uBF. Predictions of uLEA had an  $R^2$  of 0.75, 0.64, and 0.65 for the pooled, wether and doelings data, respectively, with BW and HipShL included in the pooled and doelings model and BW and FntLC in the wether model. Predictions of uBF had an  $R^2$  of 0.59, 0.23, and 0.68 for the pooled, wether and doelings data, respectively, with BW and HipShL included in the pooled and wether models and BW and HH in the doelings model. Results showed that RTU body composition traits are highly correlated to body linear measurements in yearling Boer  $\times$  Spanish yearling goats and that different measurements within gender accounted for more variation within the RTU traits. More research is needed to refine the models and improve accuracy of prediction.

**Key Words:** ultrasound, body composition, goat

**633 An investigation in to the interaction between ewe BCS and litter weights at key times of the production cycle.** Francis P. Campion<sup>\*1</sup>, Fiona M. McGovern<sup>1</sup>, Philip Creighton<sup>2</sup>, Alan G. Fahey<sup>1</sup>, and Tommy M. Boland<sup>1</sup>, <sup>1</sup>*University College Dublin, Dublin, Ireland*, <sup>2</sup>*Teagasc Athenry, Co. Galway, Ireland*.

Ewe body reserve accumulation and mobilization is an essential part of the production cycle. The interaction between breed, ewe body condition score (BCS) and animal performance is an area that warrants investigation. Using 3 commercial flocks which are part of a national progeny testing scheme, BCS data was collected from twin bearing ewes at mating, mid-pregnancy, mid-lactation (~d 40 of lactation) and weaning (~d 100 postpartum). This data was then combined with ewe breed, maternal age, and litter weight at birth (BW), d 40 of lactation (D40) and weaning (WE) data. The objective of this study was to investigate if litter weight from birth to weaning of twin bearing ewes was influenced by ewe BCS. Lamb weights collected from each lamb at BW, D40 and WE were summed to calculate litter weight. Regression coefficients were estimated using PROC MIXED (SAS v9.4). The model included the fixed effects of flock, breed, time point and maternal age, and previous years litter size along with the continuous variables of lambing date and lambing difficulty, and BW, D40 or WE. Variables with a  $P$ -value  $> 0.10$  were removed from the final model. Initially a correlation analysis was carried out on BCS and weight. Mid-lactation BCS and weaning BCS had a correlation of 0.63; all other time points had correlations less than 0.40. There was strong correlation between D40 and WE litter

weight (0.80) but not between BW and D40 (0.40) or BW and WE (0.23) litter weights. Changes in BW and WE litter weight were influenced by mid-pregnancy BCS ( $P < 0.05$ ) and there was a tendency toward a relationship between D40 litter weight and BCS at mid-pregnancy and weaning ( $P < 0.10$ ). There was no relationship between litter weight at any stage during the first 14 weeks of life and mating or mid-lactation BCS ( $P > 0.10$ ). Farm, ewe maternal age and breed, time point and breed by time point interaction all had a significant effect on the relationship between BCS and litter weight ( $P < 0.05$ ). These findings show that mid pregnancy BCS is the key BCS measurement linked to litter weight of twin bearing ewes at birth and weaning.

**Key Words:** BCS, litter weight, lamb.

**634 Development of a low-density single nucleotide polymorphism panel for prolificacy in sheep.** Thaisa Lacerda<sup>1</sup>, Harvey Blackburn<sup>2</sup>, Michel Yamagishi<sup>3</sup>, Concepta McManus<sup>1</sup>, Alexandre Caetano<sup>4</sup>, and Samuel Paiva<sup>\*5,2</sup>. <sup>1</sup>Universidade de Brasilia, Brasilia, DF, Brazil, <sup>2</sup>USDA-ARS National Center Genetic Resources Preservation, Fort Collins, CO, <sup>3</sup>Embrapa Informatica Agropecuaria, Campinas, SP, Brazil, <sup>4</sup>Embrapa Recursos Geneticos e Biotecnologia, Brasilia, DF, Brazil, <sup>5</sup>Embrapa Secretaria de Relacoes Internacionais, Brasilia, DF, Brazil.

High-density SNP panels (e.g., 50,000 and 600,000 SNPs) have been used in exploratory population genetic studies with commercial and minor sheep breeds. Routine genetic diversity evaluations of large numbers of samples and panels are in general cost-prohibitive for gene banks. Lower cost panels based mostly on SNPs known to be associated with production traits of interest and may be more efficient for genetic diversity assessment and improvement of gene bank collections. The first phase of the study was to develop and validate a small SNP panel (29 SNPs) based on known prolificacy genes. SNP selection was based on known polymorphisms in major genes affecting sheep prolificacy (*GDF9*, *BMP15* and *BMPR1B*), as well as new polymorphisms mined from whole genome resequencing data from *GDF9* and *BMP15* by the International Sheep Genome Consortium. A total of 125 animals from 15 breeds with litter size information collected by the National Animal Germplasm Program were tested. Genotyping was performed with PCR-based KASP chemistry and 27 SNPs had a call rate higher than 98%. All *BMP15* and *BMPR1B* SNPs were monomorphic. In *GDF9* 8 SNPs were polymorphic - 3 are located in introns and 5 in exons. Three SNPs were non-synonymous AA changes. No significant allele frequency differences were found for GDF9\_G4:E241K among from single or multiple births. The GDF9\_G6:V332I allele was related to high prolificacy in 5 breeds with the following frequencies: Lincoln (0.416), Polypay (0.32), Navajo Churro (0.27), St Croix (0.125) and Suffolk (0.083). Allele GDF9\_G7:V371M was found only in Polypay at a frequency of 0.35 which agrees with recent studies with the hyper-prolific Norwegian White Sheep. The other 10 breeds did not differ significantly. Our results suggest that the developed panel will be useful to identify possible genes/mutations associated with prolificacy in worldwide sheep breeds. Additional SNPs will be included as more information becomes available. In the next phase of this study, SNPs related to other economically important traits will be added to the panel to improve the characterization and management of gene bank sheep collections.

**Key Words:** *Ovis aries*, conservation genetics, gene bank

**635 Influence of surgical castration on biochemical profile of rams.** V. M. Storillo<sup>1</sup>, M. B. R. Alves<sup>1</sup>, E. C. C. Celeghini<sup>1</sup>, B. Barcelos<sup>\*2</sup>, D. B. Birgel<sup>2</sup>, V. F. P. Rispoli<sup>2</sup>, W. C. Garcia<sup>2</sup>, P. S. Silva<sup>2</sup>, and E. H. Birgel Junior<sup>2</sup>. <sup>1</sup>Faculdade de Medicina Veterinária e Zootecnia da Universidade de São Paulo, São Paulo, São Paulo, Brazil, <sup>2</sup>Faculdade de Zootecnia e Engenharia de Alimentos da Universidade de São Paulo, Pirassununga, São Paulo, Brazil.

Castration is often used in male sheep to prevent losses in meat palatability, improve subcutaneous adipose tissue and prevent undesirable pregnancies. But, there is insufficient research on how long for the changes in biochemical profile to take place and finally stabilize as well as which parameters are definitely altered after castration in adult rams. To elucidate this issue, this research was carried on using 31 White Dorper males at 36 mo of age, when biochemical changes are not expected, since they reached adulthood. Animals were surgically castrated, with removal of the scrotum apex, under tranquilization, local anesthesia and using sterile materials. Antibiotic and analgesia treatments were performed for 3 d. Blood samples for measurement of urea, creatinine, albumin, total protein, GGT, AST, CK, HDL, LDL, VLDL, cholesterol,  $\beta$ -hydroxybutyrate (BHB), NEFA and triglycerides, were collected days before castration (D0) and, after that, on d 1, 3, 7, 15, 30, 60 and 9 mo. Castration had no influence on GGT, creatinine, total protein and cholesterol. Up until 15 d after castration, the others parameters were altered because of stress and surgery. Urea increased ( $P < 0.0001$ ) from 19,85 (D0) to 32,43mg/dL (9 mo). Albumin decreased after surgery, possibly due to exudation, and returned to the same values as D0 on D60. AST increased after surgery, then decreased gradually and returned to normal on D15. CK decreased ( $P < 0.0001$ ) from 230,1 to 103,4 U/L, which can be explained for the tranquil temper of rams after surgery, with the absence of fights, that used to be common. The HDL, BHB and NEFA decreased on D3 and remained low. Triglycerides and VLDL decreased on D1 until D15, then raised on D30 to the same values as before castration. The LDL values only increased on D1 and on D3 they were similar to D0. The results showed that surgical castration, even under ideal surgical conditions, causes important changes in biochemical profile, especially in the first 15 d after surgery, with stabilization of values in 30 d. The castration alters the behavior of rams, decreases HDL, BHB and NEFA values and raises urea amounts.

**Key Words:** orchietomy, sterilization, metabolism

**636 Effects of dairy slurry application and bale moisture concentration on voluntary intake and digestibility of alfalfa silage by sheep.** Jessica K. Clark<sup>\*1,2</sup>, Bruce C. Shanks<sup>1</sup>, James D. Caldwell<sup>6</sup>, Ken P. Coffey<sup>2</sup>, Wayne K. Coblenz<sup>3</sup>, R. E. Muck<sup>4</sup>, Dirk Phillip<sup>2</sup>, M. A. Borchardt<sup>3</sup>, Robert T. Rhein<sup>2</sup>, Ashley N. Young<sup>2</sup>, Marshal D. Basham<sup>2</sup>, W. E. Jokela<sup>3</sup>, Elizabeth A. Backes<sup>2</sup>, Keith A. Center<sup>2</sup>, M. G. Bertram<sup>5</sup>. <sup>1</sup>Lincoln University, Jefferson City, MO, <sup>2</sup>University of Arkansas, Fayetteville, AR, <sup>3</sup>USDA-ARS, Marshfield, WI, <sup>4</sup>USDA-ARS, Madison, WI, <sup>5</sup>University of Wisconsin, Arlington, WI, <sup>6</sup>Land O' Lakes.

Dairy slurry is used commonly as a fertilizer in agriculture. However, residual effects of slurry application on intake and digestibility of alfalfa silage from subsequent harvests are not well known. The objective of this study was to determine if moisture concentration of alfalfa silage and timing of dairy slurry application relative to subsequent harvest affect intake and digestibility by sheep. Pregnant crossbred ewes (n = 18; 3–5 yr old; 47.6  $\pm$  5.34 kg) were stratified by BW and allocated randomly each period to 1 of 6 treatments arranged in a 2  $\times$  3 factorial consisting of high (HM; 46.8%) or low (LM; 39.7%) moisture at baling after no slurry application (NS), slurry applied to stubble immediately



after removal of the previous cutting (S0), or slurry applied 14 d after the previous cutting (S14). Period 1 consisted of a 14-d adaptation and a 7-d fecal collection. Period 2 immediately followed period 1 and consisted of an 11-d adaptation with a 7-d fecal collection. Ewes were housed individually in 1.4 × 4.3-m pens equipped with rubber mats and feces were swept from the floor twice daily, weighed, and dried at 50°C. Ewes had ad libitum access to water, were offered chopped silage based on 10% refusal, and were offered commercial sheep mineral (14 g) daily. Intake (g/d) of DM and OM and NDF digestibility (%) did not differ ( $P \geq 0.13$ ) across moisture or slurry application treatments. However, DM and OM digestibility (%) tended ( $P < 0.10$ ) to be greater from LM vs. HM. Total white blood cell concentrations, hematocrits, and red blood cell concentrations were greater ( $P < 0.05$ ) from S0 and S14 vs. NS. Lymphocytes were greater ( $P < 0.05$ ) from LM vs. HM and from NS vs. S0 and S14. Serum urea N concentrations did not differ ( $P > 0.13$ ) across treatments. Therefore, moisture level of alfalfa silage and time of dairy slurry application may not affect voluntary intake or NDF digestibility, but moisture concentration may have a slight effect on DM and OM digestibility. Also, moisture concentration of alfalfa silage and time of dairy slurry application may affect specific blood hemograms.

**Key Words:** digestibility, silage, dairy slurry

**637 Comparison of delayed weaning and mineral form on lamb growth and parasitism.** Jefferson McCutcheon\*, David Clevenger, Gary Lowe, and Francis Fluharty, *The Ohio State University, Columbus, OH.*

The objectives were to 1) compare a 60 d of age and a 120 d of age weaning on lamb growth and parasitism on pasture and lamb growth in a feedlot and 2) compare form of mineral (BLOCK and LOOSE) offered ad libitum on pasture on lamb growth and parasitism in lambs. Each

weaning treatment consisted of 4 replicate fields/pens of 6 lambs per field/pen blocked by gender and initial starting weight. Twin lambs were used with one randomly weaned (WEAN) and one left with its mother for the pasture phase (NURSE). Subdivided perennial pastures were rotationally stocked. Paddocks size matched stocking density between ewes with lambs and weaned lambs. Mineral form was randomly assigned to both weaning groups. After the 63 d pasture phase, nursing lambs were weaned and intact groups were placed in feedlot pens and fed an alfalfa haylage based diet to a pen average final live weight of 54 kg. Statistics were run using SAS Proc Mixed with PDIF for mean separation. At the end of the pasture phase lamb final live weight was greater for the NURSE lambs (LSM ± SEM) ( $39.64 \pm 0.57$  kg) compared with the WEAN lambs ( $30.26 \pm 0.57$  kg) ( $P < 0.05$ ). The pasture ADG was greater for the NURSE ( $254 \pm 6$  g/d) than the WEAN ( $100 \pm 6$  g/d) lambs ( $P < 0.05$ ). Packed Cell volumes were lesser for the WEAN lambs ( $2.6 \pm 0.6$ ) at 63 d than the NURSE lambs ( $1.1 \pm 0.6$ ) ( $P < 0.05$ ). Log-transformed fecal egg counts were greater for the WEAN lambs ( $3.42 \pm 0.29$ ) at 42 d compared with the NURSE lambs ( $1.88 \pm 0.29$ ) ( $P < 0.05$ ). Mineral form revealed greater ADG for the LOOSE lambs ( $195 \pm 6$  g/d) compared with ( $159 \pm 6$  g/d) for the BLOCK lambs ( $P < 0.05$ ). Lamb ADG in the feedlot was similar ( $P > 0.05$ ) for the WEAN ( $217 \pm 6$  g/d) and NURSE ( $233 \pm 6$  g/d) lambs, while days on feed was greater for the WEAN ( $71 \pm 5$  d) than the NURSE ( $96 \pm 5$  d) lambs ( $P < 0.05$ ). Weaning at 120 d produced greater gains and lower measures of parasitism on pasture compared with weaning at 60 d. In the feedlot this translated to shorter time to final weight. Loose mineral produced greater ADG than block mineral when offered on pasture.

**Key Words:** lamb, growth, weaning

# Teaching/Undergraduate and Graduate Education Symposium: Teaching graduate students to teach and be successful at teaching

**638 Engaging introductory animal science students through free-range learning.** Frank E. Robinson\*, Dana C. Penrice, and Martin J. Zuidhof, *University of Alberta, Edmonton, Alberta, Canada.*

Student success in post-secondary education is largely assessed by their competency assessed by exams and written reports. Curricula may be updated frequently or not. In some cases students are active learners, while in others, a lecture lab format leaves the students learning what they are told to. Often, first year students take primarily large basic science or arts classes with little direct connection to agriculture. An animal science course could serve to “warm-up” the first year for agriculture students. For 11 years, the introductory animal science class at the University of Alberta has carried about 50% of the marks through open-ended project work, known to us as “free-range learning.” Through a series of individual and group projects, over 1,100 students have learned the basics of animal science. Lectures, lab tours and exams are still held. The value-added part of the courses have been the offering of 2 to 4 projects per term, designed to allow creativity, embrace the arts, allow opportunity for in depth science discovery and to construct and maintain a learning community. In these projects, students become the experts, with encouragement to contact local individuals as well as those at other universities for up-to-date information. Student deliverables have included videos, live drama presentations, and in-class presentations. For some students, success in this environment can be a challenge so we have senior undergrad alumni return as “learning coaches.” Some projects have focused on tools and technologies used in animal production and processing and value-added food production. The program “There’s a Heifer in Your Tank” provided groups of students opportunity to answer “questions you didn’t know you had about food production” in a public venue reminiscent of an agriculture-based show. In an effort to build ag-fluency, a Rural Café provided 78 students a chance to converse one-on-one with 18 farmers in a speed-dating format. In our experience, engaging students as active learners has improved student satisfaction and retention and resulted in a stronger student-institution bond.

**Key Words:** student engagement, undergraduate learning, teaching

**639 Teaching graduate students to teach: An integrated approach.** Donald L. Gillian-Daniel\*, *University of Wisconsin-Madison, Madison, WI.*

A key leverage point for improving undergraduate education is preparing graduate students and post-doctoral researchers to teach effectively. In this session, I will present a model professional development program for future faculty. I’ll discuss the conceptual basis behind the program and its effects on the local institution. The Center for the Integration of Research, Teaching and Learning Network (CIRTL) is a national association of research universities collaborating to change graduate education by training graduate students and post-docs to teach. Three core ideas form CIRTL’s conceptual framework: Teaching-As-Research – using disciplinary research skills to address questions about student learning; Learning Communities – engaging participants in shared learning around effective teaching; and Learning through Diversity – viewing diversity as an asset that can enrich learning. The Delta Program in Research, Teaching and Learning (Delta) at the University of Wisconsin-Madison is CIRTL programming. It involves credit-based graduate courses, facilitated discussion groups, workshops, community events, a Teaching-As-Research Internship and a Certificate. Delta’s

use of CIRTL’s conceptual framework has affected the campus. For example, (1) curricular innovations are developed and evaluated through Teaching-As-Research projects, which also promote cross-institutional collaborations (Gillian-Daniel and Walz, 2015). This type of capstone program has common elements that can be replicated at different institutions (Gillian-Daniel et al., 2015; submitted); (2) grant proposals that leverage Delta programming for broader impacts receive more research funding; (3) interdisciplinary STEM learning communities develop and thrive; (4) program graduates experience improved hiring and early-career success. CIRTL future faculty learn to apply research-based, high-impact teaching practices and to acquire data about student learning to advance their teaching (Pfund et al., 2012; Benbow et al., 2011). CIRTL’s conceptual framework provides a solid foundation for training graduate students and post-docs how to teach effectively to promote learning.

**Key Words:** graduate education, teaching

**640 Assessing teaching skills when hiring new faculty members, and rewarding faculty and graduate students for teaching well.** Kenneth G. Odde\*, *Kansas State University, Manhattan, KS.*

Outstanding teaching has long been a priority for animal science departments. Sustaining excellence in teaching has become a challenge for animal science departments because of declining state funding, the changing nature of our student bodies, loss of livestock units that support teaching, and increased emphasis on extramurally funded research. Excellence in teaching requires that the organization has a culture that respects teaching equal to other mission areas. Attributes of outstanding teachers include compassion for students, a commitment to helping students learn and grow, excellent listening and communication skills, and competence in the subject matter. When recruiting new faculty with teaching responsibilities, it is essential that the evaluation process include the potential for effective teaching. Most candidates for new faculty positions do not have extensive teaching training or experience. Therefore, the evaluation of teaching is primarily one of assessing teaching potential. Many departments use a teaching demonstration as part of the interview for faculty positions that have teaching responsibilities. Properly rewarding faculty and graduate students for excellence in teaching requires a reward system where teaching is effectively evaluated and respected. Student evaluation of teaching is a useful tool, but it has limitations. Peer evaluation of teaching can also be an effective tool in improving teaching quality.

**Key Words:** assessing teaching potential, rewarding teaching

**641 Scholarship of teaching: What are the best methods of conducting research in teaching?** Michel A. Wattiaux\*, *University of Wisconsin, Madison, WI.*

Instructors are encouraged to bring some level of scholarship of teaching and learning (SoTL) to their instructional activities. Although years of experience may serve as a basis for scholarship, the informal or formal but purposeful collection of data, which is core to SoTL, provides a way to document efforts to improve teaching quality (which may contribute to reward system). However, the ultimate goal of research in teaching is student learning (i.e., achievements). Classroom assessment techniques

(CAT) are informal and formative tools providing instructors with an opportunity to modify instruction based on students' feedback (i.e., perception) when it matters the most (within a semester). Research has shown, however, that people are poor at evaluating their own learning. In contrast, a systematic data collection protocol can be deployed to generate replicated data addressing specific issues with the goal of producing generalizable knowledge. Unfortunately measuring learning outcomes is extremely complex demanding high level of expertise and investment in educational research. Reviewing a large body of literature, Carl Weiman (Nobel laureate in physics) and a colleague have proposed recently a simple teaching practice inventory tool, which they claim can be used as a proxy for students learning outcomes (measured as improvement in grades). This tool may serve as a pedagogical framework to initiate research projects. Evidently, innumerable factors influence the type of research an instructor can realistically engage in successfully. The literature abounds with examples of qualitative (words as data) or quantitative (numbers as data) approaches, experimental designs (pre- and posttest, survey) and statistical methods (parametric, non-parametric). Regardless of challenges and limitations inherent to research in teaching, it is essentially a goal-oriented discovery process that has the potential to transform profoundly one's own teaching practice. Thus, the "best" research methods might be the ones that that the instructor deems most adequate to shed light on the degree to which the learning environment of their classes promotes student achievements.

**Key Words:** undergraduate education

**642 Is *Natural Sciences Education* the journal for you?** Ken Barbarick\*, *Colorado State University, Fort Collins, CO.*

*Natural Sciences Education* (NSE) is a peer-reviewed international journal published online continuously during the year. Articles are written by and for educators in the areas of animal science, ecology, natural resources, agronomy, the environment, entomology, and more. Articles cover extension, university, industry, administration, and grades K-12 education. NSE is published by the American Society of Agronomy. NSE provides quick turnaround (49 d from submission to first decision in 2014) and an editorial board that represents all cooperating societies and that works closely with authors to improve the chances for publication (76% acceptance rate on 29 submissions in 2014). We welcome pedagogical studies that include supporting data replicated in time or space, review papers, philosophical discussions, K-12 activities, and profiles of legendary scientists/teachers. We hope members of ADSA and ASAS will consider NSE as their outlet for educational papers.

**Key Words:** teaching



## Animal Behavior and Well-Being II

**W1 The relationship between equine temperament and behavior as affected over time by the skill level of the rider.** Taylor Huffman and Katherine Koudele\*, *Andrews University, Berrien Springs, MI.*

The goal of this study was to determine if there was a relationship between horse temperament and incidence of misbehavior due to the stress of being ridden by riders with a wide range of ability during the course of a camp season (9 weeks). It was hypothesized that horses with a social or aloof temperament would be able to adjust more easily while those with a fearful or challenging temperament would have higher incidence of misbehavior. At the beginning of the summer season at Timber Ridge Camp, Spencer, Indiana, the temperament of each horse ( $n = 15$ ) was determined using on a rubric based on criteria developed by Barteau (2007). During the camp season, each rider was evaluated for riding skill level, and each incidence of equine misbehavior on trail rides was recorded as were the lengths of the trail rides. The scale for misbehavior was 0–4 with 0 = was not ridden, 1 = no incidents, 2 = annoyance (i.e., head tossing, ear-pinning, moving out of line), 3 = disruptive (i.e., balking, kicking, moderate shying), 4 = hazardous (i.e., bucking, bolting, rearing). The rider skill level was evaluated using a rubric based on Blokhuis et al. (2008) with a scale of 1–4, 1 = an advanced rider and 4 = total novice. The skill level of the rider was multiplied by the length of the rides in minutes to develop a stress index. One-way ANOVA showed a highly significant relationship ( $P < 0.0001$ ) between the stress index and incidence of misbehavior. The behavior scale data for misbehaviors were combined due to lower frequency of severe misbehaviors 3–4. Horses with a challenging temperament exhibited significantly more misbehavior ( $P = 0.007$ ) as did horses with a fearful temperament ( $P < 0.0001$ ). Horses with social or aloof temperaments did not show any significant increase in misbehavior. The results support that in a summer camp horse program a positive relationship exists between stress and misbehavior of horses which varies with their temperament ( $P = 0.02$ ). Horses with fearful or challenging temperaments appeared to be more stressed than horses with a social or aloof temperament based on their greater incidence of misbehavior. It is therefore recommended that horses be temperament tested before entering a summer camp program.

**Key Words:** horse temperament, rider skill level, misbehavior

**W2 Better welfare outcomes in lambs subjected to gradual weaning when compared with abrupt weaning.** Cristiane G. Titto\*<sup>1</sup>, Fábio L. Henrique<sup>1</sup>, Evaldo A. L. Titto<sup>1</sup>, Adroaldo J. Zanella<sup>2</sup>, Henrique B. Hooper<sup>1</sup>, Lina F. P. Rodriguez<sup>1</sup>, Ana Luisa S. Longo<sup>1</sup>, Thays M. C. Leme-dos Santos<sup>1</sup>, Raquel F. Calviello<sup>1</sup>, Jessica C. Veronezi<sup>1</sup>, and Alfredo M. F. Pereira<sup>3</sup>, <sup>1</sup>*Faculdade de Zootecnia e Engenharia de Alimentos, Universidade de São Paulo, Pirassununga, São Paulo, Brazil*, <sup>2</sup>*Faculdade de Medicina Veterinária e Zootecnia, Universidade de São Paulo, Pirassununga, São Paulo, Brazil*, <sup>3</sup>*Instituto de Ciências Agrárias e Ambientais Mediterrânicas, Universidade de Évora, Évora, Alentejo, Portugal*.

Weaning is a stressful process during the early-life of lambs. The aim of this study was to compare 2 types of weaning on cortisol levels and rectal temperature of 47 crossbreed lambs. During gradual weaning (GW,  $n = 12$  males,  $n = 10$  females), for 6 d, from d 39 to 44, the mothers were removed from the paddock at 7:00AM, and the lambs were kept separated from them until 17:00h. During the GW the same animal groups were maintained and lambs had free access to clean and fresh

water, pasture and concentrate. On d 45, the lambs were placed in the feedlot and blood samples for cortisol levels evaluation were taken at 3 different moments: the first during the last contact with the mother (T1), the second 60 min after entering the feedlot (T2) and the third 180 min after entering the feedlot (T3). On d 46 and 47, the samples were taken once a day, 24 h and 48 h after the entry into the feedlot, respectively. The abrupt weaning (AW,  $n = 9$  males,  $n = 15$  females) began at d 45 of age, and the samples were collected in a similar way as reported for GW. Statistical analyses of variance were divided in 2 steps: the first at d 45 for both GW and AW, with fixed effects of sex, type of weaning and sample time and interactions; the second one analyzed the d 45, 46 and 47 and had fixed effects of sex, days of total separation of the mother and type of weaning as their interactions. Means were compared by F test and *t*-test (PDIFF). On d 45, AW lambs had higher cortisol levels ( $1.74 \mu\text{g} \cdot \text{dL}^{-1}$ ) compared with GW lambs ( $0.93 \mu\text{g} \cdot \text{dL}^{-1}$ ;  $P < 0.05$ ), and time had no effect on type of weaning. However, the first sample, just after the separation, was 66.6% higher than the T2 and 76.2% higher than T3 ( $P < 0.05$ ). GW lambs had lower cortisol levels ( $1.38 \mu\text{g} \cdot \text{dL}^{-1}$ ) than AW lambs ( $2.22 \mu\text{g} \cdot \text{dL}^{-1}$ ) on d 45, 46 and 47 ( $P < 0.05$ ) when they were in the feedlot. Sex had no effect on cortisol levels ( $P > 0.05$ ). When weaning is done early and lambs are allowed to have a period to adapt to solid feed and to the absence of their mother, there is less activation of the stress axis after total separation and on the beginning of the feedlot period.

**Key Words:** cortisol, sheep, stress

**W3 Assessing cow-calf welfare.** Gabrielle E. Simon\*<sup>1</sup>, Bruce R. Hoar<sup>2</sup>, and Cassandra B. Tucker<sup>1</sup>, <sup>1</sup>*University of California, Davis, Davis, CA*, <sup>2</sup>*University of Wyoming, Laramie, WY*.

Certification programs are one way beef producers can assess and communicate information about animal welfare to the public. Programs have been created to monitor cattle all the way through production (e.g., Global Animal Partnership) or at individual stages (e.g., slaughter; American Meat Institute), but to date, there is no certification program addressing welfare specifically at the cow-calf level. The objectives of this study were to gather animal-based prevalence information, describe management practices and facility characteristics, and identify welfare risk factors on cow-calf operations. A welfare assessment was designed using features of similar programs in the beef and dairy industries and was applied to 25 California ranches that varied in herd size (mean  $\pm$  SD, range:  $669 \pm 751$ , 28 – 2450 cows), specialization (i.e., commercial, purebred, grass-finished), and geographic location. Cow health and behavior and stockperson handling measures were collected during a chute-side procedure, management practices were evaluated through an interview-guided questionnaire, and facility characteristics were recorded at the chute and water access points. Cattle health problems were rare and only seen on specific ranches (e.g., prevalence of lame cattle:  $1.5 \pm 1.6$ , 0 – 7.1%). Conversely, cattle behavior and stockperson handling varied between ranches (e.g., cattle balking prevalence:  $16.5 \pm 16.2$ , 1.6 – 71.4%, electric prod use:  $21.5 \pm 21.1$ , 0 – 73.0%). Although some management characteristics were shared by all (e.g., all producers reported having a veterinary/client/patient relationship), a variety of practices were reported (e.g., weaning age:  $8.1 \pm 1.5$ , 6 - 11 mo). Likewise, some facility features were common (e.g., 20 curved head gate stanchions vs. 4 straight) and other aspects varied (e.g., 14 restraints without louvers vs. 10 with louvers). A risk factor evaluation

will identify which inputs (i.e., stockperson handling, management practices, facility design) influence welfare on cow-calf operations.

**Key Words:** welfare assessment, cow-calf, risk factor

**W4 Identifying the principal problems of animal welfare on intensive dairy farms in the central zone of Chile.** Maria Sol Morales\*, Lorena Carmona, Tamara Tadich, and Carlos Alvear, *Facultad de Ciencias Veterinarias y Pecuarias, Universidad de Chile, Santiago, RM, Chile.*

Animal welfare (AW) on 19 intensive dairy farms (DF) from the central zone of Chile was studied using Welfare Quality protocol (WQP); 18 were qualified between acceptable and enhanced, one as non-classified, and none as excellent. The objective of the present study was identify the specific problems of AW on these DF using the data on which the evaluations were made. WQP assesses AW on animal-based observations (cow or herd level) and on indirect information from farm records. For each observation or record, an average value/DF was obtained and compared with the alarm value (AV) for each indicator; AV is considered as the minimum value acceptable. For some indicators, AV was provided by WQP, otherwise this was calculated as 25% of the distance between maximum and minimum value found on the evaluation. Then the percentages of DF that exceeded the AV for each indicator were obtained and ranked to identify the principal problems. Problems were related to animal health, where 78.9, 52.6, 53, 40 and 40% of DF exceeded the AV on: % cows with areas of hairless patch, Total % of lame cows, % of cows with dystocia, mortality and downer cows, respectively; although some sanitary indicators as: % of cows with oral, nasal or vulvar secretions, as well % of cows with diarrhea did not show problems at all. Housing problems are mainly related to dirtiness (68.5, 57.9, 52.6% of DF had problems on: % of cows with udder dirty, % of cows with flank/upper leg dirty and % of cows with dirty lower legs, respectively). With regard to animal behavior, social interaction among cows did not seem to be a major problem; however, at human-cow interaction there is poor AW: 64.7 and 58.8% of DF showed problems with flight zone and % of cows that can be touched, respectively. Good feeding aspects did not appear as a major AW problem. The results show diverse aspects of management and housing, which affect the AW of dairy cows. This study has identified those problems that deserve greater attention in training of farm workers. Funded by project No. FOOD-CT-2004-506508 and CONICYT.

**Key Words:** animal welfare, dairy cow

**W5 Identifying farm-level factors affecting milking attendance and productivity in automated milking systems.** Meagan T. M. King\*<sup>1</sup>, Ed A. Pajor<sup>2</sup>, Stephen J. Leblanc<sup>3</sup>, and Trevor J. DeVries<sup>1</sup>, <sup>1</sup>*Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada,* <sup>2</sup>*Faculty of Veterinary Medicine, University of Calgary, Calgary, AB, Canada,* <sup>3</sup>*Department of Population Medicine, Ontario Veterinary College, University of Guelph, Guelph, ON, Canada.*

The objective of this study was to determine how barn design, herd management, and lameness prevalence relate to automated milking system (AMS) attendance and productivity. Data were collected from 26 AMS farms in Eastern Ontario, Canada. Farms averaged  $108 \pm 13$  (mean  $\pm$  SE) lactating cows and  $2.3 \pm 0.3$  AMS units. The majority of farms used free-flow cow traffic ( $n = 24$ ), while 2 farms employed a mixture of free-flow and milk-first traffic for different groups. Physical barn characteristics and stocking densities were recorded, and comple-

mented by a questionnaire regarding feeding and bedding management. At each farm, 40 cows were gait scored (or 30% for herds  $> 130$  cows) using a 5-point numerical rating system (1 = sound to 5 = lame). Cows were defined as lame with gait scores  $\geq 3$  ( $28.4 \pm 2.8\%$ ) and severely lame as  $\geq 4$  ( $2.9 \pm 0.7\%$ ). For a 6-d period, milking activity and production parameters for all cows at each farm were extracted from the respective AMS computers. Data were summarized across all cows and the 6-d observation period for each farm and analyzed in multivariable mixed-effect regression models. Milking frequency ( $2.9 \pm 0.1$  milkings/cow/d) increased with herds of greater average parity ( $P = 0.04$ ; mean =  $2.2 \pm 0.4$  lactations), greater frequency of feed push up/d ( $P = 0.04$ ; mean =  $9 \pm 1.2 \times/d$ ), and when stalls were raked  $\geq 3 \times/d$  vs.  $2 \times/d$  ( $P = 0.01$ ), while it decreased with a greater prevalence of severe lameness ( $P = 0.008$ ). Milk yield ( $32.6 \pm 0.5$  kg/d) increased with herd size ( $P = 0.01$ ) and feed space/cow ( $P < 0.001$ ; mean =  $70.4 \pm 2.6$  cm/cow). Lying time ( $706 \pm 11.8$  min/d) was greater with concrete flooring ( $P = 0.045$ ) as opposed to rubber flooring, and tended to decrease with lying stall neck rails placed further from the back curb ( $P = 0.07$ ; mean =  $172.0 \pm 1.6$  cm). Lying bouts ( $8.9 \pm 0.3$  no./d) were less frequent with a greater prevalence of lameness ( $P = 0.01$ ), and tended to be less frequent with longer stall lengths ( $P = 0.07$ ; mean =  $248.9 \pm 3.4$  cm). These results demonstrate that behavior and productivity in AMS herds may be positively affected by modifiable housing and management practices and are negatively affected by a higher prevalence of lameness.

**Key Words:** automatic milking, dairy cow, behavior

**W6 Differences in cow comfort between traditional bedded pack and compost bedded pack for dairy housing systems.** Anna Fernández, Eva Mainau, Xavier Manteca, Cinta Sol\*, and Lorena Castillejos, *Animal Nutrition and Welfare Service, Animal and Food Science Department, Universitat Autònoma de Barcelona, Bellaterra, Spain.*

The objective of this research was to compare cow welfare between 2 different types of dairy cattle housing systems, compost bedded pack (CB) and traditional bedded pack (TB), using the Welfare Quality protocol. Both behavior indicators (time needed to lie down) and health indicators (dirtiness of the cows, body condition, hairless patches, lesions and swellings, lameness, nasal and ocular discharge, hampered respiration, diarrhea, and vulvar discharge) were evaluated as indicators of cow welfare. The study was conducted on 2 commercial dairy farms (816 and 394 lactating cows) between the spring 2013 and the summer 2014. Data from behavior and health indicators were collected from 451 lactating cows ( $11.28 \pm 0.29$  cows for each farm, season and treatment) and 1222 lactating cows ( $30.5 \pm 0.6$  for each farm, season and treatment) respectively. Statistical analyses were carried out with the SAS 9.2 software applying a MIXED procedure for behavior and GLIMMIX procedure for health data. Regarding behavior, cows took a similar amount of time to lie down in CB and TB ( $4.8 \pm 0.08$  and  $4.7 \pm 0.07$  s;  $P > 0.05$ ). For health indicators, the percentage of cows with dirtiness on the udder was higher ( $P < 0.05$ ) in CB ( $50.7 \pm 2.61$ ) than TB ( $44.0 \pm 3.33$ ) in summer and spring. Conversely, in winter, the percentage of cows with dirtiness on the hindquarters was higher ( $P < 0.05$ ) in TB ( $61.7 \pm 2.93$ ) than CB ( $50.3 \pm 2.99$ ). Cows allocated in TB showed higher prevalence of hairless patches on the flank, side and udder than in CB ( $10.8 \pm 1.57$  and  $9.6 \pm 1.55$ ;  $P < 0.05$ ) in summer. There were no other differences in health measures between cows housed in CB and TB throughout the year. Most of the welfare parameters evaluated did not differ between the 2 loose housing systems, with the exception of dirtiness and hairless patches.

**Key Words:** compost bedded pack, cow comfort, lactating cow

**W7 Behaviors of Holstein heifers associated with large follicles during the estrous cycle.** Cassandra S. Skenandore\* and Felipe C. Cardoso, *University of Illinois, Urbana, IL.*

Estrus detection in heifers is pivotal for reproductive success. The objective of this study was to identify behaviors associated with 2 different periods of the estrous cycle of Holstein heifers. Eighteen heifers at breeding age were selected, balanced by age, BW, and BCS (average  $13.7 \pm 1$  mo,  $394 \pm 32$  kg, and  $3.43 \pm 0.1$  respectively), and observed for 6 wk. The experimental period was divided into 3 periods of 14 d each. All heifers received the same basal diet to meet requirements (NRC 2001), fed once daily. Visual observations were performed in 30 min segments every 2 h from 6AM to 6PM. Video recordings were used to confirm observations. Twelve behaviors were recorded (Social Lick, Rump Lick, Tail Paint Lick, Body Butt, Head Butt, Push, Chin Rest, Anogenital Sniff, Play Rub, Winner, Mount, and Attempt Mount). A synchronization protocol starting on d 1 of each period (Ovsynch: 100mg GnRH at d 1, 25mg PGF $_{2\alpha}$  at d 7, and 48 h later an application of 100mg of GnRH) was used to stimulate periods of high and low interactions. Observations were summarized to daily counts of interactions for each behavior. Ovaries were examined via ultrasound imaging on d 1, 8, and 10 of each period. The presence of follicles or a corpus luteum (CL) was recorded with their respective sizes. Lying time, standing time, and bouts /d were recorded using accelerometers (Onset HOBO Pendant G) at 1 min intervals for 14 d. Statistical analyses were performed using the GLIMMIX procedure of SAS (v9.3). During periods of low activity per the synchronization protocol, social ( $P < 0.01$ ) and paint ( $P < 0.05$ ) licking were more likely to be expressed than estrus-related behaviors such as rump licking ( $P < 0.05$ ), chin resting ( $P < 0.002$ ), and mounting ( $P < 0.003$ ). The aforementioned estrus related behaviors were also performed in more frequency than other behaviors when a large follicle is present ( $P < 0.03$ ). Heifers spent more time standing and less time lying per day when they had a large follicle ( $P < 0.006$ ) compared with a small follicle. In conclusion, increased activity and specific behaviors may be used in the future to facilitate estrus detection in heifers.

**Key Words:** heifer, behavior, heat detection

**W8 Behavioral responses of periparturient Jersey cows treated with recombinant bovine somatotropin during the periparturient period.** Henrique F. Soares<sup>1</sup>, Daniela N. Liboreiro\*<sup>1</sup>, Gabriel D. Bombardelli<sup>1</sup>, Paula R. B. Silva<sup>1</sup>, and Ricardo C. Chebel<sup>1,2</sup>, <sup>1</sup>University of Minnesota, St Paul, MN, <sup>2</sup>University of Florida, Gainesville, FL.

Objectives were to determine the effects of recombinant bovine somatotropin (rbST) treatment of Jersey cows from -21 to 21 d relative to calving on rumination, activity, and lying behavior. Cows were assigned randomly to control and rbST (125 mg of rbST every 7 d from -21 to 21 d relative to calving) treatments. Cows (n = 50) were fitted with rumination/activity collars (SCR) and lying behavior loggers (HOBO) from -21 to 21 d relative to calving. Rumination and activity were recorded every 2 h and lying behavior was recorded every 1 min. Data were analyzed in 24 h intervals. Continuous data were analyzed by ANOVA using the PROC MIXED procedure for repeated measures with cows as the experimental unit. The model included treatment (control vs rbST), time (days relative to calving), and the interaction between treatment and time. Daily rumination time was not ( $P = 0.74$ ) different between treatments in the prepartum, but rbST cows had reduced daily rumination time in the postpartum (rbST $_{125} = 428.5 \pm 17.0$ , control =  $484.3 \pm 16.7$  min/d,  $P = 0.03$ ). The interaction between treatment and time affected ( $P < 0.01$ ) daily rumination time because from 0 to 7 d postpartum treatments did not differ, but in most days from 8 to 21 d

postpartum control cows had greater daily rumination time. Treatment did not affect activity during the prepartum ( $P = 0.97$ ), but control cows tended to have greater activity during the postpartum ( $540.3 \pm 33.7$  vs  $499.1 \pm 36.1$  arbitrary units;  $P = 0.12$ ). Lying time was not affected by treatment (prepartum- $P = 0.34$ , postpartum- $P = 0.72$ ). Treatment by time interaction affected number of lying bouts in the prepartum ( $P < 0.01$ ) and rbST cows tended to have greater number of lying bouts in the postpartum (rbST =  $7.2 \pm 1.1$ , control =  $6.5 \pm 1.6$  bouts/day). Lying bout duration was not different in the prepartum ( $P = 0.86$ ), but rbST cows tended to have shorter lying bout duration in the postpartum (rbST =  $83.9 \pm 3.8$ , control =  $93.5 \pm 3.7$  min/bout;  $P = 0.08$ ). Reduced rumination and increased lying bouts during the postpartum period among rbST treated cows may be associated with changes in metabolic and health status resulting from rbST treatment.

**Key Words:** Jersey cow, behavior, recombinant bovine somatotropin

**W9 Preference of flavored concentrate premixes by dairy cows.** Michael T. Harper\*<sup>1</sup>, Joonpyo Oh<sup>1</sup>, Fabio Giallongo<sup>1</sup>, Juliana C. Lopes<sup>1</sup>, Alexander N. Hristov<sup>1</sup>, and Joelle Faugeron<sup>2</sup>, <sup>1</sup>The Pennsylvania State University, University Park, PA, <sup>2</sup>Pancosma, Geneva, Switzerland.

Flavor preferences may stimulate feed intake in dairy cows, which may improve energy balance in early lactation and lead to increased milk production. A cafeteria experiment was designed to determine if dairy cows have flavor preferences. Sixteen lactating Holstein cows averaging  $197 \pm 32$  DIM,  $1.9 \pm 0.8$  lactations,  $27.8 \pm 4.2$  kg/d DMI, and  $41.5 \pm 7.4$  kg/d milk yield were involved in the experiment. Cows were offered 7 flavored and a control concentrate premixes (FCP). The FCP flavors were: anise, fenugreek, honey, orange, thyme, molasses, vanilla, and the absence of flavor acted as a control. The inclusion rate of the flavors in FCP was 250 to 300 g/t, as-is basis. Cows were not adapted to the flavors before the experiment. Cows were housed in a tie-stall barn and were offered each d 4 different FCP of 1 kg each in plastic bins placed in front of each cow. The duration of the experiment was 6 consecutive days. Each FCP was presented to each cow once every 2 d, 2 h after the morning feeding. Thus, each flavor was presented to each cow 3 times during the experiment. Flavors and position of the bins in front of the cows were randomized. Each cow had access to the FCP for 5 min from the time they started eating. Eating times were recorded. Amount consumed and rate of FCP consumption were analyzed using the MIXED procedure of SAS with cow as random effect. The vanilla and fenugreek FCP were consumed the most ( $P = 0.03$ ) at 408 and 371 g per 5 min offering, respectively, while the orange and anise FCP were consumed the least at 264 and 239 g, respectively. Similarly, cows spent the most ( $P = 0.002$ ) time eating the vanilla and fenugreek FCP at 99 and 75 s, respectively, and the least amount of time eating the orange and anise FCP at 49 and 50 s, respectively. There was an effect of bin position with the 2 center FCP being consumed more ( $P < 0.001$ ) than the outer 2 FCP. There was a trend for flavor by bin position interaction ( $P = 0.07$ ) for the amount consumed, but not ( $P = 0.11$ ) for the time spent eating. Flavor had no effect on consumption rate. In conclusion, dairy cows may prefer vanilla or fenugreek flavors, when offered a novel choice.

**Key Words:** feed intake, flavor preference, dairy cow

**W10 Sorting of TMR by dairy calves is affected by availability of an alternative source of grain.** Joao H. C. Costa\*, Nicola A. Adderley, Daniel M. Weary, and Marina A. G. von Keyserlingk, *Animal Welfare Program, University of British Columbia, Vancouver, BC, Canada.*



We performed 2 trials on sorting behavior in individually raised dairy calves ( $n = 18$ ). For both, calves were provided free access to TMR [ $49.1 \pm 1.5\%$  DM (Mean  $\pm$  SD); chemical composition shown as % of DM, CP  $16.9 \pm 0.95\%$ , NDF  $31.8 \pm 2.68\%$ , ADF  $20.4 \pm 1.77$ ] and calf starter [with an overall DM of  $89.5\%$ ; chemical composition shown as % of DM, CP  $20.6 \pm 1.13\%$ , NDF  $16.5 \pm 0.39\%$ , ADF  $7.84 \pm 0.45\%$ ] for the first 2 mo of life. Sorting was assessed at 65 d, when calves still had access to calf starter, and again at d70 when starter was no longer available. On each day, sorting was measured by comparing diets before feeding with orts after 24 h of feed access. Particle sizes were measured using the Penn State Particle Separator with 3 screens (19, 8, and 1.18 mm) and a bottom pan to separate the TMR into long, medium, short, and fine fractions respectively. The predicted intake of each particle size fraction in the orts was tested between treatment for a difference from 100 using *t*-tests. When able to access the calf starter, calves sorted the TMR selecting for long particles ( $133 \pm 9\%$ ;  $P = 0.0038$ ) and against small particles ( $92 \pm 3\%$ ;  $P = 0.0240$ ), with no differences for the remaining particle fractions ( $99 \pm 5\%$  [ $P = 0.90$ ] for medium;  $107 \pm 5\%$  [ $P = 0.14$ ] for fine). When calves no longer had access to the starter these preferences reversed; calves preferentially selected the fine particles ( $113 \pm 4\%$ ;  $P = 0.01$ ), with no selection for the other fractions ( $101 \pm 11\%$  [ $P = 0.94$ ] for long;  $99 \pm 6\%$  [ $P = 0.80$ ] for medium;  $97 \pm 4\%$  [ $P = 0.51$ ] for short). These results indicate that young dairy calves are capable of sorting TMR and adjust their sorting behavior in response to changes in feed offered.

**Key Words:** feeding behavior, sorting behavior, motivation

**W11 Predicting filching: A logistical approach.** Kristina A. Weld\*, Louis E. Armentano, and Amy L. Stanton, *University of Wisconsin, Madison, WI*.

Understanding the feeding behavior of dairy cattle in greater detail can lead us to broader conclusions about animal welfare. One feeding behavior of interest is filching, in which cows steal feed from an automatic gate which they do not have access to, rather than taking the easier route and eating out of a gate that they have access to. The objective of this study was to determine factors that predict filching behavior. Sixty-three cows (28 primiparous, 35 multiparous, average 76 DIM) were assigned to 4 mixed parity groups. All cows were in the same physical pen containing 32 Insentec Roughage Intake Control feed gates (Insentec BV, Marknesse, the Netherlands), but each group had access to a different set of 8 randomly assigned gates. For the first 6 weeks, cows were fed a common diet, followed by a second 3 week period in which cows were fed 1 of 2 diets that differed only in the variety of soybean that they contained. Data were analyzed with logistic regression (Proc Glimmix) in SAS 9.4. For the first 6 weeks, filching at or above 0.5% of diet was positively associated with hip height ( $P = 0.01$ ). Cows that filched above 1% in the second period consumed 92% (Confidence Interval 89–95%) of their filched feed from their unassigned diet which is significantly above the amount that would be predicted if they filched randomly from unassigned gates (67%). Hip height was positively associated with filching and meal length was shorter for cows that filched in the second period ( $P < 0.05$ ). Filching in period 1 was not predictive of filching during period 2. Filching was not associated with the number of bunk displacements. These models indicate that height is a requirement for filching in this gate arrangement. However, competition does not necessarily drive filching indicating that filching may not be driven by necessity. Because cattle are willing to work for a novel feed, regardless of competition, this may indicate that cattle are willing to work to experience novelty. This might indicate that provision

of novelty, in the form of alternative feeds, plays a larger role in cattle welfare than previously thought.

**Key Words:** filching, welfare, novel feed

**W12 Validation of methodology for assessing heat abatement strategies in dry-lot cattle.** Grazyne Tresoldi\*, Karin Schütz<sup>2</sup>, and Cassandra Tucker<sup>1</sup>, <sup>1</sup>Center for Animal Welfare, Department of Animal Science, University of California, Davis, Davis, CA, <sup>2</sup>Farm Systems North, AgResearch Ltd., Ruakura Research Centre, Hamilton, New Zealand.

Identifying dairy cattle experiencing heat stress can improve welfare and profitability. However, it is unclear how often we need to observe the animals for this purpose, nor the relevance of specific aspects of the heat stress response, particularly in terms of panting (e.g., closed vs. open mouth panting). Our objectives were to refine and validate methodology used to assess heat load in dairies by (1) determining sampling intervals to measure usage of heat abatement resources (HAR; sprayed water or shade); and (2) evaluating the relationship between respiration rates (RR) and panting characteristics (PC; drooling, open mouth, and tongue outside the mouth). High-producing lactating cows ( $>40$  kg milk/d) were chosen from 4 California dry-lot dairies (8 cows/dairy,  $n = 32$ ) and observed for at least 6 h (1000 to 1800 h, excluding milking) when air temperature, humidity and the combined index averaged  $33^{\circ}\text{C}$ , 30% and 79, respectively. Use of HAR by individual cows was recorded continuously. Every 5 min, RR and presence/absence of PC (observed for 10 s at a time) were recorded. For use of HAR, estimates from different sampling intervals (1, 5, 10, 15, 20, 30, 60, 90 and 120 min) were derived from continuous data. Linear regression was used to compare these estimates against the continuous measurements. Sampling intervals were considered accurate if they met 3 criteria:  $R^2 \geq 0.9$ , intercept = 0 and slope = 1. The relationship between RR and each PC was analyzed using mixed models with random effects for cow and cow  $\times$  PC, to generate appropriate error degrees of freedom. When the sampling interval for HAR was  $\leq 60$  min,  $R^2$  values were  $\geq 0.9$  and both the intercept and slope did not differ from 0 and 1, respectively ( $P > 0.05$ ). RR was higher when PC were present (least squares means  $\pm$  standard error, breaths/min: with vs. without drool present,  $97 \pm 3$  vs.  $74 \pm 3$ ; open vs. closed mouth,  $100 \pm 4$  vs.  $80 \pm 3$ ; tongue outside vs. inside the mouth,  $96 \pm 4$  vs.  $81 \pm 3$ ;  $P < 0.001$ ). In summary, use of HAR is best measured when using sampling interval  $\leq 60$  min. Panting was accompanied by higher RR, and further analysis is planned to determine appropriate sampling intervals for these measures.

**Key Words:** well-being, behavior, cooling

**W13 Infrared thermography as a tool to diagnose foot rot and digital dermatitis in feedlot cattle.** Sonia Marti<sup>1</sup>, Randy E. Wilde<sup>1</sup>, Diego Moya\*<sup>1,2</sup>, Eugene D. Janzen<sup>2</sup>, Michael J. Jelinski<sup>3</sup>, Craig L. Dorin<sup>3</sup>, Karin Orsel<sup>2</sup>, Ed Pajor<sup>2</sup>, Jan Shearer<sup>4</sup>, Suzanne T. Millman<sup>4</sup>, Johann F. Coetzee<sup>4</sup>, Dan Thomson<sup>5</sup>, and Karen S. Schwartzkopf-Genswein<sup>1</sup>, <sup>1</sup>Agriculture and Agri-Food Canada, Lethbridge, AB, Canada, <sup>2</sup>University of Calgary, Faculty of Veterinary Medicine, Calgary, AB, Canada, <sup>3</sup>Veterinary Agri-Health Services Ltd., Airdrie, AB, Canada, <sup>4</sup>Iowa State University, College of Veterinary Medicine, Ames, IA, <sup>5</sup>Kansas State University, College of Veterinary Medicine, Manhattan, KS.

Diagnosis of lameness due to infectious claw lesions in beef cattle can be extremely challenging under commercial feedlot conditions resulting in inappropriate medical treatment and unnecessary drug and labor costs.

The objective of this study was to determine if infrared thermography could be used to make a differential diagnosis between foot rot (FR) and digital dermatitis (DD) based on claw area temperatures. Over a 16-mo period, 470 lame cattle from the same commercial feedlot in Southern Alberta were examined while restrained in a squeeze chute. Thermographic images were taken using a Flir i40 infrared camera, and processed with ThermCam QuickView 1.3 (Flir Systems Inc., Burlington, ON, Canada) from the anterior and posterior views of each affected hoof, and the laterally adjacent unaffected hoof within the same animal, to obtain temperature differentials. Limb position (fore or hind) of the imaged hoof was also recorded. Temperatures from the underside view of the affected limb were also measured. Ambient temperature and relative humidity within the examination barn were recorded using a Hobo U23 Pro v2 logger (Onset Computer Corporation, Bourne, MA). After images were obtained, the affected hoof and lesion were cleaned with a brush and a physical exam was conducted to determine the actual cause of lameness. Data were analyzed using a mixed effect model with diagnosis, affected limb and their interaction as a main effects, and ambient temperature and relative humidity as covariates. No differences were observed in temperatures of unaffected and affected hooves for either FR or DD cases ( $-1.3 \pm 0.22^\circ\text{C}$  and  $-0.7 \pm 0.37^\circ\text{C}$ , respectively ( $P = 0.15$ ) for the anterior view, and  $-0.9 \pm 0.16^\circ\text{C}$  and  $-1.2 \pm 0.24^\circ\text{C}$ , respectively ( $P = 0.41$ ) for the posterior view). In addition, temperatures from the underside view were not different ( $P = 0.67$ ) between FR ( $35.9 \pm 0.12^\circ\text{C}$ ) and DD ( $35.8 \pm 0.18^\circ\text{C}$ ). Under the conditions of this study, infrared thermography was not a useful tool for differentiating between beef feedlot cattle diagnosed with foot rot and digital dermatitis.

**Key Words:** lameness, feedlot, cattle

#### **W14 Continuous recording versus scan-sampling in behavioral studies with growing heifers fed high-concentrate diets.**

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Continuous observation is an accurate method for behavioral measurements in ruminants but is very time-consuming. An alternative is scan-sampling (SC), in which behavior is recorded at regular intervals. The aim of this study was to compare the loss of information when different SC intervals were chosen to record heifer behavior. Behavior of 8 growing heifers fed a high-concentrate diet was recorded on 6 non-consecutive days. Continuous sampling (CS) was considered the control treatment and was used to evaluate scan-sampling intervals of 2, (SC2), 5 (SC5), 10 (SC10), 15 (SC15), 20 (SC20) and 30 (SC30) min. Two additional sampling techniques were considered: a time sampling of the first 10 min of every hour (TS), and the continuous observations during 4 h post-feeding (4PF). Behaviors were expressed as a percentage of the total daily activity. A square root-arcsine transformation was applied to achieve normal distribution. Differences were analyzed by using the GLIMMIX procedure of SAS. The model contained the fixed effect of treatment, and the random effects of heifer and day. Pearson correlations were used to compare CS with SC treatments. Daily time in each behavioral activity using the SC and TS treatment did not differ from CS ( $P > 0.10$ ). In contrast, 4PF affected behavioral measurements. Time spent ruminating, social behavior and self-grooming were 50, 41 and 38% lower ( $P < 0.001$ ), while eating and resting times were 28 and 17% higher ( $P < 0.001$ ) in 4PF than CS. For all behaviors, high correlations were observed between CS and SC2 ( $r > 0.93$ ;  $P < 0.001$ ) and between CS and SC5 ( $r > 0.75$ ;  $P < 0.001$ ). For eating, drinking,

social, self-grooming, tongue-rolling, licking and biting fixtures, and rummaging in wood shavings, the longer the scan interval, the lower was the correlation. For ruminating and resting, correlations between CS and SC30 were greater than 0.90 ( $P < 0.001$ ). In conclusion, the use of SC or the TS treatment did not result in any loss of information. However, 4PF is not an accurate technique for measuring behavioral activities in ruminants.

**Key Words:** beef cattle, behavior, scan-sampling method

**W15 Evaluation of technologies to predict the onset of calving in Holstein dairy cows.** Véronique Ouellet\*<sup>1</sup>, Elsa Vasseur<sup>2</sup>, Wolfgang Heuwieser<sup>3</sup>, Onno Burfeind<sup>3</sup>, Xavier Maldague<sup>4</sup>, and Édith Charbonneau<sup>1</sup>, <sup>1</sup>*Département des Sciences Animales, Université Laval, Québec, QC, Canada*, <sup>2</sup>*Organic Dairy Research Center, University of Guelph, Alfred, ON, Canada*, <sup>3</sup>*Clinic for Animal Reproduction, Faculty of Veterinary Medicine, Freie Universität Berlin, Berlin, Germany*, <sup>4</sup>*Département de Génie électrique et de Génie Informatique, Université Laval, Québec, QC, Canada*.

Dystocias are common in dairy cows and are known to have labor, production, reproduction, welfare and economic implications. A technology that would accurately predict the onset of calving would help minimize the effects of dystocia by allowing producer intervention in a timely matter. The aim of this study was to assess the performances to predict calving using a decrease in vaginal temperature, rumination time, and lying time or an increase in number of lying bouts measured by 3 technologies. The combination of technologies allowing simultaneous measure of the variables was also tested. Forty 2 multiparous Holstein cows housed in tie-stall were fitted with a temperature logger, a rumination sensor and an accelerometer 7  $\pm$  2 d before their expected calving date. Data collected during the last 120 h before calving were summarized per day and in 6 h periods. Differences between days and between periods were analyzed using proc GLIMMIX of SAS. Test performances (sensitivity, specificity, predictive values) were conducted to predict calving within the next 24, 12 or 6 h. Vaginal temperature were 0.3°C lower ( $P < 0.05$ ) whereas rumination and lying time were respectively 41 and 52 min lower ( $P < 0.05$ ) on calving day compared with the 4 d precalving. Cows had 2 more lying bouts on calving day ( $P < 0.05$ ). While comparing tested variables, a decrease of vaginal temperature achieved the best performance to predict calving within the next 24, 12, and 6 h. Between those periods, the best performance was achieved for a prediction within the next 24 h with a sensitivity, specificity, positive and negative predictive values respectively of 74, 74, 51, and 89%. Combining the technologies enhanced the performance to predict calving with best results obtained by the combination of the 3 technologies for a prediction within the next 24 h (sensitivity: 77%, specificity: 77%, positive and negative predictive values: 56 and 90%). These results suggest that technologies are better at identifying events during which the cow did not calve than calving events. Therefore, a device that would be able to measure the 4 variables may not be able to accurately predict calving time but would provide insightful information for calving monitoring.

**Key Words:** technologies, calving, dystocia

**W16 Barrow behavioral reactivity to a human or novel object when fed low versus high fiber diets.** Jessica D. Colpoys\*<sup>1</sup>, Nicholas K. Gabler<sup>1</sup>, Caitlyn E. Abell<sup>2</sup>, Aileen F. Keating<sup>1</sup>, Suzanne T. Millman<sup>1</sup>, Janice M. Siegford<sup>3</sup>, and Anna K. Johnson<sup>1</sup>, <sup>1</sup>*Iowa State University, Ames, IA*, <sup>2</sup>*DNA Genetics, Columbus, NE*, <sup>3</sup>*Michigan State University, East Lansing, MI*.

Low energy, higher fiber diets (HFD) are becoming more prevalent in the US swine industry due to fluctuating corn-soy diet prices. In sows, HFD are reported to increase satiety and reduce stereotypic behavior, aggression, and activity. However, little is known about how fiber content in diets contributes to behavioral reactivity in grow-finish pigs. Therefore, the objective of this study was to determine if diet influences behavioral reactivity using a human approach test (HAT) and novel object test (NOT). We hypothesized that pigs reared on HFD would be less reactive to HAT and NOT compared with pigs reared on high energy, low fiber corn-soy diets (CD). Forty Yorkshire barrows ( $48 \pm 8$  kg BW) were randomly allocated to 2 treatments: HFD ( $n = 20$ ) and CD ( $n = 20$ ). The barrows were evaluated once using HAT and once using NOT utilizing a crossover experimental design. Each pig was individually tested within a  $4.9 \times 2.4$  m test arena for 10 min between 1300 and 1900 h. Behavior was evaluated using live and video observations. The video was watched continuously by one trained observer for latency, frequency, and duration of human and novel object (orange traffic cone) touches, frequency of escape attempts, frequency of freezing postures, activity (number of arena line crossings), urination, and defecation. Data were analyzed using the Glimmix procedure of SAS with fixed effects of diet and test week, covariate of body weight, and random effect of pen. Diet did not alter latency to first touch, touch frequency, or duration of touches with the human or novel object ( $P > 0.10$ ). Similarly, frequency of escape attempts, freezing, activity, and urination did not differ between diets during HAT or NOT ( $P > 0.10$ ). Barrows fed HFD defecated more during NOT ( $P = 0.01$ ), and tended to defecate more during HAT compared with CD barrows ( $P = 0.06$ ). Differences in defecations are likely due to high fiber content of HFD resulting in more waste excretion. These results suggest that feeding high fiber diets did not alter grow-finisher barrow behavioral reactivity.

**Key Words:** approach, fear, high fiber diet

**W17 A novel objective chute score interacts with monensin to affect growth of receiving cattle.** Kelsey A. Bruno\*, Eric S. Vanzant, Alex W. Altman, Monoj Kudupojje, and Kyle R. McLeod, *University of Kentucky, Lexington, KY.*

Temperamental animals often have lower gains associated with reduced intake and/or efficiency. Temperament is ill defined. Thus, different temperament measures may relate differently to production traits. Also, hepatic oxidation theory (HOT) suggests that propionate can exacerbate intake depression in stressed cattle. To examine relationships between diet, temperament, growth, and health, 160 crossbred steers were used in a 56-d RCBD experiment with a  $2 \times 2 \times 2$  factorial treatment structure. The experimental unit was pen (5 pens/treatment). Steers were pen fed a corn silage-based diet with or without monensin, ad lib. Temperament treatments (assigned on d -7) were exit velocity (EV; slow vs. fast) and objective chute score (OCS; low vs. high), a novel temperament measure, the CV of weights collected at 5 Hz for 10 s while an animal's head was restrained in a chute. Both were measured on d -7, 1, 14, 28, 55, and 56. Subjective chute scores (SCS) were measured on d -7 and d 56 by 4-5 observers. Jugular blood samples were analyzed for antibody response to leptospirosis vaccine. There was a positive correlation between SCS and OCS ( $P < 0.01$ ;  $R^2 = 0.38$ ) and SCS was moderately repeatable among observers (Krippendorff's  $\alpha = 0.58$  to  $0.67$ ). Treatment x day

effects ( $P < 0.10$ ) for EV and OCS indicated that initial measures may be better proxies of growth than average measures. There were no interactions between EV and OCS ( $P \geq 0.24$ ) and no interactions between treatments ( $P \geq 0.12$ ) on intake (%BW). Monensin decreased intake ( $P < 0.01$ ) without effect of temperament. Thus, our hypothesis regarding HOT was not supported. Gains, antibody titer, and gain:feed responses to monensin depended on OCS ( $P < 0.10$ ) but not ( $P \geq 0.34$ ) EV. Gain was reduced ( $P < 0.10$ ) by monensin with low, but not high OCS and gain:feed was increased ( $P < 0.10$ ) and titer response decreased ( $P < 0.10$ ) by monensin on high, but not low OCS. Gain and intake tended ( $P \leq 0.15$ ) to be reduced in fast, compared with slow EV steers. Results provide novel indications that certain temperament measures can interact with dietary manipulation to influence animal performance.

**Key Words:** temperament, growth, monensin

**W18 Could forming uniform body weight groups at entrance result in improved performance, behavior, health and carcass in fattening Holstein bulls?** Marçal Verdú\*<sup>1</sup>, Alex Bach<sup>2,1</sup>, Armando Pérez<sup>3</sup>, and Maria Devant<sup>1</sup>, <sup>1</sup>*IRTA-Ruminant Production, Animal Nutrition, Management, and Welfare Research Group, Caldes Montbui, Spain*, <sup>2</sup>*ICREA, Barcelona, Spain*, <sup>3</sup>*Grup Alimentari Guissona, Guissona, Spain.*

The purpose of the present study is related to the criterion that should be followed to form animal groups according to BW, and to evaluate the effect of this practice on productive, behavior, or health outcomes when animals are mixed at the onset of fattening. A total of 160 Holstein bulls ( $162 \pm 0.3$  kg BW and  $124 \pm 1.1$  d age) were allocated in 8 pens with similar mean BW, and were randomly assigned to 1 of 2 treatments: homogeneity of initial BW that corresponded to a CV of BW within a pen of 7% (HO), and heterogeneity with a CV of BW within a pen of 21% (HE). Concentrate intake and health incidences were recorded daily, straw consumption weekly, and BW every 14 d. Animal behavior was registered on d 0, 1, 3, 5, 8, 14, 28 and every 28 d by scan sampling. Animals were slaughtered after 206 d, and HCW and carcass quality were recorded. Data were analyzed using a mixed-effects model with repeated measures. Performance and carcass data were not affected by the BW uniformity of the groups at the onset. However, while the CV of BW within HO was maintained ( $P > 0.10$ ) throughout the study ( $6.1 \pm 0.73\%$ ), the CV of BW within HE was reduced ( $P < 0.01$ ) from 21.5 to  $10.7 \pm 0.86\%$  at the beginning and at the end, respectively. During the 1st month HO exhibited ( $P < 0.05$ ) lesser completed mounts than HE ( $0.5$  and  $1.0 \pm 0.09$  times/15 min). Throughout the study HO tended ( $P = 0.10$ ) to perform more self-grooming compared with HE ( $13.6$  and  $11.6 \pm 0.13$  times/15 min). Whereas 80% of veterinary therapies applied in HE were chronic ( $\geq 3$  consecutive treatments in the same animal), 90% of therapies in HO were acute ( $\leq 2$  consecutive treatments in the same animal,  $P < 0.05$ ). Moreover, 6 HE bulls were removed from the study for health reasons. In conclusion, even if bulls from pens with a non-uniform BW at the onset had greater frequency in completed mounts and worse health status than bulls from pens with uniform BW at the onset, animal growth and carcass was not impaired.

**Key Words:** bull, BW uniformity, performance



# Animal Health: Dairy calves and heifers

**W19 Effect of housing on the leukocyte transcriptome of beef calves.** Robmay Garcia\*, Dianelys Gonzalez-Pena, and Sandra L. Rodriguez-Zas, *University of Illinois at Urbana-Champaign, Urbana IL.*

Movement of beef calves from a pasture to a housing environment induces a stress response including changes in leukocyte physiology. Understanding the molecular profiles underlying these changes can help to develop better management practices and treatments to minimize the deleterious effects of stress. The objective of this study is to characterize the leukocytes transcriptome changes during housing stress after moving calves from outdoor to indoor environments. Transcriptome of blood leukocytes from 6 beef calves in outdoor pasture (d 0) was compared with that of d 1, 2 and 7 after being moved indoors. Individual calf RNA-seq libraries were sequenced using Illumina HiSeq analyzer platform. Single-end reads were mapped to the *Bos taurus* reference genome (UCSC\_bosTau7) using Tophat v2.0.12. In total, 26,163 isoform transcripts pertaining to 8,606 genes were identified and 1,636 isoform transcripts pertaining to 1,320 genes were differentially expressed at d 1, d 2 and d 7 relative to d 0 (False Discovery Rate-adjusted P-value <0.05) using Cufflinks v2.2.1. The number of differentially expressed genes increased with time indoors: d 0 vs d 1 (32 genes), d 0 vs d 2 (434 genes), and d 0 vs d 7 (1170 genes). Among the top 50 differentially abundant transcripts CCAAT/enhancer binding protein C/EBP epsilon (CEBPE) and Dual Specificity Phosphatase 1 (DUSP1) were overexpressed in leukocytes collected on d 1, d 2, and d 7, relative to d 0. DUSP1 plays a role in the cellular response to environmental stress, and CEBPE is essential for terminal differentiation and functional maturation of leukocyte cells. Functional analysis of the differentially expressed genes using DAVID identified 8 enriched functional category clusters pertaining to Biological Processes, Molecular functions and KEGG Pathways (Enrichment score >2): protein synthesis and transport, cell regulation and reorganization, RNA transport, leukocyte activation, proliferation and differentiation. These categories confirm that housing stress elicits changes in the leukocyte transcriptome associated with inflammatory response. The permanency of these changes and the impact on inflammatory response to other stress sources including infection and management requires further evaluation.

**Key Words:** transcriptome, stress, leukocyte

**W20 Short-term  $\alpha$ -tocopherol treatment during neonatal period modulates pro-inflammatory response to endotoxin (LPS) challenge in the same calves several months later.** S. Kahl\* and T. H. Elsasser, *USDA, ARS, Animal Biosciences and Biotechnology Laboratory, Beltsville, MD.*

Vitamin E, a major natural antioxidant, has been previously shown to attenuate pro-inflammatory response to immune challenge in cattle. Our objective was to evaluate the effect of short-term treatment with  $\alpha$ -tocopherol in newborn calves on selected elements of the pro-inflammatory response to LPS challenge in the same calves ~8 mo later. In 2 separate trials (T<sub>1</sub>, T<sub>2</sub>), newborn Angus  $\times$  Hereford calves (T<sub>1</sub>, n = 41; T<sub>2</sub>, n = 17) were assigned to  $\alpha$ -tocopherol (E, 1000 IU/d, ~1 g, i.m.) or control (C, placebo, canola oil, ~1 g) treatments. Injections of E/placebo (21/20, 8/9, in T<sub>1</sub> and T<sub>2</sub>, respectively) started on the day of birth and were repeated every other day for the first 14 d of life. At the age of 237  $\pm$  3 d (BW 276  $\pm$  5 kg) all calves were challenged with 2 consecutive LPS injections (LPS1, LPS2) 4 d apart (0.25  $\mu$ g *E. coli* 055:B5/kg

BW, i.v.). Blood samples were obtained at 0, 1, 2, 3, 4, and 24 h relative to each LPS injection. In T<sub>1</sub>, plasma concentration of tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) was determined by RIA with TNF- $\alpha$  response to LPS calculated as area under the time  $\times$  concentration curve (AUC). In T<sub>2</sub>, plasma total antioxidative capacity (TAC), xanthine oxidase activity (XO), as well as concentration of acute phase proteins, haptoglobin (Hg) and serum amyloid-A (SAA), were measured. No differences between treatment groups were found after LPS1 and LPS2 in plasma XO and Hg, and after LPS1 in TNF- $\alpha$  and SAA responses. However, compared with C, E treatment decreased plasma TNF- $\alpha$  AUC (5.9 vs. 9.3 ng/mL  $\times$  h,  $P < 0.05$ ) and increased SAA concentration (246 vs. 151  $\mu$ g/mL,  $P < 0.01$ ) after LPS2. Plasma TAC declined 4 d after LPS1 in all calves (116 vs. 168  $\mu$ mol/L,  $P < 0.01$ ) although the overall values during LPS2 were greater in E than C calves (125 vs. 106  $\mu$ mol/L,  $P < 0.05$ ). The data suggest that the i.m. treatment of calves with E during a critical first 14 d of life may serve to condition the animals toward a modified immune response later in life. Whether this implies a potential to alter the development of the immune system to improved life-long health remains to be determined.

**Key Words:** beef calves, endotoxin,  $\alpha$ -tocopherol

**W21 Influence of a feed additive on support of calves during initial growth phase.** Jorge A. Saltijeral Oaxaca<sup>1</sup>, Luis Galicia<sup>1</sup>, Juan E. Guerra<sup>2</sup>, and H. I. Rogge\*<sup>3</sup>, <sup>1</sup>Universidad Aut3noma Metropolitana, Mexico, DF, Mexico, <sup>2</sup>Universidad Aut3noma de Sinaloa, Culiacan, Sinaloa Mexico, <sup>3</sup>Phytobiotics Futterzusatzstoffe GmbH, Eltville, Germany.

The objective of this study was to analyze the effect of Sangrovit CS, plant-derived quaternary benzophenanthridine and protopine alkaloids (QBA+PA) on growth performance and health status of calves. A total of 160 Holstein Friesians calves obtained from 2 commercial farms of Tizayuca/Mexico were randomly assigned into 2 groups: a) control, b) Treatment (control + 5 g/meal of QBA+PA product added into the milk replacer (MR) from the 3rd until the 23rd day of life). Calves were fed twice daily 2 L of MR/meal and concentrate and alfalfa ad libitum. Colostrum, MR, bacteriological and parasitic fecal analysis were conducted. The coproparasitoscopic analyses were performed on 3 and 23 d of age of each calf. In the treatment group no antibiotic of any kind was used, whereas in control the routine farm protocols including antibiotic treatments (ATB) were conducted. The following management factors that influence the health status of the calves were considered: birth management, colostrum and feeding management, housing and hygiene of the calves. Overall, morbidity and mortality rates were 21.8% and 5.66%, respectively, both below the national benchmark parameters, with no significant differences among the groups. In the control group, 2 calves died from diarrhea whereas in the treatment no deaths were observed. The average duration of diarrhea tended to be lower in the treatment group with 2.4 d vs. 3.2 d in the control group (SD 1.25;  $P = 0.07$ ). Parasitized calves were lower in treatment group ( $P < 0.01$ ; Table 1). The starting weight was 38.83 kg ( $\pm$ SD 6.12 kg). Weight gain during the trial was 5.596 and 4.045 kg for treatment and control groups respectively ( $P = 0.07$ ). The QBA+PA had a positive influence on the well-being and weight gain of the calves. Furthermore, it supported to remedy intestinal disorders leading to lower persistence of diarrhea and no death due to diarrhea.

**Table 1 (Abstr. W21).** The most common pathogens found in feces

Species	Control	Experimental
<i>Entamoeba</i> spp.	21	17
<i>Eimeria</i> spp.	3	0
<i>Cyptosporidium parvum</i>	0	1
<i>Giardia</i> spp.	1	0
<i>Bacillus</i> spp.	6	3
<i>Escherichia coli</i>	6	6

**Key Words:** calves, feed additive

**W22 Influence of sex and temperament on response to *Salmonella* Newport extract vaccine in crossbred beef calves.** Anita M. Snell<sup>1,2</sup>, Jason P. Banta<sup>2</sup>, Brittini P. Littlejohn<sup>1,2</sup>, Sara D. Lawhon<sup>3</sup>, Tom H. Welsh Jr.<sup>1</sup>, Ron D. Randel<sup>2</sup>, and Rhonda C. Vann<sup>4</sup>, <sup>1</sup>Department of Animal Science, Texas A&M, College Station, TX, <sup>2</sup>AgriLife Research and Extension Center, Texas A&M, Overton, TX, <sup>3</sup>Department of Veterinary Pathobiology, Texas A&M, College Station, TX, <sup>4</sup>MAFES-E.G. Morrison Brown Loam Exp. Station, Mississippi State University, , Raymond, MS.

The effect of sex and temperament on response of crossbred beef calves to a commercially available *Salmonella* vaccine was studied. Exit velocity (m/s) and pen score (1 = calm and 5 = excitable) data collected from calves 28 d before weaning were used for assignment to 1 of 3 temperament classes (calm, n = 31; intermediate, n = 44; temperamental, n = 31). Calves, stratified by temperament class and sex, were assigned to non-vaccinated (control, n = 54) and vaccinated (n = 52) groups. Vaccinated calves received 2 mL of *Salmonella* Newport extract vaccine (Zoetis, Florham Park, NJ) twice: at weaning (d 0) and 28 d after weaning. Body weights were recorded on d 0, 28, and 56 and blood samples were taken at 7-d intervals to determine serum cortisol concentration by RIA and Ig ratio specific to the vaccine by ELISA (EpiToxip, MN). Weight and age data were analyzed with mixed model procedures of SAS; sex, temperament class, and vaccine group were included as fixed effects. Average age at weaning did not differ between treatments ( $P = 0.2$ ;  $200 \pm 3.9$  d). Vaccination did not affect weight gain from d 0 to 28 ( $P = 0.33$ ;  $7.21 \pm 0.9$  kg) or d 0 to 56 ( $P = 0.4$ ;  $24.61 \pm 1.1$  kg). As there was no difference in performance due to vaccination only vaccinated calves were used in the remaining analysis. A specific Ig profile of each calf was assessed for maximum height for both initial (Ig1; d 0 to 21) and booster vaccination (Ig2; d 28 to 56). Neither sex ( $P > 0.19$ ) nor temperament ( $P > 0.07$ ) influenced peak Ig1 ( $0.34 \pm 0.06$ ), peak Ig1d ( $15 \pm 1.5$  d), peak Ig2 ( $0.70 \pm 0.03$ ), or peak Ig2d ( $41 \pm 2.7$  d). Cortisol and Ig ratio during the study were analyzed as repeated measures; the models included temperament, sex, and day as fixed effects. Temperament did not affect average cortisol ( $P = 0.29$ ) or average Ig ratio ( $P = 0.50$ ). Male relative to female calves had lower average cortisol ( $P < 0.01$ ;  $31.5 \pm 2.8$  vs  $55.7 \pm 2.8$  ng/mL) and greater average Ig ratios ( $P = 0.04$ ;  $0.44 \pm 0.03$  vs  $0.34 \pm 0.03$ ). Sex of calf influenced the adaptive immune response to *Salmonella* Newport extract vaccine.

**Key Words:** *Salmonella*, cortisol, calves

**W23 Passive transfer of immunity in dairy heifer calves on Costa Rican dairy farms.** J. A. Elizondo-Salazar<sup>\*1</sup>, D. Benavides-Varela<sup>2</sup>, A. Vargas-Ramirez<sup>1</sup>, and C. M. Campos-Granados<sup>3</sup>, <sup>1</sup>Estación Experimental Alfredo Volio Mata, Facultad de Ciencias Agroalimentarias, Universidad de Costa Rica, San José, Costa Rica, <sup>2</sup>Alimentos del Norte S.A.-DIPCR, Costa Rica, <sup>3</sup>Centro de

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The objective of this study was to determine the transfer of passive immunity of dairy heifer calves in 4 provinces of Costa Rica. The data presented correspond to total serum protein (TSP) measurements obtained in 50 dairy farms. A total of 2500 heifer calves were sampled. Dam breeds were classified into Holstein, Jersey, Holstein × Jersey and other. Blood samples were collected between d 1 and 7 of age into serum Vacutainer tubes, refrigerated overnight, centrifuged, and the serum separated from clot within 24 h of collection. A hand-held refractometer was used to measure TSP. For the purpose of this study, failure of passive immunity was considered when TSP concentration was less than 5.5 g/dL. GLM procedure was used to establish differences between parity and breed of the dams. Descriptive statistics were generated to define percentage of failure of passive transfer by breed and parity of the dam. TSP concentration ranged from 2.0 to 10.0, with an overall mean of 5.9 g/dL. Of all the calves evaluated, 38.8% presented failure of passive transfer of immunity. Calves born to Jersey and Holstein × Jersey crosses had significantly ( $P < 0.05$ ) higher TSP concentrations than calves born to Holstein and other breeds. When considering parity of the dam, there were no significant differences ( $P > 0.05$ ) on TSP concentration; however, offspring born to first lactation heifers showed the lowest percentage of animals with inadequate transfer of immunity. Calves that were allowed to suckle their dams showed a 44% failure of passive immunity against 33% of calves that were given colostrum by bottle. The findings of this study suggest that minimizing the risk of calves with failure of passive transfer of immunity is an important task to perform in dairy herds of Costa Rica.

**Table 1 (Abstr. W23).** Effect of dam breed over total serum protein (TSP) concentration in 2500 heifer calves from 1 to 7 days old in 50 dairy farms located in four provinces of Costa Rica<sup>1</sup>

Dam breed	n	TSP, g/dL	Animals with FPTI, %
Holstein	769	$5.8 \pm 0.06^b$	39 <sup>b</sup>
Holstein × Jersey	287	$6.4 \pm 0.09^a$	25 <sup>a</sup>
Jersey	743	$6.3 \pm 0.07^a$	27 <sup>a</sup>
Other	740	$5.4 \pm 0.07^c$	55 <sup>c</sup>

<sup>1</sup>n = number of animals; FPTI = failure of passive transfer of immunity.

**Key Words:** total serum protein, colostrum, passive immunity

**W24 Prevalence of bovine respiratory disease in pre-weaned dairy calves in Northern California.** Betsy M. Karle<sup>\*1</sup>, Sasha A. Dubrovsky<sup>2</sup>, William J. Love<sup>3</sup>, Deniece R. Williams<sup>3</sup>, Jeffery W. Stackhouse<sup>4</sup>, Randall J. Anderson<sup>5</sup>, Alison L. Van Eenennaam<sup>2</sup>, Terry W. Lehenbauer<sup>3,5</sup>, and Sharif S. Aly<sup>3,5</sup>, <sup>1</sup>University of California Cooperative Extension, Orland, CA, <sup>2</sup>University of California Department of Animal Science, Davis, CA, <sup>3</sup>UC Davis Veterinary Medicine Teaching and Research Center, Tulare, CA, <sup>4</sup>University of California Cooperative Extension, Eureka, CA, <sup>5</sup>California Department of Food and Agriculture, Animal Health Branch, Sacramento, CA, <sup>6</sup>Department of Population Health and Reproduction, School of Veterinary Medicine, University of California, Davis, Davis, CA.

Bovine respiratory disease (BRD) is one of the most important diseases affecting the health of dairy calves. The objective of this cross-sectional study was to estimate the prevalence of BRD in pre-weaned dairy calves and identify management practices that may be risk factors for BRD. A total of 31 dairies in 3 regions in Northern California were surveyed by convenience sample. Represented counties included Humboldt and

Del Norte (Region 1, average herd size 788, n = 10), Sonoma, Marin and Mendocino (Region 2, average herd size 305, n = 11), and Glenn, Tehama and Yuba (Region 3, average herd size 1,010, n = 10). A comprehensive questionnaire focusing on calf management practices and relevant demographic information was administered via in-person interviews at each dairy. Calves currently raised on each dairy were randomly sampled and evaluated using the California BRD scoring system. A total of 1,438 calves were evaluated, and mean BRD prevalence was 10.2% (95% CI 8.7, 11.6), which did not differ significantly across regions ( $P = 0.67$ ; 10.7% Region 1, 8.9% Region 2 and 12.5% Region 3). Overall BRD prevalence was 10.5% on organic dairies and 10.8% on conventional dairies ( $P = 0.93$ ). Herd size, volume of colostrum fed within 12 h of birth, testing colostrum for IgG concentration or age that calves were moved to group pens were not significantly associated with BRD prevalence at the herd level. Additionally, age, sex, and airborne ammonia concentrations within 1 foot of the hutch or housing floor were not significantly associated with BRD in calves. Group housing of pre-weaned calves was common in Northern California, with pens containing between 2 to 40 calves, and significantly increased the odds of BRD (OR 1.07,  $P < 0.001$ ). Additionally, the prevalence of BRD in Jersey calves was 2.2 times higher than in Holstein calves ( $P < 0.001$ ). Respiratory disease clearly affects calves throughout Northern California. While factors affecting the prevalence of BRD are apparent, further research is needed to identify additional management practices that affect prevalence of respiratory disease in pre-weaned dairy calves.

**Key Words:** BRD, preweaned calves, scoring system

**W25 Risk factors for diarrhea and pneumonia in Holstein calves offered colostrum of different quality in a hot environment.** Edir Torres-Rodriguez<sup>\*1</sup>, Miguel A. Mellado-Bosque<sup>2</sup>, Jose E. Garcia-Martinez<sup>2</sup>, and Francisco G. Veliz-Deras<sup>1</sup>, <sup>1</sup>Universidad Autonoma Agraria Antonio Narro, Torreon, Coahuila, Mexico, <sup>2</sup>Universidad Autonoma Agraria Antonio Narro, Saltillo, Coahuila, Mexico.

Intake of non-contaminated and high immunoglobulin content colostrum by neonatal calves is an important factor in successful calf programs. The objectives of this study were to determine the risk factors for the occurrence of diarrhea and pneumonia by evaluating colostrum quality (Ig content determined with colostrometer and refractometry) and bacterial contamination in 300 Holstein calves born in June 2014. A second objective was to describe the effect of feeding colostrum of different quality on growth and physiological parameters in neonatal calves. Calves were removed from their dams shortly after parturition and fed 2.8 L of colostrum by bottle; calves were placed in individual portable roofed pens. Colostrum variables were total bacterial counts, coliform bacteria counts, refractometry values, and Ig content. Multivariable logistic-regression analyses were performed. None of the variables included in the model affected the occurrence of diarrhea. The calves with highest odds of pneumonia were those receiving colostrum with total standard plate counts  $>15,000$ /mL (odds ratio = 1.8). Calves fed colostrum with  $< 15,000$  counts/mL (n = 145) had greater weaning weights (66.3 kg  $\pm$  6.7 vs. 63.4 kg  $\pm$  6.7;  $P < 0.01$ ) and preweaning daily weight gain (0.515 kg/d  $\pm$  116 vs. 0.477 kg/d  $\pm$  123;  $P < 0.01$ ) than calves fed colostrum with  $>15,000$  counts/mL (n = 155). Calves fed colostrum with the highest bacterial counts presented higher ( $P < 0.01$ ) values for fecal consistency (1.5  $\pm$  0.8) than calves receiving colostrum with low bacterial counts (1.2  $\pm$  0.5; scale 1 to 4, with 1 being normal feces to 4 being severe diarrhea;  $P < 0.01$ ). Calves receiving colostrum with higher Ig content ( $>85$  mg/mL, measured with colostrometer) presented feces more solid ( $P < 0.01$ ) than calves receiving colostrum with Ig  $< 85$  mg/

mL (1.2  $\pm$  0.6 vs 1.5  $\pm$  0.9). There was no difference between groups when examining the effect of different characteristics of colostrum on height to withers, respiration condition and rectal temperature. Given the conditions of this trial, feeding colostrum with  $<15,000$  total bacterial counts/ml reduced the odds of pneumonia and enhanced growth rate of Holstein calves in a hot environment.

**Key Words:** colostrum, total standard plate count, calves

**W26 Preweaning plane of nutrition and *Mannheimia haemolytica* dose influence metabolic responses to a combined bovine herpesvirus-1 and *Mannheimia haemolytica* challenge in post-weaned Holstein calves.** K. P. Sharon<sup>\*1,2</sup>, Y. L. Liang<sup>1</sup>, N. C. Burdick Sanchez<sup>2</sup>, J. A. Carroll<sup>2</sup>, P. R. Broadway<sup>2</sup>, and M. A. Ballou<sup>1</sup>, <sup>1</sup>Texas Tech University, Department of Animal and Food Sciences, Lubbock, TX, <sup>2</sup>USDA-ARS, Livestock Issues Research Unit, Lubbock, TX.

To determine whether previous plane of milk replacer nutrition (PON) and *M. haemolytica* (MH) dose influences metabolic responses to a combined viral-bacterial respiratory challenge, Holstein calves (1d of age; n = 30) were assigned to treatments in a 2  $\times$  3 factorial with pre-weaned PON and dose of MH as main effects (n = 5/treatment). Calves were fed either a low (LPN; n = 15) or a high PON (HPN; n = 15) from birth through weaning. Calves fed the LPN were fed 445g DM/d of milk replacer until weaning, and HPN calves were fed 830g DM/d of milk replacer from 1 to 10d and 1080g from 11d until weaning. Calf starter and water were offered ad libitum. Calves were step-down weaned beginning at 54d and moved into an enclosed barn at 70d. Indwelling jugular catheters were inserted at 80d. Calves were challenged with 1.5  $\times 10^8$  PFU/mL/nostril of bovine herpesvirus-1 (BHV-1) at 81d and with 10<sup>6</sup>, 10<sup>7</sup>, or 10<sup>8</sup> cfu of MH at 84d. Blood samples were collected at varying intervals respective to BHV-1 and MH challenges. Body weights at 70d were different ( $P < 0.01$ ) between LPN and HPN calves (62.2 vs 81.2  $\pm$  2.67 kg, respectively). Although HPN calves consumed more calf starter ( $P < 0.01$ ) during the challenge, there were no differences ( $P \geq 0.42$ ) in ADG or when intake was expressed per kg BW<sup>0.75</sup>. Glucose concentrations were greater ( $P \leq 0.02$ ) in HPN compared with LPN calves during both BHV-1 (61.1 vs 57.5  $\pm$  1.01 mg/dL, respectively) and MH challenges (57.8 vs 53.5  $\pm$  1.13 mg/dL, respectively). Following the MH challenge, calves receiving 10<sup>8</sup> MH had the greatest ( $P = 0.05$ ) NEFA concentrations. During the BHV-1 challenge, there was a time  $\times$  PON interaction ( $P = 0.02$ ) for plasma urea N concentrations, where LPN calves had greater ( $P = 0.02$ ) concentrations at 72 h. Glucose concentrations were reduced among LPN calves throughout the respiratory challenge, and calves receiving 10<sup>8</sup> MH mobilized more lipid reserves after the MH challenge suggesting calves fed a LPN during preweaning and calves receiving a 10<sup>8</sup> MH dose results in a more severe response to a viral-bacterial respiratory challenge.

**Key Words:** health, nutrition, respiratory

**W27 Effect of repeated intravenous LPS infusions in endometrium gene expression and inflammatory response in Holstein heifers.** Artur C. C. Fernandes<sup>\*1,2</sup>, Juliana S. Souza<sup>1</sup>, Douglas Veira<sup>1</sup>, Audrey Nadalin<sup>1</sup>, Lúcio E. H. Melo<sup>2</sup>, and Ronaldo L. A. Cerri<sup>1</sup>, <sup>1</sup>Faculty of Land and Food Systems, University of British Columbia, Vancouver, BC, Canada, <sup>2</sup>Federal Rural University of Pernambuco, Recife, Pernambuco, Brazil.

This study aimed to evaluate the effect of repeated intravenous lipopolysaccharide (LPS) stimulus in endometrial gene expression of candidate



transcripts of nonlactating heifers on late luteal phase. Heifers (n = 22; 11 mo of age) were synchronized by the ovsynch protocol and enrolled into control group (CON; n = 11) that received sterile saline solution i.v., or LPS group (LPS; n = 11) submitted to repeated LPS injections i.v. (0.1; 0.25; 0.5; 0.75; 1.0; 1.25 µg/kg WB) starting 2 d (d) after AI (d0), then every other d. At each injection, rectal temperatures were measured during 6 h. Blood samples were collected from the d-1 to d13, for analyses of TNF- $\alpha$ , haptoglobin, progesterone, and WBC count and differential. On d15, endometrium tissue biopsies were taken and kept at -80°C until qRT-PCR analysis of 30 target genes related to immune system, adhesion molecules and endometrium receptivity. Data were checked for normality and analyzed by ANOVA for repeated measures using proc MIXED and UNIVARIATE. After each injection, temperature was greater in the first 6h in the LPS compared with CON group ( $P < 0.05$ ). Both TNF- $\alpha$  ( $P = 0.05$ ) and haptoglobin ( $P < 0.01$ ) were increased in the LPS group with significant ( $P < 0.05$ ) treatment by d interactions. Total leukocyte count was not different between treatments ( $P = 0.29$ ), but differential count was increased for neutrophils, band cells and monocytes, but decreased for lymphocytes and eosinophil in LPS compared with CON group ( $P < 0.01$ ). Progesterone concentrations during the experimental period were not different between treatments. Out of the 30 target genes analyzed, only 3 transcripts were differentially expressed. IDO ( $P = 0.04$ ; Fold = 0.48) and PTX3 ( $P = 0.01$ ; Fold = 0.38) were downregulated, whereas MX1 ( $P = 0.02$ ; Fold = 2.85) was upregulated in the LPS group. Sequential LPS injections was able to induce a systemic pro-inflammatory state with limited, but strong effect in gene expression of transcripts related to the immune system, suggesting a possible explanation for sub-fertility related to health disorders in dairy cows.

**Key Words:** gene expression, heifer, LPS

**W28 Performance of Jersey calves born from dams treated with recombinant bovine somatotropin during the periparturient period.** Paula R. B. Silva\*<sup>1</sup>, Henrique F. Soares<sup>1</sup>, Gabriel D. Bombardelli<sup>1</sup>, and Ricardo C. Chebel<sup>1,2</sup>, <sup>1</sup>University of Minnesota, St Paul, MN, <sup>2</sup>University of Florida, Gainesville, FL.

Objectives of the current experiment were to evaluate the performance of Jersey calves born from Jersey dams treated with recombinant bovine somatotropin (rbST) during the periparturient period. Jersey dams were assigned randomly to control (n = 264) and rbST (125 mg of rbST every 7 d from -21 to 21 d relative to calving; n = 258) treatments. A sub-sample (control = 19, rbST = 19) of cows had colostrum samples collected at calving to determine IgG concentration. Male Jersey calves were sold at birth, leaving 199 and 173 calves from control and rbST cows. Calves born from cows enrolled in this experiment were fed pooled colostrum (4 L) twice within 6 h of birth, were housed in individual hutches from birth to 21 d of age, and in group pens with automated calf feeders from 22 d of age to weaning (60 d of age). Calves were weighed individually at birth (control = 198, rbST = 172) and at weaning (control = 122, rbST = 107) and average daily gain in the first 60 d of life was calculated. Calves were observed daily for diagnosis of diarrhea and respiratory illness. Data referent to health and performance to 60 d of age is reported herein. Continuous data were analyzed by ANOVA and dichotomous data were analyzed by logistic regression. Colostrum IgG concentration was not different between treatments (control = 87.6  $\pm$  6.4, rbST = 94.5  $\pm$  6.4 g/L;  $P = 0.45$ ). Treatment did not affect birth weight (control = 27.9  $\pm$  0.4, rbST = 27.4  $\pm$  0.5 kg;  $P = 0.24$ ), weaning weight (control = 70.5  $\pm$  1.8, rbST = 71.1  $\pm$  1.8 kg;  $P = 0.60$ ) or average daily gain (control = 0.74  $\pm$  0.03, rbST = 0.74  $\pm$  0.03 kg/d;  $P = 0.88$ ). There was a greater likelihood of calves born from rbST cows to be diagnosed with diar-

rhea (control = 1.0, rbST = 4.6%;  $P = 0.05$ ) but treatment did not affect the likelihood of pneumonia (control = 25.6, rbST = 27.8%;  $P = 0.61$ ) and death (control = 10.6, rbST = 13.3%;  $P = 0.41$ ) within the first 60 d of life. Treatment of periparturient Jersey cows with small doses of rbST did not affect performance and health of calves up to 60 d of life.

**Key Words:** recombinant bovine somatotropin, Jersey calves, performance

**W29 Supplementation of *Saccharomyces cerevisiae* fermentation products for the prevention of geophagia in Holstein female calves fed milk in buckets.** Sonia Vazquez-Flores\*<sup>1</sup>, Stephany Barrera-Almanza<sup>1</sup>, María de Jesús Guerrero<sup>2</sup>, Kristy Dorton<sup>3</sup>, Mark Scott<sup>3</sup>, and William Sanchez<sup>3</sup>, <sup>1</sup>Tecnológico de Monterrey, Querétaro, Querétaro, México, <sup>2</sup>Universidad Autónoma de Querétaro, Querétaro, Querétaro, México, <sup>3</sup>Diamond V, Cedar Rapids, IA.

The objective of the study was to compare 2 nutritional complements delivered in feed to diminish geophagia in neonatal calves with or without concurrent gastrointestinal diseases. Ruminal acidosis has been extensively studied in adult ruminants, and neglected in neonatal calves. Under certain feeding systems like bucket feeding milk, a common practice in most dairies in Mexico; it becomes the main factor for nutritional behavioral changes. Sand eating (geophagia) leads to tympanism, bruxism, depression, enterotoxemia, and sudden deaths. 60 Holstein female calves were randomly assigned at birth to 1 of 3 treatments: maltodextrin (C); BIOMOS (T1); and Smartcare/XPC (T2). Treatments were delivered in colostrum and in both whole UV purified milk and calf starter for up to 60 d in the morning feeding. Urine, blood and fecal samples (6, 2 and 8 respectively) were collected from each calf during the neonatal period. Analysis for bacteriological, parasitological and pH determination were performed. Binomial and repeated measures were determined by Welch ANOVA test, Duncan's test for multiple comparisons, Bartlett and Logistic Fit, nonparametric tests and odds ratio. Urine pH showed a range from 5 to 9, stratification was made taking 3 parameters from Oetzel, 2003: acid pH (5-6.9); neutral pH (7-7.9) and alkaline pH (8-9). See Table 1 for results. A statistical correlation was found with least squares means analysis for geophagia cases and days in diarrhea for the C group. No differences were found in diarrhea and *E. coli*, *Salmonella* spp., *Campylobacter* spp. and *Cryptosporidium* spp. with Tukey-Kramer test. Supplementation of T2 to neonatal calves' diet allowed the neonate to control best the metabolic acidity, being twice as often the need for buffers in the C group.

**Table 1 (Abstr. W29).**

Treatment group	Acid urine pH (5-6.9)	Geophagia mean (SD)	Odds ratio (95% CI) for no geophagia cases
C	61.7% (74)	0.23 (0.44)	
T1	73.3% (88)	0.2 (0.41)	vs. control: 0.54 (0.15-1.9) <sup>a</sup> vs. control: 0.12 (0.028-0.53) <sup>a</sup>
T2	72.5% (87)	0.1 (0.31) <sup>b</sup>	vs. T1: 0.25 (0.04-0.97) <sup>b</sup>

<sup>a</sup>Significant difference ( $P$ -value = 0.05); <sup>b</sup>highly significant difference ( $P$ -value = 0.006).

**Key Words:** neonatal, calves, acidosis

**W30 Comparison of the effect of LongRange (eprinomectin) versus Dectomax (doramectin) and fly tags on growth of post-weaned grazing dairy heifers.** M. W. Sahar<sup>1,2</sup>, J. E. Tower<sup>1</sup>, T. S. Dennis<sup>1</sup>, A. M. Mosiman<sup>1</sup>, H. F. P. Schmitz<sup>1</sup>, R. K. Tessman<sup>3</sup>, and T.

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Limited information is available on gain and structural growth of young grazing dairy heifers when treated with different parasiticides. The objective of this study was to compare the effects of LongRange (eprinomectin) (LGR) against Dectomax (doramectin) and pyrethroid impregnated fly tags (DFT) on gain, structural growth, and fecal egg counts (FEC) of grazing, post-weaned Holstein dairy heifers. Forty-eight heifers were randomly grouped into 12 paddocks according to BW ( $169.5 \pm 8.5$  Kg of BW and  $149.8 \pm 13.8$  d of age) and assigned to 1 of 2 treatments: 1) LGR or 2) DFT. The BW, hip height (HH), withers height (WH), hip width (HW), body condition score (BCS), heart girth (HG), blood samples, and fecal samples were collected every 4 wk from June until August 2014. Face flies (FF) and horn flies (HF) were counted twice/wk. Water consumption for each paddock was recorded 2 times/wk. Temperature and relative humidity were recorded hourly. Data were analyzed using PROC MIXED in SAS as repeated records using pen within treatment as a random variable. Daily midday temperatures (1200 to 1500 h) averaged  $26.9 \pm 0.14^\circ\text{C}$  during the study. The ADG did not differ between treatments (mean ADG =  $0.92(0.02)$  Kg/d;  $P = 0.79$ ) with BW at the end of the study averaging 210.2 and 208.9(1.02) Kg ( $P = 0.38$ ) for LGR and DFT, respectively. The HH and WH ( $P = 0.61$ ) averaged 117.4(0.29) and 112.3(0.39) cm, respectively, at the end of the study. The HG ( $P = 0.38$ ), HW ( $P = 0.34$ ), and BCS ( $P = 0.86$ ) were also similar between treatments. However, PUN values tended to be greater ( $P = 0.06$ ) for LGR compared with DFT (12.7 and 11.4(0.28) mg/dL, respectively). Water intakes averaged 18.8 L/d for DFT and 20.1(2.07) L/d for LGR ( $P = 0.67$ ). Fecal egg counts were lower ( $P = 0.02$ ) for heifers treated with LGR compared with DFT (95.2 and 246.7(60.7) epg, respectively) and HF counts tended ( $P = 0.08$ ) to be lower for DFT than LGR (9.7 and 17.2(0.08) flies/heifer, respectively) with no differences in FF counts ( $P = 0.24$ ). Treating post-weaned grazing Holstein dairy heifers with either LGR or DFT resulted in similar growth performance; however, LGR reduced FEC and DFT reduced the presence of horn flies.

**Key Words:** doramectin, dairy heifer, eprinomectin

**W31 Associations between fecal pathogens, growth, and clinical signs of diarrhea in dairy heifer calves.** Elizabeth S. Binversie\*, Melissa C. Cornett, Catie C. Cramer, and Amy L. Stanton, *University of Wisconsin-Madison, Department of Dairy Science, Madison, WI.*

Diarrhea can affect the health, performance, and welfare of preweaned dairy calves. The objectives for this project were to determine (1) associations between a positive test for a fecal pathogen and the probability of a calf showing clinical signs of diarrhea and (2) the impact of number of positive fecal tests on calf growth. The study population included dairy heifer calves ( $n = 54$ ) on a commercial dairy that were enrolled in a  $2 \times 2$  factorial study design examining a direct fed microbial and 2 milk programs. Feces were collected weekly for the first 4 weeks of life and tested for presence of 4 fecal pathogens: Rotavirus (ROT), Coronavirus (COR), *E. coli* K99 (F5) (ECO) and *Cryptosporidium* (CRP) (Enterichex). Clinical diarrhea was identified using a standardized fecal score that combined observations of odor, consistency, fluidity and color. Calves were weighed at birth (BW) and 5 weeks of age (WT5). The association between pathogen and diarrhea was evaluated with a logistic regression model (PROC GLIMMIX, SAS) with calf as a random effect and an autoregressive covariance structure. The association between weight at 5 weeks and number of positive fecal tests was evaluated using a linear regression model (Proc Mixed), controlling for initial weight

and calf age. At 2 weeks of age calves were 5.8 times [95% CL: 2.2 to 15.3] as likely to test positive for at least one fecal pathogen compared with 4 ( $P < 0.001$ ). Controlling for age, calves that tested positive for CRP were 2.8 times [95% CL: 1.2 to 6.4] as likely to show clinical signs of diarrhea ( $P < 0.05$ ). Rotavirus and ECO were not associated with clinical signs of diarrhea. *E. coli*, CRP, and COR were not significantly associated with WT5. Rotavirus was associated with WT5 ( $P < 0.05$ ). Calves with ROT at 1 or 2 time points weighed  $5.1 \pm 1.9$  kg (LSM  $\pm$  SEM) and  $5.3 \pm 1.9$  kg (LSM  $\pm$  SEM) less than calves that never tested positive, respectively ( $P < 0.001$ ). In conclusion, although considered to be associated with a single diarrheal complex, specific pathogens may have different effects on growth, and knowledge of pathogens present may provide greater insight into treatment and management of calves with diarrhea.

**Key Words:** calves, growth, diarrhea

**W32 Efficacy of feeding First Day Formula CR versus maternal colostrum on calf serum immunological parameters.**

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Commercially available colostrum replacers (CR) are commonly used for convenience, when maternal colostrum (MC) is unavailable, of questionable quality or in disease control programs. The objective was to determine the efficacy of a colostrum-derived CR, First Day Formula CR (Accelerated Genetics, Baraboo, WI), versus raw MC on immunological parameters. A total of 1,220 female and male Jersey and crossbred calves born in a California Central Valley dairy were systematically assigned to either CR or MC based on birth order. Calves assigned to CR were tube fed the CR product, reconstituted with 1.9 L of water and calves assigned to MC were tube fed 2.8 L of MC at 1 h  $\pm$  5 min after the calf was born. Every sixth calf born between 1:00 to 7:00 and 9:00 to 15:00 h that was enrolled was weighed and bled before first feeding and all calves born during those time intervals were bled at 22 to 28 h after birth. Immunoglobulin G (IgG) concentration was determined in serum samples using a radial immunodiffusion kit (Triple J Farms, Bellingham, WA). IgG was also measured in MC fed to every sixth calf born during the previously mentioned time intervals. The CR provided  $\sim 150$  g IgG per dose and MC contained an average of 63.6 mg/mL of IgG. Outcome variables included serum IgG at 22 to 28 h after birth ( $n = 592$ ), failure of passive transfer (FPT) defined as serum IgG  $< 10$  mg/ml ( $n = 592$ ), and apparent efficiency of absorption (AEA) ( $n = 96$ ), calculated as grams of IgG absorbed into blood circulation. Only one calf in each treatment group (CR and MC) experienced FPT. The mean serum concentration of IgG was 19.8 mg/mL for calves fed CR and 23.4 for MC ( $P < 0.05$ ). However, AEA of IgG did not differ between treatments and was 34.4% for CR and 35.9% for MC ( $P = 0.52$ ). Therefore, the slightly higher mean blood concentration of IgG for calves fed MC was due mainly to a higher IgG intake. Overall, IgG absorption and serum concentration of calves were within acceptable ranges when fed either CR or MC. In this study the parameters measured indicated that the CR used was comparable to MC in eliciting passive transfer.

**Key Words:** calves, colostrum, immunoglobulin

**W33 Relationship between serum total proteins and immunoglobulin G for calves fed either First Day Formula CR or maternal colostrum.** Alfonso Lago\*<sup>1</sup>, Claudia Leonardi<sup>1</sup>, Cedric Blanc<sup>2</sup>,

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Total serum proteins concentration (TP) is frequently measured in calves to evaluate adequacy of passive transfer of immunoglobulin G (IgG) from colostrum. However, calves fed some commercially available colostrum-derived colostrum replacers (CR) may have lower serum TP at equal IgG concentrations than calves fed maternal colostrum (MC). This can be due to removal of some proteins during manufacturing of CR. The objective was to determine the linear relationship between TP and IgG for CR and MC. Serum IgG and TP measurements from blood collected at 22 to 28 h after birth were available from 299 calves fed CR (First Day Formula CR, Accelerated Genetics, Baraboo, WI) and 292 calves fed MC. Concentration of IgG was determined using a radial immunodiffusion kit (Triple J Farms, Bellingham, WA) and TP using a digital refractometer (Misco, Model DD-2, Solon, OH). The linear relationship between TP and IgG was investigated for MC and CR using the Reg procedure of SAS (SAS Institute Inc., Cary, NC). The following are the linear relationships between IgG and TP in serum for the CR [IgG mg/mL =  $-9.87(\pm 1.88) + 5.77(\pm 0.36) \times \text{TP g/dL}$ ] and MC [IgG mg/mL =  $-45.30(\pm 2.58) + 12.45(\pm 0.44) \times \text{TP g/dL}$ ]. For both CR and MC, a linear relationship between TP and IgG was observed ( $P < 0.01$ ). However, for CR, a smaller percentage of the variability in IgG was accounted by TP ( $R^2 = 0.465$  for CR and  $R^2 = 0.738$  for MC). The TP value that resulted in IgG equal to 10 mg/mL (90% CI) was 3.44 g/dL (2.48, 4.38) for CR and 4.44 g/dL (3.65, 5.23) for MC. The 90% CI for MC includes the classical reference value of 5.2 g/dL indicating that the value estimated in the present study is not significantly different from the classical reference. Only one calf fed CR and one calf fed MC had a serum IgG value less than 10 mg/mL, making the estimation of the TP cut point for failure of passive transfer potentially inaccurate. In conclusion, lower serum TP values at equal IgG concentrations were found when feeding CR compared with MC. Therefore, a lower TP cut point indicative of successful passive transfer of IgG should be used when using CR.

**Key Words:** calves, replacer, total protein

**W34 Prophylactic efficacy of an engineered biotherapeutic fusion protein against *Cryptosporidium parvum* in experimentally challenged neonatal calves.** Travis J. De Wolfe\*<sup>1</sup>, Sheila M. McGuirk<sup>1</sup>, Nicholas S. Keuler<sup>1</sup>, Robert D. Bremel<sup>2</sup>, Jane Homan<sup>2</sup>, Michael Imboden<sup>2</sup>, Deborah A. Schaefer<sup>3</sup>, and Benjamin J. Darien<sup>1</sup>, <sup>1</sup>*University of Wisconsin-Madison, Madison, WI*, <sup>2</sup>*ioGenetics, Inc., Madison, WI*, <sup>3</sup>*University of Arizona, Tucson, AZ*.

*Cryptosporidium parvum* is a parasitic pathogen that causes gastroenteritis in cattle and humans. The host innate immune response to *C. parvum* infection is associated with expression of enteric antimicrobial peptides that may represent a useful prophylactic to prevent *C. parvum* infection. The aim of this study is to evaluate a model of *C. parvum* infection and to evaluate the prophylactic efficacy of a *C. parvum*-specific monoclonal antibody, 4H9, fused to the human cathelicidin LL-37 (4H9-G1-LL37) in *C. parvum* challenged calves. Holstein bull calves ( $n = 18$ ) were randomly assigned to receive either 0 mg (control), 20 mg, or 100 mg of fusion protein 4H9-G1-LL37 orally upon birth. Approximately 36–44 h later, calves were challenged with  $1 \times 10^7$  *C. parvum* oocysts. Fecal consistency, daily total fecal volumes, daily oocyst concentration (oocysts/mL), and total daily oocysts shed were assessed over a 10-d post-challenge period. Prior to experimental *C. parvum* challenge

all calves tested negative for *C. parvum* and by d 3 post-challenge all calves were shedding *C. parvum* oocysts in feces and had developed diarrhea. The control calves exhibited features of a natural infection, with diarrhea occurring between 4 and 7d post-challenge. The daily oocyst concentration was determined by immunofluorescence microscopy. Enumeration of total daily oocysts shed, was calculated as the product of the oocyst concentration and the total fecal volume collected for the corresponding day. The daily oocyst concentration was highly correlated with total daily oocysts shed for d 2–10 in the control calves, validating both quantification methods as robust measures of shedding even during peak oocyst production. Prophylactic treatment (20 mg) of calves before challenge tended to reduce total oocyte shedding on d 6 compared with control calves ( $P = 0.085$ ). These results validated the oocyst quantification technique and demonstrate the potential efficacy of the prophylactic fusion protein 4H9-G1-LL37 in reducing *C. parvum* shedding. These results also suggest that additional studies with a larger sample size are warranted.

**Key Words:** *Cryptosporidium parvum*, cathelicidin, fusion protein

**W35 Association between plasma haptoglobin concentration and bovine respiratory disease status in preweaned dairy calves.** Sonia J. Moisa\*<sup>1</sup>, Sharif S. Aly<sup>2</sup>, William J. Love<sup>2</sup>, Terry W. Lehenbauer<sup>2</sup>, Alison L. Van Eenennaam<sup>3</sup>, and Lindsey E. Hulbert<sup>1</sup>, <sup>1</sup>*Department of Animal Sciences and Industry, Kansas State University, Manhattan, KS*, <sup>2</sup>*School of Veterinary Medicine, University of California-Davis, Tulare, CA*, <sup>3</sup>*Department of Animal Sciences, University of California-Davis, Davis, CA*.

The objective of this case-control study was to assess the association between bovine respiratory disease score, determined by a scoring system (BRDs), and plasma Haptoglobin concentration (Hp). Four dairy operations located in the central San Joaquin Valley of California participated in this study (A, B, C, D) during the months of April to September, 2013. At each location, heifer or bull dairy calves were identified using as treatments BRDs-positive ( $n = 150$ ); and randomly chosen calves within the same age group, sex, and breed (Jersey or Holstein) served as controls (Con;  $n = 356$ ). Whole blood was collected, and plasma Hp concentrations were measured using a colorimetric method based on peroxidase activity. In addition, each calf had a respiratory examination via thoracic auscultation (Au) and Ultrasound (Us). Haptoglobin data were analyzed using the MIXED procedure of SAS using age as a covariate with BRDs as the fixed effect in the statistical model with breed, sex, Au, Us, or location analyzed separately. Some Con calves had abnormal Au ( $n = 22$ ; 15%) and some of the BRDs calves had normal Au ( $n = 113$ ; 32%). Preliminary results showed that BRDs-positive calves with abnormal Au and Us tended to have higher Hp ( $P = 0.10$  and  $P = 0.13$  respectively). Location accounted for variation in Hp ( $P = 0.01$ ); Control calves at D had greater concentrations of Hp than Con calves at the other locations. In addition, BRDs-positive calves at A had lower Hp than BRDs-positive calves at the other locations. Further investigation is needed to determine if Hp can serve as a biological marker for respiratory disease in BRDs-positive or negative calves with abnormal Au and Us. Also, baseline concentrations for haptoglobin may need to be established for specific locations.

**Key Words:** bovine respiratory disease, haptoglobin, preweaned dairy calves

**W36 Cosinor analysis of CRT in heifers.** Alexander W. Altman\*<sup>1</sup>, Nicole C. Burdick-Sanchez<sup>2</sup>, Jeffery A. Carroll<sup>2</sup>, Ty B. Schmidt<sup>3</sup>, Kyle R. McLeod<sup>1</sup>, Glen E. Aiken<sup>4</sup>, and Eric S. Vanzant<sup>1</sup>,



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Cosinor analysis can detect changes in phase, amplitude, or means of circadian rhythm of body temperature (CRT). Two experiments (E1: 22 Angus heifers, 292 ± 9 kg BW; E2: 16 Angus and 8 Hereford × Angus heifers, 335 ± 11 kg BW) evaluated changes to CRT due to a minor, chronic stressor (endophyte) and a major, acute stressor (LPS). Diets, starting 10 d before LPS challenge and fed at 1.8 × NE<sub>m</sub> in individual stalls (3.0 × 3.7 m), contained 20% of either endophyte-free (E-) or endophyte-infected (E+) fescue seed, 30% cottonseed hulls, 36% cracked corn, and 14% supplement and were balanced to meet protein and mineral requirements. E2 included MGA (0.5 mg/hd/d) and evaluation of weaning exit velocity (wEV). Vaginal (E1 and E2) and rectal (E2 only) temperature probes recorded body temperature every 5 min. Cosinor analysis was used to analyze data in each of 4 phases: pre-treatment diet (P1), treatment diet, pre-LPS (P2), post-LPS temperature spike 1 (P3), post-LPS temperature spike 2 (P4). Disruption to CRT occurred during P2 of both experiments, as evidenced by 39% of curves failing to fit a cosine model (i.e., less than 40% R<sup>2</sup>) compared with only 5% of curves failing to meet inclusion criteria for all other phases, suggesting that abrupt dietary shifts can affect temperature regulation. Amplitude differences across days ( $P = 0.05$ ) occurred in P1 of E1, when mean temperatures did not differ ( $P > 0.10$ ). Amplitude ( $P = 0.80$ ) and mean temperature ( $P = 0.66$ ) between treatment groups in P3 of E1 were similar, but shifts in timing of the peak ( $P = 0.09$ ) occurred. Endophyte affected vaginal temperature amplitudes in P2 ( $P = 0.07$ ) and P4 ( $P = 0.08$ ) of E2 without influencing mean temperatures. Shifts in timing of peak vaginal temperatures during P1 ( $P = 0.01$ ) of E2 across days may indicate adaptation of CRT to shifts in environment. Treatment × wEV interactions were significant for mean rectal, but not vaginal, temperatures during P1 ( $P = 0.10$ ), P2 ( $P = 0.10$ ), and P3 ( $P = 0.04$ ) of E2, indicating different temperature responses between sites. These data demonstrate that cosinor analysis can be used to detect shifts in CRT responses that would not be detected by evaluation of mean temperature responses alone.

**Key Words:** endophyte, LPS, CRT

**W37 Effect of bovine genotype on heifer response to repeated lipopolysaccharide (LPS) administration.** Georgina Cousillas<sup>\*1</sup>, Aimee Benjamin<sup>2</sup>, Wanda J. Weber<sup>1</sup>, David Kerr<sup>2</sup>, Theodore H. Elsasser<sup>3</sup>, Stanislaw Kahl<sup>3</sup>, and Brian A. Crooker<sup>1</sup>, <sup>1</sup>University of Minnesota, St. Paul, MN, <sup>2</sup>University of Vermont, Burlington, VT, <sup>3</sup>USDA-ARS, Beltsville, MD.

Heifers (n = 4/genotype) from unselected (stable milk yield since 1964, UH) and contemporary (CH) Holsteins that differed in milk yield (6,200 and 11,100 kg milk/305 d) or from Red Angus cows (RA) were fed the same diet ad lib and housed together for 47 d before being challenged with 0.5 µg LPS/kg BW. Heifers were 20 mo old and pregnant except for 2 CH heifers that were synchronized to be at d 8 of their cycle at the first LPS challenge (C1). Progesterone exceeded 3.5 ng/mL at C1. Jugular catheters were implanted 24 h before LPS (*Escherichia coli* O111:B4). Blood samples were collected at -1, -0.5, 0, 1, 2, 3, 4, 6, 8, and 24 h relative to LPS administration and plasma harvested. Body temperatures (BT) were determined at these times and at 5 and 7 h. A second identical LPS challenge (C2) was administered 4 d later. Data were analyzed by repeated measures using PROC MIXED (SAS). Means differed when  $P < 0.05$ . Cortisol, interleukin-6 (IL-6), xanthine oxidase

(XO), and tumor necrosis factor α (TNFα) were greater ( $P < 0.01$ ) in C1 than C2, BT and IGF-1 were less ( $P < 0.01$ ) and glucose and nitric oxide (NOX) did not differ ( $P > 0.11$ ) between C1 and C2. There were genotype × challenge × time interactions ( $P < 0.05$ ) for BT, glucose and TNFα. During C1, BT increased earlier and peaked higher in RA than UH or CH, glucose increased less in RA than UH or CH, and TNFα increased more in CH than UH and RA. Glucose, TNFα, and BT did not differ ( $P > 0.10$ ) among genotypes during C2. There was an interaction of genotype and challenge for IL-6 as response in UH was greater than in CH or RA in C1 but there was no effect of genotype on IL-6 in C2. There was a trend for NOX ( $P = 0.10$ ) to be less in RA than in UH or CH and a trend ( $P = 0.10$ ) for a genotype × challenge interaction for XO as XO was greater in UH than in CH and RA in C1 but not in C2. Results indicate the reduced response during a repeated challenge decreases the ability to detect an effect of genotype on LPS. Regardless, results indicate genotype affects bovine response to LPS and that the effect of genotype differs among the response variables assessed in this study.

**Key Words:** innate immunity, bovine genotype, lipopolysaccharide

**W38 The effects of plane of milk replacer nutrition on the health and performance of high-risk Holstein bull calves from a commercial calf ranch.** K. P. Sharon<sup>\*1,3</sup>, L. E. Hulbert<sup>2</sup>, J. A. Carroll<sup>3</sup>, and M. A. Ballou<sup>1</sup>, <sup>1</sup>Department of Animal and Food Sciences, Texas Tech University, Lubbock, TX, <sup>2</sup>Department of Animal Sciences and Industry, Kansas State University, Manhattan, KS, <sup>3</sup>Livestock Issues Research Unit, USDA-ARS, Lubbock, TX.

To determine if preweaning plane of milk replacer nutrition (PON) influences health and performance of high risk Holstein bull calves, 36 Holstein bull calves (1d of age) from a commercial calf ranch were assigned to either a high (HPN; n = 18; 20% and 28% DM lipid and protein) or a low plane of milk replacer nutrition (LPN; n = 18; 20% DM lipid and protein) through weaning. Total serum protein concentrations confirmed that 72 and 76% of calves had failure of passive transfer (<5.2g/dL) in HPN and LPN, respectively. All calves were bottle fed twice daily, the LPN were fed 445 g DM/d of milk replacer until weaning, and the HPN calves were fed 830 g DM/d of milk replacer during the first 10 d and 1080 g from 11 d until weaning. Calf starter and water were offered ad libitum. Calves were step-down weaned beginning at 51 d and ending on 57 d. Data are reported as HPN vs. LPN throughout, respectively. Two of the 18 calves died in both treatments. There was a tendency ( $P = 0.100$ ) for more HPN calves to bloat during the study (29.4 vs 6.7%). The HPN calves had greater ( $P = 0.007$ ) incidence of scouring (66.7 vs. 22.2%), but there was no difference ( $P = 0.688$ ) in the percentage of calves that received systemic antibiotics (47 vs. 40%). There was a time × PON interaction ( $P < 0.001$ ) in weekly starter intake, where LPN ate more ( $P \leq 0.004$ ) starter during wk 6, 7, 8, and 9. There was a time × PON interaction ( $P = 0.017$ ) in water intake, where LPN had greater ( $P \leq 0.001$ ) water intake throughout the study (3.6 vs. 2.3 ± 0.21L). There was a time × PON interaction ( $P < 0.001$ ) in ADG, whereas HPN had greater ( $P \leq 0.001$ ) ADG from 0 to 25 d (0.53 vs. 0.08 ± 0.040kg/d), 0 to 49 d (0.60 vs. 0.23 ± 0.040kg/d), and 0 to 68 d (0.58 vs. 0.33 ± 0.049kg/d). There was also a time × PON interaction ( $P < 0.001$ ) in F:G, whereas HPN had greater ( $P \leq 0.054$ ) efficiency from 0 to 25 d (1.7 vs. 5.8 ± 0.38), 0 to 49 d (1.8 vs. 3.7 ± 0.38), and 0 to 68 d (2.2 vs. 3.3 ± 0.38). Risk for bloat and scouring were greater for calves fed the HPN, but there was no difference in antibiotic treatment or mortality. Further, the HPN calves had greater performance over the preweaning period.

**Key Words:** calf, health, nutrition

**W39 The effect of diarrhea, fever, and respiratory disease on post-weaning weight of Holstein dairy heifers in an automated feeding system.** Gabriela A. Zaldumbide\*, Melissa C. Cornett, and Amy L. Stanton, *University of Wisconsin-Madison, Madison, WI.*

Automated feeder systems (AFS) are gaining popularity in the dairy industry. Considering that calves intermingle in this system, it is important to evaluate and understand the risk of diseases and their impact on calf weight. This study examined the effect of cumulative health events (HE), respiratory disease (BRD), fever (F), and diarrhea (D), on 1-wk post-weaning weight (FWT) in calves. Holstein heifers ( $n = 96$ ) from a  $2 \times 2$  factorial study were used for this trial. Factorials were direct-fed microbial (DFM) efficacy and 2 pasteurized milk feeding plans that offered 543 L over 53 feeder days (FD). Plan 1 offered (7L/d) and peaked at 28 FD (13L/d). Plan 2 offered (11L/d) and peaked at 21 FD (15L/d). Both plans weaned calves at 53 FD. Researchers conducted complete health exams twice weekly from birth until FWT. Calves were weighed at  $3 \pm 2$  d (Mean  $\pm$  SD) of age and at FWT  $66 \pm 3$  d. The effect of HE on FWT was evaluated by PROC MIXED in SAS, controlling for

initial weight, age, and milk plan with study month as a random effect. The HE of calves were categorized into a score of 0, 1, 2, or 3 based on number of HE for each of the following: F, BRD, and D. A score of 0 indicated no HE, a score of 1 was 2–3 HE, a score of 2 was 4–5 HE, and a score of 3 was 6+ HE. For F, D, and BRD 22%, 20.8%, 19.8% of calves had a score of 0, respectively. For F, 52.1%, 18.8%, and 7.2% were categorized as 1, 2, and 3, respectively. For D, 53.1%, 24.0%, and 2.1% of calves scored 1, 2 and 3, respectively. For BRD 41.7%, 29.1%, and 9.4% of calves scored 1, 2, and 3, respectively. Final post weaning weight was  $93.6 \pm 12.9$  kg (LSM  $\pm$  SEM). Initial weight and final age were significantly associated with FWT ( $P < 0.0001$ ), ( $P < 0.0001$ ), respectively. For every increase in D HE score, FWT were lowered by  $-1.54 \pm 0.62$  kg ( $P < 0.02$ ). For every additional F HE, FWT was lowered  $-1.05 \pm 0.50$  kg ( $P < 0.04$ ). Final weight was not influenced by BRD ( $P = 0.54$ ). Results suggest that diarrhea and fever have a cumulative effect on growth in dairy calves reared in AFS.

## Animal Health: Monogastric health

**W40 Changes in liver and white adipose tissue metabolism induced by postnatal nutritional restriction in piglets with intra-uterine growth restriction.** Liang Hu\*, Xie Peng, Fali Wu, Chuan Yan, Qin Xu, Yan Liu, De Wu, Shengyu Xu, Yan Lin, Zhengfeng Fang, and Lianqiang Che, *Institute of Animal Nutrition, Sichuan Agricultural University, Chengdu, Sichuan, China.*

It is well known that intrauterine growth restricted (IUGR) neonates lead to escalation in the incidence of metabolic syndrome due to post-natal early catch up growth. We hypothesized that postnatal nutritional restriction may improve metabolic status of liver and white adipose tissue (WAT) in IUGR neonates. Piglets with a birth weight near the mean litter birth weight (SD 0.5) were defined as normal-birth weight (NBW), whereas those with at least 1.5 SD lower birth weight were defined as IUGR. Twelve pairs of NBW and IUGR piglets at 7 d of age were randomly assigned to adequate nutrient intake (ANI) or restricted NI (RNI) for a period of 21 d, which produced 4 experimental groups as NBW-ANI, IUGR-ANI, NBW-RNI and IUGR-RNI ( $n = 6$  per group). The NBW-ANI and IUGR-ANI piglets had free access to formula milk, the NBW-RNI piglets were provided the same amount of formula as the IUGR-ANI piglets, and the IUGR-RNI piglets were provided approximately 70% of the formula intake of the IUGR-ANI piglets. At d 28, blood, liver and WAT samples were collected at necropsy and analyzed for hormone, metabolites and gene expressions. Data were analyzed by SPSS software using the MIXED procedure. The results indicated that IUGR significantly increased leptin concentration ( $P < 0.01$ ). Compared with NBW piglets, the mRNA abundance of glucose-6-phosphatase and acetyl-CoA carboxylases were significantly decreased ( $P < 0.05$ ) while sterol regulatory element-binding protein-1c (*SREBP1c*) was markedly increased ( $P < 0.01$ ) in the liver of IUGR piglets; moreover, the mRNA abundances of *CD36*, hormone-sensitive lipase, adipose triglyceride lipase and carnitine palmitoyltransferase-1A in WAT were significantly decreased by IUGR ( $P < 0.05$ ). Irrespective of BW, the hepatic mRNA abundances of *SREBP1c*, glucose transporter 4, phosphoenolpyruvate carboxykinase, lipoprotein lipase and peroxisome proliferator activated receptor  $\alpha$  were significantly increased by RNI ( $P < 0.05$ ). In summary, postnatal nutritional restriction changed blood metabolites and hormone concentrations by the metabolic related genes in piglets with IUGR.

**Key Words:** nutritional intake, metabolism, white adipose tissue

**W41 MicroRNA expression profile of the mouse lung infected with a virulent avian H5N2 virus.** M. K. Shim\*<sup>1</sup>, E. J. Choi<sup>2</sup>, S. H. Hong<sup>1</sup>, Y. K. Choi<sup>2</sup>, and H. B. Kim<sup>1</sup>, <sup>1</sup>Dankook University, Cheonan, Chungnam, Republic of Korea, <sup>2</sup>Chungbuk National University, Cheongju, Chungbuk, Republic of Korea.

The aim of this study was to investigate the differences of the microRNA (miRNA) expression profiles in mouse lungs infected with wild type low pathogenic H5N2 avian influenza A (w81) or mouse-adapted virulent H5N2 avian influenza A (ma81) virus. Different sensitivities of influenza A virus strains to the hosts cause variations in miRNA expression. Therefore, some of miRNAs can be used as potential prognostic targets during the avian influenza A virus infections in mammalian hosts. Five-week-old C57BL/6 female mice were inoculated with 30  $\mu$ l of  $10^4$  TCID<sub>50</sub> of w81 or ma81. Lung tissues from 3 mice per group were harvested at 1 and 3 dpi. A small RNA library was constructed from the total RNAs of lung samples and sequenced using the Solexa platform. Sequence reads were normalized to determine the number of transcripts

per million, and fold changes of miRNAs were evaluated.  $P$ -value was calculated using following formula as previously described.  $p(x|y) = (N2/N1) (x+y)! / x!y! (1 + N2/N1)(x+y+1)$  where  $x$ ,  $y$ ,  $N1$ , and  $N2$  represent number of miRNAs surveyed, number of homologous miRNAs in controls, total number of clean reads in controls, and total number of clean reads in treatments, respectively. Gene ontology analysis was conducted by miRanda and DAVID. The w81 virus induced a higher number of differentially expressed miRNAs compared with the ma81 virus. It is interesting to see that only 9 miRNAs (miR-100-5p, miR-130a-5p, miR-146b-3p, miR-147-3p, miR-151-5p, miR-155-3p, miR-223-3p, miR-301a-3p, and miR-495-3p) were significantly upregulated in both lungs infected either with the w81 or ma81 strain at both time point ( $P < 0.05$ ). Especially, expression levels of 4 miRNAs were higher in the lungs of mice infected with the ma81 virus than those infected with the w81 virus ( $P < 0.05$ ). These 4 miRNAs have been implicated in immune responses (miR-223-3p and miR-147-3p), viral infection (miR-155-3p), and cell migration (miR-151-5p). Our results suggest that the mammalian adaptation of avian influenza A virus results in a different miRNA expression pattern in lungs of virus-infected mice compared with its parental strain. Thus these might be used as potential prognostic targets during the avian influenza A virus infections in mammalian hosts.

**Key Words:** influenza A virus, microRNA, virulence

**W42 Relationship between *Salmonella* translocation patterns and immune responses in orally inoculated pigs.** Paul R. Broadway\*<sup>1</sup>, Jeffery A. Carroll<sup>1</sup>, Nicole C. Burdick Sanchez<sup>1</sup>, E. V. Gart<sup>2</sup>, L. K. Bryan<sup>2</sup>, R. M. Gold<sup>2</sup>, C. Yang<sup>2</sup>, and Sara D. Lawhon<sup>2</sup>, <sup>1</sup>Livestock Issues Research Unit, USDA-ARS, Lubbock, TX, <sup>2</sup>Department of Veterinary Pathobiology, College of Veterinary Medicine and Biomedical Sciences, Texas A&M University, College Station, TX.

*Salmonella* is a pathogen of interest with broad implications ranging from animal health to food safety. Translocation patterns of *Salmonella* from the gastrointestinal tract to peripheral tissues have not been fully elucidated. Also, the mechanisms by which immunological responses influence translocation and fecal shedding are not fully understood. The objective of this study was to determine the translocation patterns of orally inoculated *Salmonella* in response to changes in immune biomarkers. Male pigs ( $n = 12$ ;  $6.1 \pm 2$  kg) were orally inoculated with  $4.7 \times 10^9$  cfu of *Salmonella* Typhimurium. Whole blood samples were collected and analyzed for serum cortisol concentrations and complete blood counts. Fecal samples were collected daily from -1 to 3 d relative to the bacterial challenge on d 0. At 72 h post-inoculation, animals were humanely euthanized and tissues were collected to determine the presence of the inoculated *Salmonella* [mesenteric lymph node (LN), subiliac LN, liver, spleen, kidney, and gallbladder]. There was a tendency ( $P = 0.06$ ) for fecal shedding quantity to be similar to mesenteric LN *Salmonella* concentrations at 72 h. There was no interaction ( $P > 0.05$ ) between the presence of *Salmonella* in LN and any of the immune parameters measured. Subiliac LN *Salmonella* concentrations were highly correlated ( $P = 0.0001$ ;  $r = 0.99$ ) with liver *Salmonella* concentrations. Fecal shedding at d 3 was negatively correlated with kidney and gallbladder *Salmonella* concentrations ( $P = 0.04$ ). Cecum concentrations of *Salmonella* were negatively correlated with white blood cell ( $P = 0.02$   $r = -0.72$ ) and neutrophil ( $P = 0.0003$ ) counts. These data suggest that orally inoculated *Salmonella* may translocate to tissue not only within the gastrointestinal tract but to organs and peripheral tissues such as musculoskeletal LN. Additionally, negative



correlations between immune biomarkers and *Salmonella* migration suggest that translocation of *Salmonella* may be inhibited following the peak immunological response. Further information is needed to fully elucidate the mechanisms by which pathogens interact with their host and how an immune response alters migration patterns.

**Key Words:** *Salmonella*, swine, immune

**W43 Isolation and characterization of *Clostridium tertium* in poultry feces.** S. H. Hong\*, S. A. Seok, M. K. Shim, and H. B. Kim, Dankook University, Cheonan, Chungnam, Republic of Korea.

The aim of this study was to isolate and characterize *Clostridium tertium* from chicken feces. *C. tertium* is found in soil as well as gastrointestinal tracts of humans and other animals. *C. tertium* is an anaerobic, motile, gram-positive, rod-shaped bacterium that forms highly resistant endospores without generating an exotoxin. Although it is considered an uncommon pathogen and its pathogenicity is often unclear, this bacterium has been implicated in bacteremia, pneumonia, enterocolitis, septic arthritis and abscess in humans and animals. Even though there is no direct evidence that *C. tertium* can be transmitted from animals to humans, it is of importance to see the prevalence and distribution of *C. tertium* in animal feces. Chicken feces samples were collected from the research farm at Dankook University, South Korea. Ten chickens per cage were housed together and 10 chicken droppings per cage (a total of 10 cages) were collected using sterile disposable culture loops starting when chickens were 1 week of age then at one-week interval until they were 4 weeks of age. After ethanol treatment (50% ethanol for 1 h), dropping samples were streaked onto TCCFA agar plates with 5% sheep blood, and incubated in anaerobic jars at 37°C for 72 h. The isolates were identified based on morphological criteria. Then identification was confirmed by the Gram staining and sequencing of the V1-V3 of 16S rRNA gene. After 72 h incubation in an anaerobic jar, a pure culture of colonies was recovered. A Gram stained smear of colonies showed weak-staining, gram-positive rods. These rod-shaped bacteria were detected from all chicken feces. The cloning and subsequent sequencing of the 16S rRNA genes from 2 representative isolates identified them as *C. tertium* with a probability of 99%. To our knowledge, this is the first isolation of *C. tertium* from chicken droppings. In depth studies on pathogenesis of *C. tertium* will be needed to better understand potential roles of *C. tertium* in poultry health.

**Key Words:** *Clostridium tertium*, chicken, feces

**W44 Antimicrobial resistance strategy and effect on the veterinary feed directive regulation.** David B. Edwards\*, Dragan Momcilovic, Sharon A. Benz, and Jo W. Gulley, Division of Animal Feeds, Center for Veterinary Medicine, Rockville, MD.

On December 12, 2013, the Food and Drug Administration (FDA) proposed to change its animal drug regulations to improve the efficiency for stakeholders using veterinary feed directive (VFD) drugs while continuing to protect human and animal health. The VFD drug class was created in 1996 by the Animal Drug Availability Act, and they are intended for use in or on animal feed under a veterinarian's order and professional supervision. The FDA established the VFD program in 2000. Changes to the VFD regulation are important as FDA implements the judicious use principles for medically important antimicrobial new animal drugs approved for use in food-producing animals, based on the framework set forth in Guidance for Industry (GFI) #209 issued April 13, 2012 and the process and timeline in GFI #213 issued December 13, 2013. Antimicrobials must continue to be available to combat disease in

animals, including treatment, control, and prevention, while preserving availability of effective drugs. The primary concern is the continued effectiveness of "medically important" drugs. One important change in GFI #213 is to include veterinary involvement in the use of these antimicrobials by changing the drug marketing status of antimicrobial drugs from over the counter to prescription or VFD drugs to be used for therapeutic uses only. This means a VFD order from a licensed veterinarian will be needed to obtain medicated feeds containing VFD drugs. This order must include information on the number and type of animals to be treated. This keeps veterinarians included in the decision-making process to use medically important antimicrobials. FDA has secured the cooperation of drug manufacturers to change the drug marketing status and remove the production uses from these drug approvals. To improve the VFD process, FDA proposed several key changes in the proposed rule such as removing the federally defined veterinarian-client-patient-relationship requirement, eliminating the automatic classification of VFD drugs as Category II, and modifying recordkeeping requirements. FDA received about 2000 comments on the proposed rule and is reviewing those comments to publish a final rule.

**Key Words:** antimicrobial, veterinary feed directive, regulation

**W45 Estimating glucose requirements of an activated immune system in growing pigs.** Sara K. Stoakes\*, Erin A. Nolan, Mohanad Abuajamieh, Maria V. Sanz Fernandez, and Lance H. Baumgard, Iowa State University, Ames, IA.

Activated immune cells are obligate glucose utilizers and a large lipopolysaccharide (LPS) IV dose causes severe hypoglycemia. Therefore, study objectives were to use the quantity of glucose needed to maintain euglycemia following an endotoxin challenge as a proxy of immune cell glucose requirement. Fifteen fasted crossbred gilts (30 ± 2 kg) were jugular catheterized bilaterally and assigned one of 2 IV bolus treatments: control (CON; 10 mL sterile saline; n = 7) or LPS-infused + euglycemic clamp (LPS-Eu; *E. coli* 055:B5; 5 µg/kg BW; 50% dextrose infusion to maintain euglycemia; n = 8). Following infusion, blood glucose was determined every 10 min and dextrose infusion rates were adjusted in LPS-Eu pigs to maintain euglycemia for 8 h. Rectal temperature was increased in LPS-Eu pigs relative to control (39.8 vs 38.8°C,  $P < 0.01$ ). After 3 h, blood glucose content gradually declined for CON pigs while LPS-Eu glucose levels remained unchanged ( $P = 0.01$ ). Plasma insulin, BUN, BHBA, and L-lactate were increased in LPS-Eu pigs compared with CON (69, 57, 21, and 60%, respectively;  $P < 0.05$ ). By 8 h, plasma LPS binding protein was increased 24% in LPS-Eu pigs relative to controls ( $P < 0.01$ ). Plasma NEFA increased with time in CON pigs, but remained unchanged in the LPS-Eu pigs ( $P < 0.01$ ). White blood cells, lymphocytes, monocytes, eosinophils, and basophils were decreased in LPS-Eu pigs relative to CON ( $P < 0.01$ ). Additionally, the neutrophil-to-lymphocyte ratio was increased in LPS-Eu pigs relative to CON (72%,  $P < 0.01$ ). During the 8 h, 232 ± 16 g of infused glucose was required to maintain euglycemia. If the amount of glucose required to maintain euglycemia can be used as a proxy, then the glucose requirements of an activated immune system are approximately 29 g/h.

**Key Words:** lipopolysaccharide, immune challenge, glucose homeostasis

**W46 The influence of sodium alendronate on performance and bone densitometry of broilers at 42 days of age.** Thays Cristina Oliveira Quadros\*<sup>1</sup>, Sarah Sgavioli<sup>1</sup>, Giuliana Milan de Andrade Rocha Garcia<sup>1</sup>, Liliana Longo Borges<sup>1</sup>, Elaine Talita Santos<sup>1</sup>, Diana

M. Correa Castiblanco<sup>1</sup>, Albaraa Hisham Sarsour<sup>2</sup>, Lizandra Amoro-oso<sup>1</sup>, Joao Batista Matos Junior<sup>1</sup>, Joao Paulo M. Chiquini<sup>1</sup>, Otto Mack Junqueira<sup>1</sup>, and Silvana Martinez Baraldi Artoni<sup>1</sup>, <sup>1</sup>*Paulista State University Julio de Mesquita Filho, Jaboticabal, Sao Paulo, Brazil*, <sup>2</sup>*North Carolina State University, Raleigh, NC*.

Sodium alendronate (SA) is a drug that shows effects on bone mass, which may contribute to better bone development of animals, allowing for more efficient production performance. This study was conducted to evaluate the influence of SA on performance and bone mineral density of Cobb broilers at 42 d of age. Twelve hundred broilers used in this study were derived from 2 treatments that included injection or no vitamin D injection during incubation. After the eggs hatched, the broilers received water containing different levels of sodium alendronate (0, 2, 4, 6 or 12 mg/mL). The water solution with SA was supplied through esophageal inoculation in broilers chicken once a week for 3 weeks. One milliliter of solution was given to each bird on 7, 14 and 35 d of age, to ensure that all animals ingested the treatment. Experimental design was completely randomized in a 2 × 5 factorial for a total of 10 treatments (injected or not × sodium alendronate level). Broiler performance was evaluated by calculating feed intake, body weight gain and feed conversion. Bone density of the tibiotarsus was analyzed at 42 d using DEXA equipment. Data were analyzed using the JMP program for ANOVA. For the performance parameters of the broilers there was no significant effect ( $P > 0.05$ ), as well as for the densitometry results, though one can observe an increased bone density value (0.242 g/cm<sup>2</sup>) in broilers derived from eggs injected with vitamin D and subsequently treated with 4mg of SA level. Treatments with SA can increase total skeletal mass, but the data suggest that despite the non-significant results of SA in this experiment, a numerical increase in bone density was observed, hence an improvement in bone development. It was concluded there was no influence on broiler performance ( $P = 0.798$ ) and for bone mineral density the level of 4 mg of SA was numerically ( $P = 0.246$ ) better when compared with other treatments, which encourages us to continue our research to establish a significant level of SA. SA may enable us to increase bone mineral density, and possibly contribute to weight gain of broilers.

**Key Words:** bone development, chicken, densitometry

**W47 Potential of a new probiotic strain, *Bifidobacterium longum* ssp. *infantis* CECT 7210, to improve health status of weaning piglets orally inoculated with *Salmonella* Typhimurium or ETEC K88.** E. Barba-Vidal<sup>1</sup>, L. Castillejos<sup>1</sup>, C. Sol<sup>\*1</sup>, M. Rivero<sup>2</sup>, JA Moreno<sup>2</sup>, and SM Martín-Orúe<sup>1</sup>, <sup>1</sup>*Animal Nutrition and Welfare Service, Animal and Food Science Department, Universitat Autònoma de Barcelona, Bellaterra, Spain*, <sup>2</sup>*Laboratorios Ordesa S.L., Parc Científic de Barcelona, Barcelona, Spain*.

A new probiotic strain of *Bifidobacterium longum* subsp *infantis* CECT 7210 (Laboratorios Ordesa, S.L.) was evaluated in 2 oral challenges against *Salmonella* Typhimurium or enterotoxigenic *E. coli* (ETEC) K88. Seventy-two piglets, 21 and 24 (±2) days old were used in each trial. Animals were distributed in 24 pens and 4 groups in a 2 × 2 design: with and without probiotic and oral challenge. The animals were fed a plain diet and the probiotic was administered orally on a daily basis (10<sup>9</sup> cfu/d). After a 1-week adaptation, a double oral inoculation was done with *Salmonella* Typhimurium (2 × 10<sup>9</sup> and 6 × 10<sup>9</sup> cfu/day) (trial 1) or ETEC K88 (5 × 10<sup>9</sup> and 6 × 10<sup>10</sup> cfu/day) (trial 2). Intake, live weight, fecal excretion of the pathogen, fecal consistency and rectal temperature were registered. Results were analyzed with a GLM and adjusted by Tukey. Microbiological frequencies were analyzed by Fisher. On d 4 and 8 post inoculation (PI), blood samples were obtained from one animal

from each pen to assess inflammatory response (TNF- $\alpha$  and Pig-Map), being euthanized afterward for intestinal content and ileal scrapings sampling for microbiological and fermentation products analysis. Performance parameters were affected by the *Salmonella* challenge with reductions in feed intake and gains (ADG 118 g challenge animals vs. 209 g,  $P < 0.001$ ), but not by *E. coli* (ADG 114g challenge animals vs. 120 g,  $P = 0.801$ ). No significant differences were found related to the probiotic. The challenge also got worse fecal consistency ( $P < 0.001$ ) and increased plasma TNF- $\alpha$  levels ( $P < 0.05$ ). Although the probiotic did not ameliorate these clinical signs it was able to reduce fecal excretion of *Salmonella* (especially at d 3 PI;  $P = 0.043$ ) and diminished the percentage of animals with countable (>10<sup>5</sup>cfu/g) coliforms in ileal scrapings at d 4 PI (58 vs. 91%;  $P = 0.010$ ). A consistent interaction was seen in both trials for colonic short chain fatty acids (SCFA), were the probiotic increased their concentration in the control animals but not in the challenged ones. Results indicate administration of the probiotic can reduce intestinal colonization by *Salmonella* and ETEC K88 in mild enteric processes.

**Key Words:** probiotic, *Salmonella*, ETEC K88

**W48 A comparison for IgG absorption between Minpig and Landrace piglets.** Shiquan Cui<sup>\*1,2</sup>, Yuan Xu<sup>1</sup>, Xuankai Huang<sup>1</sup>, Xibiao Wang<sup>1</sup>, and Yuzhi Li<sup>2</sup>, <sup>1</sup>*Northeast Agricultural University, Harbin, Heilongjiang, China*, <sup>2</sup>*West Central Research and Outreach Center, Morris, MN*.

The levels of IgG in colostrum is responsible for the natural passive immunity of piglets, so effectively obtaining colostrum is important to survival and growth of newborn piglets. A study was conducted to evaluate the ability of newborn Minpig piglets to obtain IgG from colostrum. Four pairs of Minpig and Landrace sows (3–5 parity), with each pair farrowing their first piglet within an hour, were used. The 6 first born piglets were removed from their dam immediately after birth to prevent them from suckling colostrum. Then 3 out of 6 piglets in each litter were cross-fostered to a sow of another breed, resulting in 4 treatment groups: MM (Minpig piglets nursed by Minpig sows), ML (Landrace nursed by Minpig), LM (Minpig nursed by Landrace), and LL (Landrace nursed by Landrace), with 12 piglets in each group. Blood samples were collected from all piglets at 12 h, 24 h, 36 h, 48 h, 72 h, and 7 d after birth to determine concentrations of IgG using ELISA. Serum IgG concentrations for MM, ML, LM, and LL groups were 58.95 ± 3.33, 53.81 ± 3.64, 54.03 ± 3.11, and 48.89 ± 2.60 mg/ml, respectively, at 24h after birth. Compared with Landrace piglets, Minpig piglets had higher IgG concentrations ( $P < 0.05$ ) regardless of being nursed by Minpig or Landrace sows. To further verify the efficiency of Minpig piglets to absorb IgG, a piglet from each 5 litters of Minpig and Landrace were tube-fed bovine IgG (1000mg/kg·BW) at 0 h, 6 h, 12 h, 24 h, 36 h, 48 h, and 72 h after birth, respectively. Blood samples were collected from the piglets at 6h after eating BIgG, and the absorption efficiency of BIgG was calculated for each piglet based on:  $\{[\text{serum BIgG concentration} \times \text{BW} \times 0.10 \times (1 - \text{hematocrit})] / \text{BIgG consumed}\} \times 100\%$ . Results indicate that Minpig piglets had higher absorption efficiency of exogenous BIgG than Landrace piglets (19.82 ± 1.55 vs. 17.07 ± 1.18 at 24 h,  $P < 0.05$ ; 17.25 ± 1.63 vs. 14.43 ± 1.43 at 36 h,  $P < 0.05$ ). These results suggest that Minpig piglets had a stronger capability to obtain passive immunity through milk and feed compare with Landrace piglets during the suckling period.

**Key Words:** Minpig, piglet, IgG

## Beef Species

**W49 Characterization of growth traits of Senepol heifers and bulls from birth through a year of age in the tropics.** Robert W. Godfrey\* and Henry C. Nelthropp, *Agricultural Experiment Station, University of the Virgin Islands, St Croix, VI.*

Senepol is a *Bos taurus* breed of beef cattle that is well suited to the tropics. This study was conducted to evaluate growth of Senepol bull and heifer calves from birth to a year of age using calves born in spring of 2012 (n = 11 heifers and 10 bulls) and 2013 (n = 17 heifers and 16 bulls). Hip height (HHT) and BW were measured at weaning and yearling. Pelvic area (PA) of heifers and scrotal circumference (SC) of bulls were measured at yearling. Average daily gain (ADG) was calculated from birth to weaning and weaning to yearling. Data were analyzed using year and sex of calf in the model with sire as a covariate due to unequal representation of sires across years. Age of dam was not significant ( $P > 0.10$ ) for any trait. There was no effect of year or sex on birth weight ( $P > 0.10$ ). Bulls had a greater 205-d adjusted weaning weight ( $P < 0.008$ ) than heifers ( $242 \pm 6$  vs.  $217 \pm 6$  kg, respectively) but there was no effect of year ( $P > 0.10$ ). Bulls had a greater ADG from birth to weaning than heifers ( $0.96 \pm 0.03$  vs.  $0.83 \pm 0.03$  kg/d, respectively) but there was no effect of year ( $P > 0.10$ ). Weaning HHT was greater ( $P < 0.003$ ) in bulls than in heifers ( $111.5 \pm 0.7$  vs.  $108.3 \pm 0.7$  cm, respectively) and greater ( $P < 0.002$ ) for calves born in 2012 than in 2013 ( $111.9 \pm 0.8$  vs.  $107.8 \pm 0.6$  cm, respectively). Bulls had a greater 365-d adjusted yearling weight ( $P < 0.01$ ) than heifers ( $293 \pm 7$  vs.  $268 \pm 7$  kg, respectively) but there was no effect of year ( $P > 0.10$ ). Calves born in 2012 had lower ADG from weaning to yearling ( $P < 0.004$ ) than calves born in 2013 ( $0.28 \pm 0.02$  vs.  $0.37 \pm 0.02$  kg/d, respectively) but there was no effect of sex ( $P > 0.10$ ). Bulls had greater ( $P < 0.0008$ ) yearling HHT than heifers ( $118.8 \pm 0.7$  vs.  $115.4 \pm 0.7$  cm, respectively) and calves born in 2013 had greater HHT ( $P < 0.0002$ ) than those born in 2012 ( $118.9 \pm 0.6$  vs.  $115.2 \pm 0.7$  cm, respectively). Yearling SC of bulls was not different ( $P > 0.10$ ) between years ( $24.7 \pm 1.3$  vs.  $24.4 \pm 0.9$  cm, respectively). The PA of heifers was not different ( $P > 0.10$ ) between years ( $145.5 \pm 7.7$  vs.  $136.4 \pm 5.5$  cm<sup>2</sup>, respectively). These data show that there are differences in the growth traits between Senepol bull and heifer calves reared under tropical conditions.

**Key Words:** heifer, bull, cattle

**W50 Association of skin thickness and resistance to *Rhipicephalus microplus* in Simmental heifers.** J. A. II V. Silva\*<sup>1</sup>, A. M. Maiorano<sup>2</sup>, A. C. Verdugo<sup>3</sup>, Rogerio A. Curi<sup>1</sup>, and L. A. L. Chardulo<sup>1</sup>, <sup>1</sup>Universidade Estadual Paulista, Botucatu, SP, Brasil, <sup>2</sup>Universidade Estadual Paulista, Jaboticabal, SP, Brasil, <sup>3</sup>University of Saskatchewan, Saskatchewan, Canada.

The aim of this study was to verify association among skin thickness (ST) and resistance to the *Rhipicephalus microplus* ticks on Simmental animals, artificially infested with ticks. Eighteen Simmental heifers, with about to 450 age days and belonging a farm in the Avaré/SP/Brazil were used. ST was measure on the left side behind the shoulder of animal by a tuberculin caliper. Three artificial infestations were performed using approximately 20,000 *R. microplus* larvae on each animal. Counts female ticks were measured in the 19th and 23th range days after infestations, what only left side of the animal and engorged females with sizes larger than 4.5 mm were considered. Total tick counts (TTC) and payback percentages (PP) variables were transformed, respectively, in  $\log_{10}(n + 1)$  and  $PP^{0.25}$ . MIXED procedure of SAS considering repeated

measurements was used. Models considering the skin thickness group (STG), divided into thin leather and thick leather, infestation (IN) and their interactions as fixed effects were used in the analyzes for both variables. Mean comparisons were performed using Tukey-Kramer test ( $P < 0.05$ ). Repeatability coefficient (R) of the variables was estimated by intraclass correlation between the same animal measures. The IN effect was significant ( $P < 0.05$ ) for the studied variables. The STG mean were  $7.11 \pm 0.74$  and  $11.24 \pm 0.86$  for thin leather and thick leather, respectively. Means values observed, in chronological order, were  $0.33 \pm 0.29$ ,  $1.98 \pm 0.22$  and  $2.36 \pm 0.38$  to TTC and  $1.61 \pm 1.35$ ,  $6.64 \pm 0.83$  and  $8.40 \pm 1.64$  for PP. It is probably that the amount of detached female of animals significantly increased the infesting larval on pastures, caused significantly increased ( $P < 0.05$ ) of TTC and PP in the following counts. Value of R were 0.61 for both variables, suggesting that resistance to ticks measures observed in the same animal have similar values in artificial infestations data. There was no significant effect of STG, indicating that the ST trait is not an attribute that influences in cattle resistance to ticks.

**Key Words:** artificial infestation, beef cattle, repeatability

**W51 The effect of selection using residual average daily gain and marbling EPD on growth performance and carcass traits in Angus steers.** J. B. Wells\*, J. R. Segers, J. Duggin, J. K. Bertrand, R. Rekaya, and T. D. Pringle, *University of Georgia, Athens, GA.*

Steers (n = 68) were produced at the NW Georgia REC from the mating of Angus bulls selected for high and low residual ADG (RADG) EPD and high and average marbling (MARB) EPD to Angus cows. The resulting 2 × 2 factorial design contained 2 Angus sires in the following treatments: high RADG, high MARB (Hi/Hi); high RADG, average MARB (Hi/Avg); low RADG, high MARB (Lo/Hi); and low RADG, average MARB (Lo/Avg). Steers from the matings were used to determine the effects of RADG and MARB selection on growth performance and carcass traits. Steer weights were recorded and composition was determined via ultrasound at weaning (229 d) and yearling (391 d) ages. Steers entered the feedlot and were fed ad libitum for 2 wk before completing a 65-d GrowSafe Beef feed test to measure DMI, ADG, and residual feed intake (RFI). Steers were slaughtered at approximately 1.3 cm of backfat and USDA yield and quality grade data were collected after 24 h at 4C. Data were analyzed using ANOVA with the main effects of RADG and MARB EPD levels in the sire. Significant interactions were found for weaning and yearling weights and ultrasound REA, while DMI tended to be affected ( $P < 0.10$ ). The Hi/Hi steers were heavier ( $P < 0.05$ ) than Lo/Hi and Hi/Avg steers at both ages, while Hi/Hi and Lo/Avg steers had larger REA than Hi/Avg steers. There was a tendency ( $P = 0.07$ ) for yearling ultrasound IMF to be higher in steers from high MARB sires. Hi/Avg steers consumed less feed than Lo/Avg, with the Hi/Hi and Lo/Hi steers being intermediate. No RADG effects were observed for ADG ( $P = 0.96$ ); however, RFI tended to be lower ( $P = 0.11$ ) in steers from the high RADG sires. Slaughter weights were higher ( $P < 0.01$ ) for high vs low RADG groups, however, HCW and dressing percentage were not affected. USDA yield and quality grades did not differ ( $P > 0.10$ ) across RADG or MARB groups; however, carcass REA was larger ( $P < 0.05$ ) in Hi/Hi compared with Lo/Hi steers. In conclusion, selection using RADG appears to improve growth efficiency in Angus cattle,



however, further work is needed to fully characterize the potential for this trait as a selection tool in beef production systems.

**Key Words:** residual ADG, RFI, carcass

**W52 Relationship of grazing activities with residual feed intake measured in female Angus cattle carrying different genetic marker.** Ana Ines Trujillo\*, Alberto Casal, Mariana Carriquiry, and Pablo Chilbroste, *Facultad de Agronomia, Universidad de la Republica, Montevideo, Uruguay.*

A significant difference in residual feed intake (RFI,  $-1.02$  vs.  $1.02$  kg of DM) measured under grazing conditions in beef heifers carrying different genetic marker (presence – V group or absence – C group of 3 allelic variants of the genes neuropeptide Y (NPY), IGF-1 and leptin) was reported. The objective of this study was to explore if grazing activities would be associated with the RFI measured in 2 beef cattle groups: carrying the genetic marker associated with low RFI or not carrying the genetic marker. Grazing activities were measured in beef heifers ( $370 \pm 28$  d,  $294 \pm 37.4$  kg of BW,  $n = 18$ ) grazed on unrestricted and high quality temperate pasture. Grazing activities (grazing, ruminating and idling) were determined using behavior recorders in 2 sequential moments (wk 4 and wk 5) during herbage dry matter intake (DMI) determinations. Intake rate was obtained as the ratio between herbage DMI and actual grazing time of each animal. Data were analyzed as repeated measures. The model included genetic marker group (V or C), week and their interaction as fixed effects and paddock as random effect. More efficient heifers under grazing condition (V group) tended ( $P \leq 0.14$ ) to spent 45 min less in grazing time and to graze at a lower intake rate than less efficient heifers. Our results suggest that grazing activities would contribute to the biology behind RFI in growing heifers at grazing condition. Further experiments are needed to uncover drivers for better understanding the variation in RFI under grazing condition

**Key Words:** residual feed intake, beef cattle, grazing

**W53 Reproducibility of estimators and validity of feed efficiency models.** Carl A. Old\*<sup>1</sup>, Heidi A. Rossow<sup>2</sup>, and Thomas R. Famula<sup>2</sup>, <sup>1</sup>A3 Cattle Co., LeGrand, CA, <sup>2</sup>University of California, Davis, Davis, CA.

A residual feed intake (RFI) equation ( $ADFI = a + b_1 \times BW^{0.75} + b_2 \times ADG$ ), used to estimate ADFI in individual animals, was evaluated to determine if specification was correct and if predicted ADFI was equal to observed ADFI (validity). We evaluated 2 data sets of growing beef cattle using ordinary least squares (OLS), bootstrapping (BS) or a first order (FO) model in which final BW ( $BW_f$ ) was a function of initial BW ( $BW_0$ ) and cumulative DMI ( $DMI_c$ ). The FO equation was  $BW_f = BW_0 e^{k DMI_c}$ . The RFI model was improperly specified ( $P < 0.10$ ), predicted ADFI, both OLS and BS, differed from observed ( $P < 0.05$ ), OLS and BS parameter estimates within and among data sets were not unique ( $P < 0.05$ ) and the model failed all tests of external validity ( $P < 0.05$ ). Functional forms of the variables were inconsistent with published classical energetic relationships among ADFI, BW and ADG. The intercept term,  $a$ , was either 0, negative or positive. The latter are inconsistent with biology; when BW and ADG are 0,  $a$  must likewise be 0. Parameter estimate  $b_1$  indicated that ME intake at  $NE_g = 0$  ( $ME_m$ ) was either  $0.280 \times BW^{0.75}$  or  $0.165 \times BW^{0.75}$ ; both of these are greater ( $P < 0.05$ ) than the NAS/NRC estimate of  $0.131 \times BW^{0.75}$ . Parameter estimate  $b_2$  indicated that efficiency of ME utilization for gain was either outside or within the range of biological possibility. The FO model was internally and externally invalid for both data sets ( $P < 0.05$ ) although

$R^2$  were  $> 0.90$ . It appears that initial and final BW, as state variables, improve model fit in describing the relationship among ADFI,  $BW_0$  and  $BW_f$ . Improper specification for the RFI model, parametric instability, lack of internal and external validity, irreproducibility of estimators between data sets and among published studies, and the random nature of residuals indicate the linear RFI model is a poor predictor of ADFI and of animal efficiency, for the data sets examined. A first order function (nonlinear) better describes relationships between BW change and ADFI. However, regardless of model, BW change and ADFI may be inadequate descriptors of efficiency.

**Key Words:** efficiency, beef cattle, residual feed intake

**W54 The effect of selection using residual average daily gain EPD on growth and reproductive performance in Angus heifers.** J. B. Wells\*, J. R. Segers, J. Duggin, J. K. Bertrand, R. Rekaya, and T. D. Pringle, *University of Georgia, Athens, GA.*

Forty-five heifers were produced at the Northwest Georgia Research and Education Center in Calhoun from the random mating of commercial Angus cows to Angus bulls selected for high and low residual ADG (RADG) EPD and high and average marbling EPD. The experimental design was a  $2 \times 2$  factorial design with 2 Angus sires represented in each of the following treatments: high RADG, high marbling (Hi/Hi); high RADG, average marbling (Hi/Avg); low RADG, high marbling (Lo/Hi); and low RADG, average marbling (Lo/Avg). Heifers from the matings were studied to determine the effects of selection for RADG and marbling on growth and reproductive performance. Heifer weights were recorded and composition was determined via ultrasound scanning at 229 d (weaning) and 391 d (yearling) of age. There were no differences observed in weaning and yearling weights between the groups ( $P > 0.19$ ). Weaning 12th rib fat levels were different with low RADG animals having greater fat depth ( $P = 0.01$ ) than high RADG. Interactions between RADG and marbling were present in yearling 12th rib fat ( $P = 0.04$ ), weaning IMF ( $P = 0.07$ ), and yearling IMF ( $P = 0.08$ ) with Lo/Avg heifers having greater values than Hi/Avg heifers. Blood samples were drawn at 8 mo, 10 mo, and 12 mo of age to directly quantify the level of progesterone present using the Siemens Coat-A-Count Progesterone RIA procedure. At 10 mo of age, 26% of low RADG heifers had reached puberty while no high RADG heifers had reached puberty. By 12 mo of age, 37% of low RADG and 19% of high RADG heifers had reached puberty. At approximately 14 mo of age, heifers were synchronized and artificially inseminated using the 14-d CIDR-PG & TAI protocol. Heifers that returned to estrus within 30 d were bred a second time by AI before exposure to a bull. Overall AI conception rates were Hi/Hi: 82%, Hi/Avg: 53%, Low/Hi: 89%, and Low/Avg: 80%. The average age at calving (714 d) was not different between groups ( $P = 0.73$ ). In conclusion, selection using RADG may negatively effect early reproductive performance in Angus heifers, however, further work is needed to fully characterize the potential for this trait as a selection tool in beef production systems.

**Key Words:** residual ADG, heifer, reproduction

**W55 Evaluation of serum 25-hydroxyvitamin D concentrations of beef calves during the spring and summer seasons.** Jessica L. Powell\*<sup>1</sup>, Deborah M. Price<sup>1</sup>, Matthew J. Hersom<sup>1</sup>, Joel V. Yelich<sup>1</sup>, G. Allen Bridges<sup>2</sup>, Scott Bird<sup>2</sup>, Mary E. Drewnoski<sup>3</sup>, and Wade A. Sutton<sup>4</sup>, <sup>1</sup>University of Florida, Gainesville, FL, <sup>2</sup>University of Minnesota, Grand Rapids, MN, <sup>3</sup>University of Nebraska, Lincoln, NE, <sup>4</sup>University of Idaho, Moscow, ID.

Vitamin D is known to be critical for the growth and development of young calves and contributes to activation of innate immune defenses of cattle. The 25-hydroxyvitamin D (25D) metabolite is the precursor to the active vitamin D hormone and its concentration in serum serves as an indicator of an animal's vitamin D status. Normal serum 25D concentrations originally described for cattle are between 20 and 50 ng/mL, but recent evidence has indicated that serum 25D concentrations below 30 ng/mL are insufficient for adequate health. Because little information is available regarding vitamin D status of beef calves, the objective of this study was to evaluate serum 25D concentrations of beef calves born during the spring calving season. Four separate cohorts of calves were studied where serum samples were collected at birth and again 3–4 weeks after birth or in mid to late summer. Serum 25D concentrations were determined by using a 25D ELISA. In the first cohort, the average ( $\pm$ SD) concentrations of 16 Angus and Brangus calves born in Florida were  $25.6 \pm 12.1$  ng/mL at birth in February and  $26 \pm 10.3$ ,  $53.8 \pm 12.9$ , and  $59.5 \pm 17.2$  ng/mL in March, July, and September, respectively. In the second cohort, the average serum 25D concentrations of 12 Angus and Brangus calves born in Florida were  $16.4 \pm 11.1$  ng/mL at birth in April and  $29.4 \pm 9.7$ ,  $43.8 \pm 11.8$ , and  $51.0 \pm 12.6$  ng/mL in June, July, and September, respectively. In the third cohort, the average serum concentrations of 20 Angus calves born in Minnesota were  $13.2 \pm 6.4$  ng/mL at birth in April and  $45.9 \pm 7.6$  ng/mL in July. In the fourth cohort, the average serum 25D concentrations of 15 Charolais calves born from January to April in Idaho were  $3.0 \pm 2.2$  ng/mL at birth and  $10.5 \pm 7.7$  ng/mL at 3 weeks of age. Averages of the serum 25D concentrations for the dams of the calves in cohorts 2, 3, and 4 at calving were  $69.5 \pm 16.0$ ,  $57.7 \pm 8.2$ , and  $53 \pm 8.2$  ng/mL, respectively. In summary, this study indicates that vitamin D insufficiency (serum 25D < 30 ng/mL) is prevalent in newborn beef calves, and that potential opportunities exist to improve health of newborn calves through vitamin D supplementation.

**Key Words:** beef calves, vitamin D, health

**W56 Equations to predict chemical body composition in Nellore cattle.** A. M. Castilhos<sup>1</sup>, R. H. Branco<sup>2</sup>, C. L. Francisco<sup>\*1</sup>, M. E. Z. Mercadante<sup>2</sup>, S. F. M. Bonilha<sup>2</sup>, C. M. Pariz<sup>1</sup>, M. B. Silva<sup>1</sup>, and A. M. Jorge<sup>1</sup>, <sup>1</sup>Universidade Estadual Paulista-FMVZ, Botucatu, SP, Brazil, <sup>2</sup>Centro APTA Bovinos de Corte, Instituto de Zootecnia, Sertãozinho, SP, Brazil.

Multiple linear regressions equations were developed to predict the chemical composition of the empty body of Nellore cattle. Thirty-three young bulls (Control Nellore,  $n = 11$ ; Selected Nellore,  $n = 22$ ) of  $339 \pm 35$  kg initial weight and  $448 \pm 26$  d of age were used. The characteristics used to develop the prediction models were: empty body weight (EBW), age (days), hip height (HH), and ultrasound measurements [backfat thickness (uBF), rump fat (RF) and longissimus muscle area (uLMA)]. Ultrasound measurements were collected at the end of the trial period (d 110). Chemical composition (protein, ether extract, water and ash) was performed on samples obtained after grinding and homogenization of all body tissues. No interaction ( $P > 0.05$ ) was observed between chemical components and genetic groups. Mallows Cp values were close to the ideal value of number of independent variables in the prediction equations plus one. Precise predictions ( $R^2 > 0.72$ ) were obtained for protein, ether extract, and water, whereas poor prediction for ash ( $R^2 = 0.67$ ) was revealed. Protein, ether extract, and water percentage can be calculated using the following equations: % Protein =  $52.011 - 1.415 \times RF + 0.179 \times EBW - 32.122 \times HH$  ( $R^2 = 0.91$ ; SE = 2.24); % Ether Extract =  $11.300 + 2.096 \times RF + 0.157 \times EBW - 0.061 \times \text{age}$  ( $R^2 = 0.72$ ; SE = 4.47); % Water =  $-34.156 - 2.327 \times RF + 0.550 \times EBW + 0.103 \times \text{age}$  ( $R^2 = 0.96$ ; SE = 5.81). In conclusion, multiple linear regres-

sions were detected between non-invasive measurements and chemical composition of the empty body. The equations developed can be used to estimate chemical body composition characteristics as protein, ether extract and water body percentages of Nellore cattle. Supported by FAPESP Process#2005/60042–2.

**Key Words:** empty body, equation, Nellore cattle

**W57 Effect of maternal body weight gain during mid-gestation on progeny skeletal muscle microRNA.** J. C. McCann\*, T. B. Wilson, D. W. Shike, and J. J. Looor, *University of Illinois at Urbana-Champaign, Urbana, IL.*

The objectives were to investigate the effects of maternal body gain during mid-gestation on skeletal muscle microRNAs expressed in the progeny. Post-transcriptionally, microRNAs downregulate gene expression by degrading target mRNA or translational repression. Spring-calving, multiparous cows ( $n = 26$ ) were pen fed a diet consisting of 52% corn silage, 24% soy hulls, and 24% alfalfa haylage for 82 d during mid-gestation. Cows were classified into 3 groups (Hi-gain, Med-gain, and Low-gain) based on BW gain during the feeding period. After calving, all calves were managed similarly as a single contemporary group. Longissimus muscles biopsies were taken on 99, 197, and 392 d of age. Quantitative RT-PCR was used to determine mature microRNA in LM with all reactions run in triplicate. Three reference microRNAs (let-7a, miR-191a, and miR-103) were selected due to stable expression across animals and times. Data were analyzed using the MIXED procedure of SAS with cow BW gain, biopsy time, sex, and sire as fixed effects and pen as the random effect. Pairwise comparisons were implemented to separate means. MiR-381 expression in progeny born to Med-gain BW gain cows was greater ( $P = 0.01$ ) than Low-gain with Hi-gain being intermediate. A cow BW gain  $\times$  time interaction was observed for miR-181a ( $P = 0.02$ ); while all groups decreased over time, calves from Hi-gain dams were greatest at d 392. A tendency ( $P = 0.08$ ) for a cow BW gain  $\times$  time interaction was observed for miR-26a. Despite all groups increasing from d 99 to 197, only the Hi-gain group maintained a similar level of miR-26a at d 392 while Med-gain and Low-gain were downregulated. Relative abundance of microRNAs was greatest for known muscle-specific microRNAs, miR-1 and -133a. All measured microRNAs changed ( $P < 0.01$ ) over time. Expression of miR-23a and miR-29a increased with age, while miR-376d and miR-381 decreased at each subsequent time point. Additionally, expression of miR-133a and let-7g was greatest at d 197. Overall, results suggest microRNAs have a role in the robust and coordinated regulation of skeletal muscle development and some may be regulated by epigenetic factors.

**Key Words:** fetal programming, microRNA, muscle

**W58 Physical and chemical analysis of the longissimus thoracis muscle of Nellore cattle selected for production.** Jessica Moraes Malheiros<sup>\*1</sup>, Rogério Abdallah Curi<sup>2</sup>, Josineudson Augusto Vasconcelos Silva<sup>2</sup>, Henrique Nunes de Oliveira<sup>1</sup>, Lúcia Galvão Albuquerque<sup>1</sup>, and Luis Artur Loyola Chardulo<sup>2</sup>, <sup>1</sup>College of Agriculture and Veterinary Science, College of Agriculture and Veterinary Science, São Paulo State University (UNESP), Jaboticabal, São Paulo, Brazil, <sup>2</sup>College of Veterinary and Animal Science, College of Veterinary and Animal Science, São Paulo State University (UNESP), Botucatu, São Paulo, Brazil.

Tenderness is one of the main characteristics in the producer and consumer market. Identifying and selecting beef cattle that have genetic propensity for quality meat production is necessary for zebu animals.

The aim of this study has been to evaluate the qualitative aspects of meat in *Longissimus thoracis* muscle of 2 selected contrasting groups of 718 Nellore animals finished in feedlot and from herds that are part of breeding programs. Samples were collected between the 12th and 13th ribs to perform the physical-chemical analysis of shear force (SF), rib eye area (REA), backfat thickness (BFT), visual scale of marbling scores (marbling) and cooking losses (CI). The contrasting groups were selected by SF: animals with tender meat (n = 15) and animals with tough meat (n = 15). Statistical analyzes were performed using the GLM procedure, CORR and the averages were compared using the Tukey test. The values found in the groups of animals with tender meat and animals with tough meat were, respectively to SF (3.8 ± 1.17 and 6.8 ± 1.4 kg), REA (74.8 ± 8.52 and 62.6 ± 6.55 cm<sup>2</sup>) and CI (24.2 ± 3.02 and 28.8 ± 3.81%), showed significant differences (P < 0.01). REA has a good relationship with the amount of this muscle in the carcass and thus contributes to the prediction of meat cuts and product yield. Meat with SF values below 5.5 kg may be considered potentially tender. The difference to REA between previously selected groups for meat tenderness establishes the possibility of obtaining Nellore more efficiently in production, coupled with achieving superior products. Samples of tough group showed higher values for CI, impairing the tenderness. The BFT values (3.4 ± 1.11 and 3.6 ± 0.73 mm) and marbling (2.5 ± 0.24 and 2.5 ± 0.36) for groups of tender and tough meat respectively showed no significant difference (P > 0.05). Therefore, in animals of Nellore breed for production the ability to produce meat observed through REA, BFT and marbling, has to be satisfactory. However, the wide variation in SF of the animals selected in breeding programs, sorts the final product with lower added value on meat quality characteristics.

**Key Words:** quality beef cattle, rib eye area, shear force

**W59 Characterization and identification of MyHC and their association with quality beef cattle Nellore.** Jessica Moraes Malheiros\*<sup>1</sup>, Ivan José Vechetti-Junior<sup>2</sup>, Maéli Dal-Pai-Silva<sup>2</sup>, Henrique Nunes de Oliveira<sup>1</sup>, Lúcia Galvão Albuquerque<sup>1</sup>, and Luis Artur Loyola Chardulo<sup>3</sup>, <sup>1</sup>College of Agriculture and Veterinary Science, College of Agriculture and Veterinary Science, São Paulo State University (UNESP), Jaboticabal, São Paulo, Brazil, <sup>2</sup>Institute of Biosciences, Institute of Biosciences, São Paulo State University (UNESP), Botucatu, São Paulo, Brazil, <sup>3</sup>College of Veterinary and Animal Science, College of Veterinary and Animal Science, São Paulo State University (UNESP), Botucatu, São Paulo, Brazil.

The study of myosin protein is directly related to the tenderness, growth, development and muscle contraction, and expresses different isoforms, where the way it operates has great influence on meat quality. This study aimed to relate the Nellore meat (*Bos indicus*) separating 2 contrasting groups through the identification and quantification of heavy chain isoforms of myosin (I, Ila, IIX and IIb). A total of 718 samples of the *Longissimus thoracis* from animals slaughtered in commercial abattoir were used in the shear force analysis (SF) to determine tenderness. According to SF were selected 2 groups: (1) Group of tender meat (n = 15) and (2) Group of tough meat (n = 15) who underwent extraction, identification and quantification of heavy chain isoforms myosin (MyHC) by electrophoresis (SDS-PAGE) and photo documentation of gels. Statistical analyzes were performed using the GLM procedure, CORR and averages were compared using the Tukey test using Statistical Analysis System. For MyHC was identified and quantified 3 isoforms (I, Ila and IIX), the presence of MyHC IIb isoform has not been identified. Mean values of the optical density index (IOD) in relative percentage of MyHC isoforms were observed in the tender meat group and tough respectively MyHC-I (18.2 ± 3.54 and 16.9 ± 3.14), MyHC-IIa (16.7

± 3.09 and 14.0 ± 3.38) and MyHC-IIX (64.9 ± 3.79 and 66.3 ± 5.05). It can be seen a greater amount of type II isoform (fast) compared with type I (slow). There were no differences (P > 0.05) between the extreme groups of type I and IIX, unlike Ila (P < 0.05), with the highest relative percentage in tender meat group and reported a significant negative association with SF (rp = -0.34; P < 0.05). It is concluded from the results that the Nellore have good relationship between the hypertrophy of the *Longissimus thoracis*, as measured by the concentration of MyHC Ila, and meat tenderness, measured by the SF. The study of MyHC isoforms is an appropriate type of muscle fibers identification tool having a good correlation with the meat tenderness feature.

**Key Words:** quality beef cattle, shear force, skeletal muscle

**W60 Effects of pre- and post-breeding supplementation of a ruminally protected lipid on subsequent beef cow performance.**

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We determined the effects of supplementation of a ruminally protected lipid (Megalac-R, Church & Dwight Co., Princeton, NJ) on subsequent beef cow reproductive performance. Sixty primiparous cows (428 ± 42 kg of BW) were used in a completely randomized design. Cows were stratified by breed, BCS and the previous year's calving date, and assigned to one of 2 treatments: control (CTRL; 1.36 kg·d<sup>-1</sup> of corn gluten feed) and treatment (MLAC; 1.36 kg·d<sup>-1</sup> of corn gluten feed and 0.23 kg·d<sup>-1</sup> of Megalac-R). Cows grazed a mixed winter forage pasture of rye (*Secale cereale*) and ryegrass (*Lolium multiflorum*) and had ad libitum access to water. Supplementation of CTRL and MLAC occurred from 30 d before AI until 7 d post-AI. Daily, cows were individually penned and received their respective supplementation. Cow BW and BCS were recorded 35 and 28 d before AI and 35 and 60 d after AI. At initiation of the breeding season cows were exposed to the 7-d CO-Synch+CIDR ovulation synchronization protocol. Pregnancy was diagnosed on d 30, 60, and 90 after AI. Cows pregnant to AI were monitored until calving to determine length of gestation and calf birth weight. Treatment did not affect (P > 0.05) mean cow BW (491 ± 2.9 kg) or change in cow BW. There was no effect (P > 0.05) of treatment on pregnancy rates to AI (53 ± 0.5%) or at 90 d after AI (94.5 ± 0.5%). Length of gestation was greater (P = 0.0234) for MLAC (285 ± 1.3 d) than for CTRL (281 ± 1.1 d). No effect of treatment was observed for calf birth weight (34 ± 3 kg; P > 0.05). Supplementation of 0.23 kg·d<sup>-1</sup> of a ruminally protected lipid to primiparous cows did not enhance subsequent cow reproductive performance.

**Key Words:** ruminally protected, supplementation, beef cow

**W61 Effects of prepartum supplementation of a rumen fermentation enhancer on subsequent beef cow performance.**

Darren D. Henry\*<sup>1</sup>, Francine M. Ciriaco<sup>1</sup>, Vitor R. G. Mercadante<sup>1</sup>, Danilo D. Demeterco<sup>1</sup>, Pedro L. P. Fontes<sup>1</sup>, Elliot Block<sup>2</sup>, Neil Michael<sup>2</sup>, Nicolas DiLorenzo<sup>1</sup>, and G. Cliff Lamb<sup>1</sup>, <sup>1</sup>North Florida Research and Education Center, University of Florida, Marianna, FL, <sup>2</sup>Arm & Hammer Animal Nutrition, Princeton, NJ.

We determined the effects of a prepartum negative dietary cation-anion difference (DCAD) supplement on subsequent performance of beef cows. Forty-three multiparous cows (568 ± 73 kg of BW) were used in a completely randomized design. Prepartum cows were stratified



by breed, BCS, and the previous year's calving date and assigned to one of 2 treatments: control (CTRL; 1.75 kg·d<sup>-1</sup> of 50:50 corn gluten feed (CGF):soybean meal (SBM) mixture, DM basis) and treatment (BCLR; 0.57 kg·d<sup>-1</sup> of an anion source [Bio-Chlor, Church & Dwight Co., Princeton, NJ] and 1.13 kg·d<sup>-1</sup> of 50:50 CGF:SBM mixture, DM basis). Cows had ad libitum access to bermudagrass hay (*Cynodon dactylon*) and water. Daily, cows were individually penned and received supplement (CTRL and BCLR were fed for 21.8 ± 9.3 and 24 ± 9.5 d, respectively) until calving. After calving, cows and calves were weighed within 12 h of parturition and blood samples were collected from the cow. Weekly, blood samples were collected from cows, and BW of cows and calves and BCS of cows were recorded. On d 28, 84, and 140 postpartum, milk yield was recorded and milk samples were analyzed to determine energy corrected milk (ECM). Calculated DCAD of CTRL was 469.61 mEq·kg<sup>-1</sup> DM whereas BCLR was -1295.89 mEq·kg<sup>-1</sup>. Cow weight and calf weight was not affected by treatment ( $P = 0.1154$  and  $P = 0.9598$ , respectively). The ADG of cows ( $0.1 \pm 0.043$  kg), ADG of calves ( $1.0 \pm 0.04$  kg), and BCS of cows ( $4.97 \pm 0.05$ ; 1 to 9 scale) was not different between treatments ( $P > 0.05$ ). Similarly, ECM did not differ between CTRL and BCLR ( $P > 0.05$ ). Supplemental DMI was greater ( $P < 0.001$ ) for CTRL ( $1.60 \pm 0.031$  kg·d<sup>-1</sup>) than for BCLR ( $1.23 \pm 0.033$  kg·d<sup>-1</sup>). Concentrations of plasma Ca ( $10.67 \pm 0.51$  mg·dL<sup>-1</sup>) and  $\beta$ -hydroxybutyrate ( $173.0 \pm 9.18$  mmol·L<sup>-1</sup>) did not differ ( $P > 0.05$ ) between treatments. Prepartum supplementation of 0.57 kg·d<sup>-1</sup> of an anion source to beef cows did not enhance subsequent cow or calf performance.

**Key Words:** dietary cation-anion difference, prepartum diet, supplementation

**W62 Performance effects related to administration of long-acting eprinomectin or oxfendazole near calving in spring-calving cows over a 230-d grazing period.** Elizabeth A. Backes<sup>\*1</sup>, Jeremy G. Powell<sup>1</sup>, Donald S. Hubbell III<sup>2</sup>, John D. Tucker<sup>2</sup>, William L. Galyen<sup>1</sup>, and Laura R. Meyer<sup>1</sup>, <sup>1</sup>Department of Animal Science, Division of Agriculture, University of Arkansas, Fayetteville, AR, <sup>2</sup>Livestock and Forestry Research Station, Batesville, AR.

Internal parasites have been reported to flourish in Southern states and cause detrimental economic effects to the beef cattle industry. The objective was to evaluate the effects of anthelmintic therapy administered at recommended dose to spring-calving cows approximately 5 d before the initiation of the calving season. Eighty-two ( $563 \pm 8.1$  kg) cows located at the Livestock and Forestry Research Station in Batesville, AR, were stratified by BW, BCS, and fecal egg counts (FEC), then allocated randomly to 1 of 3 treatments representing: Control (CON; no anthelmintic administered;  $n = 27$ ); Oxfendazole (OXF;  $n = 27$ ); or Long-acting eprinomectin (LAE;  $n = 28$ ). Cows were rotationally grazed within individual treatment on 2.4-ha mixed grass pastures. Body weight, BCS, FEC were taken on d 0, 14, 91, 154, and 230 and hair coat scores (HCS) taken on d 0, 91, 154. Beginning May 15, 2015, one bull was placed in each pasture for a 60 d breeding season. Performance data were analyzed using PROC MIXED of SAS, with individual cow being the experimental unit and conception rates were analyzed using Chi-squared. Body weight on any sampling date (d 230 BW 568, 573, 574 kg for CON, LAE, OXF, respectively), BCS on d 0, 14, 154, and 230, and HCS on d 0 and 154 did not differ ( $P \geq 0.25$ ) across treatments; however, BCS were greatest ( $P = 0.03$ ) for OXF (6.0), CON intermediate (5.9), and lowest for LAE (5.8) on d 91, and HCS were greater ( $P < 0.01$ ) for LAE (3.5) versus OXF (1.7) and CON (2.8) on d 91. Similar ( $P \geq 0.16$ ) overall ADG were reported (0.00, 0.08, and 0.05 kg/d for CON, LAE, OXF, respectively). Cow FEC did not differ ( $P \geq 0.11$ ) on

d 0 or 91; however, were greater for CON on d 14 and 154 versus OXF and LAE; were greater ( $P = 0.02$ ) for LAE versus CON and OXF on d 230. Conception rates did not differ ( $P = 0.17$ ) across treatments (78, 61, 81%, for CON, LAE, OXF, respectively). In this study, anthelmintic therapy did not improve BW, BCS, HCS, FEC, or conception rate, in spring-calving cows rotationally grazed over a 230-d grazing period.

**Key Words:** fecal egg counts, spring-calving cows, anthelmintic

**W63 The effects of weight gain, carcass characteristics, temperament, and Brahman influence on estrus behavior and conception rate in heifers.** Brittney L. Ivey<sup>\*</sup>, Allison C. Vail, William A. Storer, and C. Edward Ferguson, *McNeese State University, Lake Charles, LA.*

Heifers from southwest Louisiana were dry-lotted and evaluated for factors influencing estrus and conception. In October, spring born heifer calves ( $n = 121$ ,  $259 \pm 150$  kg) were received and processed with routine procedures before beginning a 120 d feeding trial. Heifers were visually assessed for temperament and percentage of Brahman influence. Temperament was evaluated as individual chute scores (CS) and denoted as 1–4 increasing with increased heifer excitability. Temperament was also evaluated as chute exit velocity (EV) using a rodeo event timer to calculate the speed of each animal exiting the squeeze chute. Brahman influence (BI) was determined by the phenotype and/or genotype of the heifers. Criteria for phenotype assessment were ear length and shape, hair type, coat color, and length of sheath. Individual BW and ADG were assessed monthly. Heifers were scanned by ultrasound at d 120 to determine RF, REA, and IMF. Intramuscular fat and RF decreased ( $P < 0.05$ ) in heifers with greater BI. Cattle that were greater than 25% BI had lower ( $P < 0.05$ ) ADG and final BW than did heifers with 0 to 25% BI. During 2 rounds of estrus detection, females were fitted with electronic estrus detection transmitters and monitored for estrus behavior. In round 1 ( $n = 121$ ), BI did not make a difference ( $P > 0.1$ ) in the duration of estrus or the estrous cycle length. Temperament, as assessed by CS and EV, did not influence ( $P > 0.1$ ) duration of estrus or estrus cycle length. In the second round of estrus detection ( $n = 59$ ), there was no relationship ( $P > 0.1$ ) between conception and estrous cycle length, duration of estrus, RF, REA, ADG, or BW. Temperament and breed also did not affect ( $P > 0.1$ ) conception rate. This aided in clarifying the relationships of weight gain, carcass characteristics, temperament and BI on estrus behavior and conception rates in heifers.

**Key Words:** temperament, Brahman, estrus

**W64 Effect of breed on the sale price of beef calves sold through video auctions from 2010 through 2014.** Michael E. King<sup>1</sup>, Kevin L. Hill<sup>2</sup>, Glenn M. Rogers<sup>3</sup>, and Kenneth G. Odde<sup>\*1</sup>, <sup>1</sup>Kansas State University, Manhattan, KS, <sup>2</sup>Merck Animal Health, Kaysville, UT, <sup>3</sup>Grassy Ridge Consulting, Aledo, TX.

The objective of this study was to quantify the effect of breed description on the sale price of beef calves marketed through video auctions while adjusting for all other factors that significantly influenced price. Information on descriptive characteristics of lots of beef calves were obtained from a livestock video auction service. Data were available on 33,811 lots of calves (3,345,826 total calves) that sold in 116 video auctions between 2010 and 2014. All lot characteristics that could be accurately quantified or categorized were used to develop a multiple regression model that evaluated the effects of independent factors on sale price using a backward selection procedure. A value of  $P < 0.05$  was used to maintain a factor in the final model. Breed description of

the calves in the lot was 1 of 18 factors included in the original model and was categorized into 5 groups: English, English crosses (EX), English-Continental crosses (ECX), Black Angus sired out of dams with no Brahman influence (AN), Red Angus sired out of dams with no Brahman influence (AR), and Brahman influenced (BR). Breed of the lot affected the sale price of beef calves ( $P < 0.0001$ ), and was 1 of 15 independent factors that remained in the final model. The number of lots in each breed group was 3,521, 9,022, 10,744, 1,373, and 9,151 for the EX, ECX, AN, AR, and BR breed groups, respectively. Lots of AR calves sold for a higher price ( $P < 0.01$ , \$366.32/100 kg) than all other breed groups. Black Angus sired calves had the second highest sale price at \$362.09/100 kg, and this price was higher ( $P < 0.01$ ) compared with the EX (\$359.04/100 kg), the ECX (\$358.36/100 kg), and the BR (\$350.05/100 kg) groups. The EX and the ECX breed groups sold for similar prices ( $P = 0.55$ ), but both of these breed groups sold for higher prices ( $P < 0.01$ ) than the BR lots. Breed composition of lots of beef calves selling through video auctions significantly influenced sale price.

**Key Words:** beef calves, breed, sale price

**W65 Economic aspects of rebreeding non-pregnant cows.** Aline Gomes da Silva\*<sup>1,2</sup> and Rick N. Funston<sup>1</sup>, <sup>1</sup>*University of Nebraska-Lincoln, West Central Research and Extension Center, North Platte, NE*, <sup>2</sup>*Universidade Federal de Viçosa, Viçosa, MG, Brazil*.

A study was conducted to evaluate the economic aspects of retaining ownership and rebreeding open spring-calving cows to be sold as pregnant fall-calving cows. Composite Red Angus × Simmental females diagnosed as non-pregnant after regular spring breeding season were

utilized over a 2-yr period (Yr 1, n = 61; Yr 2, n = 72). Hay and supplement were fed from November to February. Cows diagnosed as non-pregnant after a second breeding season were sold in March. Pregnant cows grazed Sandhills meadow pastures until April, when they were sold. Cows were synchronized with a 7-d controlled internal drug release (CIDR)-PG protocol before a 60 d natural service breeding season beginning in November, utilizing a 1:25 bull to cow ratio. Pregnancy diagnosis was determined by ultrasound 30 d after bull removal. A partial budget analysis was performed for Yr 1 to compare the economics of selling non-pregnant cows immediately after pregnancy diagnosis or retaining ownership and rebreeding them to sell as pregnant cows in more favorable market prices. Total cost was calculated by adding the purchase price (cull cow value at first pregnancy diagnosis), feeding costs, meadow grazing and management cost, breeding cost, and 6% annual interest rate on the purchase price. The net cost of 1 pregnant cow was calculated as the difference between total cost and cull value, divided by the number of pregnant cows. The overall rebreeding pregnancy rate was 90.2% for Yr 1 and 81.9% for Yr 2, the percentage of the pregnant cows that conceived in the first 21 d of the breeding season was 89.1% for Yr 1 and 79.7% for Yr 2. The total cost/female was \$1,186.38. Subtracting the cull value of the open cows sold in March, the net cost of one pregnant cow was \$1,185.08. The pregnant cows were sold for \$1,638.00, resulting in a \$452.92 net gain/pregnant cow. While conventional wisdom has held open cows should be sold after pregnancy detection, we conclude rebreeding a non-pregnant cow to be sold at higher market prices may be an economic alternative.

**Key Words:** budget analysis, cull cow, rebreeding

# Breeding and Genetics: Genomic methods and application—Beef

**W66 GWAS between single nucleotide polymorphisms with beef fatty acid profile in Nellore cattle using the single-step procedure.** Marcos V. A. Lemos\*<sup>1</sup>, Hermenegildo L. J. Chiaia<sup>1</sup>, Mariana P. Berton<sup>1</sup>, Fabiele L. B. Feitosa<sup>1</sup>, Carolyn Aboujaoude<sup>1</sup>, Adrielle M. Ferrinho<sup>2</sup>, Lenise F. Mueller<sup>2</sup>, Joyce J. M. Furlan<sup>2</sup>, Angelica S. C. Pereira<sup>2</sup>, Lucia G. Albuquerque<sup>1</sup>, and Fernando Baldi<sup>1</sup>, <sup>1</sup>State University of São Paulo, Jaboticabal, São Paulo, Brazil, <sup>2</sup>University of São Paulo, Pirassununga, São Paulo, Brazil.

The aim of this study was to determine genomic regions associated with the profile of beef fatty acid (FA) of Nellore cattle finished in feedlot using the single-step method. A total of 1,616 genotypes and 963 phenotypes were used. The FA profile was analyzed in *Longissimus thoracis* samples using a gas chromatography, with a 100-m capillary column. The following fatty acids were analyzed: lauric (C12:0), palmitic (C16:0), stearic (C18:0), oleic (C18:1 *cis*-9), linoleic (C18:2 *cis*-6), CLA (C18:2 *cis*-9 *trans*-11), CLA (C18:2 *trans*10 *cis*12), linolenic (C18:3 n3), myristic (C14:0), myristoleic (C14:1), docosahexaenoic (C22:6 n3), elaidic (C18:1 n9t), vaccenic (C18:1 t11), arachidonic (C20:4 n-6) eicosatrienoic (C20:3 n6 *cis*-8,11,14) and alpha-linolenic (C18:3 n6). The animals were genotyped with the BovineSNP BeadChip (High-Density Bovine BeadChip). After quality control of genotypes, a total of 470,000 SNPs and 1,556 samples remained. The model used for the (co)variance and genetic parameter estimation included the random genetic additive direct effect, the fixed effect of the contemporary groups, and the animal's slaughter age as a covariable. To determine the areas of QTL, segments that were  $\geq 1\%$  of the additive genetic variance were chosen. For identification and positioning of these segments, the database available in the "National Center for Biotechnology Information" and Ensembl Genome Browser were used. A total of 115 genomic regions (1-Mb SNP windows) associated with the FA profile were identified; many of these regions were previously detected in other cattle breeds, like the gene *ELOVL5* (fatty acid elongase 5) associated with the C20:4 n-6 FA, the *ESRRG* (estrogen receptor-related gamma gene) was associated with the C12:0 FA and the *PCYT1A*, *TCTEX1D* and *GALNTL6* were associated with C18:2 *cis*-9 *cis*-12 n-6, C14:0 and C16:0 FA. The genes present in these regions may help to explain the genetic basis of FA profile in Bos indicus cattle, contributing to better selection of these traits associated with improvement of human health.

**Key Words:** *Bos indicus*, fatty acid composition, genetic markers

**W67 Genotype imputation and haplotype-phase inference using trio based reference panel in Hanwoo (Korean cattle).** Dajeong Lim\*, Jung-Woo Choi, Hyung-Chul Kim, Han-Ha Chai, and Yong-Min Cho, National Institute of Animal Science, Suwon, South Korea.

In recent years, large numbers of cattle have been genotyped with SNP arrays from 3K to 800K. These platforms can be available to increase the efficiency and accuracy of breeding programs by implementing genomic selection. As for cattle, there are currently several imputation/phasing methods used in genomic selection, genome-wide association (GWA) studies, or genetic diversity analysis. Currently, many imputation and phasing methods are introduced to reduce the number of missing genotypes and to infer the haplotypes from these genotype data. Despite these efforts, imputation or phasing errors are still present. Next-generation sequencing (NGS) price has been consistently dropped, various population genomic theories and breeding programs can be now applied to

the sequencing data obtained from population of each breed of interest. For example, long-range haplotype sequencing technology can phase 99% of single-nucleotide variants (SNVs) in sequencing data without imputation process; current technologies typically phase ~95–97% in human. Therefore, we describe the phasing study using Hanwoo trio sample. First, we selected the representative trio sample from pedigree analysis in Hanwoo population. Genotyping was performed based on the Illumina 800K. Imputations for genotype data in this study were done using BEAGLE and FIMPUTE, genotype imputation tools that use a reference panel of haplotypes to estimate phase and impute missing genotypes in trio data. Second, we sequenced the trio data using Illumina Long-read haplotyping technology known as Moleculo. The short sequence reads produced from each molecule are assembled into synthetic long-reads. These fragments assign haplotype to homologous chromosomes in the phasing application. Finally, we compared accuracy of imputation/phasing based on the SNP array and sequencing data of an optimal reference panel of maternal/paternal haplotypes. These results help in improving selection and breeding value estimation and in avoiding imputation errors from SNP information.

**Key Words:** phasing, imputation, Hanwoo

**W68 Genome-wide association study analysis for meat traits of beef cattle.** Hoyoung Chung\*, National Institute of Animal Science, Suwon, KY, Korea.

To identify genomic loci with an effect on meat quality traits in Hanwoo cattle, 3,000 animals with carcass phenotypes were genotyped with a customized 56K Affymetrix SNP chip. Genome-wide association studies (GWAS) were performed for marbling (MAR), maturity (MAT), backfat thickness (BFT), loin eye area (LEA), carcass weight (CAW), meat quality grade (MQG), and meat yield grade (MYG). Multiple statistically significant SNPs were identified for MAT (674 SNP), MAR (595), CAW (754), LEA (506), BFT (440), MYG (496), and MQG (2,850) with chromosomes 14 and 23 having extreme significant associations for CAW and MYG, respectively. A 66-bp insertion in *ADIPOQ* from 81966364 to 81966419 was genotyped by agarose gel electrophoresis in 3,000 animals to verify the associations of GWAS loci located in the *ADIPOQ* region. The *ADIPOQ* insertion was significantly associated with MAR ( $P = 0.034$ ), BFT ( $P = 0.004$ ), LEA ( $P = 0.014$ ), CAW ( $P = 0.002$ ), and MYG ( $P = 0.003$ ). This study's significant SNP may be used in marker-assisted selection programs to improve meat quality traits in beef cattle.

**Key Words:** GWAS, SNP, meat trait

**W69 Admixture analysis in Brazilian synthetic cattle.** Marcos E. Buzanskas\*<sup>1</sup>, Ricardo V. Ventura<sup>2</sup>, Tatiane C. S. Chud<sup>1</sup>, Daniel J. A. Santos<sup>1</sup>, Priscila A. Bernardes<sup>1</sup>, Thiago B. R. Silva<sup>1</sup>, Mauricio A. Mudadu<sup>3</sup>, Luciana C. A. Regitano<sup>3</sup>, Marcos V. G. Barbosa da Silva<sup>4</sup>, Changxi Li<sup>5</sup>, Flavio S. Schenkel<sup>2</sup>, Mauricio M. Alencar<sup>3</sup>, and Danísio P. Munari<sup>1</sup>, <sup>1</sup>UNESP – Univ Estadual Paulista, Faculdade de Ciências Agrárias e Veterinárias, Jaboticabal, SP, Brazil, <sup>2</sup>University of Guelph, Guelph, ON, Canada, <sup>3</sup>Embrapa Southeast Livestock, São Carlos, SP, Brazil, <sup>4</sup>Embrapa Dairy Cattle, Juiz de Fora, MG, Brazil, <sup>5</sup>University of Alberta, Edmonton, AB, Canada.

The development of synthetic breeds in Brazil from crosses between *Bos taurus indicus* (Bti) and *Bos taurus taurus* (Btt) is very useful when it



is desired to combine the fitness and carcass yield. The Canchim breed (CA), which has expected proportions of 62.5% Charolais and 37.5% zebu (Nellore breed), have been the focus of several studies because this breed has high carcass quality and adaptability to extensive production system. The aim of this study was to estimate the genetic composition in the Canchim breed using single nucleotide polymorphism (SNP) data. Canchim animals (285 individuals) were genotyped with the Illumina BovineHD BeadChip (777962 SNPs). To estimate the genetic contribution of Btt and Bti, 814 animals from the Nellore breed (NE) and 405 animals from the Charolais breed (CH) were genotyped with the Illumina BovineHD BeadChip and BovineSNP50 BeadChip (54609 SNPs), respectively. The PLINK v.1.9 software was used to combine the data, perform genotype quality control, and estimate the linkage disequilibrium ( $r^2$ ). The ADMIXTURE software was used to estimate the genetic contributions. Genotype quality control resulted in 283, 811, and 405 animals from the CA, NE, and CH breeds and 29716 SNPs. The genetic contributions of Btt and Bti in the Canchim breed were, in average, 72.5% and 27.5%, respectively. Minimum and maximum proportions of Btt and Bti ranged from 66.0% to 89.0% and 11.0% to 34.0%, respectively, in Canchim cattle. The differences between the expected proportions and the estimated proportions of Btt and Bti were due to the patterns  $r^2$ , which are greater in shorter distances (0–0.04 Mb) for CH (0.20), followed by CA (0.16), and NE (0.15). When the  $r^2$  between adjacent SNPs are high, recombination rates should be low, which may be indicative of greater contribution of CH animals in the composition of CA breed. The maximum proportion of 16.0% of Btt was observed for NE, indicating remote crossbreeding, which could have contributed to higher Btt proportion in CA animals.

**Key Words:** beef cattle, genomics, genetic structure

#### W70 Genome-wide association analysis and gene ontology enrichment of meat tenderness in Polled Nellore cattle in Brazil.

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Brazil has the largest commercial cattle herd worldwide, but its meat quality is highly variable. The national herd is largely composed of *Bos indicus* breeds, which in general have less tender meat than *Bos taurus* cattle, decreasing the product value. This study was carried out to identify genomic regions and biological relevant pathways associated with meat tenderness in Polled Nellore cattle. Data consisted of Warner-Bratzler shear force (WBSF) values of *Longissimus* muscle after 7 d of aging, from 326 Polled Nellore animals born in 3 breeding seasons (2002, 2005 and 2009) at the OB ranch, located in the State of Mato Grosso, Brazil. The animals were genotyped using either the Bovine HD Chip (777k) or the GGP-Indicus Chip (77k). The imputation from the GGP to the HD Chip was performed using FImput software. SNPs were excluded when GenCall <0.7, Call rate <0.90, EHW  $P < 0.01$  (using Bonferroni adjustment), and MAF <0.05. Due to large dispersion of sires (progenies' parents), the population stratification was controlled by the 3 first genomic principal components. Genome-wide association analysis (GWAS) was performed using the Efficient Mixed-Model Association (EMMA) method. The GWAS was complemented with a gene set enrichment analysis of Gene Ontology (GO) terms using the FatiGO procedure. The most significant markers ( $P < 0.0001$ )

were located on chromosomes 2, 3, 7, 10, 11, 17, 20, 21, 24 and 25, indicating several QTLs associated with meat tenderness throughout the genome. Additionally, 48 GO terms were deemed enriched. Several of these functional categories can be related to WBSF in Polled Nellore cattle, such as activities of ion channels, membrane cell transportation, growth factors, and protein serine/threonine phosphatase complex, which participate in processes that inactivate apoptosis components. These results help to elucidate the metabolic pathways related to this trait, which is of extreme economic and social importance to Brazil as Nellore is the dominant beef cattle breed in the country. Financial support: EMBRAPA, CNPq, CAPES.

**Key Words:** shear force, GWAS, pathway

#### W71 Genomic-polygenic and genomic predictions of direct and maternal effects for growth traits in a multibreed Angus-Brahman cattle population.

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The objectives of this research were to compare variance components, genetic parameters, and EBV rankings for birth weight (BW) direct and maternal, weaning weight (WW) direct and maternal, and postweaning gain from 205 d to 365 d (WG) direct using 3 genomic-polygenic and one polygenic model. In addition, trends in EBV were evaluated for each trait and model as Brahman fraction increased from 0% to 100%. The Angus-Brahman multibreed data set included 5,264 animals born between 1987 and 2013. Genomic-polygenic models 1 (GP1; pedigree relationships for all animals; genomic relationships for genotyped animals), 2 (GP2; pedigree relationships for non-genotyped animals only; genomic relationships for genotyped animals), and 3 (GP3; no pedigree relationships; genomic relationships for genotyped animals) used actual and imputed genotypes from 46,768 SNP markers. Variance components and genetic parameters were estimated using REML procedures. Estimates of variance components and genetic parameters from GP1 were the most similar to those from the polygenic model, followed by those from GP2, and the least similar (particularly for maternal traits) were those from GP3. Similarly, the highest rank correlations were those between animal EBV from the polygenic model and GP1 (0.98 to 0.99), followed by those from GP1 and GP2 (0.82 to 0.94) and lastly by those from the polygenic model and GP2 (0.81 to 0.94). Model GP3 performed poorly for maternal traits due to ignoring calf-dam relationships (-0.12 to 0.23). These results indicated that the polygenic model and genomic-polygenic model 1 should be preferred. High genotyping costs could still make the polygenic model preferable for commercial beef cattle operations. Brahman animals tended to have higher EBV for BW direct and WW direct, and lower EBV for WG direct, BW maternal, and WW maternal. However, low regression coefficients for EBV on Brahman fraction ensured that high, medium, and low EBV animals from all breed compositions existed for all growth traits in this multibreed population.

**Key Words:** cattle, genomic, growth

#### W72 Genomic regions associated with beef fatty acid profile in Nellore cattle.

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The objective of this study was to identify genomic regions associated with beef fatty acid profile (FA) in Nelore males finished in feedlot using the single-step method. A total of 1,616 genotypes and 963 phenotypes were used. Animals were genotyped using the high-density Bovine-HDBeadChip panel, which contains 777,962 SNP markers distributed throughout the genome. Quality control (QC) criteria were: i) SNPs with minor allele frequency  $\leq 5\%$ , call rate  $\leq 90\%$  (CR); ii) Only samples with CR  $\geq 90\%$  were considered. A total of 470,007 SNPs and 1,556 samples remained in the data set. The FA profile was measured in *Longissimus thoracis* muscle using a gas chromatography, with a 100 m capillary column. The following FA were analyzed: total saturated FA (SFA), total monounsaturated FA (MUFA), total polyunsaturated FA (PUFA), omega-3 (n-3) and omega-6 (n-6). Contemporary groups (CG) were defined as farm, year of birth, and yearling management group. Variance components estimation model included random additive direct genetic effects, fixed effects of CG, and animal's slaughter age as a covariable (linear and quadratic effect). For SFA, MUFA, PUFA, n-3 and n-6, a total of 31, 19, 40, 6 and 6 genomic regions that explain 1% or more of the additive genetic variance were found, respectively. Preliminary studies showed that many of these regions were not previously detected in other cattle breeds. Genes like *PRKARIA* (protein kinase), *ELOVL5* (fatty acid elongase 5), *FASN* (fatty acid synthase) and *SCD* (esterol-CoA desaturase) were also identified. This study found several genomic regions associated with meat fat acidity profile in Nelore cattle that can be used for further studies to contribute for the production of healthier meat. This work was supported by São Paulo Research Foundation (FAPESP) grant #2009/16118-5 and grant #2011/21241-0.

**Key Words:** fatty acid composition, molecular marker, SNP

**W73 An SNP association study evaluating Brahman and Brahman-influenced steers for growth and carcass traits.** Amanda Royer<sup>1</sup>, Chris Shivers<sup>3</sup>, David Riley<sup>4</sup>, Mauricio Elzo<sup>5</sup>, and Matthew Garcia<sup>\*1,2</sup>, <sup>1</sup>Louisiana State University School of Animal Sciences, Baton Rouge, LA, <sup>2</sup>LSU AgCenter, Baton Rouge, LA, <sup>3</sup>American Brahman Breeders Association, Houston, TX, <sup>4</sup>Department of Animal Science, Texas A&M University, College Station TX, <sup>5</sup>Department of Animal Science, University of Florida, Gainesville, FL.

Brahman cattle are important in tropical regions due to the breed's ability to tolerate excessive heat and parasite presence. However Brahman cattle exhibit lower yielding, lower quality carcasses as compared with *Bos taurus* breeds. The objective of the current study was to evaluate potential SNP associations on 4 candidate genes for growth and carcass traits in a population of Brahman and Brahman-influenced steers. A total of 42 Brahman and Brahman-influenced steers born between 2009 and 2014, at Louisiana State University Central Research Station in Baton Rouge, Louisiana, were utilized. Steers were evaluated through the American Brahman Breeders Association (ABBA) carcass evaluation project in Gonzales, Texas, for growth, feedlot performance, and carcass quality and composition traits. Growth traits measured at the Central Research Station beef unit before shipment to the feedlot include birth weight, weaning weight, and hip height. Traits measured in Gonzales, Texas, included feedyard entrance weight, harvest weight, and average daily gain. Carcass traits measured include hot carcass weight, ribeye area, marbling score, yield grade, quality grade, dressing percent, and Warner-Bratzler shear force score. A mixed model design with growth, carcass traits and individual SNP genotype fit as dependent variables and breed type, year, dam fit as independent variables was utilized to

evaluate potential SNP associations. Sire was fit as a random variable in the model. Four known candidate genes were chosen for SNP analysis based on previous association with growth and carcass traits. Candidate genes include calpastatin (CAST), calpain (CAPN), thyroglobulin (TG), and adiponectin (ADIPOQ). A total of 20 SNP were chosen for each CAST and CAPN, and a total of 30 SNP were chosen for each TG and ADIPOQ. All SNP were selected equidistantly spaced across each candidate gene. Although multiple SNP in the current study were significantly ( $P < 0.05$ ) associated with growth and carcass traits, they must first be validated in much larger and diverse populations before implementation into selection strategies.

**Key Words:** *Bos indicus*, carcass trait, growth

**W74 Major loci associated with growth traits on BTA14 in Hanwoo (Korean cattle).** Seung Hwan Lee<sup>\*1,3</sup>, Ki Yong Chung<sup>1</sup>, Cedric Gondro<sup>2</sup>, Chang Gwan Dang<sup>1</sup>, Hyeong Cheul Kim<sup>1</sup>, Sidong Kim<sup>1</sup>, and Hee Ceol Kang<sup>1</sup>, <sup>1</sup>National Institute of Animal Science, Pyeongchang, Gangwon, Korea, <sup>2</sup>University of New England, Armidale, NSW, Australia, <sup>3</sup>Chungnam National University, Daejeon, Chungnam, Korea.

Genome-wide single marker regression using Bovine 50K BeadChip was performed on growth traits from 1,012 Hanwoo steers in Hanwoo (Korean Cattle). SNPs were excluded from the analysis if they failed in over 5% of the genotypes, had median GC scores below 0.6, had GC scores under 0.6 in less than 90% of the samples, deviated in heterozygosity more than 3 standard deviations from the other SNPs and were out of Hardy-Weinberg equilibrium for a cutoff  $P$ -value of  $1E^{-5}$ . Unmapped and SNPs on sex chromosomes were also excluded. A total of 32,696 SNPs were used in this analysis. To test an association between SNP and QTL, single marker regression analysis was implemented in this study. SNP was assumed to be in LD with QTL in close proximity and the effect evaluated was additive effect (QTL allele substitution effect). The Bonferroni-corrected genome wide significant association ( $P < 1.5 \times 10^{-6}$ ) was applied to detect significant SNPs for the GWAS. The GWAS identified one major QTL for body weight at 6, 12, 18 and 23 mo ranging 23Mb to 25Mb on BTA14. The most significant SNP was Hapmap32241-BTC-054753 (24Mb,  $P = 1.8 \times 10^{-6}$ ) for BW6, Hapmap27934-BTC-065223 (25Mb,  $P = 1.2 \times 10^{-10}$ ) for BW12, BW18 and BW23 in Hanwoo. The most significant SNPs accounted for 8 to 10% of additive genetic variance, which is quite large proportion against total additive genetic variance. The Hapmap27934-BTC-065223 has 12.97 kg of allele substitution effect in body weight at 12 mo (BW12). The results revealed that growth traits was affected by major QTL with large effect and many other SNP with small effects with the normal distribution.

**Key Words:** genome-wide association study, major loci, BTA14

**W75 Identification of shared copy number variation among Spanish beef cattle.** T. B. R. da Silva<sup>\*1</sup>, A. González-Rodríguez<sup>3</sup>, E. Mouresan<sup>3</sup>, J. J. Cañas-Álvarez<sup>5</sup>, L. Varona<sup>3</sup>, D. P. Munari<sup>1</sup>, M. J. Carabaño<sup>2</sup>, C. Avilés<sup>4</sup>, P. Martínez-Cambor<sup>6</sup>, and C. Díaz<sup>2</sup>, <sup>1</sup>Univ Estadual Paulista, Jaboticabal, SP, Brazil, <sup>2</sup>Instituto Nacional de Investigación y Tecnología Agraria y Alimentaria, Madrid, MD, Spain, <sup>3</sup>Univ de Zaragoza, Zaragoza, AR, Spain, <sup>4</sup>Univ de Córdoba, Córdoba, AN, Spain, <sup>5</sup>Univ Autònoma de Barcelona, Barcelona, CT, Spain, <sup>6</sup>Univ De Valladolid, Valladolid, CL, Spain.

Copy number variations (CNV) are defined as structural variation with 1 kb or larger, and are present in the genome in several forms. Gene

expression and gene dosage can be affected by CNV, therefore it can be associated with diseases and livestock economic traits. Local Spanish breed such as Asturiana de los Valles (AV); Avileña Negro-Ibérica (ANI); Morucha (Mor); Pirenaica (Pi) and Retinta (Ret) breeds play an important role in the beef market in Spain. The aim of this paper was to identify CNVs being shared among the 5 breeds and verify the functional annotation of the genes within these CNVs. The 366 animals were genotyped with the Illumina BovineHD BeadChip, which 75 are from AV; 74 from ANI; 75 from Mor; 74 from Pi and 68 from Retinta. Each breed had 25 families composed by father-mother-offspring, exception those breeds that had not 75 individuals, which had 25 incomplete families (mother-offspring). The families within breed were selected to have greater genetic variability. The PennCnv software was applied to detect and filter the CNVs, which considers the BAF (B Allele Frequency) and LRR (Log R Ratio) signals. We used the trio calling option to identify the CNVs, which consider the pedigree information after the individual CNV calling. We found CNVs being shared among breeds, varying from 87 between Pi and Ret to 514 between AV and Mor. These results suggest evidence that the formation of these breeds may be connected to common ancestry. We found the same CNV in different copy number between breeds, which means, one CNV that is a deletion in certain breed, is an insertion in another one and some of these CNVs encompassed genes. Those genes were WC1.8, PRAME, HSFY2, OAS1X, CD163L1, BOLA-DQA2, OR4P4, OR5J2, TRAC, and they are related to immune system, MHC (Major Histocompatibility Complex) and Olfactory System. Our findings speculate that some CNVs seem to be species specific and the difference of gene copy number among breeds can influence the response to environmental and pathogens challenges.

**Key Words:** genomic, single nucleotide polymorphism, structural variant

**W76 Whole-genome resequencing analysis for identifying genome-wide SNPs and signatures of selection.** Dajeong Lim\*, Jung-Woo Choi, Bong-Hwan Choi, Won-Hyong Chung, and Seung-Soo Lee, *National Institute of Animal Science, Suwon, South Korea.*

Over the last 30 years, Hanwoo has been selectively bred to improve economically important traits. Hanwoo is currently the representative Korean native beef cattle breed, and it is believed that it shared an ancestor with a Chinese breed, Yanbian cattle, until the last century. However, these 2 breeds have experienced different selection pressures during recent decades. Here, we whole-genome sequenced 10 animals each of Hanwoo and Yanbian cattle (20 total) using the Illumina HiSeq 2000 sequencer. A total of approximately 3.12 and 3.07 billion sequence reads were mapped to the bovine reference sequence assembly (UMD 3.1) at an average of approximately 10.71- and 10.53-fold coverage for Hanwoo and Yanbian cattle, respectively. A total of 17,936,399 single nucleotide polymorphisms (SNPs) were yielded, of which 22.3% were found to be novel. By annotating the SNPs, we further retrieved numerous nonsynonymous SNPs that may be associated with traits of interest in cattle. Furthermore, we performed whole-genome screening to detect signatures of selection throughout the genome. We located several promising selective sweeps that are potentially responsible for economically important traits in cattle; the *PPP1R12A* gene is an example of a gene that potentially affects intramuscular fat content. These discoveries provide valuable genomic information regarding potential genomic markers that could predict traits of interest for breeding programs of these cattle breeds.

**Key Words:** whole-genome sequencing, Hanwoo, Yanbian

**W77 Genome-wide association on growth traits in Nellore Cattle.** Rafael M. O. Silva\*<sup>1</sup>, Daniela A. L. Lourenco<sup>2</sup>, Breno O. Fragomeni<sup>2</sup>, Luciana Takada<sup>1</sup>, Rafael Espigolan<sup>1</sup>, Maria E. Z. Mercadante<sup>3</sup>, Fernando Baldi<sup>1</sup>, Guilherme C. Venturini<sup>1</sup>, Joslaine N. S. G. Cyrillo<sup>3</sup>, and Lucia G. Albuquerque<sup>1</sup>, <sup>1</sup>Univ Est Paulista Julio de Mesquita Filho-FCAV-UNESP, Jaboticabal, SP, Brazil, <sup>2</sup>The University of Georgia, Athens, GA, <sup>3</sup>APTA Center for Beef Cattle, Animal Science Institute, Sertãozinho, SP, Brazil.

The purpose of this study was to identify genomic regions which could explain the genetic variation in growth traits in a Nellore cattle population. The data set contained 8702, 8004, 3828, and 3942 records for birth weight (BW), weaning weight (WW), one year weight (YA) and yearling weight (YW), respectively. The animals were genotyped using panels of high-density SNP (Illumina High-Density Bovine BeadChip, 700k). After genomic data quality control, 437,197 SNPs for 631, 635, 342, and 299 animals for BW, WW, YA, and YW, respectively, were also available. SNP solutions were estimated by genome-wide association study using a single-step BLUP approach (ssGWAS). Before the ssGWAS the data was analyzed by a single-step genomic BLUP. Variances were calculated for windows of 200 SNP. Fixed effects in the model included month of birth, age of dam (linear and quadratic effect), contemporary group (sex, year of birth, and pen), plus animal and maternal additive random effects. Moreover, maternal permanent environmental effects were considered as random for all traits but BW. The results showed the top 10 SNP windows for each trait explained a total of 7%, 2.5%, 1.5%, and 3.5% of variance of BW, WW, YA, and YW, respectively. For all of analyzed traits the SNP windows with greatest influences were at chromosome number 14 (BTA14). In all regions of top SNP windows many genes that have been associated with growth in beef cattle were found. Various authors have recommended caution to interpret the results. Even though many SNP windows explained part of variance of all studied traits, it does not necessarily mean those regions cause the phenotypic variation. These results suggest that there are many regions on chromosome 14 associated with growth traits in Nellore cattle. São Paulo Research Foundation (FAPESP) grant 2013/01228-5 associated to grant #2009/16118-5

**Key Words:** beef cattle, GWAS

**W78 Genome-wide association study for flight speed in Nellore cattle.** Tiago S. Valente\*, Fernando Baldi, Aline C. Sant'Anna, Lucia G. Albuquerque, and Mateus J. R. Paranhos Da Costa, *São Paulo State University, Faculty of Agriculture and Veterinary Sciences, Jaboticabal, São Paulo, Brazil.*

The aim of this study was to identify single nucleotide polymorphisms (SNPs) that are significantly associated with temperament, measured by flight speed (FS), in Nellore cattle using the single step procedure (ssGWAS). Temperament was assessed by the speed (m/s) at which each animal exited the crush after yearling weighing. Data were from 16,119 animals with phenotypes and a pedigree file with 162,644 animals. A total of 1,405 animals were genotyped with BovineHD BeadChip. Quality control was performed to exclude SNP markers with unknown genomic position, located on sex chromosomes, monomorphic, MAF < 1%, call rate < 90%, and animal call rate (with less than 90% of SNPs called). After edits, 455,374 SNPs and 1,384 genotyped animals remained. The association analyze was performed by ssGBLUP, a modification of BLUP with numerator relationship matrix  $A^{-1}$  matrix replaced by  $H^{-1}$ , that uses the GEBV to estimate the SNPs effects. Variance components and genetic parameters were estimated by Bayesian inference via Gibbs sampling using the softwares GIBBS2F90, PREGSF90 and POSTGSF90, assuming a linear animal model for FS which included



direct additive genetic and residual effects as random effects and contemporary groups as fixed effect. The effects were calculated to segments of 10 sequential SNPs and results interpreted as the percentage (%) of total genetic variance explained by each SNP window. Segments that explained 1% or more of the total genetic variation were considered as candidate region associated with FS. Ten regions were associated with FS: one on BTA1 (1:73354330–73406566, 2.07%), 2 on BTA5 (5:22596661–22604723, 3.04% and 5:119291684–119306475, 1.44%, respectively), one on BTA9 (9:98759214–98767952, 3.33%), BTA11 (11:67385287–67404876, 1.39%), BTA15 (15:16598639–16662233,

1.45%), BTA17 (17:639678–671693, 4.62%), BTA18 (18:34146668–34168795, 1.22%) BTA22 (22:32886184–32904212, 1.08%) and BTA26 (26:47061401–47095621, 1.86%). This result confirms the polygenic architecture related to expression of cattle temperament, resulting from the influence of numerous genes interacting with each other and with environmental factors. Future approaches are required to identify the gene expression associated with the SNP windows found in this study.

**Key Words:** GWAS, temperament

## Breeding and Genetics: Genomic methods and application—Dairy

**W79 Common copy number variation regions affecting dairy traits in Gyr cattle.** Gerson A. Oliveira Junior<sup>\*1</sup>, Adriana S. Carmo<sup>2</sup>, Adam T. H. Utsunomiya<sup>3</sup>, Tatiane C. S. Chud<sup>3</sup>, Fernando S. B. Rey<sup>3</sup>, Jose Bento S. Ferraz<sup>1</sup>, and Marcos Vinicicus G. B. da Silva<sup>2</sup>, <sup>1</sup>University of São Paulo, Pirassununga, São Paulo, Brazil, <sup>2</sup>Embrapa Dairy Cattle, Juiz de Fora, Minas Gerais, Brazil, <sup>3</sup>São Paulo State University, Jaboticabal, São Paulo, Brazil.

The Gyr cattle (*Bos indicus*) is a very important dairy breed in tropical countries like Brazil, mainly due to its tolerance to heat stress and parasites infestations as well as their use in crossbreeding schemes with other specialized dairy breeds, such as Holstein. In addition to the single nucleotide polymorphism (SNP), genomic structural variants such as copy number variants (CNV) have been revealed to be a substantial source of genetic and phenotypic variation in cattle, being an alternative to explain the missing heritability of complex traits. The aim of this work was to investigate common CNV regions associated with dairy traits in Gyr cattle. The Log R ratio profiles of 481 Gyr animals were determined using a high density SNP chip (Illumina BovineHD BeadChip assay), and the phenotypes evaluated were: age at first calving (AFC), milk (MY), fat (FY), protein (PY) and total solids yields (TSY), and protein (PP), fat (FP) and total solids percentage (TSP). The Log R ratio, predicted from Genome Studio software, was used in Golden Helix SNP & Variation Suite (SVS) 8.1, and the copy number analysis module (CNAM – multivariate algorithm) was used to identify common CNVs among animals. Linear regression was employed to identify CNVs associated with each production trait with significance level of FDR >0.05. A total of 47 CNV regions were detected that affected at least one trait, with 22 regions affecting 3 or more traits where one of them, located at chromosome 7, affected all traits except MY. Under these 22 regions, 38 structural variation, 18 genes and one pseudo-gene annotated in bovine genome (Biomart tool of Ensembl) were observed. The Panther Classification System divided these genes into 10 biological process categories highlighting localization, immune system and metabolic processes. The results suggest that common CNV regions can be biologically involved with more than one dairy trait.

**Key Words:** structural variant, genomics, dairy cattle

**W80 Genome-wide association study on conception rate, milk production, and SCS in different stages of lactation for first three parities in US Holsteins.** Shogo Tsuruta<sup>\*1</sup>, Daniela A. L. Lourenco<sup>1</sup>, Ignacio Aguilar<sup>2</sup>, and Ignacy Misztal<sup>1</sup>, <sup>1</sup>University of Georgia, Athens, GA, <sup>2</sup>INIA, Las Brujas, Canelones, Uruguay.

The objectives of this study were to conduct genome-wide association studies (GWAS) on conception rate (CR), production traits, and SCS for Holstein cows and determine if the genetic architecture of these traits is different in 3 stages of lactation for the first 3 parities in US Holsteins. Genome-wide association studies were conducted for conception rate (CR), test-day milk, fat and protein yields, and test-day SCS. The data were split into 3 sets: early (<14wk), middle (14wk ≤ DIM ≤ 29wk), and end (29wk <). Heritability estimates for CR were lowest (0.03) in the middle and highest (0.05 to 0.08) in the end of lactation in all parities. Genetic correlations of CR with other production traits were low and all negative (−0.1 to −0.5). The SNP marker effects were divided into equal segments of 30 SNP. A segment on chromosome 14 was associated with CR only in early and middle stages of lactation in the third parity; the proportion of the total genetic variance explained by

this segment for CR were 2.7% and 2.5%, respectively. The proportions for test-day milk and fat yields were highest in the middle of lactation (7% for milk and 8% for fat) in all 3 parities, whereas the proportions for test-day protein and SCS were low (<2%). The results suggest that gene expression for CR and milk production traits is stronger in early and middle lactation stages and similar over the lactations.

**Key Words:** GWAS, conception rate, US Holsteins

**W81 Single nucleotide polymorphisms associated with thermoregulation in lactating dairy cows exposed to heat stress.** Serdal Dikmen<sup>\*1,3</sup>, Xian-zhong Wang<sup>2,3</sup>, and Peter J. Hansen<sup>3</sup>, <sup>1</sup>Department of Animal Science, Faculty of Veterinary Medicine, University of Uludag, Bursa, Turkey, <sup>2</sup>College of Animal Science and Technology, Southwest University, Chongqing, China, <sup>3</sup>Department of Animal Sciences, University of Florida, Gainesville, FL.

Dairy cows with increased rectal temperature during heat stress experience lower milk yield and fertility. Given that rectal temperature during heat stress is heritable in dairy cattle, genetic selection for regulation of body temperature should reduce effects of heat stress on production. One goal of the study was to validate the relationship between genotype and heat tolerance for SNPs previously related to resistance to heat stress. A second goal was to identify new candidate gene SNPs related to resistance to heat stress. Thermotolerance was assessed in 625 lactating Holstein cows during the hottest part of the day in summer by measuring rectal temperature (a direct measurement of body temperature regulation), respiration rate (an indirect measurement body temperature regulation) and sweating rate (the major evaporative cooling mechanism in cattle). Specific genetic markers and candidate genes responsible for genetic variation in these variables were identified. For SNPs previously related to heat tolerance in genome-wide analysis, a region of BTA6 was related to rectal temperature and 3 closely-located genetic markers on BTA24 and another on BTA29 were associated with sweating rate. New candidate gene SNPs were identified for rectal temperature (n = 7), respiration rate (n = 9), and sweating rate (n = 6). The largest effect on rectal temperature was for *PGR*, which explained 2.1% of the phenotypic variation after adjustment for dry bulb temperature. This SNP could affect heat loss via cutaneous cooling because progesterone regulates vasodilation in the skin during local heating. *ACAT2* (involved in lipid metabolism) explained the largest variation in respiration rate (3.3%) and *SERPINE2*, which regulates the enzyme thrombin that regulates epithelial cells in sweat glands, explained the largest variation in sweating rate (2.1%). These genetic markers could prove useful in genetic selection for heat tolerance in Holstein cattle.

**Key Words:** heat stress, thermoregulation, SNP

**W82 Multi-generational imputation of SNP genotypes and accuracy of genomic selection.** Sajjad Toghiani<sup>\*</sup> and Romdhane Rekaya, *The University of Georgia, Athens, GA.*

Superiority of genomic selection (GS) is possible only when high density single nucleotide polymorphism (SNP) panels are used to track QTLs affecting traits. Even with the continuous decrease in genotyping costs, only a small fraction of the population has been genotyped with these high-density panels. To reduce the cost of GS, it is often the case that a larger portion of the population is genotyped with low-density SNP panels and then imputed to a higher density. Accuracy of SNP genotype

imputation tends to be high when minimum requirements are met. Nevertheless, a certain rate of errors is unavoidable. Such rate of errors tends to increase with the increase of the generational interval between reference and testing generations. Thus, it is reasonable to assume that accuracy of GEBVs will be affected by imputation errors. To evaluate the impact of multi-generational selection on the accuracy of SNP genotypes imputation on the reliability of resulting GEBVs, a simulation was carried out under varying updating of the reference population, distance between training and validation sets, and the approach used for the estimation of GEBVs. Using fixed reference populations, imputation accuracy decayed by around 0.5% per generations. In fact, after 25 generations, the accuracy was only 7% lower than the first generation. When the reference population was updated by either 1% or 5% of the top animals in the previous generations, decay of imputation accuracy was substantially reduced. These results indicate that low-density panels are useful, especially when the generational interval between reference and testing population is small. As the generational interval increases, the imputation accuracies decay, although not at an alarming rate. In absence of updating of the reference population, accuracy of GEBVs decays substantially in 1 or 2 generations with a decrease rate of around 20–25% per generation. When the reference population is updated by 1 or 5% every generation, the decay in accuracy was only 8 to 11% after 7 generations. These results indicate that imputed genotypes provide a viable alternative, as long the reference and training populations are appropriately updated.

**Key Words:** genotype imputation, genomic selection, accuracy

**W83 Genome-wide association study for milk production traits in Russian dairy cattle.** Alexander A. Semyagin\*<sup>1</sup>, Elena A. Gladyr<sup>1</sup>, Sergei N. Kharitonov<sup>1</sup>, Alexander N. Ermilov<sup>1,2</sup>, Ivan N. Yanchukov<sup>2</sup>, Nikolai I. Strekozov<sup>1</sup>, and Natalia A. Zinovieva<sup>1</sup>, <sup>1</sup>L.K.Ernst Institute of Animal Husbandry, Dubrovitsy, Moscow, Russia, <sup>2</sup>Regional Information Selection Center, Noginsk, Moscow, Russia.

Genome-wide association study has been proven as a powerful tool for identifying genomic variants associated with economically important traits in domestic animal breeds. Our study is the first step toward the creating the reference population of Russian Holsteins to utilize the genomic information in the dairy cattle breeding programs in Russia. The objective of our study was to evaluate the association between SNPs and estimated breeding values (EBVs) for milk production traits. The genomic data were obtained by genotyping 195 progeny-tested and 61 young bulls using the Illumina Bovine SNP50 v2 BeadChip. The SNP's quality control was performed by using Plink (1.07) software. BLUP AM approach has been used to estimate the marker effects which were applied then to calculate the genomic EBVs for young bulls to increase the prediction reliability of the associations. Direct genomic and genomic EBVs (by GBLUP) were estimated for 305-d milk yield (MY), milk fat yield (FY), milk protein yield (PY), fat percent (FP) and protein percentage (PP). After the quality control, 41370 SNPs were selected for the association analysis. The average number of daughters per sire was about 240 and the reliability of EBVs amounted 87%. The linkage disequilibrium was  $r^2 = 0.41$ . The Bonferroni correction for detection significant associations was applied as  $P < 1.2 \times 10^{-6}$ . Two SNPs which had the most significant effect for MY were identified: ARS-BFGL-NGS-50172 on BTA17 ( $P = 7.6 \times 10^{-8}$ ) and Hapmap54246-rs29017970 on BTA13 ( $P = 1.7 \times 10^{-7}$ ). The association analysis for milk components revealed 3 SNPs significantly associated with FP: BTA-104917-no-rs on BTA9 ( $P = 4.1 \times 10^{-8}$ ), ARS-BFGL-NGS-107379 on BTA14 ( $P = 6.0 \times 10^{-7}$ ) and BTB-01604502 on BTA9 ( $P = 1.1 \times 10^{-6}$ ). The effect on

PP was shown for SNP Hapmap43278-BTA-50082 on BTA20 ( $P = 8.0 \times 10^{-7}$ ). Few SNPs were found to have the effects on FY and PY traits. The significant effects of SNPs explained from 9.0 to 11.3% of additive genetic variances. Supported by the Russian Ministry of Education and Science (RFMEFI60414X0062).

**Key Words:** genome-wide association, breeding value, milk production

**W84 Identification of copy number variable gene families in Holstein and Jersey cattle.** Derek M. Bickhart\*<sup>1</sup>, Lingyang Xu<sup>2,1</sup>, Jana L. Hutchison<sup>1</sup>, Harris A. Lewin<sup>3</sup>, and George E. Liu<sup>1</sup>, <sup>1</sup>United States Department of Agriculture, Agricultural Research Service, Animal Genomics and Improvement Laboratory, Beltsville, MD, <sup>2</sup>University of Maryland, Department of Animal and Avian Sciences, College Park, MD, <sup>3</sup>University of California, Department of Evolution and Ecology, Davis, CA.

Copy number variants (CNV) represent a large proportion of genetic variation within the cattle genome that has yet to be accurately characterized by SNP genotyping arrays. While significant progress has been made in the identification of CNVs within individual animals using next generation sequence data, CNV frequencies within larger populations have not yet been estimated in cattle. In this study, we sequenced 28 individual bulls from 2 dairy breeds of cattle (22 Holstein bulls; 6 Jersey bulls) to identify dairy breed-specific copy number variation. Using a read depth method of CNV detection, we identified 1359 non-redundant CNV regions within all 28 animals. The number of variable bases contained within these CNV regions accounts for ~2% of the cattle genome, and the average CNV region frequency was 37.67%. This high average frequency suggests that a large proportion of CNVs were present in the ancestral population of both breeds of cattle rather than as a result of a large number of de novo events arising in subsequent generations after breed formation. We also assigned copy number values to each gene within each individual sequenced using the normalized sequencing read depth of non-overlapping genomic windows. Using a Vst approach on these gene copy number values, we identified 27 gene families with breed specific copy number expansions/contractions. We identified a Jersey-exclusive expansion of the CLEC5A gene, which is a regulator of osteoclastogenesis. Additionally, we identified a Holstein-exclusive duplication of the ASAP1 gene, which may be involved in cell membrane trafficking and the differentiation of fibroblasts into adipocytes. CNVs identified by this survey intersected gene families that may play a role in productive traits in dairy cattle and are therefore good candidates for novel genetic marker design.

**Key Words:** copy number variant, genomics, sequencing

**W85 Single nucleotide polymorphisms in specific candidate genes are associated with phenotypic differences in days open for first lactation in Holstein cows.** M. Sofia Ortega\*<sup>1</sup>, Anna C. Denicol<sup>1</sup>, Daniel J. Null<sup>2</sup>, John B. Cole<sup>2</sup>, and Peter J. Hansen<sup>1</sup>, <sup>1</sup>Department of Animal Sciences, University of Florida, Gainesville, FL, <sup>2</sup>Animal Genomics and Improvement Laboratory, Agriculture Research Service, United States Department of Agriculture, Beltsville, MD.

Previously, a candidate gene approach identified 51 single nucleotide polymorphisms (SNP) associated with genetic merit for reproductive traits and 26 associated with genetic merit for production in dairy bulls. We evaluated association of these 77 SNP with days open (DO) for first lactation in a population of Holstein cows grouped based on predicted



transmitting ability for daughter pregnancy rate (DPR):  $\leq -1$  ( $n = 1220$ ) and  $\geq 1.5$  ( $n = 1053$ ), and located on 11 farms in Florida and California. Cows were genotyped using a Sequenom MassARRAY assay. To evaluate phenotypes, farm records were retrieved from on-farm computers and combined with records from the national genetic evaluation system. The association of the genetic variants with DO was evaluated using the MIXED procedure of SAS V9.4 (SAS Institute, Inc., Cary, NC). The model included farm, number of copies of the minor allele, and the numerator relationship matrix to account for (co)variances among animals. For each SNP, the genotype was treated as a categorical variable to estimate additive and heterosis effects. Days open was lower ( $P < 0.0001$ ) for cows in the high DPR group as compared with the low DPR group ( $97.8 \pm 2.6$  d vs  $163.0 \pm 2.9$  d). There were 6 SNP with significant additive effects ( $P < 0.05$ ) on DO (*COQ9*, *FCER1G*, *FST*, *GPLD1*, *MRGPRF* and *OCNL*) and an additional 6 SNPs with a tendency ( $P < 0.10$ ) for an association (*ACAT2*, *CD14*, *PCCB*, *PMM2*, *RABEP2* and *SREBF1*). For example, DO for cows with 0, 1, or 2 copies of the minor allele for *COQ9* averaged  $139.4 \pm 3.5$ ,  $134.3 \pm 2.8$ , and  $123.6 \pm 3.5$  d, respectively. The DO for cows with 0, 1, or 2 copies of the minor allele for *FST* averaged  $124.9 \pm 3.3$ ,  $134.8 \pm 2.6$  and  $135.8 \pm 4.4$  d, respectively. For 9 of 12 genes, the favorable allele for DO was also the favorable allele in the earlier report based on bulls. The SNP related to genetic and phenotypic estimates of fertility are likely to be informative markers for genetic selection. Moreover, the study of the role of these genes could provide new insights into the physiological regulation of fertility in dairy cattle (USDA AFRI 2013–68004–20365).

**Key Words:** single nucleotide polymorphism, days open, dairy cattle

**W86 Animal selection for whole-genome sequencing by quantifying the unique contribution of homozygous haplotypes sequenced.** Jana L. Hutchison\*, John B. Cole, and Derek M. Bickhart, *United States Department of Agriculture, Agricultural Research Service, Animal Genomics and Improvement Laboratory, Beltsville, MD.*

Major whole-genome sequencing projects promise to identify rare and causal variants within livestock species; however, the efficient selection of animals for sequencing remains a major problem within these surveys. The goal of this project was to develop a library of high accuracy genetic variants found within diverse haplotypes that were in a homozygous state identified from animal genotypes in the national database. An inverted weight function that calculated the value of sequencing an animal based on the sum of the rarity of the haplotypes it had in its SNP-based genotype was used to calculate the estimate, as more common haplotypes would likely be represented within animals already sequenced in subsequent iterations. A weight value was assigned to each 75-SNP haplotype based on the inverse of its frequency within genotyped animals in the national database. Each individual's haplotype weights were summed, and the highest scoring animal was selected for sequencing. Haplotypes from selected animals were removed from future consideration, and the cumulative scores of all remaining animals were recalculated in the absence of those selected haplotypes. This iteration continued until all haplotypes above a frequency threshold of 4% had been selected for sequencing. There were a total of 3,680 75-SNP haplotypes above a frequency of 4% in the national database and 484,522 genotyped Holstein animals. We compared this method against the selection of animals for sequencing based on 3 additional algorithms: (1) an ascending relatedness weight function, (2) an unbiased predictor of imputation accuracy, and (3) a random selection of animals from the population. By calculating an iterative summed score based on the inverse value of an animal's unsequenced haplotypes, one can

quickly determine the value of sequencing a new individual and avoid data redundancy that plagues projects that focus on sequencing highly related individuals in a population.

**Key Words:** sequencing, haplotype

**W87 A GWAS on heat tolerance phenotypes for Italian Holstein bulls.** Stefano Biffani<sup>1</sup>, Umberto Bernabucci<sup>2</sup>, Nicola Lacetera<sup>2</sup>, Andrea Vitali<sup>2</sup>, Paolo Ajmone Marsan<sup>3</sup>, Nicolo PP Macciotta\*<sup>4</sup>, and Alessandro Nardone<sup>2</sup>, <sup>1</sup>*IBBA-CNR, Lodi, Italy*, <sup>2</sup>*Dipartimento di Scienze e Tecnologie per l'Agricoltura, le Foreste, la Natura e l'Energia Università degli Studi della Tuscia, Viterbo, Italy*, <sup>3</sup>*Istituto di Zootecnica, Università Cattolica del Sacro Cuore, Piacenza, Italy*, <sup>4</sup>*Dipartimento di Agraria, Università di Sassari, Sassari, Italy.*

Heat stress is a key factor that negatively affects livestock productive and reproductive performance. A genome-wide scan was performed on a sample of 1,592 Italian Holstein bulls using 2 different measures of heat tolerance for milk yield and protein percentage. The first was a temperature-humidity index breeding value (THI EBV) recently proposed for the Italian Holstein breed. The latter was obtained by a principal component analysis carried out on milk test-day records corrected for environmental effects except for THI. Only the second principal component (PC2), which describes the individual patterns of corrected production data across different THI levels, was considered. Animals were genotyped with the Illumina BovineSNP 50 BeadChip. Monomorphic SNPs (7,140) and SNPs with a call-rate  $< 95\%$  (1,045) were discarded. In total, 45,546 SNPs were retained for the analysis. All bulls had a THI EBV, whereas the PC2 was available only for a sub-sample of bulls (423). Genome-wide scan was performed fitting the GRAMMAR approach through the GenABEL R package. Then, a Gene discovery analysis was carried out considering windows of 0.5 Mb surrounding the significant marker (0.25Mb up and down stream respectively). No significant associations were detected for milk THI EBV, apart from a weak signal on BTA 2 at about 32 Mb. In this region is located the solute carrier family 38, member 11 (SLC38A11) gene, reported to be involved in folliculogenesis in cattle. For PC2, 3 SNPs were detected on BTA 6, 16 and 26, respectively. The SNP on BTA26 is located in a region that hosts genes involved in the ovarian activity (FGF8). An interesting candidate for the SNP located on BTA16 at approximately 42.1 Mb is the dehydrogenase/reductase member 3 (DHRS3), involved in the embryonic development in humans. No significant associations were found for protein percentage THI EBV. However, 3 significant markers were detected for PC2 on BTAs 20, 14 and 8. Interestingly the BTA14 region hosts the junctophilin 1 (JPH1) gene, whose expression has been found to be upregulated in the hypothalamus of chickens subjected to heat stress. These preliminary findings suggest potential genomic regions linked to heat stress resistance in dairy cattle.

**Key Words:** heat stress, GWAS, principal component analysis

**W88 A genome-wide association study of mastitis in US Holstein and the relationship to mammary microbiome profile identifies novel QTL.** Heather Huson\*<sup>1</sup> and Rodrigo Bicalho<sup>2</sup>, <sup>1</sup>*College of Agriculture and Life Sciences, Cornell University, Ithaca, NY*, <sup>2</sup>*College of Veterinary Medicine, Cornell University, Ithaca, NY.*

One of the most prevalent and costly obstacles facing dairy producers is the occurrence of mastitis. Mastitis is a worldwide endemic disease causing both short and long-term cow health and economic repercussions with production losses in terms of reduced milk yield, clinical treatment, culling of animals, and discarded milk. The objective of this study was

to conduct a genome-wide association study (GWAS) for mastitis and explore the use of milk microbiome profiles to identify novel mastitis QTL using the Illumina Bovine High-Density (777K SNPs) Beadchip on a cohort of US Holstein dairy cows. To this end, 2 GWAS were run comparing Holstein cows; the first compared cows phenotypically characterized as having clinical mastitis (CM) or healthy based on their somatic cell score (SCS) and the second GWAS used a linear score based on their mammary microbiome profile. The mammary microbiome profile was generated sequencing 16S rRNA of the milk microbiota, directly correlated with mastitis incidence, to generate a cow specific profile. Four genomic regions demonstrating significant association to CM incidence were identified on BTA 5, 16, 26, and 29 using SCS and 5 regions on BTA 5, 9, 16, 26, and 29 were significantly associated with microbiome profile GWAS (corrected  $P$ -value  $< 0.05$ ). Of particular interest to this study was to identify regions of similar association and those different between the 2 trait GWAS, CM as opposed to microbiome profile. Regions on BTA 5, 9, and 26 validated previous mastitis QTL findings. Both GWAS approaches in this study identified novel QTL on BTA 16 and 29 potentially being the product of a more generalized immune response to infection. In contrast, marker associations on BTA 1, 2, and 11 show more variation between the 2 GWAS and may reflect a genetic predisposition to specific microbiota. In all, the study validated previously discovered QTL and identified novel QTL with both similarities and variation in the comparison of the 2 trait GWA studies. Further studies are required to validate specific candidate genes and their influence on mastitis and the potential identification of diagnostic markers for the selection of mastitis resistant dairy cows.

**Key Words:** mastitis, GWAS, microbiome

**W89 The accuracy of genomic predictions for Japanese Holsteins using by GBLUP and ssGBLUP methods.** Yusaku Gotoh<sup>\*1</sup>, Toshimi Baba<sup>1</sup>, Satoshi Yamaguchi<sup>2</sup>, and Takayoshi Kawahara<sup>1</sup>, <sup>1</sup>Holstein Cattle Association of Japan, Hokkaido Branch, Sapporo, Japan, <sup>2</sup>Hokkaido Dairy Milk Recording and Testing Association, Sapporo, Japan.

Several strategies including GBLUP and ssGBLUP methods have been proposed to predict genomic breeding values (GEBV) for dairy cattle. Many studies have reported that the accuracy of GEBV predicted from ssGBLUP was higher than from GBLUP. There is no report applying same analysis to Japanese Holsteins. The objective of this study was to compare the accuracies of GPI predicted by GBLUP and ssGBLUP. The traits used in this analysis were 305-d milk, fat and protein yields, and feet and legs, udder and final score. In total, 3,787 bulls genotyped by the Illumina BovineSNP50 BeadChip were used. Validation bulls were prediction bulls with no daughters in 2009 data but more than 20 daughters in 2013 data. In multistep evaluation, training bulls were required to have 10 daughters in 2009 data. Number of training bulls were 517 and 554 for the analysis of milk and type traits, respectively. For multistep evaluation, GPI were estimated by selection index blending after calculating DGV by linear GBLUP. Coefficients of determination ( $R^2$ ) by regression analysis of GPI for 2009 data on deregressed EBV for 2013 data were used as indicator of accuracy. The  $R^2$  ranged from 0.19 (protein) to 0.27 (milk) and from 0.17 (feet and legs) to 0.28 (milk) for DGV and GPI predicted by GBLUP and from 0.21 (protein) to 0.30 (milk) for GEBV predicted by ssGBLUP, respectively. Hence, it is suggested that the GEBV predicted by ssGBLUP is higher accuracy than DGV and GPI predicted by GBLUP.

**Key Words:** GBLUP, ssGBLUP, accuracy

**W90 Identification of loci associated with fertility in Holstein heifers.** Joao G. N. Moraes<sup>\*1</sup>, Joseph Dalton<sup>2</sup>, Thomas E. Spencer<sup>1</sup>, Jennifer N. Kiser<sup>1</sup>, Gregory W. Burns<sup>1</sup>, Andrzej Wojtowicz<sup>1</sup>, Mahesh Neupane<sup>1</sup>, and Holly L. Neibergs<sup>1</sup>, <sup>1</sup>Department of Animal Science, Washington State University, Pullman, WA, <sup>2</sup>Department of Animal and Veterinary Sciences, University of Idaho, Caldwell, ID.

Selection for higher milk production in United States dairy cattle has been very successful during the past 50 years, however modern lactating dairy cows exhibit a high incidence of subfertility and infertility with a national pregnancy rate of only 15 to 20%. The objective of this study was to identify genomic loci associated with fertility in nulliparous Holstein heifers. Breeding and health records of Holstein heifers ( $n = 2,333$ ) were analyzed from a commercial heifer raising facility in Southern Idaho. Of these, 1,114 heifers were classified as highly fertile (conceived on first AI service) and 209 were identified as subfertile (did not conceive until after the fourth AI service or culled due to failure to conceive). Blood samples were obtained from the fertility-classified heifers, and DNA was extracted from 497 high fertile and 209 subfertile heifers. The DNA was genotyped with the Illumina Bovine HD Genotyping BeadChip. Quality control consisted of removing SNPs with  $< 90\%$  call rate, and a MAF  $< 1\%$  and removing heifers with a genotyping rate  $< 90\%$ , leaving 575,959 SNPs and 470 fertile and 189 subfertile heifers for analysis. A genome wide association analysis (GWAA) and heritability estimate was conducted with the Efficient Mixed-Model Association expedited (EMMAX) software. This mixed model program empirically estimated a genomic relationship matrix and used it to model the correlation between the fertility phenotypes. Correction for population stratification was done by variance components and resulted in  $\lambda_{GC} = 1.0$ . The GWAA identified a QTL on BTA4 with a strong association with fertility ( $P = 2.9 \times 10^{-9}$ ), while loci on BTA1, BTA2, BTA5, BTA6, BTA10, BTA11, BTA18, BTA23, BTA26, BTA27 and BTA28 were identified with a moderate association with fertility ( $P < 5.0 \times 10^{-5}$ ). The heritability estimate for fertility in Holstein heifers was 0.52. These results indicate that there is ample opportunity to make significant gains in fertility in nulliparous Holstein heifers with genomic selection. This project was supported by Agriculture and Food Research Initiative Competitive Grant no. 2013–68004–20365 from the USDA National Institute of Food and Agriculture.

**Key Words:** genomics, fertility, heifer

**W91 Accuracy of genomic imputation in a Thai multi-breed dairy cattle population.** Danai Jattawa<sup>\*1,2</sup>, Skorn Koonawootrittriron<sup>1</sup>, Mauricio A. Elzo<sup>2</sup>, and Thanathip Suwanasopee<sup>1</sup>, <sup>1</sup>Kasetsart University, Chatuchak, Bangkok, Thailand, <sup>2</sup>University of Florida, Gainesville, FL.

The objective of this study was to investigate the accuracy of imputation from low (LDC) to moderate density SNP chips (MDC) in a Thai Holstein-Other multibreed dairy cattle population. Dairy cows with complete pedigree information ( $n = 1,110$ ) from 129 dairy farms were genotyped with GeneSeek GGP20K ( $n = 570$ ) and GGP26K ( $n = 540$ ) BeadChips. After checking for genotypic quality, 16,387 SNP in common between the GGP20K and GGP26K were used to represent MDC in this study. Cows were divided into 2 groups, a reference group ( $n = 778$ ) and a test group ( $n = 332$ ). The SNP genotypes chosen for the test group were those SNP located in positions corresponding to GeneSeek GGP9K ( $n = 7,356$ ). The LDC to MDC genomic imputation was carried out using 3 different methods, namely a population-based algorithm in the Beagle software (PBG), a population-based algorithm in the FImpute software (PFI), and a combined family and population-based algorithm in FImpute (CFI). Imputation accuracies within and

across chromosomes were calculated as ratios of correctly imputed genotypes to overall imputed genotypes. Imputation accuracy for the 3 methods ranged from 76.31% to 93.91%. The CFI had slightly higher imputation accuracy (93.91%) than PFI (93.56%) and both methods were substantially more accurate than PBG (76.31%). Noticeably most chromosomes that showed either high or low imputation accuracies were the same chromosomes that had high and low average linkage disequilibrium (defined here as the correlation between pairs of adjacent SNP within chromosomes less than 5 MB apart). This suggested that choosing sets of SNP with high levels of average linkage disequilibrium would improve imputation accuracy. Results clearly indicated that FImpute software (population or combined family-population) were more suitable than Beagle for genotype imputation in this Thai multibreed population. Perhaps additional increments in imputation accuracy could be achieved by discarding SNP with low levels of average linkage disequilibrium, and by increasing the completeness of pedigree information.

**Key Words:** imputation accuracy, linkage disequilibrium, multibreed dairy cattle

**W92 Identification of copy number variation in Brazilian synthetic dairy cattle breed.** T. C. S. Chud<sup>1</sup>, M. V. G. B. da Silva<sup>2</sup>, A. S. Carmo<sup>2</sup>, T. B. R. Silva<sup>\*1</sup>, G. A. Oliveira Junior<sup>3</sup>, F. S. Baldi Rey<sup>1</sup>, and D. P. Munari<sup>1</sup>, <sup>1</sup>Univ Estadual Paulista "Júlio de Mesquita Filho," Jaboticabal, SP, Brazil, <sup>2</sup>Embrapa - Brazilian Corporation of Agricultural Research, Juiz de Fora, MG, Brazil, <sup>3</sup>Universidade de São Paulo, Pirassununga, SP, Brazil.

Copy number variation (CNV) refers to genomic segments that present a type of structural variation, such as duplications or deletions. CNVs have been observed as an important source of genetic and phenotypic variation for production traits and animal health. The aim of this work was to identify CNVs in a synthetic breed (Gyr × Holstein) dairy cattle population (Girolando cattle). The data set contained 417 animals genotyped with the Illumina 50K SNP panel (~54,609 SNPs). An algorithm based on the Hidden Markov Model was implemented using PennCNV software (Wang et al., 2007) for CNV identification. PennCNV perl script was used to eliminate calls from low quality samples, based on the standard deviation of LRR (<0.30), the BAF drift (<0.01) and waviness factor (less than 0.05). The final data set was composed of 384 animals. Gene content of cattle CNV was assessed using Ensembl genes. We used the PANTHER classification system for testing the hypothesis ( $P < 0.05$ ) that the GO terms of the molecular function, biological process, and pathway terms were under or overrepresented in the CNV. An account of 1,986 CNVs were found along the genome, of which 84% were duplications and 16% were deletions. The chromosomes BTA3, BTA17 and BTA23 presented higher frequencies (10.52%, 11.53%, 8.30%, respectively) of CNV. Chromosomes that showed lower frequency of CNV (<1%) were BTA27, BTA14 and BTA29. A total of 861 genes were found within these regions and they are involved in biological processes, such as development (105 genes), growth (2 genes), immune system (83 genes), metabolism (343 genes) and reproduction (12 genes). This study showed evidences of structural variations in the genome of Girolando cattle and the genes found in CNV may be involved in the expression of production and animal health traits.

**Key Words:** genomics, single nucleotide polymorphism, structural variation

**W93 Linkage disequilibrium in a Thai dairy cattle population with different Holstein fractions.** Thawee Laodim<sup>1</sup>, Skorn Koonawootrittriron<sup>\*1</sup>, Mauricio A. Elzo<sup>2</sup>, and Thanathip Suwanasopee<sup>1</sup>,

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Linkage disequilibrium (LD) is important for gene mapping, accuracy of genomic prediction, and understanding of recombination biology in dairy cattle populations. The level of LD can vary among populations depending on their genetic structure, selection and recombination rates. The objective of this study was to estimate and compare levels of LD in dairy cattle with different Holstein fractions under tropical conditions. Blood samples of 2,643 dairy cattle (89 bulls and 2,554 cows) from 304 farms located in Central, Northern, Western and Southern Thailand were extracted for DNA. The DNA samples were genotyped with one of 4 GeneSeek Genomic Profiler BeadChips (9K, 20K, 26K, or 80K). Only SNPs from autosomes in common among the 4 chips were considered. In addition, SNPs with a minor allele frequency (MAF) lower than 0.01 and a call rate lower than 90% were excluded. This resulted in a set of 7,123 SNPs used in this study. Animals were classified into 7 groups based on their Holstein fraction (HF): HF <75%, 75% ≤ HF <80%, 80% ≤ HF <85%, 85% ≤ HF <90%, 90% ≤ HF <95%, 95% ≤ HF <100%, and purebred HF. Distribution of MAF and estimation of LD were done using Haploview. All HF groups had similar patterns of MAF across autosomes (fraction of SNPs increased with an increase in MAF). However, means of MAF across autosomes differed among HF groups and it tended to decrease with an increase in H fraction (from 0.376 for HF <75% to 0.362 for purebred HF). Conversely, the mean  $r^2$  across autosomes tended to increase as HF increased from 0.081 for HF <75% to 0.109 for purebred HF. Results from this study will be useful for genome wide association studies and for genomic prediction and selection of crossbred Holstein cattle in tropical regions.

**Key Words:** linkage disequilibrium, Holstein, tropics

**W94 Improving the genotyping-by-sequencing (GBS) approach for the identification of SNPs associated with Johne's disease.** Émilie Constant<sup>1,2</sup>, Eveline M. Ibeagha-Awemu<sup>1</sup>, Filippo Miglior<sup>3,4</sup>, Gilles Robitaille<sup>5</sup>, and Nathalie Bissonnette<sup>\*1,2</sup>, <sup>1</sup>Dairy & Swine Research and Development Centre Agriculture and Agri-Food Canada, Sherbrooke, Quebec, Canada, <sup>2</sup>Department of Biology, Université de Sherbrooke, Sherbrooke, Quebec, Canada, <sup>3</sup>Canadian Dairy Network, Guelph, Ontario, Canada, <sup>4</sup>CGIL, University of Guelph, Guelph, Ontario, Canada, <sup>5</sup>Food Research and Development Centre, Agriculture and Agri-Food Canada, Saint-Hyacinthe, Québec, Canada.

Bovine paratuberculosis is a disease caused by *Mycobacterium avium* ssp. *paratuberculosis* (MAP). Most infected cows are culled before they reach clinical stage, leading to the premature slaughter of many animals and significant economic losses. Several genetic variants have been reported associated with host susceptibility to MAP. The objective of this study was to validate a whole genome genotyping-by-sequencing (GBS) method to identify single nucleotide polymorphisms (SNPs) associated with bovine paratuberculosis. Animals were selected from 10 farms in the province of Quebec. Fecal and blood samples were collected to identify 24 MAP infectious status by fecal culture and serum ELISA and 24 healthy cows. Two GBS methods were compared: a conventional (restriction enzymes *Pst*I and *Msp*I at 5'/3' used to construct DNA libraries) method (CM) and CM with more selective primers to reduce the complexity of the libraries (RM). DNA was extracted from isolated peripheral blood monocyte cells. Multiplexed libraries (48 libraries per lane) were subjected to 100-bp single-end sequencing on an Illumina HiSeq 2000 system. Reads that passed all filtering criteria were mapped to the bovine genome (Bta\_4.6.1). The SNPs were called using the Universal Network-Enabled Analysis Kit and 30,266 and 82,593 passed



the quality control steps for the CM and the RM methods, respectively. Using the SVS GoldenHelix software, 3,653 (CM) and 17,413 (RM) SNPs were associated with regions that are not intergenic. Associated genes are linked to biological processes related to the immune system and to responses to stimuli, such as defense responses to bacteria. Analysis confirmed that 8 (CM) and 19 (RM) SNPs were associated with MAP infectious status ( $P < 0.05$ ). Validation of their genetic association with bovine paratuberculosis are currently performed by SEQUENOM mass spectrometry using a larger group of animals ( $n = 800$ ). The reduction of the complexity of the genome produced a larger number of qualified

genotypes compared with the conventional method suggesting that this GBS strategy provides a greater reading depth, which increases the quality of each genotype.

**Key Words:** genotyping-by-sequencing, *Mycobacterium avium* ssp. *paratuberculosis*, genetic predisposition to disease

## Companion Animals: Nutrition

**W95 Identifying sources of *Salmonella* contamination in animal feed and pet food facilities.** Andrea M. Jeffrey\*<sup>1</sup>, Cassandra K. Jones<sup>1</sup>, Greg Aldrich<sup>1</sup>, Anne R. Huss<sup>1</sup>, and Carl Knueven<sup>2</sup>, <sup>1</sup>Kansas State University, Manhattan, KS, <sup>2</sup>Jones-Hamilton, Walbridge, OH.

*Salmonella* is a potential biological hazard in animal food, and may contaminate livestock feed and pet food through cross contamination at manufacturing facilities. The FDA has evaluated *Salmonella* concentrations in classes of feed ingredients, but has not evaluated pathogen concentrations based on location throughout a facility. The objective of this experiment was to investigate sources of *Salmonella* contamination from various equipment and environmental locations in 2 livestock feed manufacturing plants and 2 pet food manufacturing plants on a specific sampling day. Up to 40 environmental swab samples were collected at each facility using sterile, prepackaged swab vials containing buffered peptone water. Samples were collected from a variety of equipment and structural surfaces, including concrete, dust, plastic, rubber, and broom bristles and analyzed for qualitative *Salmonella* determination as described by FDA's Bacteriological Analytical Manual. Data were categorized by facility (1 to 4), type (equipment or structural), and surface (concrete, metal, plastic, rubber, or dust) and analyzed using the GLIMMIX procedure of SAS. There were no interactions, so all were removed from the model ( $P > 0.05$ ). There were no differences in *Salmonella*-positive samples among facilities, but facility 1, a livestock feed manufacturer, doubled the locations testing positive for *Salmonella* as compared with the other 3 facilities ( $P = 0.11$ ; average positive-*Salmonella* concentration of 44.1, 20.1, 19.0, and 20.1% for facilities 1, 2, 3, and 4, respectively). There were no differences in percentage of *Salmonella* samples testing positive based on location of swab ( $P = 0.57$ ; average *Salmonella* concentration of 22.9 and 28.7% for equipment and structural, respectively). Finally, there was a tendency for rubber and concrete to have greater *Salmonella*-positive samples than plastic, dust, or metal ( $P = 0.10$ ; 40.9, 35.2, 24.4, 20.4, and 8.2%, respectively). With these results in mind, it may be important to consider the types of surfaces present and the appropriate sanitation to best control *Salmonella* in livestock and pet food manufacturing facilities to prevent cross-contamination into animal food.

**Key Words:** *Salmonella*, animal feed, pet food

**W96 Dog ownership increases the richness of the cutaneous microbiome.** Celia S. Sobelman\*<sup>1</sup>, Jessica K. Suagee<sup>2</sup>, and Cristina Caldari<sup>1</sup>, <sup>1</sup>Centenary College of Louisiana, Shreveport, LA, <sup>2</sup>The Ohio State University, Wooster, OH.

Maintaining a normal microflora is imperative for the modulation of immune responses by the host and prevents the overgrowth of opportunistic microorganisms. Changes in cutaneous microflora could result in skin diseases. It has been shown that individuals that co-habitate harbor similar cutaneous microbial communities. The purpose of this study was to determine if humans that co-habitate with dogs have different cutaneous microbial richness compared with humans that are rarely exposed to dogs. Microbial richness was defined as the number of morphologically distinct colonies in a sample. Humans that co-habitate with dogs (dog owners) were defined as humans living with  $\geq 1$  indoor dogs. Humans having rare contact with dogs (non-dog owners) were defined as humans that were in contact with dogs  $\leq$  once a week. Cutaneous microbial samples were obtained by using a sterile cotton

swab to swab the top of the hands, forearms and foreheads of humans and the dorsal thoracic area, nose area, and chest of dogs. The cotton swabs were stored in sterile phosphate buffered saline (PBS) for 24 h after which time 100 mL of the PBS were plated onto a tryptic soy agar plate with 5% sheep blood. After a 48-h incubation period at 37°C, distinct microbial colonies were counted. Distinctness of the colonies was determined based on colony morphology, cell morphology and Gram staining. Microbial richness varied among dogs, dog-owners, and non-dog owners (Levene's test,  $P$ -value = 0.007), with dogs having the most variation and non-dog owners having the least variation. The average cultured cutaneous microbial richness was significantly higher in dogs and dog owners ( $6.73 \pm 4.23$  and  $5.60 \pm 3.20$  microbial species, respectively) compared with non-dog owners ( $2.73 \pm 0.96$  microbial species; dog vs. non-dog owner:  $P = 0.0001$ , dog owner vs non-dog owner:  $P = 0.002$ ). There was no significant difference in the cutaneous microbial richness of dogs and dog-owners. These results suggest that frequent contact with dogs leads to an increase in cutaneous microbial richness compared with infrequent contact with dogs. The implications of these findings on health and disease warrant further investigation.

**Key Words:** cutaneous microbiome, dog, dog ownership

**W97 Effects of thiamine type, species meat versus livers, and sulfite addition on water-soluble B-vitamins in a canned cat diet.** Shelby D. Tribble\*, Charles G. Aldrich, and Cassandra K. Jones, Kansas State University, Manhattan, KS.

There has been little work published concerning the effect of processing on the degradation of water-soluble B-vitamins in canned pet foods. The objectives of these experiments were to evaluate the effect of thiamine type (mononitrate vs hydrochloride), protein type (species meat and livers), and sulfite (yes or no) addition on B-vitamin retention. All diets were produced at a batter temperature of 60°C and moisture of 78%. The cook time was one hour at 121°C and 21psi. In Exp. 1 ( $n = 26$ ) thiamine type tended ( $P = 0.12$ ) to influence retention, wherein thiamine mononitrate retention (140.68 mg/kg) was higher than that of thiamine hydrochloride (128.37 mg/kg). In Exp. 2 ( $n = 14$ ), protein type had an effect on retention of several B-vitamins. Thiamine retention was highest in fish relative to chicken or liver diets ( $P < 0.05$ ; 202.26 vs 140.72, and 131.19 mg/kg, respectively). Riboflavin retention was highest in diets containing liver, intermediate with fish, and lowest for chicken ( $P < 0.05$ ; 200.83, 153.46, and 113.66 mg/kg, respectively). Niacin was at its lowest retention in diets containing chicken compared with fish and liver diets ( $P < 0.0015$ ; 238.42, 327.90, and 444.64 mg/kg, respectively). Pyridoxine retention was highest in diets containing fish versus those with liver or chicken ( $P < 0.05$ ; 135.89 vs 113.34, and 100.59 mg/kg, respectively). Cobalamin retention was at its lowest in diets containing chicken, and intermediate for fish and highest for liver ( $P < 0.05$ ; 0.45, 0.62, and 1.53 mg/kg, respectively). The addition of sulfites indirectly from dehydrated potatoes in Exp. 2 had a negative effect on pantothenic acid ( $P < 0.05$ ; 234.80 mg/kg vs 360.29 mg/kg), and pyridoxine ( $P < 0.05$ ; 108.02 mg/kg vs 134.34 mg/kg) relative to the controls, respectively. The dehydrated potatoes with sulfites tended to influence thiamine retention ( $P = 0.07$ ; 137.80 mg/kg vs 188.22 mg/kg) and riboflavin retention ( $P = 0.09$ ; 147.89 mg/kg vs 188.25 mg/kg) relative to the control. Vitamin form, protein type, and presence of sulfites may influence their retention in canned pet foods.

**Key Words:** B-vitamin, wet pet food, sulfites

**W98 Chemical composition, nutrient digestibility, and true metabolizable energy of commercially available protein sources using the precision-fed cecectomized rooster assay.** Ping Deng<sup>\*1</sup>, Pamela Utterback<sup>1</sup>, Carl Parsons<sup>1</sup>, and Kelly Swanson<sup>1,2</sup>, <sup>1</sup>*Department of Animal Sciences, University of Illinois, Urbana, IL*, <sup>2</sup>*Department of Veterinary Clinical Medicine, University of Illinois, Urbana, IL*, <sup>3</sup>*Division of Nutritional Sciences, University of Illinois, Urbana, IL*.

A wide variety of animal protein-based ingredients are commonly used in the pet food products. The raw ingredients and processing procedures used may greatly affect protein quality and digestibility. Testing the quality of alternative protein sources is necessary and contributes to the sustainability of pet foods. The objective of this study was to determine the chemical composition, nutrient digestibility, and nitrogen-corrected true metabolizable energy (TMEn) of 4 protein sources intended for use in dog and cat foods (pork peptone, calamari meal, chicken meal, and duck meal) using the precision-fed cecectomized rooster assay. Calamari meal and pork peptone had lower ash [4.4% and 3.6% of dry matter (DM), respectively], but greater crude protein (CP; 88.1% and 80.5% of DM, respectively) and gross energy (GE; 5.55 and 5.32 kcal/g of DM, respectively) compared with chicken meal (11.8% ash; 65.9% CP; 4.79 kcal/g) and duck meal (17.3% ash; 60.8% CP; 4.62 kcal/g). Acid-hydrolyzed fat (AHF) was lower in calamari meal (8.7% of DM) compared with the other proteins tested (15.5–15.9% of DM). Nutrient digestibility was variable among the protein sources [64 to 79% of DM, 76 to 83% of organic matter (OM), 86 to 92% of AHF, 83 to 89% of GE] with pork peptone having the highest DM, AHF, and GE digestibility, and calamari meal having the highest OM digestibility. Essential amino acid (AA) true digestibility was highest for calamari meal, with all AA having a digestibility greater than 90%. Except for histidine, all essential AA had a digestibility over 85% for pork peptone. All essential AA had a digestibility over 80% for duck meal, but chicken meal had 3 essential AA with digestibilities less than 80%. The TMEn of calamari meal (4.82 kcal/g DM; 86.8% of GE) was greater ( $P > 0.05$ ) than that of pork peptone (3.83 kcal/g DM; 71.9% of GE), chicken meal (3.46 kcal/g DM; 72.2% of GE), and duck meal (3.46 kcal/g DM; 74.9% of GE). This study demonstrates the considerable variability that exists not only in the chemical composition, but also the nutrient digestibility, among protein sources intended for use in dog and cat foods.

**Key Words:** protein source, nutrient digestion, rooster assay

**W99 The effect of low-bloom gelatin on physical properties of extruded pet food.** Analena E. Manbeck<sup>\*</sup>, C. Greg Aldrich, and Sajid Alavi, *Department of Grain Science and Industry, Kansas State University, Manhattan, KS*.

Gelatin is an animal-based protein that has been used to improve pellet quality. Previous work with low-bloom gelatin in extruded kibble demonstrated improved hardness and durability but decreased expansion. The objective of this experiment was to clarify the relationship of expansion on product hardness and durability resulting from gelatin inclusion. Two complete diets (30% protein) were produced: a control with no gelatin (OG) and another with 10% low-bloom gelatin (10G) added at the expense of chicken by-product meal. Diets were produced on a Wenger X-20 extruder through a circular die (4.6mm diameter). The extruder screw speed and throttle valve opening were adjusted to achieve 2 densities (HD and LD). Products were analyzed for bulk and piece density, radial expansion, specific length, hardness, and pellet durability index (PDI). Results were summarized with the aid of statistical analysis software (SAS 9.4). Hardness was not affected by treatment ( $P > 0.05$ ). The main effect means of PDI decreased 33% with the inclusion

of gelatin ( $P < 0.05$ ; 77.57% v 52.25%) and decreased 20% between HD and LD ( $P < 0.05$ ; 72.17% v 57.65%). The main effect means of radial expansion and specific length increased with gelatin inclusion ( $P < 0.05$ ; 2.86 v 3.56mm<sup>2</sup>/mm<sup>2</sup> expansion and 4.12 v 4.47cm/g length), but were unaffected by HD or LD. The main effect means of piece and bulk density were lower with gelatin inclusion (0.52 v 0.39g/cm<sup>3</sup> piece density and 351.8 v 280.7g/L bulk density) and also reduced from HD to LD (0.46 v 0.44 g/cm<sup>3</sup> piece density and 345.8 v 266.7g/L bulk density). The means for PDI were affected by the interaction of gelatin level and target density. PDI decreased from HD to LD within the OG diet ( $P < 0.05$ ; 92.0% v 63.14%) and continued to decrease with the 10G diets. However, there was no difference between HD and LD for the 10G diets ( $P > 0.05$ ; 52.34% v 52.16% for HD and LD, respectively). The decrease in PDI between OG and 10G may be due to the increased expansion, but the gelatin may prevent a further decrease in PDI between HD and LD. Low-bloom gelatin may improve the physical properties of high protein pet food without compromising expansion.

**Key Words:** pet food, gelatin, extrusion

**W100 Effects of age and diet on colonic mucosa microbiota of dogs.** Ana Paula J. Maria<sup>\*1</sup>, Ping Deng<sup>2</sup>, Hannah D. Holscher<sup>2</sup>, Franz N. Yoshitoshi<sup>3</sup>, Thaila C. Putarov<sup>1</sup>, Kelly S. Swanson<sup>2</sup>, and Aulus C. Carciofi<sup>1</sup>, <sup>1</sup>*São Paulo State University (UNESP), Jaboticabal, SP, Brazil*, <sup>2</sup>*University of Illinois at Urbana-Champaign, Urbana, IL*, <sup>3</sup>*Endoscopy and Surgery, São Paulo, SP, Brazil*.

The objective of this study was to identify the effects of age and diet, with particular focus on carbohydrate and protein sources, on the colonic mucosa microbiota of dogs. Thirty-six healthy beagles were used in a 3x2 factorial design. Dogs were separated in 2 age groups, young adult (2.6 ± 0.9 yr) and geriatric (10.2 ± 1.1 yr), and were assigned to 3 isonutritive kibble dietary groups containing: (1) a non-fermentable insoluble fiber (IF; 34% poultry meal; 8% sugarcane fiber); (2) a fermentable fiber (FF; 35% poultry meal; 10% beet pulp); and (3) soybean meal (SM; 30% SM; 11% poultry meal; no additional fiber source). Dogs were fed the experimental diets for 30-d, followed by sample collection on d 31. Mucosal biopsies from the colon were performed by colonoscopy procedure with dogs under anesthesia. DNA was extracted and the V4 region of the 16S rRNA gene was amplified and subjected to Miseq Illumina sequencing. Data analysis was performed using QIIME 1.8.0. The resulting operational taxonomic units (OTU) were aligned to the Greengenes 13\_8 database (97% similarity threshold). Firmicutes (44.1%) was the predominant bacterial phylum, followed by Bacteroidetes (39.2%), Fusobacteria (6.5%), Proteobacteria (5.6%), Actinobacteria (4.3%) and Deferribacteres (0.2%). There was an interaction between age and diet ( $P < 0.05$ ) for *Prevotella*, *Sutterella*, and an undefined genus in the Mogibacteriaceae family. The abundance of *Slackia*, *Bacteriodes*, *Plesiomonas*, and an undefined genus in the Paraprevotellaceae family were lower ( $P < 0.05$ ) in geriatric dogs compared with the young adult dogs. *Peptococcus* and *Slackia* genus were in a higher ( $P < 0.05$ ) abundance in dogs fed the IF and FF diets than those fed the SM diet. Dogs fed the IF diet had lower ( $P < 0.05$ ) colonic mucosa *Megamonas* and *Suturrella* abundance when compared with the dogs fed FF and SM diets. In conclusion, both dietary fiber fermentability and age may impact the microbial communities present on the colonic mucosa of dogs. More research is needed to identify the relevance of these microbial shifts in regards to gastrointestinal health.

**Key Words:** age, carbohydrates, microbiota



**W101 Digestibility of the crude corn oil in dogs.** Tabyta T. Sabchuk<sup>\*1</sup>, Karoline Vanelli<sup>1</sup>, Larissa Barrile<sup>2</sup>, Fabiane Y. Murakami<sup>1</sup>, Alex Maiorka<sup>1</sup>, Simone G. Oliveira<sup>1</sup>, and Ananda P. Félix<sup>1</sup>, <sup>1</sup>*Federal University of Paraná, Curitiba, Paraná, Brazil*, <sup>2</sup>*Cargil Agrícola SA, Uberlândia, Minas Gerais, Brazil*.

There are many fat sources used in dog nutrition, such as poultry fat, beef tallow, and soy oil. In addition, there are several factors that can determine the choice of the fat source, such as price, availability, fatty acid profile, chain saturation. One alternative fat source can be the crude corn oil (CCO) that is a co-product of maize, produced worldwide. Besides, CCO has a large amount of linoleic acid, an essential fatty acid for dogs. Thus, the objective was to evaluate the apparent total-tract digestibility (ATTD) and ME of CCO in adult dogs. Three diets were evaluated: a control diet (with 8% beef tallow, CD), and 2 diets containing 92% of the ingredients of the CD and 8% of CCO and another with 8% poultry fat (PF). Diets were sprayed with the fat sources. Nine Beagle dogs were randomly assigned in 2 blocks (2 periods), totaling 6 replicates per treatment. The experimental diets in kibble form were offered for a 5-d adaptation period, followed by 5 d of total fecal collection per period. The ATTD and ME of the tested ingredients (CCO and PF) were calculated according to the substitution method. The results were submitted to Student's *t*-test ( $P < 0.05$ ). The ATTD of DM (98.7% vs 96.0%), acid-hydrolyzed fat (AHF, 98.6% vs 97.4%), GE (99.7% vs 98.6%), and ME (38.08 MJ/kg vs 37.93 MJ/kg) of CCO and PF, respectively, did not differ ( $P > 0.05$ ). Normally we evaluate the digestibility of diets and not the digestibility of the ingredient itself, such as in this study. Consequently, specific information on the nutritional quality of ingredients is lacking, particularly in fat sources to dogs. We found that the digestibility and ME of CCO was similar to the PF, which is the most frequently used fat source in dog nutrition. Thus, the CCO has high digestibility and can be a co-product with potential use in diets for dogs.

**Key Words:** companion animal nutrition, fat source, ingredient digestibility.

**W102 A high protein intake allows the preservation of lean mass and prevents the increase of fat mass, compared with a moderate protein intake, in neutered cats.** Agnès André<sup>1</sup>, Isabelle Leriche<sup>2</sup>, Gwendoline Chaix<sup>3</sup>, and Patrick Nguyen<sup>\*1</sup>, <sup>1</sup>*Nutrition & Endocrinology Unit, National College of Veterinary Medicine, Nantes, France*, <sup>2</sup>*Virbac Nutrition, Vauvert, France*, <sup>3</sup>*Virbac Medical Department, Carros, France*.

Cats are strict carnivores and have a high dietary protein requirement. Rich-protein diets are often intended to prevent obesity or manage weight loss, as they help preserve the lean body mass. The aim of this study was to assess the effect of an experimental high-protein low-carbohydrate maintenance dry diet (HP) on body composition (BC), compared with a commercial moderate-protein high-carbohydrate dry diet (MP) in neutered cats. Twelve (12) young adult neutered cats ( $19.6 \pm 0.4$  mo old;  $3.56 \pm 0.2$  kgBW) were randomized in 2 groups and received, for 20 weeks, either a HP (3,320 kcal/kg of DM; 50.2 CP %DM) or a MP (3,590 kcal/kg of DM; 33.7 CP %DM) diet. Main protein sources and amino acid content (DM basis) were: HP diet: meat meal, pea; Lys 2.3%, Met 1.0%, Try 0.4%, Thr 1.7%; MP diet: poultry meal, corn gluten meal, corn; Lys 2.0%, Met 1.0%, Try 0.3%, Thr 1.3%. Animals were fed according to their estimated energy requirement to maintain their BW. Body composition (BC) was determined using deuterium oxide dilution at the beginning then at the end of the study. Tukey's test was used to detect the effect of each diet and a Wilcoxon test to evaluate the differences between groups, with a 5% significance level. The mean protein intake during the study was  $7.2 \pm 0.6$  g/kgBW/d in the HP

group, and  $4.6 \pm 0.3$  g/kgBW/d in the MP group. On d 1, the 2 groups were similar regarding their BW and BC. In both groups, no change in BW was observed. BC was unchanged in the HP group whereas body fat mass increased ( $P < 0.05$ ) and lean body mass decreased ( $P < 0.01$ ) in the MP group. The lean mass/fat mass ratio changed from 74/26 to 75/25 and from 77/23 to 69/31, in the HP and MP groups respectively. Although the protein content of the MP diet was higher than the recommended allowance (20% ME according to NRC 2006), it appeared not high enough to maintain lean body mass in these cats. Our results are in accordance with another study showing that adult cats would require at least 5.2 g protein/kg BW/d to maintain their lean body mass.

**Key Words:** cat nutrition, protein intake, body composition

**W103 The effect of processing and elevated storage temperatures on omega-3 fatty acid stability in pet food.** Alaina K. Mooney<sup>\*</sup>, C. G. Aldrich, C. K. Jones, and S. Alavi, *Kansas State University, Manhattan, KS*.

Essential fatty acid research has shown that omega-3 fatty acids such as eicosahexaenoic acid (EPA) and docosahexaenoic acid (DHA; 22:6n3) may help maintain normal body structure, function and aid in long-term health and wellbeing. Common sources of omega 3 fatty acids include flax seed, fish oil, fishmeal, and more recently, purpose grown algae. This commercially produced source of omega 3 fatty acids has been evaluated as a supplement to animal diets and for its impact on metabolism; however, questions regarding the effect of processing and storage in pet foods are unanswered. The objective was to determine the effect of processing on stability of an algal source of DHA, (DHAgold S17-B; DSM Nutritional Products) added to the diet by premix, extrusion-drying processing, and extended storage. Three nutritionally complete pet diets at protein levels 21.7, 25, and 30% CP (Low, Medium and High, respectively) were produced with equal levels of DHA supplied by DHAgold S17-B, fishmeal and fish oil. Diets were produced on a Wenger X-20 single screw extruder (Wenger Mfg, Sabetha, KS) and dried at 104°C for 10 min at each pass in a triple pass dryer (Wenger Mfg, Sabetha, KS). Samples from each treatment were analyzed immediately following production for moisture and fatty acids. Shelf-life samples were collected in whirlpaks with a pin-hole and stored at 40°C and 75% relative humidity for analysis at 3, 6, 12, 18 and 24 weeks following production. Retention of EPA and DHA at production time was not affected by CP level ( $P > 0.05$ ), but was impacted by DHA source ( $P < 0.05$ ). The total omega 3 fatty acids were affected by DHA source and CP level ( $P < 0.05$ ). As time in storage progressed through 0, 3, 6, 12, 18 and 24 weeks EPA ( $P < 0.05$ ; 12.53, 10.45, 10.19, 8.9, 8.4, and 8.2 mg/kg, respectively) and DHA ( $P < 0.05$ ; 7.7, 7.2, 6.9, 6.1, 6.4, and 6.0 mg/kg, respectively) declined slightly; but, total omega 3 fatty acids ( $P < 0.05$ ; 35.6, 47.7, 47.9, 44.4, 43.3, and 41.8 mg/kg, respectively) were greater at all times than the start. These results suggest that elevated temperatures during storage for 24 weeks could result in slight EPA and DHA sacrifice. DHAgold S17-B appears to be a stable source of DHA when compared with fish oil and fishmeal.

**Key Words:** extrusion, pet food, omega-3 fatty acid

**W105 The impact of rendered protein meal level of oxidation on shelf life and acceptability in extruded pet foods.** Morgan N. Gray<sup>\*</sup>, Charles G. Aldrich, Cassandra K. Jones, and Michael W. Gibson, *Kansas State University, Manhattan, KS*.

Increasing pressure has been put on ingredient suppliers to assure a low level of oxidation, as measured by a low peroxide value. Our objective was to determine the effect of increasingly oxidized protein meals on

the shelf life of extruded pet foods. Approximately one metric ton each of unpreserved chicken by-product meal (C) and unpreserved beef meat and bone meal (B) were collected and left unpreserved (U) or preserved with either ethoxyquin (E), or mixed tocopherols (T). These were allowed to oxidize at ambient conditions (25°C and 51% RH) while being monitored for peroxide value (PV) and anisidine value (AV) until they plateaued (41 and 63 d, respectively) at a PV of 88.44, 4.43, 2.22 mEq/kg and AV of 1.08, 0.55, 0.00 g/g for CU, CT, CE, respectively and at a PV of 86.42, 8.88, 2.23 mEq/kg and AV of 12.23, 7.14, 0.00 g/g for BU, BT, BE, respectively. Each meal was then incorporated into a model extruded cat food diet (~30% protein). Samples of kibble for each treatment were collected and stored at an elevated temperature and humidity (40°C and 70%) for 18 weeks. At time 0, PV and AV were greater for CU and BU ( $P < 0.05$ ; 14.41, 10.07 mEq/kg and 15.56, 10.08 g/g, respectively) versus the preserved treatments CT, CE, BT, and BE (2.78, 2.22, 2.22, 2.22 mEq/kg and 3.85, 1.79, 9.62,

3.03 g/g, respectively). At elevated storage temperatures, the PV for CE remained low (4.44 mEq/kg), CT was intermediate (23.21 mEq/kg) and CU increased to 53.15 mEq/kg by 18 weeks ( $P < 0.05$ ). The AV for C followed a similar pattern. The PV of B under elevated temperatures behaved differently; wherein, BE was low (3.33 mEq/kg), but BT had the highest PV (15.48 mEq/kg) and BU was intermediate (6.66 mEq/kg) by 18 weeks ( $P < 0.05$ ). BE had the lowest ( $P < 0.05$ ) AV and BT and BU were greater, but did not differ from each other (average 16.75 g/g) at 18 weeks. The results from this study demonstrate that oxidation occurred regardless of treatment; but, was rapid and extensive in meals without preservative. The ingredient oxidation levels were diluted by food production and their oxidation may not completely account for later food product deterioration.

**Key Words:** pet food, oxidation, rendered protein meals

# Comparative Gut Physiology

**W106 Effect of complex and prebiotic diets on intestinal health of nursery pigs inoculated to porcine circovirus 2 (PCV2).** Marco M. Lima<sup>\*1,2</sup>, Dana M. van Sambeek<sup>2</sup>, Huyen Tran<sup>2</sup>, Daniel C. Ciobanu<sup>2</sup>, Phillip S. Miller<sup>2</sup>, and Thomas E. Burkey<sup>2</sup>, <sup>1</sup>Univesidade Estadual Paulista, Jaboticabal, Sao Paulo, Brazil, <sup>2</sup>University of Nebraska, Lincoln, NE.

To investigate the effects of complex and prebiotic diets on nursery pigs inoculated for or vaccinated against porcine circovirus 2 (PCV2) on the intestinal health, 96 weaned barrows (age 27 to 40 d; BW 7.1 kg) were housed (4 pigs/pen) in an environmentally controlled nursery with ad libitum access to feed and water over a 28-d study. Forty-eight pigs were vaccinated (VAC) for PCV2 before arrival, while remaining pigs (PCV) were inoculated for PCV2 on d 0. Pigs were randomly assigned to 1 of 3 diets: complex (CO; lactose, spray-dried plasma, spray-dried whey), simple (SI; corn and SBM), or simple + 2.5% Grobiotic-S (GS). After 28 d of experiment, 1 pig per pen was euthanized by exposure to CO<sub>2</sub> for collection of samples. Two-cm segments of duodenum and ileum were collected for the determination of villous height (VH), villous area (VA), crypt depth (CD), and villous height: crypt depth (VH:CD) ratio. Images were recorded using an Olympus DP71 camera (Olympus, Center Valley, PA). The VH, CD, VH:CD ratio, and VA were measured using Cell Sense standard software (Olympus, Center Valley, PA). For each intestinal sample, a minimum of 10 to 15 full villi and crypts were measured and averaged for statistical analysis. All data were analyzed as a completely randomized design using the MIXED procedure of SAS. Pen was considered the experimental unit and the model included treatment as a fixed effect. All measurements were performed without treatment awareness. No significant PCV-status effects were observed in the duodenum histological measurements; however, a significant diet × PCV-status interaction ( $P = 0.0251$ ) on VH:CD in duodenal samples was observed. The highest VH:CD result observed was in pigs fed GS and inoculated with PCV. Diet had an effect ( $P = 0.0066$ ) on duodenal CD, with deeper crypts being observed for GS pigs. With respect to VH and CD, a tendency for a diet × PCV-status interaction ( $P < 0.07$ ) was observed whereby PCV inoculated pigs and fed simple or GS diets had greater CD and VH, respectively. No significant effects were observed for ileum histological measurements. Results indicate that diet complexity may affect duodenal health in pigs inoculated with PCV.

**Key Words:** diet, health, intestine

**W107 Mitotic and apoptosis rate of intestinal cells of nursery pigs fed with a blend of organic acids and medium chain fatty acids.** Marco M. Lima<sup>\*1</sup>, Maria C. Thomaz<sup>1</sup>, Fabricio F. Castro<sup>1</sup>, Rosemeire S. Filardi<sup>2</sup>, Daniela J. Rodrigues<sup>1</sup>, Maryane S. F. Oliveira<sup>1</sup>, Manuela V. Marujo<sup>1</sup>, Everton Daniel<sup>1</sup>, Antonio C. Laurentiz<sup>2</sup>, and Thomas E. Burkey<sup>3</sup>, <sup>1</sup>Univesidade Estadual Paulista, Jaboticabal, Sao Paulo, Brazil, <sup>2</sup>Univ Estadual Paulista, Ilha Solteira, Sao Paulo, Brazil, <sup>3</sup>University of Nebraska, Lincoln, NE.

To investigate the effects of a blend of organic acids and medium chain fatty acids on gut health, 96 weaned barrows (21 d; BW 5.2 kg) were housed (3 pigs/pen) with ad libitum access to feed and water over a 28-d study. The diets were comprised primarily of corn, soybean meal and a source of lactose, with inclusion of 0, 0.15, 0.30 and 0.45% of a mixture of acetic acid, citric acid, formic acid, lactic acid, caprylic acid and caproic acid (BLEND). Piglets ( $n = 4$  pigs/treatment/time point) were euthanized on d 14 and 28 for sample collection. Two-cm

segments of duodenal and jejunal tissue were collected for analysis of apoptotic and mitotic rates. Rate of apoptosis and mitosis were evaluated immunohistochemically in paraffin wax-embedded tissue sections using polyclonal antibodies (anti-Caspase 3 and anti-PCNA, respectively; Biocare Medical, Concord, CA). The immunohistochemical procedure was conducted per the manufacturer's instructions. Images were recorded using an Olympus BX41 camera. For each intestinal sample, a minimum of 20 to 30 full villi and crypts were measured. ANOVA was generated using the mixed-model procedure of SAS. Linear, quadratic, and cubic polynomials were used to detect the response to dietary inclusion of the BLEND. All measurements were performed without treatment awareness. A cubic effect was observed with respect to the number of cells in mitosis in duodenal ( $P < 0.0001$ ) and jejunal ( $P = 0.0062$ ) tissue segments obtained on d 14 with the greatest rate of mitosis observed in tissues obtained from pigs fed 0.30% BLEND. In addition, a quadratic effect in mitosis:apoptosis ( $P = 0.0171$ ) ratio was observed in duodenal tissue obtained from piglets on d 14 with minimal inflection point at 0.18% inclusion of BLEND. No significant effects were observed with respect to mitotic or apoptotic rates in tissues obtained on d 28. In conclusion, feeding a blend of organic acids and medium chain fatty acids may have a short-term positive effect on mitosis and apoptosis, possibly contributing to intestinal epithelial restitution and gut health.

**Key Words:** apoptosis, intestine, mitosis

**W108 Effects of organic acids and medium-chain fatty acids on gut health of nursery pigs.** Marco M. Lima<sup>\*1</sup>, Maria C. Thomaz<sup>1</sup>, Fabricio F. Castro<sup>1</sup>, Rosemeire S. Filardi<sup>3</sup>, Daniela J. Rodrigues<sup>1</sup>, Maryane S. F. Oliveira<sup>1</sup>, Manuela V. Marujo<sup>1</sup>, Dana M. van Sambeek<sup>2</sup>, and Thomas E. Burkey<sup>2</sup>, <sup>1</sup>Univesidade Estadual Paulista, Jaboticabal, Sao Paulo, Brazil, <sup>2</sup>University of Nebraska-Lincoln, Lincoln, NE, <sup>3</sup>Universidade Estadual Paulista, Ilha Solteira, Sao Paulo, Brazil.

To investigate the effects of a blend of organic acids and medium chain fatty acids on gut health, 96 weaned barrows (21 d; BW 5.2 kg) were housed (3 pigs/pen) with ad libitum access to feed and water over a 28-d study. The diets were comprised primarily of corn, soybean meal and a source of lactose, with inclusion of 0, 0.15, 0.30 and 0.45% of blend (acetic acid, citric acid, formic acid, lactic acid, caprylic acid and caproic acid). For sample collection, piglets ( $n = 16$ ) were euthanized at 14 and 28-d study. Two-cm segments of duodenum and jejunum were collected for the determination of villous height (VH), crypt depth (CD), villous height: crypt (VH:CD) depth ratio and count goblet cell by methods of Schiff-PAS and Alcian Blue. Images were recorded using an Olympus BX41 camera. Cell Sense standard software was used for measurement of the respective morphological aspects. For each intestinal sample, a minimum of 20 to 30 full villi and crypts were measured and in assessing the number of goblet cells, 15 counts were performed. ANOVA was generated using the mixed-model procedure of SAS. Linear, quadratic and cubic polynomials were used to detect the response to dietary inclusion of the blend. Was observed a cubic effect in duodenal VH:CD ( $P < 0.0001$ ) ratio and jejunal goblet cell count ( $P = 0.0034$ ) by method of Schiff-PAS at 14-d study. Was observed a cubic effect on CD ( $P = 0.0009$ ), goblet cell count ( $P = 0.0014$ ) by method of Schiff-PAS, goblet cell count ( $P = 0.006$ ) by method of Alcian Blue, and an increasing linear effect on the VH:CD ( $P = 0.0006$ ) ratio in jejunum of pigs at 28-d study. Results indicate the blend had little effect at 14-d study; however,



positive effects of blend were observed at 28-d of study. In conclusion, the blend may contribute to intestinal restitution.

**Key Words:** health, organic acid, pig

**W109 Effect of turmeric, ginger and garlic extracts on performance, microbial load, and gut morphology of weaned pigs.**

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Producing swine without in-feed antibiotics requires a combination of different strategies. Some of the approaches that are available to improve the pigs ability to reduce the impact of intestinal pathogens are phytogetic, which improve intestinal health and promote productivity. Therefore, the objective of this experiment was to assess the effect of turmeric, ginger and garlic extracts on performance, microbial loads and gut morphology of weaned pigs. A total number of 20 weaned pigs were randomly allotted into 4 treatments with 5 replicates arranged in a completely randomized design. The treatments are: T1 (control), T2 (2g turmeric extract/kg feed), T3 (2g ginger extract/kg), T4 (2g garlic extract/kg). The feed was formulated to contain 19% crude protein and 2950 kcal/kg ME which meets the nutritional requirements of the weaner pigs. The experiment lasted for 49 d. No significant ( $P < 0.05$ ) difference was observed in the performance attributes of the weaned pig that were fed the different diets. The feed conversion ratio ranges from 3.59 in pigs fed T2 diet to 3.92 in pigs fed the control diet. Enterobacter count was significantly ( $P < 0.05$ ) higher in pigs fed T1 (6.38 cfu), T2 (6.29 cfu) and T3 (6.27 cfu) diets, compared with their counterpart on T4 (5.12 cfu). The least value of 6.63 cfu for total bacteria count was also observed in pigs fed T4 (garlic extract) diet. The ileum section of the gastro-intestinal tract of the pigs revealed significant ( $P < 0.05$ ) increase in the villus height for pigs on T4 (1419.75  $\mu$ m) than what was observed on pigs fed other diets (709.25  $\mu$ m, 1156.50  $\mu$ m and 1068.00  $\mu$ m in T1, T2 and T3 respectively). However, no significant ( $P > 0.05$ ) variations was observed in the villus width and crypt depth despite the different treatments. It can be concluded that inclusion of garlic extract at 2g/kg feed resulted in improved intestinal morphology and reduction in the microbial load in weaned pig.

**Key Words:** pig, extract, performance

**W110 Effect of heat stress on endogenous intestinal loss of amino acids in growing pigs.**

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Heat stress (HS) appears to increase intestinal cell death in pigs, which in turn may increase endogenous intestinal losses (EIL) of proteins and amino acids (AA). Experimental objectives were to analyze the effect of HS on the AA composition of intestinal endogenous proteins and the EIL of AA in pigs. Eight pigs (24 kg initial BW) were surgically implanted with T-type cannulas at the end of the small intestine. After surgery recovery, all pigs were adapted to a protein-AA-free diet and trained to consume the same amount of feed twice a day for 7d in thermo-neutral (TN) conditions (22  $\pm$  2°C). On d 8 of TN conditions ileal content was collected during 12 consecutive h. On d 9 pigs were exposed to natural HS conditions (31 to 37°C) for 8 d. During the HS period ileal content

was collected again on d2 (HSd2) and d8 (HSd8). Chromic oxide was used as indicator of the intestinal digesta flow. Data were analyzed using PROC MIXED of SAS; the model considered the TN pigs as no HS and the fixed effect of HS-sampling day was used to compare TN with HSd2 and HSd8. The AA composition of endogenous intestinal protein was not affected by HS ( $P > 0.10$ ). The EIL of indispensable AA of TN, HSd2 and HSd8 pigs were: Arg, 314, 345, 335; His, 144, 163, 153; Ile, 169, 196, 173; Leu, 378, 434, 387; Lys, 238, 275, 250; Met, 099, 113, 110; Phe, 298, 337, 296; Thr, 489, 533, 481; Val, 335, 354, 343 mg/d, respectively. The EIL of dispensable AA of TN, HSd2 and HSd8 pigs were: Ala, 377, 423, 383; Asp, 639, 684, 621; Glu, 478, 553, 509; Gly, 977, 838, 872; Pro, 763, 892, 1,941; Ser, 375, 394, 422; Tyr, 219, 251, 227 mg/d, respectively. The EIL of Thr and Phe tended to be increased ( $P \leq 0.10$ ), and Arg and His were enhanced at HSd2 ( $P < 0.05$ ), and Pro increased at HSd8 ( $P = 0.01$ ) compared with TN conditions. The EIL of the remainder AA was not affected by HS. Although HS increased the EIL of Arg and His within the first 2 d, it appears that normal EIL of AA is quickly reestablished. In summary, HS does not appear to affect the AA composition of intestinal endogenous proteins and suggest that the EIL of AA may not be critical in chronic HS pigs.

**Key Words:** pig, heat stress, endogenous amino acids loss

**W111 Effect of dietary fiber and a multicarbohydrase enzyme blend on net glucose and lactate fluxes, insulin production, and oxygen consumption by the portal-drained viscera and by the whole animal in growing pigs.**

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The objective was to determine the effects of supplementing a high-fiber (HF) diet with a multicarbohydrase enzyme (MC; 4000 units/g xylanase, 1000 units/g  $\alpha$ -amylase, 500 units/g protease, 150 units/g  $\beta$ -glucanase) on net portal fluxes of glucose and lactate, insulin production and oxygen consumption by the portal-drained viscera (PDV) and whole animal (WAOC) in growing pigs. Three isocaloric/nitrogenous diets based on corn and soybean meal (SBM) with either 0% (CTRL) or 30% distillers dried grains with solubles (DDGS; 1:1 corn and wheat mixture; HF) were used; the third diet was supplemented with MC in addition to the 30% DDGS (HF+MC). Five 22.8  $\pm$  1.6 kg gilts fitted with permanent catheters in the portal vein, ileal vein (to infuse para-amino hippuric acid to measure blood flow rate) and carotid artery were fed the 3 diets at 4% BW once daily at 0900 h for 7 d in a replicated 3  $\times$  3 Latin square design. On d 7, pigs were placed in an open-circuit indirect calorimeter to measure WAOC and sample blood for 7 h postprandial. Net glucose and insulin production were calculated from portal-arterial differences  $\times$  flow and PDV O<sub>2</sub> consumption was calculated as arterial-portal O<sub>2</sub> differences  $\times$  flow. Diet had no effect ( $P > 0.10$ ) on postprandial WAOC, flow rate, and lactate flux. Also, diet had no effect ( $P > 0.10$ ) on overall postprandial PDV O<sub>2</sub> consumption, but altered ( $P < 0.05$ ) postprandial PDV O<sub>2</sub> consumption from 240 to 360 min postprandial. Pigs fed CTRL had higher ( $P < 0.05$ ) portal insulin and glucose fluxes, from 90 to 300 min, and net glucose flux from 90 to 240 min postprandial. However, pigs fed CTRL and HF+MC had similar ( $P > 0.10$ ) postprandial net glucose flux, which was higher ( $P < 0.05$ ) than pigs fed HF. Therefore, it was concluded that adding MC to the HF diet improved net glucose portal flux in pigs.

**Key Words:** high-fiber diet, multicarbohydrase, net portal flux

**W112 Effect of a post-weaning diet supplemented with gut health-enhancing feed additives on ileum transcriptome activity and serum cytokines in piglets challenged with lipopolysaccharides (LPS).** Nathalie Bissonnette\*, Xian-Ren Jiang<sup>1,4</sup>, Jacques J. Matte<sup>1</sup>, Guylaine Talbot<sup>1</sup>, Frédéric Guay<sup>2</sup>, Joshua Gong<sup>3</sup>, Qi Wang<sup>3</sup>, Valentino Bontempo<sup>4</sup>, and Martin Lessard<sup>1</sup>, <sup>1</sup>Dairy & Swine Research and Development Centre Agriculture and Agri-Food Canada, Sherbrooke, Quebec, Canada, <sup>2</sup>Département des Sciences Animales, Université Laval, Québec, Québec, Canada, <sup>3</sup>Guelph Food Research Centre, Agriculture and Agri-Food Canada, Guelph, Ontario, Canada, <sup>4</sup>Department of Health, Animal Science and Food Safety, University of Milan, Milan, Italy.

This study aimed to evaluate the impact of dietary feed additives with supplemental vitamins and trace minerals (DS) on the ileum and on systemic immune responses to a LPS challenge in low- (LW) and high-weight (HW) weaned piglets. At 20 ± 1 d of age (weaning), litters (12 piglet each) from 32 sows were randomly assigned to 4 dietary groups: control diet (CTL), CTL diet + chlortetracycline (ATB) or + DS, or DS + bovine colostrum instead of spray-dried animal plasma (DS-BC). Within each litter, 2 LW, 2 medium-weight (MW) and 2 HW piglets were identified. At 37 d, LW and HW piglets received intra-peritoneal LPS injection (200 µg). Piglets were bled before the injection (T0) and at slaughter, 4 h (T4; 1 LW and 1 HW) and 18 h (T18; 1 LW and 1 HW) post-injection. Microarray analysis was performed on LW, MW, and HW ileal tissues. The dietary feed treatments had no effect on the basal transcriptome level of the in MW piglets or on blood concentrations at T0 of tumor necrosis factor-α (TNF-α), IL-8, and IL-10. At T4, LW piglets secreted more TNF-α ( $P = 0.05$ ) and more (tendency) IL-8 ( $P = 0.11$ ) and IL-10 ( $P = 0.10$ ) than HW piglets. LPS altered the expression of 3,078 ileal genes with the false discovery rate (FDR) at 0.05.

There was no diet × LPS interaction on ileal gene expression on T4 vs T18 vs unchallenged MW piglets. However, a diet effect was observed on gene expression for LW piglets using fold change > 1.5 for the T4/MW ratio. Confirmed by QPCR, T4/MW in DS+BC ileal tissue of the apoptotic *CD163* gene was lower than in ATB's for LW piglets ( $P = 0.02$ ), whereas the corresponding effect in HW was a tendency ( $P = 0.07$ ). The T4/MW ratio of the acute phase (*SAA2* and *S100A12*), *SOD2*, and heat shock protein (*DNAJB1* and *HSP90AA1*) genes was also lower in DS than ATB ( $P < 0.05$ ) for LW piglets. The chemokine (*CXCL9*) T4/MW ratio was lower in both DS and DS+BC than ATB's ( $P < 0.02$ ) for LW and HW piglets. In conclusion, LW piglets developed a more pronounced inflammatory response than HW piglets and dietary feed additives supplemented with micronutrients attenuated the ileal gene response to LPS challenge.

**Key Words:** weaned swine feeding, gut health-enhancing feed additive, intestinal disease

**W113 Chitoooligosaccharide improves intestinal barrier function in the jejunum of weaning piglets.** Y. Yao\*, G. Tian, D. W. Chen, B. Yu, X. H. Song, J. Yu, P. Zheng, X. B. Mao, J. He, and Z. Q. Huang, *Animal Nutrition Institute, Sichuan Agricultural University, Chengdu, Sichuan, China.*

The recognition that chitoooligosaccharide (COS) possess potent anti-inflammatory, antioxidant and antimicrobial properties has prompted studies investigating its efficacy for animal growth and intestinal health. The objective of this study was to examine the effect of COS on growth and jejunal barrier function of weaning piglets. A total of 18 piglets (6.55 ± 0.32 kg) at 21 d of age, were randomly assigned to 1 of 3 groups (n = 6/group). Piglets in group 1 continued to be nursed by

sows, while piglets in groups 2 and 3 were weaned to a corn and soybean meal-based diet that was supplemented with 0 mL or 10 mL COS (5 ml/time, 2 times/d). At 28 d of age, growth performance, jejunal structure and epithelial integrity were examined. Data were analyzed by ANOVA procedure of SPSS. Differences between means were determined by using the Duncan multiple comparison method. The results indicated that final BW and ADG were markedly reduced in weaning piglets when compared with suckling piglets ( $P < 0.05$ ), while supplementation with COS restored ADFI, final BW and ADG in the weaned piglets ( $P < 0.05$ ). Weaning stress did not affect villous height but increased crypt depth and decreased the ration of villous height: crypt depth ( $P < 0.05$ ), whereas, this alteration can't be reversed by COS supplementation. Peripheral D-lactate concentration and diamine oxidase activities were not affected by weaning stress or COS supplementation. Weaning stress led to greater jejunal cell apoptosis index and sIgA content ( $P < 0.05$ ), and COS supplementation partially restored this changes ( $P < 0.05$ ). Jejunal gene abundance of occludin was not affected by weaning or COS supplementation, while the weaning-induced decrease in jejunal ZO1 ( $P < 0.05$ ) and ZO2 mRNA levels were elevated by COS supplementation ( $P < 0.05$ ). Collectively, our results support an important role of COS in regulating jejunal barrier function of weaning piglets.

**Key Words:** chitoooligosaccharide, weaning stress, piglet

**W114 Exogenous enzymes blend (DigeGrain Delta) improve growth performance and gut health of weaning pigs fed corn-soybean based diet in absence of antibiotic growth promoters.** Y. H. Kim<sup>1</sup>, S. L. Ingale\*<sup>2</sup>, P. C. Rathi<sup>2</sup>, S. H. Lee<sup>1</sup>, and B. J. Chae<sup>1</sup>, <sup>1</sup>College of Animal Life Sciences, Kangwon National University, Chuncheon, Gangwon-do, Republic of Korea, <sup>2</sup>Advanced Enzyme Technologies Ltd., Thane, Maharashtra, India.

The present study investigated the effects of dietary supplementation of an exogenous enzyme blend (DigeGrain Delta; a blend of amylase, protease, β-mannanase, xylanase and phytase) on growth performance, apparent total-tract digestibility (ATTD) of nutrients, and gut health of weaning pigs. A total of 192 weaned piglets (Landrace × Yorkshire × Duroc, initial body weight: 7.3 ± 0.7 kg) were randomly allotted to 4 treatments on the basis of body weight (4 pens per treatment with 12 pigs in each pen). The dietary treatments included corn-SBM based basal diet supplemented with 0 (control), 0.025, 0.050, or 0.075% exogenous enzyme blend. The experimental diets were fed in a meal form for 2 phases (d 0–14, phase I and d 14–28, phase II). Pigs fed dietary increasing levels of enzyme blend had linear improvement ( $P < 0.05$ ) in overall (d 0–28) average daily gain (ADG), gain:feed (G:F), and ATTD of dry matter, gross energy, and crude protein. Also, overall ADG and G:F of pigs fed diets supplemented with 0.050 and 0.075% enzyme blend were greater ( $P < 0.05$ ) than pigs fed the control diet. The ATTD of Ca and P was tended to increase ( $P < 0.10$ ) with dietary increase in enzyme blend. At d 21, pigs fed dietary increasing levels of enzyme blend had greater (linear,  $P < 0.05$ ) *Lactobacillus* spp. (ileum and cecum) and fewer (linear,  $P < 0.05$ ) *Clostridium* spp. (ileum and cecum) and coliform (ileum) populations. On d 28, villus height of the jejunum and ileum were linearly increased ( $P < 0.05$ ) with dietary increase in enzyme blend. In addition, crypt depth (duodenum and ileum) and villus height: crypt depth (duodenum, jejunum and ileum) tended to increase (linear;  $P < 0.10$ ) with increase in dietary level of enzyme blend. Results obtained in the present study indicates that dietary supplementation of amylase, protease, β-mannanase, xylanase, and phytase blend had potential to improve the growth performance, ATTD of nutrients, and

gut health of weanling pigs fed a corn-SBM based diet in the absence of antibiotics growth promoters.

**Key Words:** exogenous enzyme blend, performance, weanling pig

**W115 Endoplasmic reticulum stress pathway is involved in weaning stress induced jejunum cell apoptosis in piglets.** Y. Yao\*, G. Tian, D. W. Chen, B. Yu, X. H. Song, J. Yu, P. Zheng, X. B. Mao, J. He, and Z. Q. Huang, *Animal Nutrition Institute, Sichuan Agricultural University, Chengdu, Sichuan, China.*

Unresolved endoplasmic reticulum (ER) stress can lead to cell apoptosis. We therefore tested the role of ER stress in intestinal epithelial barrier disruption in piglets post-weaning. A total of 24 piglets (21 d of age and  $6.52 \pm 0.29$  kg) from 4 litters were randomly continued to be nursed by sows (suckling piglets, 3 piglets/sow) or weaned to a corn and soybean meal-based diet. On d 3 and 7 post-weaning, blood and jejunum tissue were collected at necropsy. Thus, this produced 4 experimental groups (management/time) as suckling/3 d, suckling/7 d, weaning/3 d and weaning/7 d ( $n = 6$ /group). Data were analyzed by ANOVA procedure of SPSS including management, time, interaction between management and time as the main factors. Differences between means were determined by using the Duncan multiple comparison method. Weaning stress induced significant elevations in plasma cortisol level, D-lactate content and diamine oxidase (DAO) activity ( $P < 0.001$ ). Moreover, this management-related increase in blood index was higher in 3 d than in 7 d ( $P = 0.03$  for cortisol,  $P = 0.03$  for D-lactate and  $P = 0.02$  for DAO). Enterocyte apoptosis index in jejunum was increased in weaning piglets ( $P < 0.001$ ), this alteration was accompanied by increasing jejunal gene expression of caspase3 ( $P = 0.03$ ), without any difference between piglets at different time. Weaning stress did not affect Bcl-2 expression but decreased caspase9 expression ( $P = 0.04$ ). Meanwhile, weaning stress upregulated mRNA expression of fas and caspase8 that were related to death receptor signal ( $P = 0.03$  and  $P < 0.001$ ). Marked weaning modifications were also observed for gene expression of ER stress pathway (glucose regulated protein 78kDa, +16%,  $P = 0.02$ ; eukaryotic translation initiation factor 2-subunit 1  $\alpha$ , +17%,  $P = 0.02$ ; activating transcription factor 4, +16%,  $P = 0.002$ ; x-box binding protein1, -19%,  $P = 0.004$ ; ER degradation enhance- mannosidase  $\alpha$  like 2, -11%,  $P = 0.10$ ). However, most of these genes were not affected by time or interaction between management and time. In conclusion, weaning stress induced jejunal enterocyte apoptosis and barrier disruption possibly via regulation of ER stress pathway and death receptor signal.

**Key Words:** endoplasmic reticulum stress, weaning stress, apoptosis

**W116 The expression of genes encoding gastrointestinal enzymes, microbial populations, and volatile fatty acids in pigs differing in feed efficiency.** Stafford Vigors\*, Torres Sweeney, Cormac J. O'Shea, and John V. O'Doherty, *College of Agriculture, Food Science and Veterinary Medicine, Dublin, Ireland.*

The objective of this study was to examine the effect of divergent selection for feed efficiency using residual feed intake (RFI) as a model on nutrient digestibility, bacterial populations, enzyme gene expression and volatile fatty acid (VFA) production. Male pigs ( $n = 75$ ; initial BW 22.4 kg) were fed a standard finishing diet (9.21 g/kg of standard ileal digestible lysine and 14 MJ/kg digestible energy) for 43 d before slaughter to evaluate feed intake and growth for the purpose of calculating RFI. Phenotypic RFI was calculated as the residuals from a regression model regressing average daily feed intake (ADFI) on average daily gain (ADG) and mid-test metabolic BW0.75 (MWT). Data were

analyzed using the GLM procedure of SAS. Sixteen pigs (85 kg, SEM 2.84 kg), designated as high RFI (HRFI;  $n = 8$ ) and 8 low RFI (LRFI) were killed and digesta was collected from the cecum and colon to determine microbial populations and VFAs. Digesta was collected from the ileum and rectum to measure coefficient of apparent ileal (CAID) and total-tract nutrient digestibility (CATTD). Mucosal scrapings from the duodenum, jejunum and ileum were used for RNA extraction and subsequent analysis of mRNA expression of several enzymes using RT-PCR. As expected LRFI pigs had lower ADFI (2.44kg vs. 1.87kg, + 0.07) and improved feed conversion ratio (1.96 vs. 2.48, SEM 0.02) than HRFI pigs ( $P < 0.001$ ) with no difference in ADG or MWT. The LRFI pigs had improved CAID of GE ( $P < 0.05$ ). Similarly, LRFI pigs had improved CATTD of GE, while also having improved CATTD of nitrogen and dry matter ( $P < 0.05$ ). In the jejunum RFI was correlated ( $r = -0.46$ ;  $P < 0.10$ ) with the gene expression of the enzyme sucrase-isomaltase which is located in the intestinal brush border cells. In the colon HRFI pigs had increased concentration of acetic acid ( $P < 0.05$ ). In the cecum LRFI pigs had increased molar proportions of butyric acid ( $P < 0.10$ ). LRFI pigs had increased cecal lactobacillus ( $P < 0.05$ ) compared with HRFI pigs. In conclusion differences in GE digestibility, sucrase-isomaltase gene expression, and populations of lactobacillus may contribute to differences in feed efficiency between RFI groups.

**Key Words:** feed efficiency, pig, RFI

**W117 Diets containing flaxseed-meal and oat hulls modulate fat digestibility, production, and excretion of bile acids and neutral sterols in growing pigs.** Saymore P. Ndou\*<sup>1</sup>, Elijah Kiarie<sup>1,2</sup>, Sijo J. Thandapilly<sup>3</sup>, Nancy Ames<sup>3</sup>, and Charles M. Nyachoti<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Manitoba, Winnipeg, MB, Canada, <sup>2</sup>DuPont Industrial Biosciences-Danisco Animal Nutrition, Marlborough, Wiltshire, United Kingdom, <sup>3</sup>Richardson Centre for Functional Foods and Nutraceuticals, Agriculture and Agri-Food Canada, Winnipeg, MB, Canada.

Effects of dietary fiber type on fat digestibility, intestinal and fecal concentrations of bile acids and plant sterols were determined. Forty-eight Genesus [(Yorkshire-Landrace)  $\times$  Duroc] barrows ( $25.0 \pm 0.32$  kg BW) were housed in pairs and assigned to 3 diets; corn-soybean meal-based diet (control), 12% flaxseed meal (FM)- and 10% oat hulls (OH)-containing diets. All diets were replicated 8 times in a completely randomized design. Titanium oxide was added in the diets as an indigestible marker. Diets were formulated to be iso-energetic; FM and OH were used as sources of soluble and insoluble fiber, respectively. The analyzed dietary NDF content (% as fed) was 9.4, 18.0 and 18.7% in the control, FH and OH, respectively. After 28 d of ad libitum feeding, fecal samples were collected from each pen, and then pigs were slaughtered to sample ileal and cecal digesta for the assessment of fat, bile acids and sterols concentrations. Fat digestibility in pigs fed FM-based diets was 24% lower ( $P = 0.02$ ) than the control, whereas that of OH-based diets fed pigs was intermediate. The ileal deoxycholic acid concentrations in pigs fed OH- and FM-containing diets were increased ( $P < 0.01$ ) by 46.3% and 30.1%, respectively, compared with the control. In the cecal contents, the total bile acids concentration was increased ( $P < 0.01$ ) by 114 and 75% in pigs fed FM- and OH-containing diets, respectively. The total bile acids excreted in feces of pigs fed FM- and OH-rich diets were respectively 2.47- and 2.03-fold higher than in control fed pigs. Compared with the control, intestinal bioconversion of cholesterol (coprostanol: cholesterol) was enhanced more in pigs fed FM- and OH-containing diets ( $P = 0.05$ ). Pigs fed FM- and OH-containing diets had higher cecal ( $P < 0.01$ ) and fecal ( $P = 0.002$ ) cholesterol concentrations than control fed pigs. In summary, soluble fiber reduces fat digestibility



more than insoluble fiber, and malabsorption of fat can be attributed to deconjugation of bile acids.

**Key Words:** dietary fiber, fat digestibility, bile acids

**W118 The gut and vaginal microbiota profile of pregnant sows and their contribution to neonatal piglet gut microbiota development.** Alessi A. Kwawukume\*, Hein M. Tun, Martin C. Nyachoti, and Ehsan Khafipour, *University of Manitoba, Winnipeg, Manitoba, Canada.*

The establishment of an initial gut microbiota is identified as a critical stage in neonatal development and is believed to influence health throughout life. Neonates are believed to receive their first bacterial colonizers while passing through the birth canal, from the environment as well as from milk during suckling; however, the relationship between maternal microbiota and development of neonatal gut microbiota is not clearly understood. To establish the contribution of maternal microbiota (gut and vaginal) to neonatal gut microbiota development, swab samples were taken from the posterior and anterior vagina 4 d before expected farrowing in pregnant sows housed on concrete slatted floors (CSF) (n = 12) and on straw (ASF) (n = 15). Fecal samples were obtained from sows at the same time as vaginal sampling, and from 5 piglets born to each sow (n = 135 piglets) on d 0, 1 and 3 after birth. DNA was extracted and subjected to V3-V4 bacterial 16S rRNA Illumina sequencing. Firmicutes (46.5%) and Bacteroidetes (24.8%) dominated the vaginal bacterial community of pregnant sows while Proteobacteria were in low numbers (14.5%). Among Firmicutes, Clostridia (34.6%) were found to predominate while Lactobacilli (3.8%) were few. No differences in  $\beta$ -diversity ( $P = 0.447$ ) of bacterial communities were observed between the anterior and posterior vagina in sows from the 2 housing types. Firmicutes and Bacteroidetes dominated sows fecal microbiota (48.7% and 37.13% respectively) with relatively low numbers of Proteobacteria (0.9%). Fecal samples of piglets differed in percent composition during the first 3 d after birth. Proteobacteria were found to be predominant on d 0 (63.8%), Firmicutes on d 1 (55.2%), and Firmicutes and Bacteroidetes on d 3 (60.1% and 15.15% respectively). Although piglet fecal microbiota was significantly different from both sow vaginal, and fecal microbiota ( $P = 0.001$ ), the principal coordinate analysis showed a moving trend of piglet microbiota toward the sow vaginal microbiota as piglets grew from d 1 to d 3. Results indicate that maternal microbiota may contribute to piglet gut microbiota development.

**Key Words:** piglet, sow, vaginal and fecal microbiota

**W119 Enteral bile acids modulate intestinal immune response and gut microbiota in early-weaned piglets challenged with LPS.** Alessandro Mereu\*, Nuria de Diego-Cabero<sup>2</sup>, Jose Javier Pastor Porras<sup>1</sup>, David Menoyo<sup>2</sup>, and Ignacio Ipharraguerre<sup>3,1</sup>, <sup>1</sup>*Lucta SA, Montornés del Valles, Barcelona, Spain*, <sup>2</sup>*Departamento de Producción Animal, Universidad Politécnica de Madrid, ETS Ingenieros Agrónomos, Madrid, Spain*, <sup>3</sup>*Institute of Human Nutrition and Food Science, University of Kiel, Kiel, Germany.*

Bile acids (BA) have recently emerged as regulators of intestinal immune homeostasis and mucosal integrity. We examined the effects of administering enterally deoxycholic acid (DCA) to early-weaned pigs challenged with lipopolysaccharide (LPS) on gut BA profile, immune response and microbiota. Twenty-four piglets were weaned at 21 d, acclimatized for 14 d, and subsequently grouped (n = 8) to be intragastrically infused with either deionized water (C+, C-) or 15 mg of DCA·kg<sup>-1</sup> initial BW (DCA) daily during 14 d. On d 28, C+ and

DCA piglets were injected i.p. with 150  $\mu$ g LPS·kg<sup>-1</sup> BW. Three h later, all animals were bled and killed for organ measurement and sampling. Blood samples were analyzed for endotoxin (EDT), interleukin (IL)-6, and tumor necrosis factor  $\alpha$  (TNF- $\alpha$ ). Expression of occludin, IL-6 and IL-10 genes and the concentration of individual BA were measured in the ileal mucosa. Colonic microbiota was characterized by sequencing bacterial 16S ribosomal-RNA. Individual BW and feed intake were recorded weekly. Data were analyzed with a mixed-effects model in which pig was treated as random effect and treatment as fixed effect. Compared with C-, LPS decreased total BA concentration ( $P < 0.01$ ) and increased plasma TNF- $\alpha$  ( $P < 0.01$ ) and EDT ( $P < 0.02$ ) as well as hepatic ( $P < 0.01$ ) and intestinal ( $P < 0.03$ ) weight. Interestingly, DCA infusion increased the proportion of BA with the greatest ability to induce BA-signaling pathways ( $P < 0.05$ ), prevented alterations ( $P > 0.10$ ) in plasma EDT and intestinal weight, decreased expression of IL genes ( $P < 0.04$ ) in the ileum, and enhanced ( $P < 0.05$ ) feed intake. In addition, treating pigs with DCA increased ( $P < 0.05$ ) *Peptostreptococcaceae* and *Clostridiaceae* whereas decreased ( $P < 0.05$ ) *Lactobacillaceae* in the colon. In conclusion, DCA acted locally to prevent LPS-induced inflammation and barrier disruption of the intestinal mucosa. These effects were associated with changes in the intestinal BA signature.

**Key Words:** deoxycholic acid, bile acid signaling, gut permeability

**W120 Impact of xylanases on gut microbiome of growing pigs fed with corn-based and wheat-based diets.** H. M. Tun\*, R. Li<sup>1</sup>, E. Kiarie<sup>1,2</sup>, M. Nyachoti<sup>1</sup>, and E. Khafipour<sup>1</sup>, <sup>1</sup>*University of Manitoba, Winnipeg, MB, Canada*, <sup>2</sup>*DuPont Industrial Biosciences-Danisco Animal Nutrition, Marlborough, Wiltshire, UK.*

This study was carried out to assess the effects of exogenous xylanases on production performance and gastrointestinal microbiota of growing gilts fed corn-based or wheat-based diets. A total of 96 individually penned gilts (22.7  $\pm$  0.65 kg initial BW) were used in a CRD experiment with 2  $\times$  6 factorial arrangement and fed experimental diets for 42 d. Gilts (n = 8 per treatment) received one of the 2 basal diets: (a) corn with 40% corn distillers dried grains, or (b) wheat with 25% wheat co-products, each without or with one of the 5 xylanases (XA, XB, XC, XD and XE). Xylanases were supplemented at 75 mg/kg of feed and all diets contained phytase. All xylanases were produced by different fungal organisms, except for xylanase-C, which originated from bacteria. On d 42, all pigs were euthanized to obtain ileal and cecum digesta for microbiome analysis using pyrosequencing of V1-V3 regions of 16S rRNA and downstream bioinformatic analyses using QIIME. Spearman's correlation was used to correlate microbiome composition data with performance characteristics, apparent total-tract digestibility (ATTD), and colon digesta volatile fatty acids (VFA) concentration. The XA yielded the best growth performance in both corn and wheat based diets ( $P < 0.05$ ) when compared with other xylanases. Based on PERMANOVA analysis on UniFrac distances, XA significantly affected the  $\beta$ -diversity of ileum microbiota compared with other xylanases and controls ( $P < 0.05$ ). Furthermore, xylanases influenced ATTD and colonic digester volatile fatty acids diet dependently ( $P < 0.05$ ). The XA increased *Lactobacillus* species in the cecum in both diets, whereas other xylanases drastically decreased *Lactobacillus* species when compared with control ( $P < 0.05$ ). In the wheat-based diet with xylanases, cecal bacterial community composition changes significantly correlated with ATTD of DM ( $r = -0.5$ ), gross energy digestibility ( $r = -0.49$ ) and colon VFA concentrations ( $r = -0.67$ ) ( $P < 0.02$ ). However, in the corn-based diet with xylanases, ileal bacterial community changes correlated with ATTD of DM ( $r = -0.4$ ) and gross energy digestibility ( $r = -0.43$ ), whereas cecal bacterial community changes correlated with

ATTD of fat digestibility ( $r = -0.58$ ) and colon VFA concentration ( $r = -0.41$ ) ( $P < 0.03$ ).

**Key Words:** pig, xylanases, gut microbiome

**W121 Effects of sulfur amino acids to lysine ratio on the expression of selected genes from piglets challenged with enterotoxigenic *Escherichia coli* K88.** Roseline Kahindi\*<sup>1</sup>, Alemu Regassa<sup>1</sup>, John Htoo<sup>2</sup>, and Martin Nyachoti<sup>1</sup>, <sup>1</sup>*University of Manitoba, Winnipeg, MB, Canada*, <sup>2</sup>*Evonik Industries AG*.

The sulfur AA (SAA; methionine and cysteine):Lys ratio for protein accretion is 55% for 7 to 11 kg pigs (NRC, 2012). The use of SAA to support immunity in addition to accretion of lean tissue may exert additional requirements in immune stimulated pigs. Moreover, due to negative whole body protein balance resulting from muscle wasting during an immune challenge, performance responses may not be suitable response criteria for determining SAA requirements. A study was conducted to determine the standardized ileal digestible (SID) SAA:Lys ratio of weaned pigs under an enterotoxigenic *Escherichia coli* (ETEC) challenge using gene expressions of methionine adenosyltransferase 1 and 2- $\alpha$  (MAT1A and 2A), 5-methyltetrahydrofolate-homocysteine methyltransferase (MTR), and cystathionine  $\gamma$ -lyase (CTH) as response criteria. Thirty-five [Duroc  $\times$  (Yorkshire  $\times$  Landrace)], 7-kg piglets were assigned to 5 dietary treatments in a completely randomized design. The corn-wheat-soybean-meal based antibiotic-free diets had graded SID SAA:Lys ratios (48, 54, 60, 66, and 72%) and 1.18% SID Lys. Feed was offered at 4% BW and fed twice a day at 08.00 and 16.00 h. Piglets had a 6-d diet adaptation period and were all orally challenged with 6 and 15 mL of ciprofloxacin-resistant ETEC K88+ ( $5 \times 10^9$  cfu/mL) on d 7 and 10, respectively. Blood samples were collected before and after challenge. On d 13 all pigs were killed to collect liver and ileal tissues for gene expression of MAT1A and 2A, MTR, and CTH using quantitative real-time PCR. Serum TNF- $\alpha$  concentration 6 h after inoculation was higher ( $P < 0.05$ ) than pre-challenge and was improved with increasing SAA:Lys ratio. Data were subjected to ANOVA using Proc Mixed of SAS 9.2. Increasing SAA:Lys ratio linearly decreased ( $P < 0.01$ ) liver MTR, while quadratically increasing ( $P < 0.01$ ) CTH and MAT1A gene expression. Ileal expression of MTR and MAT2A linearly increased, whereas, CTH and MAT2A were quadratically increased ( $P < 0.05$ ) with increasing SAA:Lys ratio. Maximum transmethylation, transsulfuration, and remethylation was at SAA:Lys ratio of 60% indicating that this ratio was enough to support piglets during an immune challenge.

**Key Words:** pig, *Escherichia coli* K88, SAA:Lys ratio

**W122 Messenger ribonucleic acid abundance of intestinal enzymes and nutrient transporters in chickens fed with vitamin D<sub>3</sub> and 1,25-dihydroxycholecalciferol.** Cristiane R. A. Duarte\*, Alice E. Murakami, Ana F. Q. M. Guerra, and Iván C. Ospina-Rojas, *Departamento de Zootecnia, Universidade Estadual de Maringá, Maringá, Paraná, Brazil*.

Vitamin D plays an essential role in regulating the bone formation and mineral balance in chickens and it is also related to functional development of the small intestine in rats. This study compared the effects of cholecalciferol and 1,25-dihydroxycholecalciferol on gene expression of vitamin D receptor, intestinal enzymes and nutrient transporters in chickens in the starter phase (1 to 21 d). A total of 1,008 1-d-old male Cobb chicks were used, distributed in a completely randomized factorial design 2  $\times$  3 (2 sources of vitamin D<sub>3</sub>: D<sub>3</sub> and 1,25(OH)<sub>2</sub>D<sub>3</sub>  $\times$  3 levels: 200; 950; 1,700 IU/kg feed) with 6 replicates and 28 birds

each one. Relative mRNA abundance of vitamin D receptor (VDR), aminopeptidase N (APN), maltase, sucrase-isomaltase (SI) complex, vitamin D-1- $\alpha$ -hydroxylase, sodium/glucose cotransporter 1 (SGLT1), glucose transporter 2 (GLUT2), peptide transporter 1 (PEPT1), sodium/phosphate cotransporter type IIb (NaPi-IIb), plasma membrane Ca<sup>2+</sup> ATPase (PMCA-1b) were assayed using real-time PCR. All data were analyzed by SAS GLM and differences among means were separated using Tukey's multiple range tests. There was an interaction ( $P < 0.05$ ) between sources and levels of vitamin D<sub>3</sub> only for mRNA abundance of SI. The mRNA abundance of maltase, NaPi-IIb, PMCA-1b, PEPT1 and GLUT2 were higher ( $P < 0.05$ ) in jejunal mucosa of animals fed with vitamin D<sub>3</sub>, while the mRNA abundance of 1- $\alpha$ -hydroxylase and SGLT1 were lower ( $P < 0.05$ ) in these animals. The levels of vitamin D (independently of source) affected the mRNA abundance of 1- $\alpha$ -hydroxylase, PEPT1 and SGLT1. The mRNA abundance of 1- $\alpha$ -hydroxylase in jejunal mucosa of animals fed with 200 IU/kg feed of vitamin D was higher ( $P < 0.05$ ) compared with 950 IU/kg feed. The mRNA abundance of PEPT1 was higher ( $P < 0.05$ ) with 200 IU/kg feed of vitamin D compared with other levels, while the mRNA abundance of SGLT1 was higher ( $P < 0.05$ ) in animals fed with 200 IU/kg feed of vitamin D compared with 1,700 IU/kg feed. It can be concluded that vitamin D and 1,25(OH)<sub>2</sub>D<sub>3</sub> can differently influence the gene expression of intestinal enzymes and nutrient transporters in chicken.

**Key Words:** cholecalciferol, gene expression, intestine

**W123 Early supplementation of alfalfa to starter diets altered electrophysiological properties and permeability of the gastrointestinal tracts in growing lambs.** Bin Yang\*<sup>1</sup>, Shan-shan Wang<sup>1</sup>, Bo He<sup>1</sup>, Jian-xin Liu<sup>1,2</sup>, and Jia-kun Wang<sup>1</sup>, <sup>1</sup>*Institute of Dairy Science, Hangzhou, Zhejiang, China*, <sup>2</sup>*Zhejiang University, Hangzhou, Zhejiang, China*.

To investigate the effects of early supplementation of starter pellets with alfalfa in pre-weaning period on the permeability, expressions of tight junction (TJ) proteins and cytokines of gastrointestinal tract during the pre- and post-weaning period, 6 of 66 male Hu lambs at the age of 10 d were slaughtered as control, and the other 60 lambs were randomly allocated to 2 dietary treatments: milk replacer and starter pellets without (STA) or with free-choice provision of chopped alfalfa (S-ALF). The animals were offered 300-g/d concentrate mixture and had free access to alfalfa after weaning at the end of wk 4 (age of 38 d). The plasma concentrations of D-lactic acid, lipopolysaccharides, IgA, IgM and IgG were measured by ELISA kit (RB). Ruminal epithelium and duodenal mucosa were subjected to Ussing chamber to measure the transepithelial electrical resistance (TER) and conductance. The mRNA expressions of TJ proteins and cytokines of gastrointestinal tract were determined with qRT-PCR. Results were analyzed as a completely randomized design. All data were analyzed by 2-way ANOVA using PROC GLM SAS (version 9.2; SAS Institute Inc., Cary, NC). Data of TER and conductance showed that alfalfa supplementation increased or decreased permeability of ruminal epithelium and duodenal mucosa numerically in pre- or post-weaning period, respectively, while the plasma concentrations and the morphological appearance of duodenum were not different between 2 treatments ( $P > 0.05$ ). Compared with STA group, alfalfa supplementation increased ( $P = 0.057$ ) the ruminal occludin expression, enhanced ( $P < 0.01$ ) the duodenal expression of claudin-1, and numerically enhanced ileal expression of claudin-1 and claudin-4 during the pre-weaning period, and inhibited ( $P < 0.05$ ) the weaning-caused compensatory increase in ruminal epithelial expression of claudin-4. The change in ruminal expression of TNF- $\alpha$  with weaning was similar to claudin-4. In summary, early supplementation of alfalfa

to starter diet could maintain the normal function of the gastrointestinal barrier, and helped to release the stress from weaning and feed transition in growing lambs.

**Key Words:** alfalfa, weaning, gastrointestinal tract

**W124 Heat-treated colostrum feeding promotes beneficial bacteria colonization in the small intestine of neonatal calves.** Nilusha Malmuthuge\*, Guanxiang Liang, Yanhong Chen, Laksiri Goonewardene, and Le L. Guan, *University of Alberta, Edmonton, AB, Canada.*

The feeding of heat-treated colostrum is a common practice to increase passive transfer of immunity and decrease enteric infections in calves. However, its influence on gut microbial colonization has not been well studied. The present study investigated the effect of heat-treated colostrum feeding on the bacterial colonization in calf small intestine, within the first 12 h of life. Newborn Holstein bull calves (n = 32) were removed from dams and fed with either fresh colostrum (FC, n = 12) or heat-treated colostrum (HC, n = 12) soon after birth, while the control (NC, n = 8) group did not receive colostrum. Small intestinal samples (tissue and contents) were collected from proximal jejunum, distal jejunum and ileum at 6 and 12 h after birth, following euthanasia.

Quantitative real time PCR was used to explore the colonization of total bacteria, lactobacilli, and bifidobacteria. The feeding of colostrum soon after birth increased total bacteria density (FC- $1.4 \pm 0.7 \times 10^{10}$  16S rRNA copy/g of sample, HC- $9.8 \pm 3.4 \times 10^9$  16S rRNA copy/g of sample) in calf gut within the first 12 h compared with NC ( $5.3 \pm 2.2 \times 10^8$  16S rRNA copy/g of sample). Although there were bacteria in FC ( $1.8 \times 10^3$  16S rRNA gene copy/mL), bacteria were not detectable in HC. In contrast, the prevalence of lactobacilli was lower in HC ( $0.007 \pm 0.004\%$ ) and FC ( $0.014 \pm 0.008\%$ ), comparing to NC ( $0.031 \pm 0.015\%$ ). Remarkable changes in the prevalence of small intestinal tissue-attached bifidobacteria were observed with the feeding of HC soon after birth, but not that in gut contents. The prevalence of bifidobacteria was 3.2-fold and 5.2-fold higher in HC ( $28.6 \pm 10.3\%$ ) than FC ( $8.9 \pm 2.5\%$ ) and NC ( $5.5 \pm 1.5\%$ ), respectively, at 6 h. The feeding of FC increased the prevalence of tissue-attached bifidobacteria gradually, reaching to that of HC at 12 h. Thus, the present study suggests that feeding of HC enhances the colonization of bifidobacteria on small intestinal tissue immediately postpartum compared with that of FC and NC, which may prevent the colonization of enteric pathogens, and thereby decrease enteric infections in neonatal calves.

**Key Words:** newborn calves, gut colonization, bifidobacteria



## Dairy Foods: Cheese

**W125 Determining economic feasibility for artisan cheese companies.** Cathy Durham<sup>2</sup>, Andrea Bouma<sup>1</sup>, and Lisbeth Goddik\*<sup>1</sup>, <sup>1</sup>*Department of Food Science, Oregon State University, Corvallis, OR*, <sup>2</sup>*Food Innovation Center, Oregon State University, Portland, OR*.

Artisan cheese makers lack access to valid economic data to help them evaluate business opportunities and make important business decisions such as determining cheese pricing structure. An economic model was developed in Excel following close collaboration with current and future artisan cheese companies. The objective of this study was to utilize this economic model to evaluate the net present value (NPV), internal rate of return, and pay back period for artisan cheese production at different annual production volumes for a given cheese type. The model is also used to determine the minimum retail price necessary to assure positive NPV for 5 different cheese types produced at 4 different production volumes. These 2 scenarios demonstrate important business considerations facing artisan cheese makers. For example, a small size cheese maker with annual production volume at 3,401 kg (7,500 lb) cannot be economically viable (negative NPV) if selling cow milk Gouda for \$48.50/kg (\$22/lb); by doubling the production size, the business would obtain a positive NPV. Due to differences in cheese yield, investment in aging facility, labor required during aging, and raw milk purchase price, fresh cow milk cheeses such as fresh mozzarella can be sold for about half the price of hard, aged, goats' milk cheeses at the largest volume or about 2-thirds the price at the lowest volume examined. For example, for the given model assumptions, at an annual production of 13,608 kg cheese (30,000 lb), a fresh cows' milk mozzarella should be sold at a minimum retail price of \$27.29/kg (\$12.38/lb) while a goats' milk gouda needs minimum retail price of \$49.54/kg (\$22.47/lb) for the business to have NPV at or above zero. The model is utilized within the OSU Extension program and has gone through 2 major updates. The observations derived from the model are consistent with the current business situation for artisan cheese companies.

**W126 Effect of *terroir* for raw and pasteurized milk Cheddar on nonstarter lactic acid bacteria.** Christopher Baird, Lisbeth Goddik\*, Gregory Turbes, Elizabeth Tomasino, Juyun Lim, and Joy Waite-Cusic, *Department of Food Science, Oregon State University, Corvallis, OR*.

*Terroir* is a connection to the land and producer that influences the organoleptic properties in many products including cheese. Non-starter lactic acid bacteria (NSLAB), which are present in the environment, are thought to be one driving component of *terroir* in cheese. The effect of *terroir* was explored through differences in NSLAB between Cheddars made from different milk source locations. The effects of heat treatment on the importance of *terroir* on NSLAB present in cheddar was also investigated. Cheddar was produced with raw and low-temperature long-time (LTLT) pasteurized milk at Oregon State University. Milk was sourced from 3 individual farms, and 2 commingled sites in different eco-regions of Oregon. All milk was collected within 5 weeks while the Jersey herds were on a pasture-based diet. Cheddar was aged at 5°C and 2 samples per cheese were extracted at 5 and 9 mo. Samples were homogenized and grown anaerobically on MRS at 30°C for 48 h. Five random colonies were selected per for further identification. Isolates were speciated using API50 fermentation test kits. At 5 mo, the majority of isolates identified in raw and LTLT pasteurized cheddar were *Lactobacillus paracasei* and *Lactobacillus plantarum* respectively. Fermentation patterns between similar identifications showed

wide variation at 5 mo with no overlap between regions. *L. paracasei* remained the dominant NSLAB in raw cheeses after 9 mo of aging. Each Cheddar showed reduced variety in NSLAB fermentation patterns at 9 mo. The unique fermentation patterns suggest that milk source location influenced the NSLAB profile of Cheddars. Further variability of isolates will be performed by pulsed field gel electrophoresis (PFGE). The NSLAB profile of cheddar is a reflection of the milk source location. This connection between NSLAB and location demonstrates one facet of *terroir* that affects Cheddar.

**Key Words:** nonstarter lactic acid bacteria, Cheddar, *terroir*

**W127 Effect of *terroir* on flavor for raw and pasteurized milk Cheddar.** Gregory Turbes, Lisbeth Goddik\*, Christopher Baird, Juyun Lim, Joy Waite-Cusic, and Elizabeth Tomasino, *Department of Food Science, Oregon State University, Corvallis, OR*.

*Terroir* is a term that in the United States has come to be known as "taste of place." It builds off the idea that a food's organoleptic properties are defined by the environment, climate, and production practices. The effect of *terroir* and heat treatment effect on *terroir* to Oregon Cheddar flavor were explored. Cheddar was produced with raw and low-temperature long-time (LTLT) pasteurized milk at Oregon State University. Milk was sourced from 3 individual farms, and 2 commingled sites in different eco-regions of Oregon. Dairy farms were selected with similar herd management styles. Collection of milk occurred within a 5-week period while the Jersey herds were on a pasture-based diet. Cheddar was aged at 5°C and 2 samples per cheese were extracted at 5 and 9 mo of aging. Flavor compounds were analyzed using gas chromatography-mass spectrometry (GCMS). A total of 54 flavor compounds were detected. At 5 and 9 mo 46 and 30 flavor compounds were identified respectively, consisting of acetates, alcohols, aldehydes, alkanes, alkenes, esters, fatty acids, ketones, lactones, nitrogen compounds, sulfur compounds, and terpenes. Principal component analysis (PCA) was performed for all samples. At 5 mo, samples separated based on milk source location and at 9 mo samples separated based on heat treatment. Results suggest that the flavor of Cheddar is affected by both *terroir* and LTLT pasteurization; 5 mo cheddar flavor is best characterized by *terroir* and 9 mo Cheddar flavor is best characterized by heat treatment. This connection between Cheddar flavor and milk source location suggest that *terroir* is part of what characterizes a Cheddar.

**Key Words:** flavor, Cheddar, *terroir*

**W128 The effects of *terroir* and heat treatment on consumers' perception of Cheddar cheese flavor.** Gregory Turbes, Lisbeth Goddik\*, Tyler Linscott, Elizabeth Tomasino, Joy Waite-Cusic, and Juyun Lim, *Department of Food Science, Oregon State University, Corvallis, OR*.

The objective of this research was to investigate the effect of *terroir* (geographical location of milk source) on Cheddar cheese flavor. Milk from 5 locations, including single dairy farms and commingled sites, were collected from around the state of Oregon: 3 individual dairy farms from the mid-valley and 2 commingled sites at coastal and high desert plateau. These farms/sites were selected based on their representation of Oregon's dairy industry as well as similarities in herd and herd management characteristics. Using raw and pasteurized counterparts of the standardized milk, Cheddar cheese was made and aged. At 5 and 9 mo

into aging, Cheddar cheese consumers were asked to sort the samples based on perceived similarity/dissimilarity of cheese flavor and also to describe the characteristics of each cheese groups. Grouping data were subjected to multidimensional scaling and subsequent cluster analysis. Correspondence analysis was also performed on consumer descriptions as a simplified exploratory approach to discover underlying characteristics. Results at 5 mo into aging revealed that consumers could differentiate cheese samples based on dairy farm origin, while cheeses produced from commingled milk samples were perceived to be similar. At 9 mo into aging, consumers differentiated the cheeses based on heat treatment; that is, raw vs. pasteurized milk cheeses. These data suggest that for younger cheeses, the geographical location of the milk source has a significant effect on the flavor of Cheddar cheese but that the practice of milk commingling may overcome the effects of geographical location. Thus we can source cheese flavor to individual farms and thereby demonstrate the existence of *terroir*. In contrast, the older cheeses are primarily characterized by their history of heat treatment.

**Key Words:** *terroir*, cheese, sensory

**W129 Sensory and functional properties of cheese across three storage temperatures.** Ni Cheng<sup>\*1</sup>, P. D. Gerard<sup>2</sup>, and M. A. Drake<sup>1</sup>, <sup>1</sup>*Southeast Dairy Foods Research Center, North Carolina State University, Raleigh, NC*, <sup>2</sup>*Dept. Mathematical Sciences, Clemson University, Clemson, SC*.

Cheese quality influences sales and applications. Storage time and temperature affect cheese quality and may play a crucial role in cheese designated for export markets as optimum quality must be maintained. A mild flavor profile is expected in export markets concurrent with optimum functional properties. The objective of this study was to evaluate sensory and functional properties of 4 cheese types across different storage temperatures. Triplicate lots (18 kg blocks) of cheeses (Cheddar, Monterey Jack, Mozzarella and Gouda) were obtained from 2 commercial facilities and assigned to 4°C, -1°C and -18°C storage. Cheeses were evaluated every 3 mo through 1 year storage. At each time point, sensory evaluation (flavor and texture) and functional tests (moisture, pH, shreddability, sliceability, color, melting, viscosity, and fork stretch) were conducted. Two-way ANOVA (storage time, temperature) as well as multivariate analyses were applied to evaluate the data. Time and storage temperatures had no practical effects on cheese composition or color ( $P > 0.05$ ). Storage time increased shreddability and melt and decreased sliceability, viscosity and fork stretch values ( $P < 0.05$ ). Storage at -18°C resulted in smaller increases in shreddability and melt than -1°C or 4°C. There were no differences in shreddability and melt properties for cheeses stored at 4°C or -1°C ( $P > 0.05$ ). Across storage time, cheeses increased in intensities of typical aged cheese flavors (sulfur and brothy), cheeses stored at lower temperatures maintained young/undeveloped cheese flavors (cooked, whey, milkfat and diacetyl) longer than cheeses stored at 4°C. Texture properties by sensory analysis were unchanged through 6 mo storage, regardless of temperature. These results indicate that a lower temperature (-1°C) may be applied to cheeses for up to 6 mo to maintain sensory quality (young, mild flavor) without detrimental effects on functional properties.

**Key Words:** cheese, storage, quality

**W130 Microstructural arrangements observed using electron microscopy during the manufacture of cheese and the influence of cheese pH.** Almut H. Vollmer<sup>\*</sup>, Nabil N. Youssef, and Donald J. McMahon, *Western Dairy Center, Utah State University, Logan, UT*.

It is known that the texture of cheese changes as a function of pH. Our objective was to determine the differences in the arrangements of protein and fat between a medium pH cheese (Cheddar cheese) and a low pH cheese (feta cheese). Cheese was made using 16-kg aliquots of milk and the make procedure modified to produce cheddar cheeses with a range of pH values as well as making a feta cheese. Samples of curd were collected throughout the cheese making processing, starting from before cutting the curd and during cheese storage. Samples were fixed in glutaraldehyde, then dehydrated, heavy metal stained, embedded and sectioned, and multiple fields observed using transmission electron microscopy. During initial curd formation the characteristic fine stranded network of aggregated casein micelles was observed with fat globules entrapped between the protein strands. As the cheese manufacture continued, and the curd particles contracted and whey was expelled, the initial protein strands apparently coalesced into thicker strands and the casein micelles tended to lose their individual identity. This same general pattern continued throughout manufacture and into the storage period. After 3 mo of aging the cheddar cheeses ranged from pH 4.9 to pH 5.2. There were variations in protein arrangements observed within individual cheese samples as is typical in microstructure of dairy foods. No observable differences in structural arrangements outside this natural variability were apparent between the Cheddar cheese of lowest pH to the highest pH. In contrast, the microstructure arrangements of the proteins in feta cheese were very different to those in Cheddar cheese. In feta cheese, there is a more openness present in the protein network with the appearance of the protein regions having a looser nature; that is, not having large coherent regions of connected protein. This was related to feta cheese having a pH below that at which the caseins are acid precipitated and as they become less soluble in the surrounding serum phase of the cheese. These differences in protein arrangements observed using electron microscopy helps explain the crumbly texture associated with low pH rennet-set cheeses.

**Key Words:** Cheddar, feta, microstructure

**W131 Effect of salting pH, salting rate, and stretching temperature on proteolysis in Mozzarella cheese.** Ananya C. Biswas<sup>\*1</sup>, Anil Kommineni<sup>1</sup>, Praveen Upreti<sup>2</sup>, and Lloyd E. Metzger<sup>1</sup>, <sup>1</sup>*Dairy Science Department, South Dakota State University, Brookings, SD*, <sup>2</sup>*Nestle R&D Center Inc., Solon, OH*.

The level of proteolysis during refrigerated storage of Mozzarella cheese can have an effect on functionality including stretching and melting characteristics. Proteolysis in Mozzarella cheese could be influenced by several manufacturing parameters including: salting pH (SP), salting rate (SR), and stretching temperature (ST). The objective of this study was to evaluate the effects of SP, SR and ST on Mozzarella cheese proteolysis during storage. Thirty-two batches of Mozzarella cheese were manufactured using a partial split plot design that included a range of protein-to-fat ratios (1 to 4.5) in the main plot. In the sub plot, 2 salting pH (5.2 and 5.6), 2 salting rates (1.5% and 2.2%), and 2 stretching temperatures (65°C and 85°C) were used. After manufacture Mozzarella cheese was vacuum packed and stored at 4°C for 28 d. After storage, proteolysis was determined by measuring the soluble protein at pH 4.6 as a percentage of total protein content. Proteolysis and composition data were analyzed using simple linear regression analysis to establish potential relationships among the cheese compositions (moisture, fat, and protein), SP, SR, ST and proteolysis. Linear regression analysis indicated that proteolysis was primarily influenced by ST and cheese manufactured with the high ST always resulting in less proteolysis as compared with the low ST. Subsequently the GLM procedure of SAS was utilized to conduct an ANOVA. The results determined that milk

fat and protein content, and ST had a significant effect ( $P < 0.05$ ) on cheese proteolysis, whereas no significant effects were observed for SP and SR. These results demonstrate that in this study milk composition and stretching temperature were the primary factors influencing proteolysis of mozzarella cheese. This information can be used by cheese manufacturers to control the level of proteolysis in Mozzarella cheese.

**Key Words:** milk protein-to-fat ratio, Mozzarella cheese, proteolysis

**W132 Effect of selenium supplementation on Se status and Mozzarella quality in dairy cattle feed.** Weizheng Zhu, Hongyun Liu\*, Daxi Ren, and Jianxin Liu, *Institute of Dairy Science, College of Animal Science, Zhejiang University, Hangzhou, Zhejiang, China.*

Selenium (Se) is an important trace element and its deficiency has been associated with human and animal health. The human recommended dietary allowance (RDA) for adult Se intake is about 55 µg per day; however, many places in the world are lower than that level so need a dietary supply. The objective of this study was to research the effect of feed Se supplementation on Se status in milk and mozzarella cheese quality. Thirty multiparous Holstein dairy cows (3 groups) with similar condition were raised in local dairy farm. First group of dairy cow was fed with selenium yeast (0.5 mg Se/kg dry matter), second group was fed with sodium selenite at the same Se concentration, third group without Se supply as control. After 2 mo feeding, milk samples were collected for Se analysis and cheese-making. Data were analyzed by one-way ANOVA using SAS 9.3. Results showed that the Se content in milk was increased to 47.9 µg/L by selenium yeast group, significantly higher than the 25.6 µg/L in sodium selenite group when both supplied with 0.5 mg Se/kg DM. The fat, casein and whey protein of Se-milk were separated by ultracentrifuge and the Se content was determined by atomic absorption spectroscopy. The results were 9.82%, 45.56% and 44.62% respectively. When Se-enriched mozzarella cheese was made from high Se-milk and compared with control, no significant difference was found in composition or texture. But the functional properties (meltability, flowability and stretchability) were better in Se-cheese after 8 weeks storage. Additionally, the pH and  $a_w$  were lower in Se-cheese, leading to lower total plate count. The Se content increased more than 4 times in mozzarella cheese than milk, and additional Se was found in whey, hot water and brine collected during cheese-making. Organic and inorganic Se was found in Se-cheese after 8 weeks storage. Most of the Se-peptide produced during the storage was SeMet (215 ng/g) and SeCys (123 ng/g). The results of this study showed that the Se content in milk and cheese can be increased by dietary Se supplement, and the mozzarella cheese quality was not affected by feed Se supply.

**Key Words:** selenium status, Mozzarella cheese

**W133 Microbiological characteristics and mineral content of probiotic low sodium Minas cheese with added arginine.** Taissa Felicio, Marcia Cristina, Renata Raices, Luciana Nogueira, and Adriano Cruz\*, *Federal Institute of Science and Technology of Rio de Janeiro (IFRJ), Rio de Janeiro, Brazil.*

The objective of the present study was to develop technology for the production of probiotic fresh Minas cheese with a reduced sodium content and the addition of arginine, and evaluate any changes in the quality parameters occurring throughout the shelf life. Gross composition and mineral content (moisture, protein, lipid, sodium, calcium and potassium). With the exception of the lipid content, differences were observed between the treatments during the storage period ( $P > 0.05$ ) for all the parameters evaluated. Cheese QIII (50/50 NaCl/KCl reduction

plus 1% w/w arginine) showed the highest moisture content (variation of 60 to 61.6% w/w,  $P < 0.05$ ) and a progressive decrease in protein during storage (variation from 17.1 to 16.6% w/w,  $P < 0.05$ ). With respect to the sodium and potassium levels, there was a progressive reduction in sodium in relation to the substitution level, that is, between the treatments, varying from 315 to 275 mg/100g (Qc, 100% NaCl) and from 193 to 215 mg/100g (QIII, 50/50 NaCl/KCl plus 1% arginine,  $P > 0.05$ ). With respect to the calcium content, the addition of arginine resulted in a higher calcium content with an average value of 735.3 mg/100g (cheese QIII), whereas the value for the cheese containing only NaCl was 587.3 mg/100g ( $P < 0.05$ ). Similarly, the concentration decreased throughout storage (variation from 591 to 566 mg/100g for cheese Qc with 100% NaCl, and from 744 to 660 mg/100g for QIII, with 50/50 NaCl/KCl added with arginine. The values observed were relevant and considerably lower than those reported on the labels of fresh Minas cheeses available on the Brazilian market, which show mean values of 437.4 mg/100g and in this context probiotic cheese with arginine seems an interesting option to functional cheese market.

**Key Words:** probiotic cheese, arginine, low sodium cheese

**W134 Probiotic Minas Frescal cheese with *Lactobacillus casei* Zhang.** Ramon Silva, Marcia Cristina, Luciana Nogueira, Renata Raices, and Adriano Cruz\*, *Federal Institute of Science and Technology of Rio de Janeiro (IFRJ), Rio de Janeiro, Brazil.*

The addition of *Lactobacillus casei* Zhang in the manufacture of Minas Frescal cheese was investigated. Minas Frescal cheeses were produced by enzymatic coagulation and direct acidification, supplemented with probiotic bacteria (*L. casei* Zhang) and subjected to microbiological (probiotic and lactic bacteria counts) and physicochemical analysis (pH, proteolysis, lactic acid, and acetic acid level). Adequate *L. lactis* and *L. casei* Zhang counts were observed in both cheeses, and as previously discussed, the highest values were observed in all cheeses throughout the refrigerated storage (about 8 log cfu / g for *L. lactis*, ranging from 8.14 to 9.02 log cfu / g,  $P < 0.05$ , respectively). However, the addition of *L. casei* Zhang provided lower pH and high proteolysis indexes during storage (from 5.38 to 4.94 and 0.470 to 0.702,  $P < 0.05$ , respectively), and these changes were more significant in the cheese produced by enzymatic coagulation. In addition, with respect to the organic acids, regardless the treatment the addition of *L. casei* Zhang led to higher lactic acid and acetic acid levels (ranging from 1.75 to 2.98 and 0.45 to 0.84 µg/mL,  $P < 0.05$ ). Consumer test reported improved overall liking for the control sample produced by direct acidification, (7.83,  $P < 0.05$ ), while the cheeses containing *L. casei* Zhang presented lower values for all sensory attribute. The optimization of *L. casei* Zhang dosage during the manufacturing of probiotic Minas Cheese should be performed.

**Key Words:** probiotic cheese, *Lactobacillus casei* Zhang

**W135 Effect of inulin and lactulose on characteristics of Iranian probiotic ultrafiltration feta cheese.** Maryam Enteshari<sup>1</sup>, Mahshid Azizi\*<sup>2</sup>, Bahram Jirsaraei<sup>3</sup>, and Mohammadreza Dolatkhahnejad<sup>4</sup>, <sup>1</sup>Department of Food Science and Technology, Shahid Beheshti University of Medical Sciences, Tehran, Iran, <sup>2</sup>Islamic Azad University of Birjand, Chemical Engineering and Food Industries Department, Tehran, Iran, <sup>3</sup>Azad University of Varamin, Faculty of Agriculture, Department of Food Science and Technology, Tehran, Iran, <sup>4</sup>Islamic Azad University of Ayatollah Amoli, Food Industries and Scientific Engineering Department, Tehran, Iran.



Nowadays, consumers' demands for foods with health effect, desirable organoleptic and mouth feel characteristics, low-caloric and low fat content have greatly increased. Meanwhile, consumption of synbiotic products (simultaneous presence of probiotic and prebiotic) has revealed functional properties. In this study, effects of prebiotic compounds including inulin (I), lactulose (L), and inulin-lactulose (IL) on physicochemical, sensorial and survival of *Lactobacillus casei* in ultrafiltration (UF) probiotic cheese in different storage days (1, 15, 30, 45 and 60) have been evaluated. Obtained results revealed that changes in pH values for all of cheese samples during storage were significant with a descending trend; the pH of IL sample declined significantly from initial value of 4.71 to 4.48 at the end of storage ( $P < 0.05$ ). In other ways, amounts of acidity for all of cheese samples increased significantly during storage ( $P < 0.05$ ); at the initial and final days of storage, the IL sample exhibited the highest acidity values (1.66 and 2.03 g/100g, respectively) while the L sample presented the lowest ones (0.98 and 1.23 g/100g, respectively). At the first and final days of storage time, the highest (65.11 and 64.8%, respectively) and lowest amounts of moisture content (63.27 and 63.12%, respectively) pertained to the C (control) and I samples, correspondingly. The cheese samples of I and IL presented the highest sensory scores. At the end of storage, the highest numbers of *Lactobacillus casei* were observed in the samples of I and L (in the range of 7.83–7.9 Log cfu/g) and the lowest one belonged to the C sample (6.63 Log cfu/g). Therefore, in this research the final synbiotic cheese which contained more than  $10^7$  cfu/g viable *Lactobacillus casei* at the end of storage, is being proposed as probiotic product and the inulin or lactulose prebiotic compounds in the single mixture (I or L samples) have protective effect on the survival of *Lactobacillus casei*.

**Key Words:** ultrafiltration cheese, probiotic, prebiotic

**W136 Volatile compounds formation in Prato cheese.** Lígia Dozena Domingos, Lilian Regina Barros Mariutti, Neura Bragagnolo, and Walkiria Hanada Viotto\*, *University of Campinas (UNICAMP), Campinas, São Paulo, Brazil.*

Prato cheese is a washed curd and semi-hard cheese produced from pasteurized cows' milk acidified by a mesophilic starter containing citrate-positive bacteria, and matured for a minimum period of 25 d. Volatiles play an important role in flavor perception of cheese and they are result of compounds formed by glycolysis, citrate metabolism, proteolysis and lipolysis. The aim of this work was to evaluate the formation of volatile compounds in Prato cheese after 30 d of refrigerated storage at 12°C. The compounds were determined by gas chromatography-solid phase micro extraction coupled to a mass spectrometer detector (GC-SPME-MS). Two grams of a mixture of cheese sample and sodium phosphate saline solution (1:3, w/v) were incubated in a sealed 20 mL vial for 15 min at 40°C and after that, the DVB/CAR/PDMS fiber was exposed to the headspace for 30 min. The fiber was desorbed in the injector (splitless mode) at 250°C for 1 min. Compounds were separated in a RTX-Wax (30 m × 0.25 mm × 0.20 µm, Restek). Electron ionization source was set at 70 eV and 250°C. MS data were acquired in SCAN mode ( $m/z$  35–350). Identification was carried out by comparison with mass spectra of authentic standards or Wiley 9 library and Kovats retention index. Volatile compounds found were organic acids ( $C_{4:0}$  to  $C_{10:0}$ ), esters (ethyl butyrate, ethyl hexanoate), ketone (2-pentanone, 2-heptanone, 2-undecanone), alcohols (ethanol, 1-pentanol, 1-butanol, 1-hexanol, 3-methyl-1-butanol, 1-heptanol, 1-octanol) and aldehydes (hexanal, heptanal, nonanal, decanal, benzaldehyde) as a result of cheese glycolysis, lipolysis and proteolysis. From the citrate metabolism, we

found acetoin, 2,3-butanediol, 2-butanone and diacetyl, which is associated with characteristic butter flavor of Prato cheese.

**Key Words:** cheese ripening, GC-MS, flavor compound

**W137 Consumer perception of reduced-fat fresh cheese in Brazil.** Juliana Zara Brondi Mendes, Jorge Herman Behrens, and Walkiria Hanada Viotto\*, *University of Campinas (UNICAMP), Campinas, São Paulo, Brazil.*

Minas Frescal, the most consumed reduced-fat cheese in Brazil, is a fresh and soft white cheese produced by the enzymatic coagulation of pasteurized milk, with high moisture content (>55%). It is industrially produced by 3 different processes that result in cheeses with different sensory characteristics, particularly texture. Cheeses manufactured by the traditional process (addition of starter) or direct acidification (by replacing the starter by lactic acid) have firmer texture than those produced by ultrafiltration which presents a gel texture, due to the incorporation of whey proteins. This work aimed to assess Brazilian consumers' perceptions regarding reduced-fat Minas Frescal cheese manufactured by three different technological processes (traditional, direct acidification, and ultrafiltration). A grocer's shop and a consumer research center in São Paulo, Brazil, served as settings for consumer test with 151 individuals. Respondents were characterized by a demographic questionnaire and purchasing habits. Conjoint analysis based on complete profile was conducted with eight different product concepts derived from a factorial design of relevant variables in local consumers' choices: texture (firm or soft), size of the pack (family or individual) and nutritional information (light or light + 40% less fat). The majority of consumers participating in the study were female, on high income and over 35 years old, and the main reason reported for consuming light cheeses was the concern for health. Conjoint Analysis revealed that texture (41.1%) was the most important attribute followed by portion size (36.2%) and nutritional information (22.7%). Consumers positively rated the firm texture, the individual portion, and the "light + 40% less fat." These results favor cheeses obtained by traditional process or direct acidification, due to their characteristic firmness that leads to higher purchase intention by the consumers. Findings of this study can assist cheese industries in adjusting process conditions, labels and packaging in order to enhance local consumers' willingness to purchase reduced fat cheese.

**Key Words:** fresh cheese, consumer, purchase intention

**W138 Effect of myrrh essential oil as a highly effective antimicrobial agent in processed cheese spreads.** A. G. Mohamed\*, J. M. Kassem, and H. M. Abbas, *National Research Centre, Dokki, Giza, Egypt*

There is a new approach to prevent the proliferation of food microorganisms using of plant essential oils. *Commiphora myrrha* is considered as highly effective, natura, and safe antimicrobial. Therefore, the current study aimed to evaluate the effects of *Commiphora myrrha* on (1) pathogenic positive bacteria including *Listeria monocytogenes*, *Staphylococcus aureus*, and *Bacillus cereus* as well as gram-negative bacteria including *Escherichia coli* O157:H7 and *Salmonella typhimurium*, and (2) the sensory properties of processed cheese spreads at 5 different levels of usage. Moreover, processed cheese spread (PCS) samples were prepared by using 5 different ratios of *Commiphora myrrha* essential oil at 0.2, 0.4, 0.6, 0.8, 1 w/w to evaluate its organoleptic acceptability. The PCSs was made by heating and mixing the ingredients at 85°C for 10 min. Gross chemical composition and some physical characteristics were estimated through 3 mo of storage at  $5 \pm 1^\circ\text{C}$ . Oil separation, penetration,

melting point and color parameters were also determined through the storage period. Results showed that all tested microbes were susceptible to *Commiphora myrrha* at minimum inhibitory concentration of 2–5  $\mu\text{L}$  of oil/mL. Moreover, results showed that using 0.2% (w/w) *Commiphora myrrha* essential oil gave satisfactory sensory properties. The appearance was shiny; gumminess and oil separation were absent. The overall preference of preferable ratio was “like very much” comparing to the other treatments. Color was more acceptable without significant differences ( $P > 0.05$ ) and it spread well compared with control. The penetration values were 33.5, 32.0 and 31.2 mm versus 33.0, 30.5 and 26.5 mm for treated PCS versus control PCS ( $P > 0.05$ ) when fresh, and after 1 mo and 3 mo of storage. Moreover, meltability 85.4, 81.6 and 80.0 mm versus 81.6, 80.5 and 78.7 mm for treated PCSs compared with control when fresh, and after 1 mo and 3 mo of storage. *Commiphora myrrha* essential oil can be used successfully as antimicrobial agent during preparing of PCS with acceptable properties and satisfied sensory behavior.

**Key Words:** *Commiphora myrrha*, processed cheese spread, antimicrobial agent

**W139 Effects of storage temperature and period on textural properties of cholesterol reduced and control Cheddar-type goat milk cheeses.** Binod P. Gupta, Brittany I. Davis, Krishna P. Bastola\*, Jolethia O. Jones, and Young W. Park, *Fort Valley State University, Fort Valley, GA.*

Consumption of fat and cholesterol reduced food products has been increasingly popular among health conscientious consumers due to its association with health problems including heart diseases and stroke. The objective of this study was to evaluate textural properties of cholesterol reduced (CR) and control (CC) caprine milk cheeses during storage. Three batches each of CR and CC Cheddar-type caprine milk cheeses were manufactured, and stored at 3 different temperatures (–18, 4 and 22°C) for 0, 3 and 6 mo.  $\beta$ -Cyclodextrin ( $\beta$ -CD) was used to remove cholesterol from milk and cream before manufacture of the CR cheeses. Textural properties of all experimental cheeses were evaluated using a texture analyzer (Model TA.XT2i, Texture Technology Corp., Scarsdale, NY). The pooled means (g force) of hardness, adhesiveness, resilience, cohesiveness, springiness, gumminess, and chewiness across batches, storage temperatures and periods of CC and CR cheeses were: 851.1, 2750.1; 11.4, 0.92; 19.3, 46.6; 0.45, 0.65; 69.9, 84.6; 388.3, 1976.2; 320.8, 1867.8, respectively. CR cheese had significantly ( $P < 0.01$ ) higher hardness, resilience, cohesiveness, springiness, gumminess, and chewiness, but lower adhesiveness compared with CC counterparts. These results may account for the difference in moisture contents (55.3 and 50.8%) in CC and CR cheeses, respectively. For 3 main factors, cheese type was significant ( $P < 0.01$ ) for all textural traits, while storage temperature and storage period were significant ( $P < 0.05$  or 0.01) except for springiness and gumminess. For 2-way interactions, cheese type  $\times$  storage temperature and cheese type  $\times$  storage period were significant ( $P < 0.05$  or 0.01) for most properties, whereas none of 3-way interactions were significant. It was concluded that textural properties of the experimental caprine cheeses were significantly affected by cheese type (cholesterol removal), followed by storage temperature and periods.

**Key Words:** goat milk cheese, cholesterol-reduced, texture

**W140 Identification of crystalline entities in the rinds of white mold ripened cheese and smear ripened cheese with powder X-ray diffractometry.** Gil F. Tansman\*<sup>1</sup>, Paul S. Kindstedt<sup>1</sup>, and

John M. Hughes<sup>2</sup>, <sup>1</sup>*Department of Nutrition and Food Sciences, University of Vermont, Burlington, VT,* <sup>2</sup>*Department of Geology, University of Vermont, Burlington, VT.*

Curd softening in wheels of surface ripened cheese occurs radially from the surface toward the center and is the result of chemical changes including decalcification. It has been theorized that calcium is removed from the system by depositing as calcium phosphate crystals in and beneath the rind. Although researchers have observed calcium-containing crystals under the bloomy rind in several cheese varieties, no work has adequately determined the identity of the crystals. The present study was conducted to identify the crystals in bloomy rind (white mold ripened) cheeses and compare them to crystals that form in the smears of washed rind cheeses. Four varieties of Vermont soft ripened cheese were obtained from a commercial source; 2 of these were bloomy rind cheeses and 2 were washed rind cheeses. Powder X-ray diffraction (PXRD) patterns were generated for each cheese from fresh samples extracted from 1 mm below the curd surface. Diffraction patterns were also generated from the smear material and surface hyphae scraped from the surfaces of washed rind and bloomy rind cheeses, respectively. The PXRD patterns revealed that small quantities of calcite ( $\text{CaCO}_3$ ) form below the rinds of both cheese classes, but the diffraction patterns were very weak, due to the limited quantity of crystalline material present. Strong PXRD patterns from smear material revealed the presence of large concentrations of crystalline material. The smear from one cheese contained crystalline ikaite ( $\text{CaCO}_3 \cdot 6\text{H}_2\text{O}$ ) and the other contained both ikaite and struvite ( $\text{NH}_4\text{MgPO}_4 \cdot 6\text{H}_2\text{O}$ ). Powder diffractions from the bloomy hyphae produced very weak diffraction patterns with characteristic peaks of calcite, which may represent crystalline material that adhered to the hyphae from the cheese-hyphae interface.

**Key Words:** cheese, crystals, powder X-ray diffraction (PXRD)

**W141 Characterization of single crystals in the rinds of white mold and smear ripened cheeses with single crystal X-ray diffractometry.** Gil F. Tansman\*<sup>1</sup>, Paul S. Kindstedt<sup>1</sup>, and John M. Hughes<sup>2</sup>, <sup>1</sup>*Department of Nutrition and Food Sciences, University of Vermont, Burlington, VT,* <sup>2</sup>*Department of Geology, University of Vermont, Burlington, VT.*

Calcium-containing crystals that form in the rinds of soft surface-ripened cheeses are thought to contribute to cheese softening through the removal of calcium from the aqueous and colloidal states. Several researchers have observed crystals near the surface of bloomy rind cheeses but none have succeeded in identifying them. The present work was conducted in parallel with a powder X-ray diffraction (PXRD) study in which fresh cheese samples were analyzed for total crystalline composition. In contrast, this study used single crystal X-ray diffraction (SCXRD) and polarized light microscopy to identify individual crystals isolated from cheese rinds and surface smears to obtain chemical information about crystal structure and atomic uniformity. Four Vermont soft surface ripened cheeses were obtained from commercial sources; 2 of these cheeses were bloomy rind (white mold) varieties and 2 were smear ripened varieties. Samples from 1 mm below the rinds of all 4 cheeses as well as scrapings of the washed rind surface smears were mounted on glass microscope slides and observed with a petrographic polarized light microscope. Rotation of the circular microscope stage allowed crystal extinction to be observed. Three distinct crystals were observed by comparing crystal morphologies and extinction variability. Two different crystals were observed in the smears (one smear exhibited only one of the crystals) and one entity was observed 1 mm beneath the rind in all 4 cheeses. Nearly all crystals that had not been damaged in the mounting process exhibited uniform extinction indicating that they were single

crystals. Single crystals from the smears and from beneath the fungal bloom were isolated with a dissection needle and mounted on a goniometer head for analysis with a single crystal diffractometer. Structures for crystals in the smears were determined for ikaite ( $\text{CaCO}_3 \cdot 6\text{H}_2\text{O}$ ) and struvite ( $\text{NH}_4\text{MgPO}_4 \cdot 6\text{H}_2\text{O}$ ), and a crystal structure for calcite ( $\text{CaCO}_3$ ) was determined from a bloomy rind crystal, thereby corroborating the

results of the parallel PXRD study. This is the first observation of single crystals in cheese.

**Key Words:** cheese, crystal, single crystal x-ray diffraction (SCXRD)



## Dairy Foods: Processing

**W142 Improved heat stability of whey protein isolate by dry-heating with inulin.** Yue He and Bongkosh Vardhanabhuti\*, *University of Missouri, Columbia, MO.*

Dry-heating whey protein with reducing saccharides via Maillard reaction has been shown to improve heat stability. Selecting proper saccharides that offer technological and nutritional benefits could create new protein ingredients that have better functional properties and are more attractive to the consumers. The aim of this study was to develop heat stable whey protein ingredients by dry-heating with inulin, a non-digestible carbohydrate with known health and technological benefits. The effects of biopolymer ratio and heating conditions on heat stability, physicochemical properties, and nutritional qualities of the conjugates were determined. Whey protein isolate (WPI) and inulin at weight ratios of 1:1 to 6:1 were dissolved in deionized water, freeze-dried, and incubated at 70 to 80°C for 12 – 72 h without or with controlled relative humidity (RH) at 44 or 80%. Heat stability was measured by heating 6% w/w protein solutions, pH 6.0 at 85°C for 15 min and the absorbance was measured at 630 nm. Particle size, zeta potential, rheological properties, and lysine content were determined. WPI or mixed WPI-inulin (no dry-heating) turned very opaque after heating. Dry-heating of WPI-inulin clearly increased heat stability as shown by a significant reduction ( $P < 0.05$ ) in turbidity (e.g., from A630 of 1.9 for WPI control to 0.8 for 12 h dry-heated 6:1 WPI-inulin) and particle size (from 160 nm in z-average diameter to 107 nm). Decreasing WPI:inulin weight ratio and increasing dry-heating temperature, incubation time, as well as RH resulted in increased heat stability; however, brown color intensity also increased. Rheological analysis revealed that all samples maintained Newtonian behavior with no significant difference in flow behavior index and consistency index. Dry-heated WPI-inulin had higher surface charge compared with WPI. The loss of lysine content ranged from 2.6% in 6:1 WPI-inulin after 12 h dry-heating to 19.2% in 2:1 WPI-inulin after 72 h dry-heating. In conclusion, dry-heating WPI with inulin creates new ingredient with improved heat stability. The new ingredient has great potential to be used in high protein beverages or other applications that require heat stability.

**Key Words:** whey protein, inulin, heat stability

**W143 Heat treatment effect on hydrolysis of sodium triphosphate in milk.** Diogo Maus, Alviélér Magalhães, and Walkiria Hanada Viotto\*, *University of Campinas (UNICAMP), Campinas, São Paulo, Brazil.*

Sodium phosphate salts are widely used in the manufacture of processed cheese for the stabilization of casein, as well as to improve the texture and functionality of the final cheese. The essential role involves calcium sequestering and pH control leading to the hydration and dispersion of casein, which in turn allows it to act as emulsifying agent resulting in the formation of a homogeneous cheese structure. During the processing phosphates may undergo hydrolysis due to heat treatment, pH and ionic environment. The formation of short chain phosphates increases the buffering capacity of the medium and therefore affects the final pH of the processed cheese. The objective of this study was to investigate the impact of temperature on the hydrolysis of sodium triphosphate (STPP) in milk. Milk with 2.3% STPP was heated to 90°C and cooled to 5°C and the nuclear magnetic resonance (NMR) spectra were obtained at 5°C intervals.  $^{31}\text{P}$  NMR spectroscopy was performed using a double resonance probe, fitted with a field gradient in the Z direction, coupled

to a Bruker AVANCE III, operating at 14.1 T static magnetic field (500 MHz for  $^1\text{H}$ ). The spectra were processed with the aid of TOPSPIN 3.1 software. STPP hydrolysis occurred for the entire temperature range during milk heating and cooling and was significantly more intense from 80°C. At 90°C, 20.5% of STPP was hydrolyzed resulting in a 33.5% increase in the amount of pyrophosphate and orthophosphate. The spectra also showed a shift from left to right, indicating that the STPP hydrolysis resulted in a more acidic environment, which can affect the rate of hydrolysis. Knowledge of the phosphates behavior during processing can lead to a better understanding of the changes associated with the protein matrix of processed cheeses and its functional properties. Acknowledgment: São Paulo Research Foundation (FAPESP), grant 14/07291–3.

**Key Words:** phosphate, hydrolysis, NMR

**W144 Inactivation of thermophilic sporeformers in milk by continuous ultrasonication.** Dikshi Bawa\*, Sanjeev Anand, and Steve Beckman, *South Dakota State University, Brookings, SD.*

Thermophilic sporeformers can survive milk pasteurization and cause spoilage in dairy foods. Our previous studies established that batch ultrasonication combined with pasteurization inactivated thermally resistant vegetative cells of spore forming *Bacillus* spp. This study investigates the effect of a continuous ultrasonication process to reduce thermophilic sporeforming bacilli in milk. We hypothesize that ultrasonication causes increase of cell membrane permeability leading to leakage of cell components thus causing cell death. Mid exponential phase vegetative cells of thermophilic *Bacillus coagulans* (ATCC 12245) were inoculated in sterile skim milk at log 5 cfu/mL. Their thermal stability was validated by exposing them to batch pasteurization (63°C for 30 min). Inoculated skim milk was passed through a continuous ultrasonicator with 22-mm sonotrode, 20 kHz frequency, and 1000 W power input (UIP1000hd, Hielscher USA, Inc.), at 86% (91.2  $\mu\text{m}$ ) amplitude and a flow rate of approximately 7.5 L/h and back pressure of 345 kPa resulting in an exposure time of 80 s per pass. Ultrasonicated samples were batch pasteurized to study the combined effect. Brain Heart Infusion agar was used for plating the survivors. Experiments were conducted as replicates of 2, and were repeated thrice. Statistical significance of the data was determined using SAS enterprise guide 7.1 software. A significant ( $P < 0.05$ ) reduction was found in the log counts after treatments. Lab pasteurization alone reduced the vegetative cells by 25%, while ultrasonication alone resulted in a greater inactivation of 92% after 12 passes (16 min total exposure time). Inactivation was further increased to 99.98% by combining ultrasonication and pasteurization. It can thus be concluded that a continuous ultrasonication process followed by pasteurization is highly effective to inactivate thermophilic vegetative cells of sporeformers such as *Bacillus coagulans*.

**Key Words:** ultrasonication, pasteurization, thermophilic sporeformer

**W145 Effect of ultrasound treatment on reconstituted deproteinized whey prior to lactose crystallization.** Steve Beckman, Lee Alexander\*, Sanjeev Anand, and Lloyd Metzger, *South Dakota State University, Brookings, SD.*

Ultrasonication (US) is an emerging technology being applied to dairy streams. The objective of this study was to determine the effect of US on reconstituted deproteinized whey (DPW) before crystallization and

isolation of lactose. Commercial DPW was reconstituted to 65% TS (wt/wt) at 80°C. Solutions were then seeded (0.027 g/100 g solution) with lactose crystals, or treated with US (10 min, 22.5 Hz) and then seeded. Each sample was then stirred for one min and transferred into a 1 L glass crystallizer (CST). The CST was immersed into a temperature controlled bath and cooled from 80 to 18°C (rate = -0.0479°C/min). Constant agitation (100 rpm) was applied to the CST by an overhead stirrer and agitator. After crystallization, approx. 225 g of solution was transferred to a centrifuge bottle. The solution was centrifuged at 10,000 × g for 30 min at 5°C. The supernatant was decanted, weighed, and a mass of deionized water (<4.0°C) equivalent to the supernatant was added to resuspend the crystals before centrifugation. The resuspended crystals were centrifuged, decanted, and resuspended 3 more times using the previous procedure (4 washes total). Recovered lactose crystals (>75 crystals) were measured microscopically (10×) to determine mean crystal size. Total solids of commingled supernatants, and of isolated crystals were measured to assess yield. Each treatment was repeated in triplicate. Mean crystal size, and lactose yield data were analyzed for statistical significance with replicate and treatment as categorical explanatory variables. Mean crystal size was larger ( $P < 0.01$ ) for the US treatment, 85.0 μm, than for the control treatment, 74.4 μm. Lactose yields, 78.7 and 78.9%, for control and US, respectively, were not different ( $P > 0.05$ ). Theoretical yield for lactose at the conditions used is 88%. Yields may be overestimated due to non-lactose impurities. Ultrasonication with seeding of reconstituted DPW before crystallization increased lactose crystal size, but did not increase lactose yield.

**Key Words:** lactose, crystallization, ultrasound

**W146 Evaluation of a Centritherm evaporator for concentrating micellar casein.** Anil Kommineni, Dustin Grossbier\*, Steven Beckman, Ananya C. Biswas, and Lloyd E. Metzger, *Midwest Dairy Foods Research Center, Dairy Science Department, South Dakota State University, Brookings, SD.*

Micellar casein is produced by micro-filtering skim milk. In a typical skim milk microfiltration (MF) process the retentate produced has approximately 10% protein and 12% total solids. After MF this retentate can be further concentrated before spray drying. In previous research we determined that micellar casein could be concentrated to approximately 25% solids using a falling film vacuum evaporator. However, concentration beyond 25% solids was challenging due to the high water binding and film forming characteristics of micellar casein. As compared with falling film evaporation, a Centritherm evaporator (CTE) is a thin film vacuum evaporator that can be used to concentrate highly viscous products. In a CTE the feed tube distributes product on the underside of a rotating heating cone through a feed nozzle. Steam is injected on the opposite side of the cone thereby heating the product stream through conduction. Centrifugal force of the rotating cone creates a thin, turbulent layer, which gives a residence time of 1 sec with a high heat transfer coefficient. The objective of this study was to evaluate the CTE for concentrating micellar casein. In this study, 3 replicates of micellar casein with 12% solids were manufactured by micro filtration of skim milk and then used as feed for the CTE. Initial trials were conducted to find the optimum evaporation conditions for minimal fouling. The heating temperature of 80°C, boiling temperature of 50°C and feed rate of 50 L/h were determined as optimal. Each replicate of micellar casein was passed through the CTE 3 times. The average water evaporation rate in each pass and the total solids of the concentrate was determined. The total solids content of the final micellar casein for each replicate was 22.49, 21.92, and 22.93%. The mean water evaporation of each pass was 6.17, 5.92, and 3.83% respectively for pass one, 2,

and 3. The decreasing water evaporation rate with each pass may be a result of increased water binding as the solids content of the micellar casein increased. The results indicate that the CTE concentrated micellar casein similar to falling film evaporation.

**Key Words:** micellar casein, Centritherm evaporator

**W147 Novel microfiltration process for the manufacture of soluble casein isolate from acidified milk.** Yanjie Lu\*, Michael Mollitor, and John Lucey, *Wisconsin Center for Dairy Research, University of Wisconsin-Madison, Madison, WI.*

Caseinates are traditionally manufactured by precipitating caseins using acid, thoroughly washing the precipitate and then neutralizing the casein curd with alkali, which requires very specialized equipment. The objective of this study was to develop an innovative process utilizing spiral filtration to produce a soluble casein product that would act as a caseinate alternative. Stable casein dispersions were prepared by sufficient acidification of pasteurized skim milk to solubilize the colloidal calcium phosphate (CCP), but avoid aggregating the caseins. The acidified skim milk was processed at ambient temperature using microfiltration /diafiltration to remove serum proteins, lactose, and the soluble minerals. Next the soluble casein product was neutralized with 40% NaOH and spray dried. Compositional analysis, SDS-PAGE, and turbidity measurement were used to characterize the casein product. Our results showed that acidification to pH ~5.3 solubilized the majority of CCP and allowed the removal of calcium during membrane processing. Acidification of the diafiltration water helped to maintain the narrow target pH range throughout the extensive diafiltration. The microfiltration process, temperature and pH were all optimized to promote casein retention and avoid temperature and /or pH-induced casein aggregation. The casein isolate powder had ~93% of crude protein (dry basis), 0.08% calcium, 0.37% lactose, 1.60% fat and casein accounted for 97% of the crude protein. When rehydrated to a total solids content of 5% (w/v), our casein product was less turbid compared with commercial sodium caseinate. This novel process looks promising for the production of a soluble casein product.

**Key Words:** caseinate, membrane filtration, acidification

**W148 Effect of some operating parameters on the hydraulic resistance developed during milk protein concentration by ultrafiltration.** Stephanie Methot-Hains\*, Alain Doyen, Laurent Bazinet, and Yves Pouliot, *STELA Dairy Research Center, Institute of Nutrition and Functional Foods, Université Laval, Québec, Canada.*

Milk protein concentrates are increasingly used as enrichment ingredients in cheese manufacturing. Ultrafiltration (UF) membranes with a molecular weight cut-off of 10 kDa are typically used for concentrating skim milk proteins content up to 57 to 84% (dry basis). However, some operating parameters such as transmembrane pressure control, temperature or multistage design have important impacts on process efficiency and functional properties of the concentrates. The objective of this work was to investigate the effect of transmembrane pressure (TMP) on the membrane performance and the protein retention by UF on skim milk using a 10 kDa polyethersulfone membrane. Skim milk was concentrated by a 4X volumic concentration factor at 50°C. Three operating modes were investigated: constant transmembrane pressure, 90 and 53 psi, and constant flux. Fouling of the membrane system was characterized using the resistance-in-series approach. It was found that operating at constant flux increases ( $P < 0.05$ ) the reversible fouling and consequently increases the energy requirement necessary to produce

milk concentrate in comparison with to operating at lower constant transmembrane pressure. Our results showed that transmembrane pressure control does not affect the irreversible fouling during milk protein concentrate production,  $66.5 \pm 0.6\%$  protein. Also, the protein retention was not affected ( $P < 0.05$ ) by changes in transmembrane pressure. This study demonstrates that milk protein concentration by UF at low transmembrane pressures limits the formation of cake layer (reversible fouling) and this improves the efficiency of the process by reducing the energy consumption during filtration and its environmental footprint.

**Key Words:** transmembrane pressure, fouling, energy

**W149 Comparative performance of two membrane configurations for the separation of casein from bovine milk by microfiltration.** Daniel Tremblay-Marchand<sup>1</sup>, Alain Doyen<sup>1</sup>, Michel Britten<sup>2</sup>, and Yves Pouliot\*<sup>1</sup>, <sup>1</sup>STELA Dairy Research Center, INAF, Université Laval, Québec, QC, Canada, <sup>2</sup>Agriculture & Agri-Food Canada, FDRC, St-Hyacinthe, QC, Canada.

Microfiltration (MF) is increasingly used for the separation of casein micelles from serum or whey proteins (SP) in milk, using a 0.1–0.2  $\mu\text{m}$  pore size membrane. One of the processing benefits of casein concentration by MF is that the co-product generated is considered as native or ideal whey. Ceramic graded permeability (GP) MF membranes ensuring long-term permeation flux and membrane selectivity have been successfully used for the separation of caseins from milk. Polymeric spiral wound (SW) MF membrane elements have also been reported to perform casein separation from milk but their performance has not yet been fully characterized. The objective of the present study was to compare the performance of 2 MF membranes configurations (SW vs GP) for the separation of casein from skim milk. The first membrane studied was a ceramic Membralox GP (model EP1940, Pall Corp.) with a 0.1- $\mu\text{m}$  pore size and a surface area of 0.72  $\text{m}^2$  while the second was a polymeric polyvinylidene fluoride (PVDF) spiral-wound membrane (model V0.2–2B-3838, Synder filtration) with a 0.2  $\mu\text{m}$  pore size and a surface area of 14.12  $\text{m}^2$  (2 elements). Both membranes were mounted on a Model 393 pilot-scale system (Tetra Pak, Champlin, MN). Experiments were performed at 50°C up to a concentration factor of 2 $\times$ , 3 $\times$ , and 4 $\times$ . Permeation flux and changes in membrane resistance during MF were compared. Membrane selectivity (separation of caseins from whey proteins) was determined by chemical analysis (TN, NCN, NPN) and validated by SDS-PAGE electrophoresis. It was found that GP membrane had an average permeation flux 3 times higher than that of SW. The CN/TP ratio of the final retentates were similar for both membranes. The water flux of fouled membranes (before cleaning) was 40% and 60% of the initial water flux for SW and GP membrane respectively. Although casein separation could be achieved using the 2 membrane configurations (GP and SW), our observations suggest that the GPMF membrane offers better process control by maintaining a higher permeability and preventing severe membrane fouling upon casein separation from milk.

**Key Words:** microfiltration, casein, membrane

**W150 Characterization of the early stages of biofouling during ultrafiltration of dairy fluids using polyethersulfone membranes in a model system.** Julien Chamberland\*, Marie-Hélène Lessard, Steve Labrie, and Yves Pouliot, STELA Dairy Research Center, Institute of Nutrition and Functional Foods, Université Laval, Québec, QC, Canada.

Bacterial biofilms are known to affect operational performance of membrane filtration, but little knowledge is available concerning the

mechanisms and factors involved in their formation. The present work aimed to characterize pioneer colonizer bacteria involved in biofilm formation on filtration membranes during dairy fluids treatments. In this context, a laboratory-scale crossflow filtration model system has been designed consisting in an assembly of 4 parallel filtration units (CF042, Sterlitech) each equipped with a UF membrane of 42  $\text{cm}^2$  flat polyethersulfone (PES) sheet with a molecular weight cut-off of 10 kDa. The dairy fluids studied were raw and pasteurized skim milk, unpasteurized cheese whey and pasteurized cheese whey from the same batch of raw milk. UF separations were performed in full-recycle mode at 40°C for 5 h. After UF operations, membranes were collected before and after cleaning procedure (pH 10.5, 150 ppm of free chlorine, 45°C, 30 min.) for DNA extraction (phenol-chloroform extraction). All experiments were performed in duplicate. A high correlation ( $R^2 = 0.93$ ) was found between DNA recovery on UF membranes after water flush (before cleaning) and the initial microbial load of the fluid; whey pasteurization reduced significantly the amount of DNA recovered on the membrane coupons before cleaning ( $P < 0.05$ ). After cleaning, genomic DNA was still detectable on membranes coupons. Indeed, preliminary results showed that even with a higher initial microbial load, unpasteurized cheese whey left the lowest amount of DNA (2.76 ng DNA/ $\text{cm}^2$ ) compared with both milk samples (3.86 and 4.71 ng DNA/ $\text{cm}^2$ ). Sequencing of the extracted metagenomic DNA is currently in preparation (MiSeq, Illumina) to establish the portrait of the rising bacterial ecosystem located on PES membranes. Our results will translate into designing processing conditions for a better control of biofouling and consequently process efficiency.

**Key Words:** dairy processing, biofouling, metagenomic

**W151 In situ monitoring of lactose crystallization using focused beam reflectance measurement (FBRM).** Karthik Pandalaneni\* and Jayendra Kumar Amamcharla, Kansas State University, Manhattan, Kansas.

Lactose is the second most abundant component present in milk. It is commercially produced from whey or whey permeate or milk permeate by crystallization. The current problem is lack of efficient tool to monitor crystal sizes and chord lengths during industrial lactose crystallization (LC). The objective of this study was to use focused beam reflectance measurement (FBRM) as a tool for in situ monitoring of LC. A 2  $\times$  3 factorial design was used, with temperature (20°C and 30°C) and concentration (w/w) (50%, 55% and 60%) as independent variables. Desired concentrations of lactose were obtained by dissolving commercial grade lactose in distilled water. The FBRM was placed in a batch crystallizer consisting of an overhead stirrer and a temperature-controlled water-bath. LC was monitored for 630 min using an in situ FBRM system to obtain chord length distributions (CLD) and crystal size distributions (CSD). CLD obtained from FBRM were recorded in the ranges of < 50  $\mu\text{m}$  (fine crystals) and 50–300  $\mu\text{m}$  (coarse crystals). At regular intervals, lactose concentration was measured using a refractometer to deduce extent of crystallization. The extent of crystallization increased rapidly during the first one hour of crystallization. The time required to reach 90% extent of crystallization at 30°C was found to be 300, 360, and 420 min for 60%, 55%, and 50% solutions, respectively. As the extent of crystallization increased, the fine crystal counts obtained from FBRM were also increased. It was observed that fine crystal counts increased with increasing supersaturated concentration (65,000 for 50% and 84,000 for 60% at 30°C) and temperature (59,000 at 20°C and 65,000 at 30°C for 60%). Square weighted CLD obtained from FBRM helped demonstrate that, as concentration increased there was a substantial decrease in chord lengths at 20°C. Mean chord lengths of lactose crys-



tals as observed by FBRM for 60, 55, and 50% at 20°C were 39.09, 40.52, and 57.64  $\mu\text{m}$ , respectively at 630 min. In conclusion, FBRM in conjunction with refractometer could be used as a potential tool for in situ monitoring of LC process.

**Key Words:** lactose crystallization, focused beam reflectance measurement (FBRM)

**W152 Influence of magnetic field exposure and clay mineral addition on the fractionation of Greek yogurt whey components.** Clinton R. Kyle\* and Jayendra K. Amamcharla, *Kansas State University, Manhattan, KS.*

Greek style yogurt in the United States is one of the largest growing sectors in the dairy industry. Greek yogurt is produced by removing a part of water and water-soluble components from yogurt. Consequently, a large quantity of Greek yogurt whey (GYW) is being produced as a co-product. The GYW is compositionally different from cheese whey, and thus posing economic and environmental challenges to the dairy industry. The objective of the present work was to evaluate the use of magnetic fluid treatment (MFT) and addition of sepiolite (IMV Nevada, NV), a clay mineral, as alternative methods for separating valuable GYW components. The MFT chamber was designed using 4 pairs of neodymium magnets arranged to produce a magnetic field strength of 0.6 Tesla. The GYW was pumped through the MFT chamber at a flow rate of 7.5 L/min. Three batches of GYW each from 2 manufacturers were procured. A  $2 \times 3$  factorial design was used with MFT or no MFT and the addition of 0, 2, or 4 g of sepiolite for 100g of GYW. The pH of GYW was adjusted to 7.2 using 5 N NaOH, and processed in the MFT chamber. The sample was split into 3 sub-samples, heated to 80°C, and sepiolite was added as per the experimental design. Subsequently, the samples were centrifuged at 1000g for 5 min. The supernatant aqueous layer was separated and analyzed for total solids, ash, lactose, protein, calcium, phosphates, sodium, and color. Data were analyzed separately for each manufacture using the MIXED procedure in SAS software. MFT and its interaction did not significantly ( $P > 0.05$ ) influence the analyzed whey components except for lactose. However, the addition of sepiolite significantly ( $P < 0.001$ ) influenced the protein content,  $a^*$ , and  $b^*$  for the top aqueous layers. Both levels of sepiolite addition resulted in about a 50% decrease in protein compared with original GYW. Adding 2g of sepiolite per 100g of GYW from manufacturer 1 resulted  $b^*$  decreasing from 25.99 to 8.16 compared with treated GYW with no sepiolite. Sepiolite was found to have possible applications in the removal of proteins and color pigments in Greek yogurt whey.

**Key Words:** Greek yogurt whey, magnetic treatment, sepiolite

**W153 Characterization and oxidative stability of oleic acid-modified chitosan/milk protein nanoparticle containing docosahexaenoic acid.** Ho-Kyung Ha\*<sup>1</sup>, Ji-Young Hong<sup>1</sup>, Jae-Young Hwang<sup>1</sup>, Won-Jae Lee<sup>1</sup>, and Mee-Ryung Lee<sup>2</sup>, <sup>1</sup>*Department of Animal Bioscience (Institute of Agriculture and Life Science), Gyeongsang National University, Jinju, Gyeongnam, Republic of Korea,* <sup>2</sup>*Department of Food and Nutrition, Daegu University, Gyeongsan, Gyeongbuk, Republic of Korea.*

The application of docosahexaenoic acid (DHA) to nonfat food is often challenging due to its poor bioavailability and susceptibility to oxidative rancidity. The aim of this study was to investigate how manufacturing variables, such as degree of substitution (DS) of oleic acid-modified chitosan and sub-ambient temperature treatment, affected the physicochemical properties of oleic acid-modified chitosan/milk

protein nanoparticle (OPN) and reduction in oxidative rancidity. Oleic acid-modified chitosan with various DS ranging from 0 to 7.4% were prepared using a carbodiimide coupling method. OPN was manufactured at various sub-ambient temperature from 5 to 25°C. The morphology and size of OPN were investigated using atomic force microscopy and particle size analyzer, respectively. Encapsulation efficiency, oxidative rancidity, and off-flavor compounds of DHA were determined by high performance liquid chromatography, peroxide value, and gas chromatography/mass spectrometry, respectively. Globular shaped particles with the size of ~190 nm were observed indicating the successful formation of OPN. As DS increased from 0 to 7.4%, a significant ( $P < 0.05$ ) increase in the size of OPN and encapsulation efficiency of DHA was observed. During storage, the peroxide value for OPN containing DHA was significantly ( $P < 0.05$ ) decreased with an increase in DS. A decrease in sub-ambient temperature from 25 to 5°C resulted in a significant ( $P < 0.05$ ) decrease in the size of OPN and increase in the encapsulation efficiency of DHA. The peroxide value for OPN containing DHA was significantly ( $P < 0.05$ ) lower in comparison to free DHA although it was not significantly affected by sub-ambient temperature treatment. No significant ( $P < 0.05$ ) off-flavor developments were observed in OPN containing DHA while free DHA exhibited a significant ( $P < 0.05$ ) increase in off-flavor development during storage. In conclusions, DS and sub-ambient temperature treatment were crucial factors affecting the physicochemical properties of OPN and oxidative stability.

**Key Words:** milk protein, nanoparticle, docosahexaenoic acid

**W154 Electrospinning casein-based fibrous mats for food applications.** Peggy M. Tomasula\*<sup>1</sup>, Shih-Chuan Liou<sup>2</sup>, Ran Li<sup>3</sup>, Laetitia M. Bonnaille<sup>1</sup>, and LinShu Liu<sup>1</sup>, <sup>1</sup>*USDA/Agricultural Research Service, Eastern Regional Research Service, Dairy and Functional Foods Research Unit, Wyndmoor, PA,* <sup>2</sup>*Chung Shan Medical University, Taichung City, Taiwan,* <sup>3</sup>*State Key Laboratory of Hollow Fiber Materials and Processes, School of Materials Science and Engineering, Tianjin Polytechnic University, Tianjin, China.*

Electrospinning is a process that produces fibrous mats from fibers with diameters on the micron or nano scales from an electrified jet of a polymer solution. If produced by electrospinning biopolymer solutions, the fibrous mats may have the same potential as edible films for protecting foods and improving food quality but also allow for preservation of sensitive nutrients. Electrospinning aqueous solutions of dextran, pullulan (PUL) and gelatin are known to result in homogeneous fibers. However, little information is available on electrospinning other food grade biopolymers. The objective of this study was to create fibers for food use from electrospinning aqueous solutions containing calcium (CaCAS) or sodium caseinate (NaCAS). A nanofiber electrospinning unit was used to generate the fibers at 40°C using voltage of 23 KV and flow rate of 0.5 mL/h. Fibers were not produced by electrospinning 5, 10, or 20% (w/w) solutions of either CAS, possibly because of little interaction among the CAS chains, but were produced when either of the 20% CAS solutions were blended with 15% or 30% (w/w) aqueous solutions of PUL in volume ratios from (2:1) to (1:4), using PUL as a spinnable carrier. The morphologies of the fibrous mats were determined using scanning electron microscopy equipped with software to sample 100 of the constituent fibers to calculate mean diameters. Electrospinning 15% PUL solutions resulted in fibers with diameters of  $190 \pm 50$  nm. CaCAS:PUL solutions in volume ratios of (1:1) and (1:2) resulted in fibers with average diameters of  $160 \pm 40$  nm and  $1020 \pm 600$  nm, respectively. NaCAS:PUL solutions in volume ratios of (1:1) and (1:2) resulted in fibers with average diameters of  $320 \pm 30$  nm and  $340 \pm 150$  nm, respectively. The mean diameter of the CaCAS:PUL (1:1) fibers

was not significantly different from the pure PUL fibers, but the increase in size of the CaCAS:PUL (2:1) fibers and the NaCAS:PUL(1:1) and (1:2) fiber sizes may stem from changes in hydrogen bonding and thus the degree of entanglement of the PUL chains in the presence of CAS, compared with pure PUL solutions. This is the first example of CAS nano- and micro-fibers prepared using a polysaccharide carrier, rendering a new dairy product with potential use in food and packaging applications.

**Key Words:** fibers, nanoscale, polysaccharide

**W155 Isolation of milk fat globule membrane (MFGM) from buttermilk.** Liza Ivanov\*<sup>1</sup>, Vladimir Shritz<sup>1,2</sup>, and Vitaly L. Spitsberg<sup>1,3</sup>, <sup>1</sup>*Astrazemcal, Raanana, Israel*, <sup>2</sup>*Baemek Advanced Technology, Afula, Israel*, <sup>3</sup>*Biovita Technologies, Bat Yam, Israel*.

Due to a great interest in application of bovine MFGM as a nutraceutical (J. Dairy Sci. 88:2289–2294, 2005), we developed a new protocol for isolation of large amounts of MFGM from buttermilk. The protocol includes the following steps: precipitation of casein and MFGM at pH 4.6, treatment of the precipitated complex casein-MFGM with sodium phosphate at pH 4.6–8.0 or with polyphosphate (Calgon) at pH 7.1 and differential centrifugation (1000–2000 × g to get a pellet of casein and 32,000 × g (Strator centrifuge) to collect a pellet of MFGM. Maximum dissociation of MFGM from casein was achieved at pH 7.1–7.4. Yield of MFGM from 1 L of buttermilk treated with sodium phosphate at pH 7.2 was 789 mg of protein and treatment with Calgon at pH 7.1 also provided a high yield of MFGM, 725 mg/L. Treatment of precipitated casein with 0.1 M Na-citrate or Na-oxalate at pH 7.1 led to the isolation of MFGM in amount of 234 mg/L and 106 mg/L, correspondingly. Treatment of precipitated casein with 0.1 M NaCl or 0.1 M Na<sub>2</sub>SO<sub>4</sub> did not produce a noticeable amount of MFGM. Recovery of MFGM from buttermilk allowed us to suggest that MFGM in buttermilk is present in association with casein through Ca- bridges between phospholipids of MFGM and phosphate groups of casein, primarily with k-casein as a peripheral protein of casein micelles. For production of large amount of MFGM on industrial scale the use of continuous-flow centrifuge and microfiltration were suggested. Baemek Advanced Technology supported this work.

**Key Words:** milk fat globule membrane (MFGM), nutraceutical, buttermilk

**W156 Effects of surface modification on bacterial and spore adhesion in dairy handling materials.** Garrett T. Walsh\* and Rafael Jimenez-Flores, *California Polytechnic State University, San Luis Obispo, CA*.

The objective of this work was to demonstrate how stainless steel microstructure, material processing and surface treatment affect the attachment of spores, and the subsequent development of biofilms on the surface of AISI 316L Stainless steel, the typical milk handling and processing material. Stainless steel coupons with different surface finishes were

prepared (as received, 220 grit, 400 grit and 800 grit) and characterized by contact profilometry and scanning electron microscopy. In addition, autogenously welded samples were produced for each surface roughness. Samples were exposed to both sterile and bioactive solutions for 5 and 20 h periods. Corrosion resistance was subsequently measured using cyclic polarization testing in each surface and solution pairing. Samples were examined using scanning electron microscopy and epifluorescence microscopy. The former was used to characterize surface damage, identify film presence, and examine spore attachments. The latter was used to quantify attachment of *Bacillus licheniformis* and to estimate the thickness of biofilms formed. The work has demonstrated that surface preparation had a significant effect on attachment and proliferation ( $P < 0.05$ ). Furthermore welding had a significant effect on attachment and electrochemical reactivity ( $P < 0.05$ ). The electrochemical reactivity of the material as measured by stable pitting potential and measured passive current is also affected by both surface roughness and by bioactivity of the solution. Metastable pitting is common in all solutions, stable pitting potential is lower in bioactive solutions and the passivation current is greater in bioactive solutions. Microstructure of the material is critical to its behavior, as is the complexity of the solution. Planktonic populations are not a direct indication of the sessile populations.

**W157 The impact of milk hauling practices on overall raw milk quality.** Emily Darchuk, Joy Waite-Cusic, and Lisbeth Goddik\*, *Department of Food Science, Oregon State University, Corvallis, OR*.

Historically, milk tankers were cleaned after every load. Consolidation of the industry has led to longer routes and use of tankers for up to 24 h in between cleans. This study focused on the effect of frequent tanker use hauling on raw milk quality. Standard tanker use, (CIP once per 24 h) served as our control and incremental cleaning treatments (water rinse after each load, water rinse after each load with a 12 h sanitizer treatment, and 12 h sanitizer treatment) were added to the study to understand if any effect could be mitigated by more frequent cleans. Two trucks were isolated for this study, which utilized a mix model with repeated measures design. Each truck was utilized for up to 9 routes per day. To understand the effect of seasonality, the 8-d study was repeated in both summer and winter. Producer samples were collected from the farm bulk tank before loading raw milk into the tanker as well as sampling the same milk directly out of the tanker truck before unloading at the manufacturer. Milk quality was quantified through commonly utilized microbiological tests: total bacteria count, thermophilic spore count and preliminary incubation count. Within our study, we defined a negative effect on milk quality as a statistically significant ( $P < 0.05$ ) difference between the producer and tanker sample in any of the 3 microbial tests conducted. Results did not identify a negative effect on raw milk quality due to hauling neither in summer nor in winter conditions. Therefore, the addition of cleaning treatments did not appear to provide a positive impact on milk quality. Based on this study, current practices appear to be effective in mitigating any measurable negative effect due to hauling.

**Key Words:** hauling, milk, quality

## Forages and Pastures: General forages and forage systems

### W158 Chemical composition, digestibility and fermentation characteristics of sorghum ensiled with soybean crop residue.

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The fermentation of silage process depends on the initial balance of nutrients in the ensiled forage and must minimize nutritional losses. Agricultural regions have high quantities of crop residues, and sometimes these residues have nutritional value and are used as co-products like soybean hulls but stalks, leaves and broken grains are frequently lost. The objective of this assay was to evaluate the effects of the soybean crop residue inclusion (0, 3 and 6% as fed basis) on sorghum silage quality. Sorghum was harvested at 30% dry matter (DM), mixed with the residues and ensiled targeting density between 600 and 650 kg as fed/m<sup>3</sup> using 6 silos/treatment. The nutritional value was evaluated after 50 d of ensiling. There was a significant effect of the soybean residue inclusion for crude protein (CP) (5.59, 6.50 and 6.74% DM), and neutral detergent insoluble protein (NDIP)/CP (42.46, 35.24 and 32.60%). The soybean residue inclusion reduced the non-fiber carbohydrates (34.51, 31.77 and 29.93% DM) and total carbohydrates (85.84, 84.84 and 83.79% DM) which caused decreases in in vitro DM digestibility (63.49, 63.35 and 59.71%) and in vitro OM digestibility (65.77, 61.64 and 59.05%). Linear increase in gas production (75.50, 101.62 and 129.22 mm<sup>3</sup>/gDM/h) was observed. The pH close to 3.8 at the moment of opening the silo, the constant ammonia-N concentration (72.8 g/kg total N), and the high recovery of DM 86.87% and OM 85.74% indicate heterolactic fermentation favoring acetic acid production which promotes aerobic stability of silage. An inclusion of soybean crop residue up to 6% as fed basis in sorghum silage is possible without effects on the fermentation characteristics of the silage.

**Key Words:** gas production, pH, silage

### W159 Effect of reduced lignin alfalfa on forage quality at three harvest intervals. Zhiqiang Li\*<sup>2</sup>, Zhenzhen Li<sup>2</sup>, David Combs<sup>1</sup>, and Daniel Undersander<sup>1</sup>, <sup>1</sup>University of Wisconsin, Madison, WI, <sup>2</sup>China Agricultural University, Beijing, China.

The present study investigates the effect of reduced lignin on alfalfa quality. Two alfalfa cultivars [Roundup Ready (RR) and double stacked Roundup Ready and Reduced Lignin (RR/RL)] were harvested at intervals of 28, 33, and 35 d. All replicated plots were harvested for 2 regrowth periods in July and August, 2014. CP, aNDF, lignin, NDFD48 value of samples on DM basis were analyzed with NIRS method (FOSS 6500) using NIRSC 2012 Alfalfa Hay Equation. Statistical analysis was done as a 2 way ANOVA using SPSS 22. CP content was not different between RR and RR/RL (27.5 vs 28.0,  $P > 0.05$ ) and declined with advancing maturity ( $P < 0.05$ ). aNDF content was higher in RR than RR/RL (31.6 vs 30.1,  $P < 0.05$ ) though differences among harvest intervals were not significant (30.8 vs 30.5 vs 31.1,  $P > 0.05$ ). Lignin content was not significantly different between RR and RR/RL (5.6 vs 5.5,  $P > 0.05$ ) or among harvest intervals (5.6 vs 5.6 vs 5.5,  $P > 0.05$ ). The NDFD48 of RR/RL was significantly higher than RR (52.2 vs 50.5,  $P < 0.05$ ) and that of 28d harvest interval was significantly higher than 33d and 35d levels (53.1 vs 50.2 vs 50.8,  $P < 0.05$ ). No significant interaction occurred between variety and harvest interval for any of the above 5 indices. The iNDF, kd and total-tract NDF digestibility (TTNDFD) are shown in the table below. The reduced lignin alfalfa tended to be lower

in iNDF and higher in kd though differences were not significant, but was significantly higher in TTNDFD ( $P < 0.012$ ). It appears that RR/RL variety reduces the NDF content and improves NDFD48, and TTNDFD.

**Table 1 (Abstr. W159).**

Item	Interval	RR	RR/RL	Mean	P-value		
					Forage	Forage harvest interval	Forage harvest × interval
iNDF	28d	34.1	29.9	31.7 <sup>b</sup>	0.086	0.001	0.470
	33d	42.1	37.0	39.9 <sup>a</sup>			
	35d	39.3	39.2	39.2 <sup>a</sup>			
	average	38.7 <sup>a</sup>	35.1 <sup>a</sup>	36.8			
Kd	28d	7.8	7.1	7.4 <sup>a</sup>	0.156	0.980	0.153
	33d	7.0	8.2	7.5 <sup>a</sup>			
	35d	6.4	8.9	7.8 <sup>a</sup>			
	average	7.0 <sup>a</sup>	8.0 <sup>a</sup>	7.6			
TTNDFD	28d	53.0	56.3	54.9 <sup>a</sup>	0.012	0.010	0.849
	33d	46.3	51.9	48.7 <sup>b</sup>			
	35d	46.8	51.1	49.2 <sup>b</sup>			
	average	48.6 <sup>b</sup>	53.3 <sup>a</sup>	51.0			

<sup>abc</sup>Means in rows or columns with different superscripts differ ( $P < 0.05$ ).

**Key Words:** alfalfa, lignin, forage

### W160 Characterization of novel polymers for alkaloid

**adsorption.** Manoj B. Kudupojje\*<sup>1,2</sup>, Eric S. Vanzant<sup>1</sup>, Alexandros Yiannikouris<sup>2</sup>, Karl A. Dawson<sup>2</sup>, and Kyle R. McLeod<sup>1</sup>, <sup>1</sup>University of Kentucky, Alltech-University of Kentucky Research Alliance, Lexington, KY, <sup>2</sup>Center for Animal Nutrigenomics & Applied Animal Nutrition, Alltech Inc., Nicholasville, KY.

A methacrylic acid-based molecularly imprinted polymer (MIP) was evaluated for physical and isothermal adsorption properties. Polymers were synthesized by suspension polymerization with (MIP) or without (NIP) ergotamine tartrate (ETA) as template. Polymer morphology was characterized by light scattering diffraction, SEM and BET. Polymer-template interactions were characterized by FT-IR, <sup>1</sup>H NMR and isotherms. For each of 4 alkaloids evaluated, adsorption difference between MIP and NIP interacted ( $P < 0.01$ ) with alkaloid concentration, but product differences were generally consistent across concentrations. With bromocryptine and methylergonovine, average adsorption was greater ( $P < 0.01$ ) for MIP than NIP (62.9 vs 37.1 and 35.8 vs 24.7%, respectively). NIP adsorption was greater ( $P < 0.01$ ) for ETA (93.1 vs 96.7%), and no difference ( $P > 0.05$ ) existed between products for lysergol adsorption (38.1 vs 37.7). The Freundlich model ( $R^2 = 0.99$ ) indicated similar adsorption intensity to ETA for MIP ( $n \sim 1.025$ ) and NIP ( $n \sim 1.011$ ) and the Qmax estimate from the Langmuir model ( $R^2 = 0.99$ ) was 388.7 and 435.4 nM/mg for MIP and NIP, respectively. Both polymers had IR spectra at ~3500 (carboxyl stretch), ~1730 (carbonyl stretch), and ~1260 cm<sup>-1</sup> (C-O stretch) indicating similarities in backbone structure. Spectral shifts observed in polymer-ETA complex samples suggest the interaction with amine groups was via H-bonding, which was confirmed using <sup>1</sup>H NMR. SEM demonstrated compactness of implanted polymer, which, after template removal turned microporous with microvoids. BET analysis showed NIPs to have a smooth surface with few pore structures, whereas MIPs exhibited greater surface area



and porosity; implying that MIPs have a larger surface for adsorption. Light scattering diffraction suggests bimodal particle size distribution for both polymers with larger percentage in 30–80 µm range. These imprinted polymers could be utilized as SPE material, or for extraction of ergot alkaloids from complex materials. Furthermore, the described MIP is a candidate for application as a feed adsorbent to reduce bioavailability of certain alkaloid compounds in the gut.

**Key Words:** molecularly imprinted polymer, ergotamine D-tartrate, isothermal adsorption

**W161 Nutritive value and in vitro degradability of Marandu palisade grass at different locations within the pasture in silvopastoral systems with different babassu palm densities.** Rosane Cláudia Rodrigues, Michelle de Oliveira Maia Parente, Xerxes de Moraes Tosta, Ana Paula Ribeiro de Jesus, Henrique Nunes Parente, Jocélio dos Santos Araújo, Sâmara Stainy Cardoso Sanchês\*, Ivone Rodrigues Araújo, Clésio dos Santos Costa, and Ivo Guilherme Ribeiro Araújo, *Universidade Federal do Maranhão, Chapadinha, Maranhão, Brazil.*

The objective of this study was to evaluate the nutritive value and “in situ” degradability of Marandu grass at different locations within the pasture in silvopastoral systems with different densities of babassu palm (Low density:39, Medium density:72 and High density:92 palm/ha). The silvopastoral system consisted of *Brachiaria brizantha*, the forage specie and Babassu palm *Orbignia phalerata* Martius, tree species. Pre-dried samples of forages were used for determination of dry matter (DM), organic matter (OM), neutral detergent fiber (NDF), acid detergent fiber (ADF), lignin, crude protein (CP), cellulose and hemicellulose. One Santa Inês male sheep, castrated, cannulated in the rumen, with an average body weight of 63 kg was used for ruminal incubation. Samples of forages (5 g) were incubated in the rumen in nylon bags. For each incubation time, 4 bags were used. The experimental design was a completely randomized design with factorial arrangement 3 × 3 (3 incubation times: 6, 24 and 96 h and 3 sampling sites within the pasture: no interference of shadow - NS, an intermediary INT and shadow interference - SI). There was no effect ( $P \geq 0.05$ ) of the density of palm trees or locations within the pasture on the content of NDF, ADF, lignin, cellulose and hemicellulose. The densities in grassland Palm did not influence the chemical composition of grass Marandu only DM content. The sampling sites influenced ( $P \leq 0.05$ ) content of CP and DM. Higher rates of disappearance of DM (RDDM) was observed in 96 h. At all densities, NS and SI had higher RDDM. Pastures with low density of palm (LDP), the location INT showed higher degradation rate and higher digestibility for passages rates 2, 5 and 8%. Locations within the SSPs reviews had a little influence on the nutritive value of grass Marandu, however, the local NS, at higher densities showed the higher protein content. In all pastoral environments, the NS and SI locations had higher DMS and location INT in SSPs with LDP and MDP smaller DMS, but higher degradation rate and effective digestibility, respectively.

**Key Words:** *Attalea speciosa* Mart, *Brachiaria brizantha*, protein

**W162 Effect of the cytokinin BAP on growth, senescence and in vitro degradation of *Lolium perenne* L.** Hilda A. Zavaleta-Mancera\*, Sergio S. González-Muñoz, Ángel H. Soto-Urano, and Omar Hernández-Mendo, *Colegio de Postgraduados, Montecillo, Estado de México, México.*

The cytokinin 6-benzylaminopurine (BAP) is a growth promoter that can delay leaf senescence. Therefore, the objective of this study was to

study the effect of BAP on growth, senescence and in vitro degradation of *Lolium perenne* L. (var. Linn). The experimental design was completely randomized with 4 treatments: 3 concentration of BAP (T:0.1 mM; T2:0.01 mM; T3: 0.001 mM); and a control without BAP (T0). Plants were grown from seed at 25 kg/ha on 68 pots containing a mix of soil:agrolite:peat-moss (1:1:1), in a greenhouse. Plants of 8 mo were cut at 5 cm above substrate. At 21 d of regrowth, plants were sprayed weekly during 65 d (7 times). The variables evaluated were: leaf growth and senescence during 112 d; soluble proteins and photosynthetic pigments during 84 d; chemical and morphological composition and in vitro degradation of the cell wall (6, 12, 24 and 48 h). Means were compared using the Tukey test ( $P < 0.05$ ). All BAP treatments increased net leaf growth (T1: 32%; T2: 18%; T3: 39%) with respect to the control, but leaf senescence and photosynthetic pigments were not significantly different ( $P > 0.05$ ). BAP 0.1 mM promoted accumulation of leaf soluble protein (3.55 mg/gFW) and reduction of acid detergent fiber by 6%, as compared with the control. In vitro degradation was increased only with 0.001 mM BAP at 60 d of regrowth (T0:0.50; T1:0.43; T2: 0.52; T3:0.53). It may be concluded that BAP increased growth and leaf protein of *Lolium perenne* L. and reduced cell wall components.

**Key Words:** *Lolium perenne* L., 6-benzylaminopurine, senescence

**W163 Effect of supplementation strategy and forage quality on in vitro digestibility of Kentucky 31 tall fescue and Tifton 85 bermudagrass.** Jeferson M. Lourenço\*<sup>1</sup>, Matthew W. Studstill<sup>1</sup>, Cathy A. Bandyk<sup>2</sup>, Dennis W. Hancock<sup>1</sup>, and Robert L. Stewart Jr<sup>1</sup>, <sup>1</sup>The University of Georgia, Athens, GA, <sup>2</sup>Westway Feed Products.

A 3 × 3 factorial experiment was conducted to evaluate IVDMD of Kentucky 31 tall fescue (KY-31) and Tifton 85 bermudagrass (T-85) with or without supplementation. The main effects included relative forage quality (RFQ) and type of supplement. Each forage type was harvested at different intervals during the yr of 2013 and 2014 to represent grazing and hay production scenarios in the state of Georgia. After harvested, the nutritive values of forages were assessed via near-infrared spectroscopy, and samples were stratified by RFQ. Forage samples were selected to represent high (HIG), medium (AVG), or poor (LOW) RFQ for KY-31 (171, 111, and 78, respectively) and T-85 (142, 101, and 80, respectively). Supplemental treatments included: 1) commercial molasses-urea based supplement (MUR) or 2) corn gluten feed (CGF) to provide the equivalent of 0.36 kg of supplemental CP; or 3) no supplement (CON). The forage-supplement mixtures were subjected to 48-h in vitro digestion to evaluate the digestibility characteristics. Two in vitro incubations were conducted and bottles (n = 140 in each incubation) were considered the experimental units. Across forage type, IVDMD increased as the incubation time was extended, and it was greatest ( $P < 0.001$ ) when samples were incubated for 48 h. The same response was observed for NDF and ADF digestibilities. No supplement × RFQ interaction was observed for KY-31 ( $P = 0.55$ ). Across RFQ for KY-31, 48-h IVDMD increased ( $P = 0.008$ ) with the addition of LIQ and CGF compared with CON (49.9, 49.6, and 47.7%, respectively). Across supplementation, IVDMD of KY-31 was greatest ( $P < 0.001$ ) for HIG and AVG compared with LOW (62.5, 50.2, and 34.5%, respectively). A supplement × RFQ interaction was observed for T-85 ( $P < 0.003$ ). In vitro dry matter disappearance was lowest for LOW × CON compared with all other supplement × RFQ combinations. These results suggest that forage quality is the main factor affecting IVDMD. However, the addition of LIQ and CGF can increase IVDMD of KY-31 and low RFQ T-85.

**Key Words:** bermudagrass, in vitro, tall fescue

**W164 Herbage accumulation of palisadegrass with variable heights in beginning of deferment period.** Manoel Eduardo Rozalino Santos\*<sup>1</sup>, Laryssa Avelino Luz<sup>1</sup>, Pedro Henrique Marçal Rodrigues<sup>1</sup>, Lucas Coelho Alves<sup>1</sup>, Wirley Duarte de Souza<sup>1</sup>, Simone Pedro da Silva<sup>2</sup>, and Dilermando Miranda da Fonseca<sup>3</sup>, <sup>1</sup>Universidade Federal de Uberlândia, Uberlândia, Minas Gerais, Brazil, <sup>2</sup>Instituto Federal Goiano, Hidrolândia, Goiás, Brazil, <sup>3</sup>Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.

The experiment was conducted to evaluate 2 different strategies for managing *Brachiaria brizantha* (palisadegrass) under deferred grazing. From January to June 2014, the palisadegrass pastures were evaluated at the Federal University of Uberlândia, located in the State of Minas Gerais, Brazil. The experimental site was located at 776 m altitude, 18°30' S and 47°50' W. Annual precipitation was around 1.584 mm. Medium temperature was 22.3°C. The experimental area was made up of 6 paddocks (experimental units) of 800 m<sup>2</sup>. Two pasture heights in the beginning of deferment period were evaluated: 15 cm and 45 cm. Before the deferment period, the pastures were managed in continuous stocking with sheep. The deferment period was 90 d. The experiment was carried out using a randomized completely design with 3 repetitions and subdivided plots. On 1, 45 and 90 d of deferment period, the tillers number m<sup>-2</sup> was quantified. In each experimental unit, 15 tillers were marked. The lengths of live portions of leaf blades and stems of tillers were measured weekly. The collected data were used to calculate the growth, senescence and herbage accumulation rates. The conversion factors were generated to transform the measures in cm tiller<sup>-1</sup> day<sup>-1</sup> to kg ha<sup>-1</sup> day<sup>-1</sup> of dry matter. The growth rate (GR) was higher at the beginning (87.1 kg ha<sup>-1</sup> day<sup>-1</sup>) than at the end (11.4 kg ha<sup>-1</sup> day<sup>-1</sup>) of deferment period. Deferred pasture with 45 cm (73.5 kg ha<sup>-1</sup> day<sup>-1</sup>) also showed higher GR, compared with deferred with 15 cm (34.9 kg ha<sup>-1</sup> day<sup>-1</sup>). Leaf senescence rate (LSR) was higher at end (78.0 kg ha<sup>-1</sup> day<sup>-1</sup>) than at the beginning (24.7 kg ha<sup>-1</sup> day<sup>-1</sup>) of deferment period in the pasture with 45 cm. The herbage accumulation rate (HAR) was higher at the beginning (37.6 kg ha<sup>-1</sup> day<sup>-1</sup>) than at end (-13.5 kg ha<sup>-1</sup> day<sup>-1</sup>) of deferment period. Deferred pasture with 45 cm showed greater HAR (22.1 kg ha<sup>-1</sup> day<sup>-1</sup>) of that with 15 cm (2.0 kg ha<sup>-1</sup> day<sup>-1</sup>). We conclude that the reduction in palisadegrass height at the beginning of deferment period results in pasture with lower growth, senescence and herbage accumulation.

**Key Words:** growth, senescence, tiller

**W165 Balance between the emergence and mortality of tiller in *Brachiaria brizantha* under variable heights in winter.** Manoel Eduardo Rozalino Santos\*<sup>1</sup>, Ludiêmilem Keith Parreira da Costa<sup>1</sup>, Bruno Humberto Rezende Carvalho<sup>1</sup>, Denis Douglas Pessoa<sup>1</sup>, Heron Alves de Oliveira<sup>1</sup>, Roger Carvalho Cardoso<sup>1</sup>, Miriã Gonçalves Simplicio<sup>1</sup>, Simone Pedro da Silva<sup>2</sup>, and Dilermando Miranda Fonseca<sup>3</sup>, <sup>1</sup>Universidade Federal de Uberlândia, Uberlândia, Minas Gerais, Brazil, <sup>2</sup>Instituto Federal Goiano, Hidrolândia, Goiás, Brazil, <sup>3</sup>Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.

The experiment was conducted to evaluate 3 different defoliation strategies of *Brachiaria brizantha* (palisadegrass) in winter. From April 2013 to March 2014, the palisadegrass were evaluated at the Federal University of Uberlândia, located in the State of Minas Gerais, Brazil. The experimental site was located at 776 m altitude, 18°30' S and 47°50' W. Annual precipitation was around 1.584 mm. Medium temperature was 22.3°C. The experimental area was made up of 12 plots (experimental units) of 9 m<sup>2</sup>. In one management strategy, palisadegrass was maintained at 30 cm high during all the trial. In the other, the palisadegrass was kept at an average of 15 cm during the winter (July to September

2013), and at 30 cm from the early spring until the summer (October 2013 to March 2014). At last, the palisadegrass was kept at an average of 45 cm during the winter and at 30 cm from the early spring until the summer. The experiment was carried out using a randomized completely design with 4 repetitions and subdivided plots. The defoliation strategies correspond to plots. The year seasons were the subplots. In each experimental unit, all tillers inside the 2 frames with 0.0625 m<sup>2</sup> were counted and marked with colored plastic. Every 30 d, all tillers were recounted and new tillers were marked with a different wire color. The collected data were used to calculate the balance between the emergence and mortality of tiller (BAL). The tillers number was quantified in each season year. The BAL was ( $P < 0.05$ ) lower in winter (-7.2%) and later spring (-5.0%), intermediate in summer (11.6%) and higher in early spring (26.2%). This response pattern was due to unfavorable weather conditions in winter. On the other hand, the high development of shoots in tillers in early spring may have generated higher shading at plant base in late spring, causing negative BAL. The defoliation strategies did not affect the BAL. We conclude that palisadegrass keep with 15 cm in winter showed higher tiller number during all experimental period, when compared with grass with 30 and 45 cm in winter.

**Key Words:** defoliation, tiller density population, seasons of the year

**W166 Effects of exogenous fibrolytic enzymes on in vitro digestibility and gas and methane production of corn silage.** Vania R. Vasconcelos\*<sup>1,2</sup>, Kathy G. Arriola<sup>2</sup>, Andressa Campos<sup>2</sup>, Felix X. Amaro<sup>2</sup>, Rafael M. Martins<sup>2</sup>, Musibau A. Bamikole<sup>2,3</sup>, and Adegbola T. Adesogan<sup>2</sup>, <sup>1</sup>Universidade Federal do Piauí, Campus Agrícola da Socopo, Teresina, Piauí, Brazil, <sup>2</sup>Department of Animal Sciences, University of Florida, Gainesville, FL, <sup>3</sup>Department of Animal Science, University of Benin, Benin City, Nigeria.

This study examined effects of enzyme type and duration of the enzyme-substrate interaction (preincubation) period before in vitro fermentation on the in vitro digestibility and gas and methane production of corn silage (CS). Exactly 1 mL of 0.1 M citrate phosphate buffer (pH 6.0; Control) or the buffer and either xylanase (XYL; 1 mg/g) or xylanase-cellulase (MIX 3.4 mg/g) fibrolytic enzyme preparations were incubated with 0.5 g of dried, ground CS at room temperature for 0, 12, 24 and 48 h in a 120-mL culture bottle. Subsequently, 40 mL of buffered-rumen fluid was added to each bottle and the contents were incubated for 24 h at 39°C. At least 3 replicates of each treatment combination were examined in each of 5 runs. Total gas and methane production, volatile fatty acids (VFA), pH and in vitro digestibility of dry matter (DMD) and neutral detergent fiber (NDFD) were measured. The data were analyzed as a completely randomized design with a 3 × 4 factorial treatment arrangement using the GLIMMIX procedure of SAS (SAS Inst. Cary, NC). There were no ( $P > 0.10$ ) enzyme by time interactions for any measure except DMD ( $P = 0.09$ ). Preincubating MIX for 12 or 24 h with CS resulted in less ( $P < 0.05$ ) DMD than that at 0 h but similar DMD to that at 48 h. Preincubating XYL with CS for 12 h resulted in greater ( $P < 0.05$ ) DMD than that at 0, 24 or 48 h. Both MIX and XYL increased NDFD but MIX had the greatest value ( $P < 0.05$ ; 58.6 and 53.6 vs 51.0%, respectively). Compared with CON, MIX increased gas production (40.1 vs. 42.9 mL) and decreased methane production ( $P < 0.05$ ; 0.023 vs. 0.019 mg/mL) but XYL had similar values to the other treatments (41.4 mL and 0.021 mg/mL, respectively). Across enzyme treatments, pH was greater after 12 h of preincubation (6.34 vs. 6.28) and gas production was greater after 48 h (45.5 vs. 40.17 mL) compared with other incubation times. The MIX enzyme was more effective than the XYL enzyme at increasing the fermentation and digestion of CS. Enzyme-substrate preincubation

period had enzyme-specific effects on DMD and no effects on NDFD or other fermentation measures.

**Key Words:** fibrolytic enzyme, gas production, digestibility

**W167 Use of two natural antimicrobial additives in solid-state fermentation of apple bagasse.** J. L. Guevara-Valdez\*, C. Rodriguez-Muela, L. A. Duran-Melendez, D. Diaz-Plascencia, E. Santellano-Estrada, and R. Marquez-Melendez, *Universidad Autonoma de Chihuahua, Chihuahua, Chihuahua, Mexico.*

The objective of this study was to evaluate the antimicrobial effect of oregano essential oil and zeolite in solid state fermentation (SSF) of apple bagasse using 5% clinoptilolite zeolite (ZEO), oregano essential oil (0.1% OEO) and both additives (ZXA), compared against the traditional method including apple bagasse, urea, ammonium sulfate and mineral mix as control (CTL). The experiment included 3 replicates per treatment in laboratory plastic containers of 1 L, with sampling at 0, 6, 12, 24, 48, 72 and 96 h. Data were analyzed by repeated measures over time and multiple means comparison. Variables analyzed were aerobic mesophilic bacteria by plate count, pH with a potentiometer, and yeast count by improved Neubauer chamber. Results showed that maximum growth of yeast were obtained at 48 h in ZXA ( $462 \times 10^6$  cells/g), while CTL achieved it at 96 h ( $470.5 \times 10^6$  cells/g), thereby reducing the fermentation time and optimizing the process by the oregano extract antimicrobial activity and the capacity of the zeolite to regulate the concentration of nitrogen compounds. The count of aerobic mesophilic bacteria was decreased significantly ( $P < 0.01$ ) at 48 h, from  $0.70 \times 10^6$  cfu/g in CTL to  $0.25 \times 10^6$  cfu/g in ZXA, being OEO the lower bacterial count treatment ( $0.11 \times 10^6$  cfu/g). There was a higher pH ( $P < 0.01$ ) in ZEO (4.80) and ZXA (4.62) versus CTL (4.06) and OEO (4.03), indicating that the adsorptive capacity of the zeolite allowed a slow and adequate release of the ammonia in the zeolite to the fermentation medium, resulting in a most efficient use of ammonia by yeasts, also regulating the pH and preventing volatile loss. We conclude that the use of both additives provides microbiological advantages over traditional SSF, reducing the fermentation time since yeasts have more available nitrogen compounds, and less foreign bacterial contamination, which could compete for substrates, production of secondary metabolites or modify pH. This guarantees a clean and free microbiological contaminants process.

**Key Words:** yeast, mesophilic bacteria, apple bagasse

**W168 In vitro fermentation and digestion characteristics of shrubs *Croton cortesianus* and *Leucophyllum frutescens* browsed by white-tailed deer (*Odocoileus virginianus*).** M. S. Alvarado<sup>1</sup>, M. A. Cerrillo-Soto<sup>1</sup>, M. Guerrero-Cervantes\*<sup>1</sup>, A. S. Juárez-Reyes<sup>1</sup>, R. G. Ramírez<sup>2</sup>, H. Rodríguez<sup>2</sup>, and T. G. Domínguez<sup>1</sup>, <sup>1</sup>Universidad Juárez del Estado de Durango, Durango, Mexico, <sup>2</sup>Universidad Autónoma de Nuevo León, San Nicolás de los Garza, Nuevo León, México, <sup>3</sup>Universidad Autónoma de Nuevo León, Linares, Nuevo León, México.

Native vegetation in northeastern Mexico is mainly composed of shrubs and small trees, which are commonly selected by white-tailed deer. The aim of the study was to determine, seasonally, the in vitro fermentation profiles of *Croton cortesianus* and *Leucophyllum frutescens*. Foliage was sampled from summer 2004 to spring 2005 for a total of 4 consecutive seasons in Los Ramones in the state of Nuevo Leon, Mexico. In vitro gas production was recorded at 3, 6, 9, 12, 24, 48, 72 and 96 h. As inoculum, rumen fluid from fistulated sheep was utilized. Kinetic parameters

such as the asymptotic gas production (B), rate of gas production ( $k$ ) and lag phase (L) were estimated by the exponential model  $G = b \times (1 - e^{-k(t-L)})$ . Microbial protein synthesis, ME content and in vitro organic matter digestibility (IVOMD) were also evaluated. Data were analyzed using ANOVA for a completely randomized design. The asymptotic gas production (B) ranged from 151 mL/g DM in *L. frutescens* in spring to 220 mL/g DM in *C. cortesianus* in winter. The rate of gas production ( $c$ ) was the lowest in *C. cortesianus* in summer (0.050%/h) while the highest (0.082%/h) in *L. frutescens* collected in winter. Values regarding lag phase (L) ranged from 0.43 to 22.06 h; in general, this variable was superior in *C. cortesianus* during summer, autumn and winter (2.06, 1.60 and 1.23 respectively). Microbial protein synthesis measured as purines was similar among shrubs and seasons. Mean values ranged from 4.04 in to 7.80  $\mu\text{mol}$  in *C. cortesianus* in spring and winter. Estimates of ME were highest in *C. cortesianus* among seasons (summer 1.73; autumn, spring 1.70 and winter 1.66 Mcal/kg DM). The same scenario was registered with the IVOMD content where values varied from 55 to 84%. Highest digestibility values were registered in *C. cortesianus*. Data suggest that *C. cortesianus* represents a better feed reserve for white-tailed deer in the semiarid regions of northeastern Mexico.

**Key Words:** gas production, white-tailed deer, native shrubs

**W169 Effect of lactic acid bacteria with bacteriocinogenic activity on the microbial populations and fermentation of alfalfa silage in tropical conditions.** V. P. Silva, O. G. Pereira\*, K. G. Ribeiro, T. C. Da Silva, M. C. N. Agarussi, L. D. A. Rufino, R. M. Martins, F. X. Amaro, and A. M. Corrêa, *Universidade Federal de Vicosa, Vicosa, Minas Gerais, Brazil.*

Lactic acid bacteria (LAB) are used as silage additives to ensure the conservation of the nutritive value of forage by reducing the pH through acid production and inhibition of undesirable microorganisms in the epiphytic flora. The objective of this work was to evaluate the effect of a commercial inoculant and 2 strains of LAB with potential bacteriocinogenic activity on the microbial populations and the fermentation of alfalfa silage. The alfalfa was harvested 82 d after sowing and was wilted for 6 h. After, it was chopped into a 1.5 cm particle size and packed in bags, with 3 replications. A  $4 \times 6$  factorial arrangement (4 inoculants and 6 fermentation periods) was used in a completely randomized design. The inoculants (I) were: 1) Control; 2) Sil All (CI; Alltech, Brazil); 3) Strain 10.4 (S-10.4; *Pediococcus acidilactici*); and 4) Strain 6.16 (S-6.16; *P. pentosaceus*). These strains have potential bacteriocinogenic activity and were isolated from *Stylosanthes* silage. The fermentation periods (P) were: 1, 3, 7, 14, 28 and 56 d after ensiling. The concentration of lactic acid and propionic acid were not affected by the interaction  $I \times P$  ( $P > 0.05$ ). There was effect of I on the DM, pH, ammonia, lactic acid and acetic acid. Silages treated with S-10.4 and S-6.16 resulted in the lowest pH. After 56 d, all silages treated with inoculants had lower ( $P < 0.05$ ) pH compared with the control. The silage treated with S-6.16 showed the highest ( $P < 0.05$ ) concentration of lactic acid (3.07% of DM) at 56 d. There was no effect ( $P > 0.05$ ) of I on the concentrations of CP, NDF, ADIN, and lignin at 56 d. Only the population of enterobacteria was affected ( $P < 0.05$ ) by P, showing a decrease overtime and the lowest number on d 14 (1.05 cfu/g). The strains evaluated improved the fermentation of alfalfa silage in tropical conditions by reducing the concentration of ammonia nitrogen, and increasing the concentration of lactic acid, leading to a faster pH drop and control of undesirable microorganisms.

**Key Words:** inoculant, lactic acid bacteria, strain



**W170 Effect of wilting and inoculant on microbial populations and fermentation profile of alfalfa silage in tropical conditions.**

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The objective of this study was to evaluate the effects of wilting (W) and microbial inoculant (I) on the fermentation characteristics and microbial populations of alfalfa silage. Data were analyzed as a  $2 \times 2 \times 6$  factorial arrangement with the effects of wilting (absence or presence), microbial inoculant (with and without) and period of fermentation (P) (1, 3, 7, 14, 28 and 56 d), and their interaction in a completely randomized design with 3 replications. Alfalfa was harvested at 56 d of regrowth and chopped into particles of 1.5 cm. Alfalfa was wilted for 6 h in the field before ensiling, in the material submitted to this treatment. The inoculant used was Sil-All 4x4 W.S. (Alltech, Brazil), applied at the rate of  $10^5$  cfu/g of fresh forage. After applying treatments the forage was packed in plastic bags of  $25 \times 35$  cm and sealed by using vacuum sealer. Wilting increased the DM content of the fresh alfalfa from 133.9 g/kg to 233.4 g/kg DM and the population of lactic acid bacteria (LAB) from  $5.28 \log$  cfu/g to  $6.88 \log$  cfu/g before ensiling. The number of enterobacteria and yeasts were affected ( $P < 0.05$ ) by P, registering a higher number of these microorganisms on the first day of ensiling. The pH and ammonia nitrogen concentration were affected by the interaction  $W \times P$  ( $P < 0.05$ ), recording the lowest values on wilted-alfalfa silages from the 7th day of ensiling. There was effect of the interaction  $W \times P$  ( $P < 0.05$ ) on the concentrations of lactic, acetic and propionic acids. Except for the first day of fermentation, we observed higher concentrations of lactic acid in the wilted silages, however in both wilted and non-wilted silages the values decreased from the 7th day of ensiling. The concentration of acetic acid in the wilted alfalfa was lower than non-wilted alfalfa in all period of fermentation. The wilting of alfalfa was not enough to promote adequate silage fermentation in presence or absence of microbial inoculant.

**Key Words:** pH, organic acid, microbial inoculant

**W171 Seeding rate affects performance of ball clover mixed with annual ryegrass in North Florida.**

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Legumes have the capacity to associate with rhizobia strains and fix atmospheric  $N_2$ . Ball clover (*Trifolium nigrescens* Viv.) has reseeding ability and when planted in mixtures with annual ryegrass (*Lolium multiflorum* Lam.) contributes to add N to the systems and to expand the grazing season. This experiment tested 3 seeding rates of ball clover (0.9, 1.8 and  $2.7 \text{ kg ha}^{-1}$ ) in a mixture with annual ryegrass compared with annual ryegrass in monoculture, fertilized ( $50 \text{ kg N ha}^{-1}$ ) or not with N. Response variables included percentage of ball clover, dry matter yield (DMY) annual ryegrass, DMY ball clover, total DMY, percentage of plant N derived from atmosphere (%Ndfa) and N fixed contained in the shoot ( $N_{\text{fix}}$ ). The study was performed in a complete randomized block design with 4 replicates. Ball clover proportion in the mixture increased ( $P = 0.02$ ) from 32% (at  $0.9 \text{ kg ha}^{-1}$  seeding rate) to 47% (at  $1.8 \text{ kg ha}^{-1}$  seeding rate), with no significant differences ( $P > 0.05$ ) observed between the 2 higher seeding rates ( $1.8$  and  $2.7 \text{ kg ha}^{-1}$ ). Ball clover DMY increased linearly ( $P = 0.02$ ) with increasing seeding rates, with DMY of 711, 1,117, and  $1,165 \text{ kg ha}^{-1}$  for 0.9, 1.8, and  $2.7 \text{ kg ha}^{-1}$  seeding rates, respectively. Annual ryegrass DMY ( $P < 0.0001$ ) and total DMY ( $P = 0.0007$ ) were greater in the N fertilized treatment

( $3,810 \text{ kg DM ha}^{-1}$ ). Average %Ndfa was 95% for ball clover and there was no difference among treatments ( $P > 0.05$ ). The  $N_{\text{fix}}$  showed linear increase with seeding rate ( $P = 0.02$ ) with values ranging from 18 to  $28 \text{ kg N ha}^{-1}$ . Treatments including seed rates of 1.8 and  $2.7 \text{ kg ha}^{-1}$  were similar in botanical composition, DMY ball clover, DMY ryegrass, total DMY, and  $N_{\text{fix}}$ . From the producer perspective, it is more economical to use  $1.8 \text{ kg seeds of ball clover ha}^{-1}$ . Increasing seed rates of ball clover in annual ryegrass mixtures allowed greater legume contribution in the pasture, improving forage N without decreasing yield.

**Key Words:** botanical composition, legume, N-fixation

**W172 Effects of hybrid and inoculant application on chemical composition and fermentation indices of barley silage.**

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This study examined the effect of hybrid and inoculant application on the chemical composition, fermentation indices and fatty acid profile of barley silage. Two Korean barley hybrids (Yuyeon, YU and Youngyang, YO) were harvested at dough stage, which were 24.9% and 27.1% of dry matter, respectively. The forages were chopped to 4–6cm lengths and treated with or without inoculant at the ratio of  $1.2 \times 10^4$  cfu/g of *L. plantarum* on fresh weight basis. Treatments had a  $2 \times 2$  factorial arrangement with 4 replicates. Each replicate was ensiled in a 10-L mini silo for 100 d and analyzed for chemical composition and in vitro DM digestibility. Data were analyzed with a model including hybrid, inoculant and the interaction using the GLM procedure of SAS. The YU silage had greater ( $P < 0.01$ ) ash, acid detergent fiber and in vitro dry matter digestibility and mold counts than YO silage, but lower ( $P < 0.01$ ) lactate and acetate concentrations. For YU but not YO, inoculation decreased the NDF concentration ( $P = 0.002$ ), lactic to acetic acid ratio ( $P = 0.004$ ), lactic acid bacteria ( $P = 0.007$ ) and yeast counts ( $P < 0.001$ ) and increased the pH ( $P = 0.032$ ) and ammonia-N concentration ( $P = 0.026$ ) (hybrid  $\times$  inoculant interaction). The YU silage had greater ( $P < 0.005$ ) C14:0, C16:0, C18:3n-3 and saturated fatty acid concentrations than YO silage, but lower ( $P = 0.001$ ) C18:2n-6 concentration. Inoculation decreased ( $P < 0.05$ ) C18:3n-3 concentrations, but increased ( $P < 0.05$ ) C18:2n-6 and saturated fatty acid concentrations. For YU but not YO, inoculation decreased total fatty acid ( $P = 0.034$ ) and polyunsaturated fatty acid ( $P = 0.003$ ) concentrations but increased C18:0 ( $P = 0.011$ ), C18:1n-9 ( $P = 0.044$ ), monounsaturated fatty acid ( $P = 0.033$ ) concentrations (hybrid  $\times$  inoculant interaction). The PCR analysis followed by gel electrophoresis showed higher concentration of *L. plantarum* DNA in the inoculated silage. In conclusion, YU silage was more digestible but it had greater mold counts than YO silage. Inoculation did not improve the fermentation but it reduced the acetate and 18:3n-3 fatty acid concentrations and increased the C18:2n-6 and saturated fatty acid concentration.

**Key Words:** barley silage, fatty acid profile, inoculant

**W173 Comparison of in situ digestion of corn stover treated by two alkali methods to untreated corn stover and soyhulls.**

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The objective of this study was to evaluate the in situ NDF digestibility of corn stover treated at ambient temperature with a solution of sodium hydroxide and ethanol co-solvent. The treatment process is patented by Cellulose Sciences International (CSI) of Madison, WI. Digestion kinetics were evaluated in situ for untreated corn stover, corn stover treated with CaOH at 70 g kg<sup>-1</sup>, corn stover treated by the CSI process and untreated soyhulls. All materials were ground through a 2 mm screen in a Wiley mill and 0.5 g was sealed into Ankom F57 bags. Bags were placed in the rumens of 2 fistulated cows for 0h, 12h, 24h, 30h, 36h, 42h, 48h, 96h, 120h, and 240 h in reverse order. Bags were then washed, dried and contents analyzed for residual NDF. The 240h time point was used as the estimate of indigestible NDF (iNDF). The rate of digestion of potentially digestible NDF (pdNDF) was determined from the slope of the natural log of the pdNDF residue verses time. The fraction of iNDF and the rate of pdNDF degradation between forages were compared by ANOVA (PROC Mixed, SAS, v 9.3) as a randomized design with cows as replicates. Least square treatment means were compared by PDIF. The digestion rate of pdNDF (kd,) of CSI-treated corn stover (5.36%/h) was higher than CaOH-treated stover (2.27%/h,  $P < 0.01$ ), and untreated corn stover (1.76%/h,  $P < 0.001$ ). The kd of soyhulls (4.93%/h) was similar CSI-treated stover ( $P = 0.56$ ). The iNDF proportions in CSI-treated stover and soyhulls, were similar (2.8 and 3.6% of NDF, respectively,  $P = 0.26$ ). The CSI-treated stover had lower iNDF than CaOH-treated stover or untreated stover (2.8, 27.3 and 35.1% of NDF, respectively,  $P < 0.0001$ ). Results indicate that the CSI-treatment process improved rate and extent of digestion of corn stover to a greater degree than the CaOH treatment process. The CSI treatment process appears to convert corn stover into a source of highly digestible fiber with digestion characteristics similar to soyhulls.

**Key Words:** in situ, NDF, NaOH

**W174 Effects of different source additive and wilt condition on the pH value, aerobic stability, and carbohydrate and protein fractions of alfalfa silage.** Lian Tao<sup>1</sup>, He Zhou<sup>2</sup>, Nai-feng Zhang<sup>1</sup>, Bing-wen Si<sup>1</sup>, Yan Tu<sup>1</sup>, Tao Ma<sup>1</sup>, and Qi-yu Diao\*<sup>1</sup>, <sup>1</sup>Feed Research Institute, Chinese Academy of Agricultural Sciences, Key Laboratory of Feed Biotechnology of the Ministry of Agriculture, Beijing, China, <sup>2</sup>Institute of Grassland Science, College of Animal Science and Technology, China Agricultural University, Beijing, China.

To improve alfalfa silage quality and reduce additive cost, the potential of applying the fermented juice of epiphytic lactobacillus (FJEL) as an additive was evaluated. The experiment was designed as a 5 × 3 2-factor (wilt condition and additive) test. The second cut alfalfa at the budding stage was harvested, exposed to sunlight for 0 (20.04% DM; type 1), 5.2 (34.15% DM; type 2) or 8.5 h (54.58% DM; type 3). Alfalfa stuff were harvested in the next morning, exposed to sunlight and then natural rainfall for 1 h (33.20%DM; type 4) or 3 h (24.58% DM; type 5) before cutting. Alfalfa forage of each type was cut to 1–2 cm, then treated with distilled water (control), FJEL, or commercial lactobacillus product (CLP) at 10 ml/kg fresh weight (FW). The FJEL was made from alfalfa juice that was anaerobically stored for 2 d. The treated forages (approximately 500 g) were ensiled in vacuum-sealed polyethylene bags for 45 d, with 3 replicates for each treatment. The statistical significance of the effects of additive, wilt condition, and additive × wilt condition on silage quality were analyzed using a repeated measures one-way ANOVA. The application of FJEL decreased ( $P < 0.01$ ) the pH value (4.44 vs. 4.93 or 4.66) and volatile fatty acid (VFA; 38.32 vs.49.55 or 44.82) content and increased ( $P < 0.01$ ) the concentrations of lactic acid (LA; 68.99 vs.51.84 or 63.29) compared with those of the control and CLP treatment. However, the FJEL treatment had the lower ( $P <$

0.01) aerobic stability (254 vs. 274 h), soluble fiber (148.89 vs. 154.85 g/kg DM), available NDF (145.39 vs. 154.25 g/kg DM), rapidly rumen degradable CP (59.91 vs. 62.33 g/kg DM) than CLP treatment. A higher pH and lower LA, sugar, starch and soluble fiber contents were found in the rain treatments compared with those of the no-rain treatments ( $P < 0.01$ ). In conclusion, the application of FJEL could improve silage quality compared with the control; in addition, its effect as a fermentation stimulant may be comparable to or even better than CLP.

**Key Words:** alfalfa silage, wilt condition, epiphytic lactic acid bacteria

**W175 Yield and nutritive value for ruminants of organic winter cereals–bard vetch intercrops.** Alexey Diaz<sup>1,4</sup>, María Dolores Carro<sup>2</sup>, Carlos Palacios<sup>3</sup>, Cristina Saro<sup>1</sup>, Iván Mateos<sup>1</sup>, María Luisa Tejido<sup>4</sup>, and María José Ranilla\*<sup>1,4</sup>, <sup>1</sup>Animal Production Department, University of León, León Spain, <sup>2</sup>Agriculture Production Department, Technical University of Madrid, Madrid, Spain, <sup>3</sup>Construction and Agronomy Department, University of Salamanca, Salamanca, Spain, <sup>4</sup>IGM (CSIC-ULE), Finca Marzanas s/n, Grulleros, León, Spain.

Organic livestock in the European Union must be fed with organic feed, and forage should be at least 60% of the diet of herbivorous animals. However, the productivity of crops in organic farming is frequently lower than that in conventional farming, and organic cultivation of winter cereals often reduces forage CP content. Intercropping of winter cereals with legumes can provide both higher forage yield and quality compared with winter cereal monocultures, but the results may differ with cultivation conditions. The objective of this study was to investigate the potential of winter oats and barley intercropping with bard vetch (*Vicia articulata*) to enhance forage yield and quality as compared with cereals as a monoculture. The experiment was carried out in the province of Zamora (Spain) in 2014, as a randomized complete block design with 4 replications. Whole-plant DM yield was determined in June and chemical composition and in vitro DM digestibility (IVDMD) of harvested forages were analyzed. In addition, samples (500 mg) of each forage were incubated with 50 mL of buffered sheep rumen fluid at 39°C for 24 h and VFA production was measured. Intercropping with bard vetch increased ( $P < 0.05$ ) forage yield by 25 and 18% for oats and barley, respectively, compared with monoculture. Intercropping also increased CP contents ( $P < 0.05$ ; mean values 629 vs. 949 g/kg DM), and decreased ( $P < 0.05$ ) NDF and ADF contents. The IVDMD was also higher ( $P < 0.01$ ) for intercrops than for cereal monocultures. The presence of bard vetch in the cultures increased total VFA production ( $P < 0.001$ ) by 27 and 15% for oats and barley, respectively. This was due to an increase in the production of acetate, propionate and butyrate ( $P < 0.05$ ), but no effects ( $P > 0.05$ ) of intercropping were detected for the acetate:propionate ratio or the molar proportions of individual VFA. The results indicate that forage yield and quality of oats and barley can be enhanced by intercropping with bard vetch under the cultivation conditions of this study. These results should be confirmed in multi-year studies before drawing up guidelines for organic farmers.

**Key Words:** organic cultivation, cereal, legume intercropping

**W176 Comparison of total tract neutral detergent fiber digestibility of different varieties of barley silage selected on the basis of in vitro NDF degradability.** Natalie G. Preston\*<sup>1,2</sup>, Jayakrishnan Nair<sup>1</sup>, Peiqiang Yu<sup>1</sup>, David A. Christensen<sup>1</sup>, John J. McKinnon<sup>1</sup>, and Timothy A. McAllister<sup>2</sup>, <sup>1</sup>University of Saskatchewan, Saskatoon,

SK, Canada, <sup>2</sup>Agriculture and Agri-Food Canada, Lethbridge, AB, Canada.

Selecting forages with increased neutral detergent fiber (NDF) digestibility has been hypothesized to increase dry matter intake (DMI) in ruminants, but this hypothesis has not been tested using barley (*Hordeum vulgare*) silage. The objective of this study was to compare the total-tract NDF digestibility of different barley silage varieties selected for increased in vitro NDF degradability at 30 h using rumen cannulated sheep. Nine cannulated wethers in a triplicated 3 × 3 Latin square design were fed a 50:50 barley silage: concentrate pellet ration on a dry matter (DM) basis. Varieties of barley silage ensiled and fed included; CDC Cowboy, CDC Copeland, and Xena, based on selection for high, intermediate, and low NDF degradability, respectively. Of the 7 ensiled barley varieties tested, NDF degradability after 30 h incubation in an ANKOM fiber analyzer, CDC Cowboy had the highest NDF degradability at 34.2% of DM, CDC Copeland and Xena were selected based on their intermediate and lower degradability of 28.3% and 26.2%, respectively ( $P < 0.05$ ). Lambs were fed ad libitum during a 12 d adaptation period. On d 12–14, lambs were restricted to 95% intake during which ruminal pH was recorded over 48 h using rumen pH loggers set to record pH every minute. On d 18, lambs were restricted to 90% intake, and housed in metabolic crates for total fecal collection over 4 d. Data were analyzed using the Proc Mixed procedure of SAS as a triplicate Latin square with day as a repeated measure within period. Lambs fed Xena had higher ( $P < 0.01$ ) DMI (773 g/d) as compared with those fed Cowboy (667 g/d), both of which did not differ from Copeland (714 g/d). Digestibility of DM was greater ( $P < 0.05$ ) for Copeland (69.3%) than Cowboy (65.6%) both of which did not differ from Xena (67.5%). Ruminal pH was reported as mean, max, min, and time < 5.8. Treatment did not affect mean, max, or min pH ( $P > 0.05$ ), but time pH < 5.8 was longer ( $P < 0.05$ ) for Xena, 301 min/d, than Cowboy, 123 min/d. Based on DM digestion and intake it can be inferred that selecting varieties based on improved in vitro NDF degradability does not improve DMI or total-tract digestibility of barley silage in sheep.

**Key Words:** barley silage, neutral detergent fiber, digestibility

**W177 Effect of reduced lignin alfalfa on forage quality at 3 harvest intervals.** Zhiqiang Li<sup>1,2</sup>, Dan J. Undersander<sup>1</sup>, David K. Combs<sup>1</sup>, and Zhenzhen Li<sup>1,2</sup>, <sup>1</sup>University of Wisconsin, Madison, WI, <sup>2</sup>Department of Grassland Science, China Agricultural University, Beijing, China.

The present study investigated the effect of reduced lignin alfalfa on forage quality. A 2 × 3 factorial design was used. Two alfalfa cultivars (Roundup Ready (RR) and double stacked RR and Reduced Lignin (RR/RL)) were harvested at intervals of 28d, 33d, and 35d. All replicated plots were established in June and harvested for 2 regrowth periods in July and August in 2014. Four replicates were used. Statistical analysis was a 2-way ANOVA using SPSS 22. Crude Protein (CP), Neutral Detergent Fiber (aNDF), Lignin, NDF Digestibility in 48 h (NDFD48) were analyzed by NIRS (FOSS 6500 using NIRSC 2012 Alfalfa Hay Equation). CP content was not significantly different between RR and RR/RL (27.5 vs 28.0,  $P > 0.05$ ) and declined with advancing maturity ( $P < 0.05$ ). aNDF content of RR was significantly higher than RR/RL (31.6 vs 30.1), though differences among harvest intervals were not significant (30.8 vs 30.5 vs 31.1). Lignin content was not significantly different between RR and RR/RL (5.6 vs 5.5) or among harvest intervals (5.6 vs 5.6 vs 5.5). The NDFD48 of RR/RL was significantly higher than RR (52.2 vs 50.5). Indigestible NDF (iNDF), Rate of fiber digestion ( $K_d$ ), and Total-tract NDF Digestibility (TTNDFD) had not been reported for the RL germplasm and were shown in the table below.

The reduced lignin alfalfa tended to be lower in iNDF and higher in  $K_d$  though differences were not significant, but was significantly higher in TTNDFD. It appears that RR/RL reduced the aNDF content and improved NDFD48 and TTNDFD.

**Table 1 (Abstr. W177).** Comparison between RR and RR/RL in iNDF,  $K_d$  and TTNDFD

Item		RR	RR/RL	Average	P-value		
					Forage	Harvest interval	Forage × harvest interval
iNDF (%NDF)	28 d	34.1	29.9	31.7 <sup>b</sup>	0.086	0.001	0.470
	33 d	42.1	37.0	39.9 <sup>a</sup>			
	35 d	39.3	39.2	39.2 <sup>a</sup>			
	average	38.7 <sup>a</sup>	35.1 <sup>a</sup>	36.8			
$K_d$ (%)	28 d	7.8	7.1	7.4 <sup>a</sup>	0.156	0.980	0.153
	33 d	7.0	8.2	7.5 <sup>a</sup>			
	35 d	6.4	8.9	7.8 <sup>a</sup>			
	average	7.0 <sup>a</sup>	8.0 <sup>a</sup>	7.6			
TTNDFD (%NDF)	28 d	53.0	56.3	54.9 <sup>a</sup>	0.012	0.010	0.849
	33 d	46.3	51.9	48.7 <sup>b</sup>			
	35 d	46.8	51.1	49.2 <sup>b</sup>			
	average	48.6 <sup>b</sup>	53.3 <sup>a</sup>	51.0			

**Key Words:** variety, harvest interval, hay quality

**W178 Microbial counts, fermentation, and aerobic stability of oats with and without combo inoculant ensiled in vacuum bags and plastic bucket silos.** Juan J. Romero<sup>\*1</sup>, Jinwoo Park<sup>2</sup>, Youngho Joo<sup>2</sup>, Yuchen Zhao<sup>3</sup>, Axel Gonzalez<sup>1</sup>, Marco A. Balseca-Paredes<sup>1</sup>, and Miguel S. Castillo<sup>1</sup>, <sup>1</sup>Department of Crop Science, North Carolina State University, Raleigh, NC, <sup>2</sup>Department of Animal Science, Gyeongsang National University, Jinju, Korea, <sup>3</sup>Department of Animal Nutrition and Feed Science, China Agricultural University, Beijing, China.

The objective was to evaluate the use of 2 types of experimental silos to characterize microbial counts, fermentation, and aerobic stability of oats ensiled with and without addition of a homolactic and heterolactic inoculant. From each of 6 sections in a field, whole-plant oats at heading stage were harvested, wilted, chopped, treated (INO) or not (CON) with inoculant, packed into 20L plastic bucket silos (BKT) or 15.2 × 30.5cm nylon-polyethylene embossed vacuum bags (BAG), and ensiled for 217d. The inoculant added contained *Lactobacillus buchneri* and *Pediococcus pentosaceus* ( $4 \times 10^5$  and  $1 \times 10^5$  cfu/g of fresh oats, respectively). Experimental design was a complete randomized design replicated 6 times. Treatment design was the factorial combination of 2 silo types × 2 inoculation treatments. At d 0 there were no differences between BAG and BKT, and between INO and CON on DM ( $44.0 \pm 1.0$ , %), pH ( $6.11 \pm 0.07$ ), and counts of lactic acid bacteria ( $9.28 \pm 0.13$ ), yeasts ( $4.88 \pm 0.09$ ), and molds ( $3.97 \pm 0.08$ ). At opening (d 217) there was no difference in DM ( $42.9 \pm 0.9$ , %) among treatment combinations. There was a lower pH ( $4.25$  vs.  $4.41 \pm 0.02$ ), yeasts ( $1.10$  vs.  $4.13 \pm 0.34$ ), and NDF ( $65.0$  vs.  $67.0 \pm 0.40$ ) for INO compared with CON, respectively ( $P \leq 0.05$ ). Lactic acid bacteria count ( $6.39$  vs.  $5.65 \pm 0.17$ ), DM recovery ( $96.1$  vs.  $92.9 \pm 0.43$ , %), and aerobic stability ( $565$  vs.  $133 \pm 29$ , h) were greater in INO compared with CON, respectively ( $P \leq 0.05$ ). Molds were similar when INO was applied to BKT (1.68) and BAG (1.42) but higher in BKT (2.82) compared with BAG ( $0.80 \pm 0.41$ ) for CON (silo type × inoculation,  $P \leq 0.05$ ). In summary, differences due to silo type were only significant for mold count at opening. The



combo inoculant used increased lactic acid bacteria, decreased pH and NDF, and improved aerobic stability by decreasing yeast count of oats silage. Silo bags are an alternative technique to buckets to adequately characterize lactic acid bacteria, yeasts, and fermentation of ensiled oats.

**Key Words:** silo, microbe, inoculant

**W179 Change in chemical and microbial composition during aerobic challenge of maize silage with and without *L. buchneri* inoculation.** Ida K. Hindrichsen\*, Nina Milora, Marianne Richelieu, and Asger Geppel, *Chr. Hansen A/S, Hørsholm, Denmark.*

Inoculation of *L. buchneri* (LB) is well recognized for extending aerobic stability of silage during feed out. The effect is mainly documented by measuring temperature change during small scale aerobic challenge. The objective of the current study was to investigate the microbial and chemical changes occurring during re-exposure of air to mini-silos inoculated with or without *L. buchneri* (LB). Newly harvested maize was collected from a farm in Denmark in September 2013. The maize was inoculated with tap water or LB and vacuum packed in 1-kg bags and stored at 25°C for 2, 7 or 15 weeks (n = 5 per treatment). For the aerobic challenge, the silage was divided into 2 containers (1.2 L; 274 kg/m<sup>3</sup>) with a 1 cm hole in the top and bottom, and kept at room temperature for 156 h. After aerobic challenge for 0, 3 and 7 d chemical and microbial analysis were performed on all samples. Data were statistically analyzed as a randomized complete block by using GLM procedure by SAS. The none-fermented maize was highly contaminated with yeast and mold of log 6 and log 5, respectively. Aerobic stability and number of yeast was not significantly ( $P > 0.05$ ) improved by inoculating with LB after 2 weeks, but LB inoculation improved the aerobic stability and reduced number of yeast significantly ( $P < 0.05$ ) compared with control, when the silage was kept anaerobic for 7 or 15 weeks. During the aerobic challenge both the microbial and chemical composition changed. Yeast enumeration significantly ( $P < 0.05$ ) increased during the aerobic challenge, except for the LB inoculated silage after 15 weeks of anaerobic fermentation, where yeast count was lower than detection level (log 2). Acetate, ethyl acetate, lactate, and ethanol levels decreased significantly ( $P < 0.05$ ) during aerobic challenge for both treatments and anaerobic fermentation time points. Considerable chemical changes occur during aerobic challenge and with increased anaerobe fermentation the effect on aerobic stability by inoculation with LB improves.

**Key Words:** volatile organic compound, maize silage, aerobic stability

**W180 Biological N<sub>2</sub> fixation and performance of cool-season legumes mixed with annual ryegrass.** Jose C. B. Dubeux Jr.\*<sup>1</sup>, Lynn E. Sollenberger<sup>2</sup>, Ann R. S. Blount<sup>1</sup>, Cheryl Mackowiak<sup>1</sup>, Erick R. S. Santos<sup>1</sup>, Hiran M. S. Silva<sup>1</sup>, and Martin Ruiz-Moreno<sup>1</sup>, <sup>1</sup>North Florida Research and Education Center, University of Florida, Marianna, FL, <sup>2</sup>Agronomy Department, University of Florida, Gainesville, FL.

Symbiotic association between forage legumes and N<sub>2</sub>-fixing microorganisms reduces the need for pasture N fertilization and increases forage N concentration. Pastures of cool-season legumes mixed with annual ryegrass (*Lolium multiflorum* Lam.) are an option to extend the grazing season in subtropical regions. We investigated the association of 4 cool-season legumes with annual ryegrass and contrasted with annual ryegrass in monoculture. Clovers tested included balansa (*Trifolium michelianum* Savi), ball (*Trifolium nigrescens* Viv.), berseem (*Trifolium alexandrinum* L.), and crimson (*Trifolium incarnatum* L.). Treatments were replicated

4 times in a randomized complete block design. Response variables analyzed included total dry matter yield (DMY), clover DMY, annual ryegrass DMY, <sup>15</sup>N grass, <sup>15</sup>N legume, botanical composition, N concentration of grass and legume components, shoot N yield for grass and legume, percentage of N derived from atmosphere (%Nd<sub>a</sub>), and N<sub>2</sub>-fixation by the legume. Legume/annual ryegrass mixtures yielded more biomass (average of 2,590 kg DM ha<sup>-1</sup>) compared with unfertilized annual ryegrass (980 kg DM ha<sup>-1</sup>). Among mixtures, crimson clover/annual ryegrass was the most ( $P < 0.01$ ) productive one (4,580 kg DM ha<sup>-1</sup>). Crimson (2,230 kg DM ha<sup>-1</sup>) and berseem (1,720 kg DM ha<sup>-1</sup>) clovers were the 2 most productive legumes. Annual ryegrass yielded more biomass ( $P < 0.001$ ) when mixed with crimson clover compared with other clover mixtures. Clover percentage in the mixtures ranged from 26 to 74%, with berseem showing the highest proportion. Total shoot N yields were 29, 41, 49, 88, and 11 kg N ha<sup>-1</sup> for balansa, ball, berseem, crimson, and unfertilized ryegrass, respectively. The %Nd<sub>a</sub> for all clovers were high, varying from 67 to 98%. N<sub>2</sub>-fixation ranged from 11 to 52 kg N ha<sup>-1</sup>, with crimson fixing the greatest amount. Crimson clover presented the best overall performance when mixed with annual ryegrass in North Florida.

**Key Words:** balansa, ball, berseem

**W181 Rumen papillae size and blood serum enzymatic cofactors concentration of bulls fattened under two feeding systems at the Mexican dry tropic.** Carlos Rodríguez-Muela\*, Nilda E. Ruiz-Holguin, Gabriela Corral-Flores, José A. Ramírez-Godínez, Alberto Flores-Mariñelarena, Pablo F. Mancillas-Flores, and Claudio Arzola-Alvarez, *Universidad Autónoma de Chihuahua, Chihuahua, México.*

The objective was to evaluate the effect of the feeding system on rumen papillae (RP) development and the animal welfare. The feeding systems used were: Intensive Silvopastoral (SPI) with grazing leucaena and Tanzania and African star grasses; Feedlot system (FLT) with a diet based on 70% of concentrate and 30% of corn stover. Eighty bulls of 2 breeding groups Brahman×Charolais (Bh×Ch) and Brahman×Brown Swiss (Bh×Ps) randomly distributed at both systems were used. The study lasted 195 d. Blood samples were taken at 0, 71, 132, and 195 d. Initial animal's body weight was 195 kg. Animals were killed at 450 kg body weight. Tissue samples of rumen were taken during slaughter of animals. Evaluated variables were length and width of RP using a Vernier caliper. The concentration of Cu, Zn and Mn in blood serum was measured by atomic absorption spectrophotometry. Data were analyzed with a completely randomized design with factorial arrangement 2 × 2. Measures repeated in time and 0d as a co-variable for the blood serum enzymatic cofactors concentration (SEC) were used. SPI favored ( $P < 0.01$ ) RP development, with values of 1.16 ± 0.05 and 1.11 ± 0.05; 0.38 ± 0.03 and 0.36 ± 0.03 cm in length and width of RP of Bh×Ch and Bh×Ps respectively (Table 1). There were a decrease ( $P < 0.01$ ) in Cu, Mn and Zn concentration for sampling day, with lowest values at 132d, due to a possible caloric stress for environmental high temperature and relative humidity. There was effect of system for SEC ( $P < 0.05$ ), with increase for Cu and Mn but decrease of Zn in SPI. We concluded that SPI allowed to a better RP development by the quality of the fiber consumed in the diet, no matter the breed group, and a larger serum concentration of Cu and Mn, although with a significant reduction of Zn concentration.

*Contd.*

**Table 1 (Abstr. 181).** Length and width of RP<sup>1</sup> and SEC of bulls fattened under two feeding systems

Variable	FLT	SPI
RP length (Bh×Ch)	0.63 ± 0.09 <sup>b</sup>	1.16 ± 0.05 <sup>a</sup>
RP length (Bh×Ps)	0.76 ± 0.07 <sup>b</sup>	1.11 ± 0.05 <sup>a</sup>
RP width (Bh×Ch)	0.20 ± 0.05 <sup>b</sup>	0.38 ± 0.03 <sup>a</sup>
RP width (Bh×Ps)	0.22 ± 0.04 <sup>b</sup>	0.36 ± 0.03 <sup>a</sup>
Cu (mg/L)	0.108 ± 0.004 <sup>b</sup>	0.124 ± 0.004 <sup>a</sup>
Mn (mg/L)	0.379 ± 0.010 <sup>b</sup>	0.424 ± 0.011 <sup>a</sup>
Zn (mg/L)	0.533 ± 0.030 <sup>a</sup>	0.406 ± 0.032 <sup>b</sup>

<sup>1</sup>Length and width of RP are expressed in cm.

**Key Words:** papillae, enzymatic cofactors, feeding system

**W182 Effect of kernel processor and theoretical length of cut on physical characteristics of forage corn harvested with one row harvester.** Gilson S. Dias Júnior<sup>\*1</sup>, Nilson N. Morais Júnior<sup>1</sup>, Ronaldo F. Lima<sup>1</sup>, Fabiana F. Cardoso<sup>1</sup>, Ozana F. Zacaroni<sup>1</sup>, Renata A. N. Pereira<sup>3,2</sup>, and Marcos N. Pereira<sup>1,2</sup>, <sup>1</sup>Universidade Federal de Lavras, Lavras, MG, Brazil, <sup>2</sup>Better Nature Research Center, Ijaci, MG, Brazil, <sup>3</sup>Empresa de Pesquisa Agropecuária de Minas Gerais, Lavras, MG, Brazil.

Adequate kernel breakage and sufficient particle size are desirable in corn silage (CS). Most Brazilian dairy farms use one-row forage harvesters. For such machines, a kernel processor (KP) was developed (MU9000462–0 U2 INPI patent). We evaluated the effect of theoretical length of cut (TLOC) and KP on CS particle size and kernel damage. Corn (Dow 2B587 Hx) was harvested at half milk line maturity (36.4% DM, 52.1% NDF, 30.0% starch). Treatments were a factorial combination of TLOC (3, 4.5, 6, 8.5 mm) and KP (with or without), and 50 m rows were harvested in triplicate. Statistical model had effects of TLOC, KP, and their interaction. Particle size distribution was measured with the Penn State Particle Separator. Visible kernels were quantified and classified in extreme (E), poor (P) or intact (I) breakage. Kernel in situ degradation in 12 h was 24.4% for E, 12.6% for P, and 11.5% for I, in 24 h 39.5%, 26.2%, and 23.6% ( $P < 0.05$  E vs. P and E vs. I), and the residue of 72 h 25.2%, 39.4%, and 48.3% (all differ at  $P < 0.05$ ), respectively. The KP reduced visible kernels in forage (44.2 vs. 70.5 g/500 g forage,  $P < 0.01$ ) and the proportion of P+I (23.2 vs. 41.0 g/500 g forage,  $P < 0.01$ ). The TLOC 3 mm without KP (traditional in practice) had more visible kernels (56 g/500 g forage) than 8.5 mm with KP (44 g/500 g forage). The reduction in visible kernels with smaller TLOC was larger when KP was not used ( $P < 0.01$  for interaction). The proportion of particles below the 8 mm screen was 22 and 28% of fresh forage for TLOC 6 and 8.5 mm with KP, respectively, and 35% for 3 mm without KP. The TLOC 3 mm without KP had smaller proportion of starch below the 8 mm screen (21% of starch) than all TLOC with KP (29 to 35% of starch). The TLOC 3 mm without KP had 72% of NDF greater than 8 mm and 6 and 8.5 mm with KP had 79% and 82%, respectively. Although KP reduced NDF particle size, the reduction was not as large as the reduction induced by small TLOC. Reducing TLOC was more effective in reducing NDF particle size when KP was not used ( $P < 0.01$  for interaction). Large TLOC with KP resulted in forage with longer NDF and increased kernel damage than small TLOC without KP, which is nutritionally desirable.

**Key Words:** forage harvester, particle size, corn silage

**W183 The relative effects of external factors on variability of in situ fiber digestion.** Abebe T. Hassen, Fredric Owens<sup>\*</sup>, Lesa Nuzback, Chris Iiams, and Mark Hinds, *DuPont Pioneer, Johnston, IA.*

Differences in fiber digestibility among high-yielding corn silage hybrids usually are small. Therefore, screening tools should be precise. This study was designed to evaluate the relative influence of several external factors on variability among in situ NDF digestibility values, and estimate the potential precision improvements from increasing the number of steers and of replicate samples per steer. Samples from 3 corn hybrids were used as check hybrids during 85 routine in situ trials conducted over several years. Dry ground whole plant from the check hybrids were weighed into sealed Dacron in situ bags; duplicate bags containing each check hybrid were placed into each of 4 larger mesh bags with other samples being assayed. Within each 48 h run, 4 steers were used. Initially data were averaged by hybrid, run, steer, and mesh bag. These then were analyzed by hybrid using a model that included random effects of run, steer, run × steer interaction, and an error term. Data pooled across hybrids were analyzed using models that included the above effects as well as hybrid. In separate models, the hybrid effect was considered fixed and random. Mean in situ fiber digestibility for check hybrids ranged from 56% to 69%; ranking of these hybrids was consistent across runs. Results from the pooled analysis showed that of the total variability, 56% could be attributed to the error term. Contributions of run, steer, and their interaction were 15%, 20%, and 9%, respectively with contributions being similar when analyzed by hybrid. Repeatability SD of hybrid measurements based on 4 data points per steer was 2.2%. Reproducibility SD of measurements based on different steers and runs was 4.4%. Intra-class correlation of measurements from a hybrid evaluated within the same run using different steers was 0.64. Current results showed that SEM would be reduced by 30 to 40% by increasing the number of steers per run from 1 to 4. Based on 4 steers per run, SEM could be reduced an additional 16% by increasing the number of mesh bag per steer from 1 to 4. Additional improvements in precision from resource re-allocation were minimal.

**Key Words:** digestibility, fiber, in situ

**W184 Characterizing corn silage in Brazil.** M. Pinho<sup>1</sup>, M. Martins<sup>1</sup>, D. Combs<sup>2</sup>, J. Goeser<sup>3</sup>, D. Meyer<sup>3</sup>, L. Meyer<sup>3</sup>, Z. Meyer<sup>3</sup>, F. Lopes<sup>4</sup>, and M. H. Ramos<sup>\*1</sup>, <sup>1</sup>3rlab, Belo Horizonte, Minas Gerais, Brazil, <sup>2</sup>University of Wisconsin, Madison, WI, <sup>3</sup>Rock River Laboratory, Watertown, WI, <sup>4</sup>Kemin Industries, Indaiatuba, São Paulo, Brazil.

A data set of 1796 commercial samples of corn silage assessed for nutritive value by NIR was utilized to generate population descriptive information for the dairy industry in Brazil. Representing silage samples were harvested in the year of 2014 and came from 13 states. Corn silage data were queried from a commercial database and population descriptive statistics were generated with MS excel. Average NDF and starch content were 50% and 24% of DM with a SD of 6.67 and 8.05, respectively, suggesting Brazil corn silage is of lesser energetic value. Typical US corn silages average 44 and 29% NDF and starch for comparison. Rumen in vitro NDF digestibility and predicted in vivo TTNDFD (total-tract neutral detergent fiber digestibility) values exhibited substantial variation, with CV ranging from 15.8 to 38.1, suggesting that opportunity exists to improve forage quality. The database presented here showed a great variation in carbohydrate content (NDF and starch) and NDF availability (TTNDFD). To our knowledge, this is one of the first studies documenting corn silage produced in Brazil variability. With the variation in important nutrients and measurements presented here, there may be potential for genetic selection and improvement for

Brazilian growing conditions. Selecting for lower NDF, greater starch and greater fiber digestibility, including greater TTNDFD, will result in hybrids that allow for greater milk production.

**Table 1 (Abstr. W184).**

Item	CP	NDF	Starch	Sugar	Fat	Ash	NDFD			TTNDFD
							at 24 h	at 30 h	at 48 h	
Median	6.9	50.5	25.1	0.9	3.4	3.3	16.4	24.8	40.2	36.9
Average	6.9	50.8	24.5	1.1	3.3	3.3	16.0	24.5	39.9	36.8
SD	1.0	6.6	8.0	1.0	0.7	0.9	6.1	5.7	7.0	5.8
CV	14.5	12.9	32.6	90.1	21.2	27.3	38.1	23.3	17.5	15.8

<sup>1</sup>All units are % of DM; NDFD = NDF digestibility.

**Key Words:** corn silage, total-tract neutral detergent fiber digestibility, milk2006

**W185 Effect of light intensity and wavelength on concentration of antiherbivory compounds in *Flourensia cernua* leaves.**

Rick E. Estell<sup>\*1</sup>, Ed L. Fredrickson<sup>2</sup>, and Darren K. James<sup>1</sup>, <sup>1</sup>USDA-ARS Jornada Experimental Range, Las Cruces, NM, <sup>2</sup>Eastern Kentucky University, Richmond, KY.

Shrub encroachment negatively affects forage production and range-land health in many semi-arid regions. *Flourensia cernua* (tarbush) has increased in the northern Chihuahuan Desert at the expense of grasslands. Our previous studies have shown a negative relationship between tarbush terpene concentrations and livestock herbivory. Concentrations of secondary compounds are affected by many biotic and abiotic factors, including amount and wavelength of solar radiation. A 3-year study was conducted to examine the impact of shade and UV light restriction on phenolic and terpene concentrations in tarbush. Our hypothesis was that UV restriction and partial shade would reduce carbon based secondary compounds. Sixty plants were randomly selected and assigned to 3 treatments (control, UV light restriction, or 50% incident light restriction) in a randomized complete block. Frames covered with clear plastic film (UV blocking treatment), black shade cloth (50% shade treatment) or frame only were placed over 20 plants each. Leaves were collected from each plant in late September each year and freeze-dried for total phenolic analysis (Folin-Denis method) or frozen for leaf surface terpene analysis (extracted with ethanol and identified and quantified with GCMS). Data were analyzed using repeated measures linear mixed effects models with treatment as the fixed effect and year as a repeated effect. Means were separated by LSD. Mean concentrations of total phenolics and total volatiles in tarbush were 82.4 and 12.5 mg/g DM, respectively. Tarbush leaves contained 102 individual terpenes (including 19 unknowns). No year x treatment interactions were detected for any variable ( $P > 0.05$ ). Total phenolics did not differ between controls and UV restricted plants, but were lower for shaded plants than the other 2 treatments ( $P < 0.05$ ). Total volatiles were higher for the UV restriction treatment than controls ( $P < 0.05$ ), with shaded plants not differing from either treatment. Treatment effects were detected for 18 compounds ( $P < 0.05$ ). Shade reduced total phenolics as predicted, but did not affect total volatiles. In contrast to our expectations, UV restriction had no effect on total phenolics, and increased total volatile concentrations.

**Key Words:** light, phenolic, terpene

**W186 In situ ruminal degradability of diets based on passion fruit (*Passiflora edulis*) flour substituting maize.** Adolfo Sánchez<sup>\*1</sup>, Emma Torres<sup>1</sup>, Leon Montenegro<sup>1</sup>, Italo Espinoza<sup>1</sup>, and Daniela

Sanchez<sup>2</sup>, <sup>1</sup>Universidad Técnica Estatal de Quevedo, Quevedo Los Rios, Ecuador, <sup>2</sup>Universidad Estatal Amazonica, Puyo Pastaza, Ecuador.

This investigation was carried out in the Nutritional Metabolism and Ruminant Laboratory, located at the Experimental Campus “La Maria,” of Quevedo State Technical University (UTEQ – Ecuador) with the objectives of determining in situ ruminal degradability of dry matter (DM), organic matter (OM), neutral detergent fiber (NDF), and acid detergent fiber (ADF), as well as of identifying ruminal pH variation in cattle fed with diets based on passion fruit flour. A complete randomized block design (CRBD) was applied with 4 treatments (T1: 0%; T2: 10%; T3: 20% and T4: 30% inclusion of passion fruit flour substituting maize in diet), (diet components: corn, rice powder, bagasse soy, CaCO<sub>3</sub>, Ca (H<sub>2</sub>PO<sub>4</sub>)<sub>2</sub> x H<sub>2</sub>O and NaCl) and 3 blocks (cattle rumen fistulated) and 7 incubation times (0; 3; 6; 12; 24; 48 and 72 h). To establish the differences between treatment averages, Tukey ( $P \leq 0.05$ ) test was applied. The highest ( $P < 0.05$ ) degradability within 48 and 72 DM - OM and NDF - ADF incubation hours was registered by treatments T1 and T2 (67.68; 69.10% MS - 65.40; 66.71% DM); (71.79; 74.14% OM - 69.28; 70.07% OM); T4 and T3 (86.46; 88.33% NDF - 80.44; 81.02% NDF); (53.94; 56.21% ADF - 47.86; 49.36% ADF respectively). The levels of passion fruit flour inclusion made it possible to keep a (6.3 – 6.8) pH after 12 h of incubation, what is considered adequate for aminolytics and fibrolytics microorganism growing.

**Key Words:** digestibility, degradability, feeding

**W187 Dry matter production of four *Brachiaria* grasses.** Vir-

ginia L. N. Brandão, Gabriel C. B. Oliveira, Bruno P. Ignacchiti, Kaik Faria, Marcos Rosa, Guilherme D. Castro, Marcos I. Marcondes<sup>\*</sup>, and Fernanda H. M. Chizzotti, Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.

Studies to determine and compare the productive potential of forages from genus *Brachiaria* under same management conditions are scarce. Therefore, we believe that cultivars with similar morphological characteristics have similar nutritional value, and that management can affect this response. Thus, we aimed to determine and compare the productivity of forage produced during dry and rainy season of *Brachiaria* pastures when managed with 28 d or 95% light interception as criteria for interrupting regrowth. The treatments consisted of 4 cultivars of *Brachiaria*: *B. decumbens* ‘Basilisk’, *B. brizantha* ‘Marandu’, *B. brizantha* ‘Piatã’ and *B. brizantha* ‘Xaraés’, managed under 2 pasture strategies: every 28 d (28D), or when the canopy reaches 95% of light interception (IL95). The experiment was designed in completely randomized blocks, in a factorial arrangement of 4 x 2, with 3 replications (blocks). The evaluated variable was dry matter production (DM) in the pre- and post-grazing. The management did not affect DM production per year ( $P = 0.696$ ), but during the rainy season the IL95 had higher DM than 28D. During dry season the 28D management provided higher DM than IL95, because the dry season had 6 grazing cycles, while IL95 had only one cycle. Xaraés was the most productive forage, with 14,961 kg DM/ha/yr; Marandu and Piatã were not different (12,766 and 13,076 kg DM/ha/yr respectively), and *B. decumbens* had the lowest productivity (10,986 kg DM/ha/yr). We conclude that management of IL95 provided higher DM during rainy season; however, they had the same productivity per year. Xaraés had the greatest total dry matter production, and *B. decumbens* had the lowest productivity.

**Key Words:** season, light interception, fixed-days management



**W188 Forage yield of four maize cultivars sown in single and double rows.** Marco A. Ramírez<sup>\*1</sup>, Pedro A. Martínez<sup>2</sup>, Jesús Jarillo<sup>3</sup>, Francisco A. Castrejón<sup>1</sup>, and Luis Corona<sup>1</sup>, <sup>1</sup>*Departamento de Nutrición Animal, Universidad Nacional Autónoma de México, FMVZ, México City, México*, <sup>2</sup>*Departamento de Zootecnia, Universidad Autónoma Chapingo, Texcoco, México*, <sup>3</sup>*CEIEGT, Universidad Nacional Autónoma de México, Veracruz, México*.

In Mexico corn silage is widely fed to dairy and beef cattle, and in the market, farmers have many maize hybrids and native cultivars with some sowing strategies to choose from. The objective of the study was to determine total forage yield and by component (leaf, stem, ear and husk, kg DM ha<sup>-1</sup>) in 4 maize cultivars under 2 sowing strategies. Maize cultivars were 2 hybrids: Gladiator and Fog, and 2 native: Red and White; sowing strategies were single and double row, the former was seeding in rows every 80 cm, the latter in pairs of rows separated 40 cm with 80 cm between consecutive pairs of rows. Seeding rate was 80 000 plants ha<sup>-1</sup>. Experimental design was a completely randomized design in a factorial arrangement 4 (maize cultivars) × 2 (sowing strategies) with 4 replicates and the experimental unit was a 4.8 × 3 m plot. Harvest was 135 d after sowing. The cultivar × sowing strategy interaction did not influence ( $P > 0.05$ ) any of the variables measured. There was a trend ( $P = 0.10$ ) for native cultivars to show higher total forage yield than the hybrids (25408 vs. 21481.5 respectively); while double-row sowing tended ( $P = 0.19$ ) to promote on the average 12% higher total forage yield than single-row (24,763 vs 22,203). Native cultivars gave 63% higher ( $P < 0.05$ ) stem yield (14635.5) than the average of the 2 hybrids (8958) which showed similar ( $P > 0.05$ ) stem yield. Gladiator showed the lowest ( $P < 0.05$ ) leaf yield (2,677), 34% lower than the average yield observed in the other 3 cultivars (3589.7) which showed no difference ( $P > 0.05$ ) among them. Both hybrids gave 48% higher ( $P < 0.05$ ) ear yield than the average of the 2 native cultivars (6939.5 vs. 4672) which gave similar ( $P > 0.05$ ) ear yield. Only in leaf yield did sowing strategy show an influence, double-row gave 18% more ( $P < 0.05$ ) leaf than single-row (3646 vs. 3078 respectively). Native cultivars might show higher total forage yield than hybrids but of lower quality as the formers have higher amount of stems and lower of ears. It was concluded that total forage yield and by component are influenced by maize cultivar and sowing strategy and that total forage yield should not be the only one attribute to decide on which maize cultivar and sowing strategy to use on the farm.

**Key Words:** maize, cultivar, sown

**W190 Effects of maturity at harvest on the nutritional value, yield and milk production potential of corn hybrids planted under tropical/subtropical conditions.** Kathy G. Arriola<sup>\*1</sup>, Rafael M. Martins<sup>1</sup>, Thiago F. Bernardes<sup>1</sup>, Fabiola M. da Silva<sup>1</sup>, Felipe X. Amaro<sup>1</sup>, Bibiana Coy<sup>1</sup>, Enrique Alias<sup>2</sup>, Ernesto Marin<sup>2</sup>, Ludwig Leyton<sup>2</sup>, Zhengxin X. Ma<sup>1</sup>, Ibukun M. Ogunade<sup>1</sup>, Yun Jiang<sup>1</sup>, Musibau A. Bamikole<sup>1</sup>, and Adegbola T. Adesogan<sup>1</sup>, <sup>1</sup>*University of Florida, Gainesville, FL*, <sup>2</sup>*University of El Salvador, San Salvador, El Salvador*.

This study examined the effect of maturity at harvest on key agronomic and nutritional traits of corn hybrids planted in the Florida Summer. Eighteen corn hybrids from 5 seed companies (Agra Tech, Croplan Genetics, Mycogen, Pioneer, and Syngenta) were grown on 4 replicate plots (1.8 × 6.1 m) in each of 4 blocks in August 2013. Hybrids were harvested at target maturity stages of 30, 35, 40, and 45% DM by removing 1.8-m sections from 2 rows in each plot. Temperatures ranged from 14.1 to 29.3°C and rainfall was 432 mm during the growing season. Disease ranking (1, low to 5, high scale), DM yield, kernel milk line

position, and chemical composition at harvest were measured. Milk-per-ton (MPT) and milk-per-hectare (MPH) values were calculated with the Milk 2006 model. Data were analyzed with a model including hybrid, maturity and the interaction using the Glimmix procedure of SAS. Mean DM concentrations at harvest were 32.3, 34.6, 40.7, and 42.5% at the respective target maturity stages above. Milk-per-hectare was unaffected by maturity (1,529–1,679 kg/t) but MPT was greater when hybrids were harvested at 30 or 35% DM than at 40 or 45% DM (1668 vs. 1531 kg/t). The NDF digestibility decreased at each maturity stage ( $P < 0.05$ ; 59.2, 58.0 and 57.2 and 55.7%, respectively), whereas DM yield did not change (22.2 t/ha) until the 45% DM stage when it increased (25.5 t/ha). For most hybrids, disease ranking and NDF concentration decreased to a nadir and starch concentration peaked when they were harvested at 40% DM (hybrid × maturity,  $P < 0.05$ ). Kernel milk line position increased quadratically with maturity for most hybrids (hybrid × maturity,  $P < 0.05$ ). Six hybrids had among the highest MPT (>1641 kg/t) and MPH (>39164 kg/ha) values and their NDF digestibility (57.3 to 60.3%) and starch concentration (27.7 to 32.0%) ranges were moderately high. Differences in MPT and MPH among hybrids of 150 kg/t and 14,132 kg/ha, respectively were evident. Yield and quality measures were simultaneously optimized when hybrids were harvested at 35 to 40% DM.

**Key Words:** corn hybrid, maturity, dry matter yield

**W191 Application of *Pedococcus pentosaceus*, *Pichia anomala*, and chitinase to high moisture alfalfa hay at baling: effects on ruminal digestibility.** Long Jin<sup>1</sup>, Lysiane Dunière<sup>1</sup>, Joseph.P. Lynch<sup>1</sup>, Eric Chevaux<sup>2</sup>, Tim.A. McAllister<sup>1</sup>, John Baah<sup>3</sup>, and Yuxi Wang<sup>\*1</sup>, <sup>1</sup>*Agriculture and Agri-Food Canada, Lethbridge, AB, Canada*, <sup>2</sup>*Lallemand animal nutrition, Blagnac, France*, <sup>3</sup>*Best Environmental Technologies Inc., Edmonton, AB, Canada*.

Baling of alfalfa hay at higher than optimum moisture level can lead to spoilage and a reduction in the nutritive quality of the forage. Organic acids such as propionic acid (PA) have been added to high moisture hay (HMH) to prevent the growth of spoilage microorganisms, but the high rates of application, costs and environmental concerns associated with these preservatives has discouraged hay producers from using them. The objective of this study was to assess the effects of applying *Pedococcus pentosaceus* (PED) and *Pichia anomala* (PIC) alone or in combinations with chitinase (CH) at baling on the nutritional quality of high-moisture alfalfa hay using in vitro and in situ procedures. Alfalfa hay was harvested at the mid-bloom stage, wilted to 73–77% DM and baled without (HMH) or with PED, PIC, PED+PIC, PED+CH, PIC+CH, and PA. The control (HMH), PED+CH, PIC+CH and PA and alfalfa hay baled at >85% DM (NMH) were repeated in 3 consecutive yrs. Other treatments were assessed only in single yr. Samples collected 90 d after baling over the 3 yr were assessed in vitro and only 1 yr samples were assessed for rumen degradability. For all 3 yr, in vitro DM disappearance (IVDMD), asymptotic gas production and rate of gas production of HMH, irrespective of treatment, was lower ( $P < 0.05$ ) than NMH. Among treatments, PED+CH and PIC+CH had higher ( $P = 0.01$ ) IVDMD than HMH and PA, but only PED+CH had higher ( $P < 0.01$ ) asymptotic and rate of gas production as compared with HMH and PA in yr 1. The potential degradable fraction of DM did not differ among treatments. However, PED+CH and PIC+CH had a higher ( $P < 0.01$ ) rapidly degradable fraction than PA. The 24-h in situ NDF digestibility of PED+CH and PIC+CH was higher ( $P < 0.001$ ) than that of all other treatments including NHM. These results suggest that application of *P. pentosaceus* or *P. anomala* combined with chitinase at baling of

high moisture alfalfa hay has potential to reduce nutrient losses and to increase fiber digestibility.

**Key Words:** high moisture alfalfa hay, additives, in situ digestibility.

**W192** *Malva sylvestris* extract affected kinetics of fermentation, methane production, and protozoa population of barley grain incubated with rumen fluid. Saba Khamooshi<sup>1</sup>, Farokh Kafilzadeh<sup>1</sup>, Hossein Jahani Aziz Abadi<sup>2</sup>, and Golnaz Taasoli\*<sup>1</sup>, <sup>1</sup>Razi University, Kermanshah, Iran, <sup>2</sup>Kurdistan University, Kurdistan, Iran.

This study was conducted to evaluate the effect of *Malva sylvestris* leaf or stem extracts on in vitro fermentation kinetic, methane production and protozoa population of barley grain. Juice from leaves and stems were prepared by extraction from crushed materials. The extracts were then centrifuged at  $454 \times g$  for 15 min before use. The extracts were added at levels of 0, 25, 50 and 100  $\mu\text{L}/250$  mg barley incubated in 30 mL mixture of buffer and rumen fluid. Addition of both extracts increased the potential gas production (mL/200 mg DM), and constant rate (mL/h) but decreased the lag time (h) of barley grain ( $P < 0.01$ ). Methane production from fermentation of barley grain decreased due to the addition of the extracts ( $P < 0.01$ ). Methane reduction potential from fermentation of barley grain were 13.15, 7.30 and 13.28% when leaf extract was added at 25, 50 and 100  $\mu\text{L}/250$  mg barley, respectively. Corresponding values for similar levels of the stem extract were -1.94, 1.80 and 9.12%, respectively. Protozoa population in incubated barley grain medium with no extract was  $9.94 \times 10^4/\text{mL}$ . The extracts particularly the extract from leaf resulted in much more protozoa reduction from barley grain fermentation (average values of protozoa population when leaf and stem extracts were added to barley grain incubation medium were  $7.67 \times 10^4$  vs.  $7.85 \times 10^4/\text{mL}$ ). The result of this study show *M. sylvestris* extract has some potential in improving the fermentation of barley grain

**Key Words:** *Malva sylvestris*, methane production, protozoa population.

**W193** Effect of a protease enzyme (papain) on in vitro NDF digestibility of alfalfa hay stem. Shahab Payandeh, Farokh Kafilzadeh, and Golnaz Taasoli\*, Razi University, Kermanshah, Iran.

An in vitro experiment was conducted to examine the effect of a protease (papain, EC 3.4.22.2) on ruminal fiber digestibility of alfalfa hay stem. Exactly 100 mg of enzyme powder was solubilized using 5 ml of water. Serum bottles containing 500 mg of alfalfa hay stem, modified McDougall's buffer and rumen fluid with addition of 0, 50, 100 or 200  $\mu\text{L}$  of the papain solution/g alfalfa hay stem DM were incubated under anaerobic conditions at 39°C for 24 and 48 h. Total gas production and NDF disappearance were determined. Partitioning factor (PF) was calculated as the ratio of NDF degraded (mg) to the volume of gas (mL) produced. Data were analyzed as a completely randomized design by ANOVA using SAS 9.2. The enzyme with a level of 200  $\mu\text{L}/\text{g}$  alfalfa hay stem DM significantly ( $P < 0.05$ ) increased in vitro NDF degradability after 24h incubation (40.2, 41.6, 40.6 and 45.9% in alfalfa hay stem, alfalfa hay stem+50, alfalfa hay stem+100 and alfalfa hay stem+200  $\mu\text{L}$  of enzyme/g alfalfa hay stem DM, respectively). No significant difference was observed after 48h incubation. The PF was also increased significantly ( $P < 0.01$ ) with addition of all levels of papain (1.57, 1.76, 1.70 and 1.77 in alfalfa hay stem, alfalfa hay stem+50 alfalfa hay stem+100 and alfalfa hay stem+200  $\mu\text{L}$  of enzyme/g alfalfa hay stem DM, respectively). From the result of this experiment, it appears that addition of papain at the highest level has a pronounced effect on the

digestion of NDF after 24h incubation. It is concluded that this enzyme has some potential on removing structural proteins in the cell wall, allowing ruminal microbes to gain faster access to digestible substrates.

**Key Words:** papain, NDF digestibility, partitioning factor

**W194** Effect of bacterial additives on fermentation quality and aerobic stability of rye silage harvested at dough stage. Young Ho Joo\*<sup>1</sup>, Seong Shin Lee<sup>1</sup>, Dong Hyeon Kim<sup>1</sup>, Hyuk Jun Lee<sup>1</sup>, Sardar M. Amanullah<sup>1,3</sup>, Ouk Kyu Han<sup>2</sup>, and Sam Churl Kim<sup>1</sup>, <sup>1</sup>Division of Applied Life Science (BK21Plus, Inst. Agric. & Life Sci.), Gyeongsang National University, Jinju, Gyeongsangnam-do, South Korea, <sup>2</sup>National Institute of Crop Science, RDA, Suwon, South Korea, <sup>3</sup>Bangladesh Livestock Research Institute, Dhaka, Bangladesh.

This study was carried out to determine the effect of different additives on chemical composition, fermentation characteristics and aerobic stability of rye silage harvested at dough stage. Rye forage was grown at National Institute of Crop Science, Suwon, South Korea, and harvested at 37% of dry matter. The harvested forages were divided into the following 4 treatments: CON (distilled water at 2 mL/kg of fresh forage); LP (*L. plantarum*  $1.5 \times 10^4$  cfu/g of fresh forage); LB (*L. buchneri* at  $1.2 \times 10^4$  cfu/g of fresh forage) and MIX (mixture of LP and LB at 1:1 ratio). The forage was chopped to 3–6 cm length and ensiled into 10 L bucket silo with 4 replications for 0, 1, 4, 7, 48 and 100 d. On 100 d of ensiling, crude protein concentration (7.61 vs. 6.63%) in LP was higher ( $P < 0.05$ ) than that in CON. The concentrations of neutral detergent fiber and hemicellulose were higher ( $P < 0.05$ ) in LB (73.7 and 24.6%) and MIX (72.9 and 24.0%) than those in CON (70.4 and 21.7%) and LP (69.5 and 22.6%), while acid detergent fiber concentration was lowest ( $P < 0.05$ ) in LP (46.9 vs. 48.7, 49.1 and 48.9%). The CON had higher ( $P < 0.05$ ) pH (4.56 vs. 4.15, 4.11 and 4.12) than all treatments applied bacterial additives. The pH in LB and MIX silage decreased more drastically than those in CON and LP on early ensiling periods (d 0–7). The concentrations of lactate and propionate were highest ( $P < 0.05$ ) in LP (13.4 and 5.72%), but acetate concentration was highest ( $P < 0.05$ ) in LB (27.5%) and MIX (22.1%). Lactate to acetate ratio (1.23 and 1.43 vs. 0.29 and 0.35) was higher ( $P < 0.05$ ) in CON and LP than that in LB and MIX. Lactic acid bacteria (6.71, 6.59 and 6.48 vs. 5.25 log<sub>10</sub> cfu/g) in CON, LP and LB were higher ( $P < 0.05$ ) than that in MIX, whereas aerobic stability (760.5 vs. 616.3, 606.5 and 634.3 h) was higher ( $P < 0.05$ ) in MIX than all other treatments. Yeast was highest ( $P < 0.05$ ) in LB (7.64 log<sub>10</sub> cfu/g), while mold was not detected in all treatments. In conclusion, LP or MIX supplementation can improve the aerobic stability by increase of acetate concentration which has strong anti-fungal effect.

**Key Words:** aerobic stability, barley silage, inoculant

**W195** Fermentation quality and chemical composition of mixed silage with shrub and corn. Bing-wen Si\*<sup>1</sup>, Zong-li Wang<sup>2</sup>, Qi-zhong Sun<sup>2</sup>, Hong-mei Wang<sup>1</sup>, and Qi-yu Diao<sup>1</sup>, <sup>1</sup>Feed Research Institute, Chinese Academy of Agricultural Sciences, Key Laboratory of Feed Biotechnology of the Ministry of Agriculture, Beijing, China, <sup>2</sup>Grassland Research Institute, Chinese Academy of Agricultural Sciences, Hohhot, Inner Mongolia, China.

With development of Chinese animal husbandry, shortage of feedstuffs is a main limit factor for livestock development. Rush bushclover (RB) and shrubby sweetvetch (SS) have the characteristics of high biomass, drought resistance, leanness tolerance. Moreover, they are of high nutritive value, rich in crude protein (CP), and potential new

resources for livestock feed. Shrub is seldom used as material to make silage as legumes are often considered hard to ensile well due to low fermentable carbohydrate content in combination with a high buffering capacity (BC). Whole-crop corn is ascribed to easy ensiling procedure and has high fermentative quality and palatability for cows, because of the chemical composition of corn, mainly in high fermentable carbohydrates. Silage produced from a mixture of whole-crop corn and 2 kinds of shrub is likely to have a better fermentation quality and nutritive value compared with silage consisting solely of herbage from RB and SS. RB and SS (315 and 274 g dry matter per kg) were cut into 2 to 5 cm by a rubbing filament machine and ensiled in 1.5 L laboratory silos with whole-crop corn as mixtures of 1, 0.7, 0.5, and 0.3 of fresh weight (FW). After 60 d, the nutrition and fermentation quality of all treatments were analyzed. Results showed that the pH of silage consisting solely of herbage from RB and SS were significantly ( $P < 0.05$ ) higher and the lactic acid content were significantly ( $P < 0.05$ ) lower than silages mixed with whole-crop corn. With the increase of the proportion of corn, lactic acid and acetic acid content also increased ( $P < 0.05$ ). Dry matter and WSC content decreased with the increase of the proportion of corn ( $P < 0.05$ ). In conclusion, RB and SS with a high BC and a low WSC concentration is best ensiled using whole-crop corn. Inclusion of 0.5 to 0.7 of whole-crop corn is recommended to ensure a good fermentation.

**Key Words:** mixed silage, fermentation quality, shrub

**W196 Late season forage yield, quality, and digestibility from mixed cropping of organic certified corn and soybean hybrids at different seeding rates.** Ishwary Acharya\* and David Casper, *Dairy Science Department, South Dakota State University, Brookings, SD.*

The production of forage resulting from the mixed cropping of corn and soybeans at planting has the potential to yield greater quantities of digestible nutrients to meet the nutrient requirements of lactating dairy cows. A field plot study was laid out using a completely randomized design to evaluate 2 organic corn hybrids [Normal (N) and MasterGraze (MG)] with 2 soybean hybrids [Regular (R) and Vining (V)] at 4 seeding rates (R1 = 65:35; R2 = 55:45; R3 = 45:55, and R4 = 35:65 of corn and soybean) having a 2 × 2 × 4 factorial treatment design replicated 3 times. Forage was hand harvested 93 d after planting during the 2014 growing season, inoculated, packed into buckets, weighed, and ensiled for 60 or 90 d. Buckets were then re-weighed, opened, and forage samples collected and submitted for nutrient analysis (Analab, Inc., Fulton, IL). Yield of DM (6.74 and 7.65 T/ha for N and MG, respectively) for main effect of corn was similar ( $P > 0.05$ ), while V yielded greater ( $P < 0.05$ ) than R (6.13 and 8.27 T/ha for R and V, respectively) for main effect of soybean, while seeding ratio main effect was similar [ $(P > 0.05)$ ; 7.91, 6.29, 7.81, and 6.77 T/ha for R1, R2, R3, and R4, respectively]. Yield of fresh digestible DM (DDM; 4.40 and 5.06 T/ha) and CP (1.04 and 1.22 T/ha) were similar ( $P > 0.05$ ) for corn, while V yielded greater ( $P < 0.05$ ) DDM (4.03 and 5.43 T/ha) and CP (0.97 and 1.29 T/ha) than R and seeding ratios were similar ( $P > 0.05$ ) in yield of DDM (5.20, 4.15, 5.08, and 4.50 T/ha) and CP (1.14, 1.05, 1.25, and 1.07 T/ha). A significant interaction ( $P < 0.05$ ) was detected for corn × soybean × seeding ratio for ensiled DDM yield at 60 and 90 d, while no other significant ( $P > 0.05$ ) interactions of main effects were detected. The combination of MG seed corn with V soybean hybrids at the ratio of 67:33 resulted in the greatest yield of DDM after 60 and 90 d of ensiling the forage. The mixed cropping of corn and soybeans holds great potential for increasing the production of forages to meet the nutrient requirements of lactating dairy cows.

**Key Words:** corn, soybean, yield

**W197 Climatic and management factors affecting the forage yield and quality of a high density mulberry forage bank established under low input tropical farming conditions.** Eliel González-García\*<sup>2</sup> and Giraldo Martín-Martín<sup>1</sup>, <sup>1</sup>*Estación Experimental de Pastos y Forrajes "Indio Hatuey," Matanzas, Cuba,* <sup>2</sup>*INRA UMR868, Systèmes d'Élevage Méditerranéens et Tropicaux (SELMET), Montpellier Cedex 2, France.*

Effects of year season (SEAS), organic fertilization (N) and harvest frequency (FREQ) on forage yield and composition were evaluated in a high density mulberry forage bank, established under low input tropical farming conditions. The experiment, replicated 2 consecutive years, was arranged in 18 treatments resulting from a 3 × 3 × 2 factorial design combining 3 FREQ (60, 90 and 120 d), 3 N rates (100, 300 and 500 kg N/ha/year) and the 2 seasons (RAIN and DRY). Forage yield, leaf to stem ratio, the edible fraction (EDIB, %), nutrient content by plant fraction (PFR; i.e., leaves, tender and ligneous stems) and the maturity indexes were monitored. Either forage yield or nutrient content was strongly affected by SEAS, FREQ, PFR, N rate and, in some cases, by their interactions. Total forage yield increased ( $P < 0.0001$ ) with FREQ (26.5, 31.2 and 40.6 t for 60, 90 and 120 d, respectively), irrespective of the SEAS (44.7 and 21.1 t for RAIN and DRY, respectively), PFR (19.2, 3.0 and 10.7 t for LEAF, TST and LST, respectively) or N (28.1, 32.0 and 38.5 t for 100, 300 and 500 kg N/ha/year, respectively). The EDIB yield was higher in RAIN (25.3 vs. 13.2 t; RAIN vs. DRY) and diminished while increasing FREQ, irrespective of SEAS or N. This decrease in EDIB yield was directly related to the decrease in the LEAF proportion and the concomitant increase of the LST fraction with FREQ. Harvesting at 60 and 90 d in RAIN and DRY, respectively, with a N rate of 300 kg N/ha/year seems the best agronomic choice as a most optimal condition between forage yield and nutritive value.

**Key Words:** forage yield and composition, harvest frequency, organic fertilization

**W198 Response to using rumen inoculum from high and low feed efficient cows on in vitro fermentation of alfalfa ensiled with different additives.** Francisco E. Contreras-Govea\*<sup>1</sup>, Richard E. Muck<sup>2</sup>, Paul J. Weimer<sup>2</sup>, and Ursula C. Hymes-Fecht<sup>2</sup>, <sup>1</sup>*Department of Dairy Science, University of Wisconsin-Madison, Madison, WI,* <sup>2</sup>*USDA-Dairy Forage Research Center, Madison, WI.*

Alfalfa from 2 consecutive harvests (H1, H2) was treated with *Lactobacillus plantarum* (LP, 10<sup>6</sup> cfu/g alfalfa), formic acid (FA, 5.4 kg/Mg), and no additive (control), ensiled in mini-silos, fermented for 60-d at room temperature (22°C), and analyzed for nutritive value and fermentation profile. Silages were wet ground to a particle size of 2–3 mm using a food processor for in vitro true DM digestibility (IVTDMD). High (HE) and low efficient (LE) groups of 3 cannulated cows each were identified based on 2 lactations and had an average energy corrected milk/dry matter intake (ECM/DMI) ratio of 1.88 and 1.61, respectively. In each in vitro run, 12 silos from a harvest were analyzed simultaneously with 6 rumen inocula, one from each cow, for 24 h. Data analysis was conducted using the MIXED procedure of SAS, as a split plot design. For silage fermentation, harvest was the whole plot and alfalfa treatment as subplot. For the in vitro, HE and LE groups were main plot and treated alfalfa as sub-plot. In both cases mean differences were declared significant at  $P < 0.05$ . Across harvests, LP had lower pH (4.47) and greater lactic acid concentration (64.3 g/kg DM) than control (4.77, 52.1) and FA (4.70, 33.9). Moreover, ammonia-N was lower in LP (3.8% total N) and FA (4.2) than control (5.4) in both harvests. IVTDMD of the H1 silages was greater with the HE cows (79.6%) than LE cows (75.13%), but there was no difference at H2 ( $P = 0.310$ ). The IVTDMD



was not different among silage treatments at H1 ( $P = 0.078$ ), but at H2 control had greater IVTDMD (79.8%) than did LP (78.1) and FA (77.6). Rumen fluid from HE cows produced greater acetate and butyrate than did LE on H1 silages, with no differences on H2. In vitro acetate and propionate were contrasting between H1 and H2 by silage treatment. At H1, LP and FA had greater acetate and propionate than control, but it was opposite at H2. It is concluded that LP and FA decrease ammonia-N during fermentation of alfalfa silage, and rumen inocula from HE cows displayed numerically higher IVTDMD, but the effect was only significant with the earlier harvested silage.

**Key Words:** alfalfa silage, additive, feed efficiency

**W199 Ruminal degradability of a *Musa* sp. fodder bank located in the central part of Costa Rica.** Pablo Chacón Hernández\*, Carlos Boschini Figueroa, and Ricardo Russo Andrade, *Universidad de Costa Rica, San Pedro, San José, Costa Rica.*

When bananas are grown for fruit, the bigger part of the plant gets wasted. In tropical areas, producers use the pseudo-stems and leaves as feed for cattle. An experiment was performed to determine the potential value of the banana plant nutrient availability for ruminants. Fifteen adult plants were randomly harvested from a fodder bank located at the University of Costa Rica's Alfredo Volio Mata Experimental Station during the rainy season of 2012. Each plant was harvested at 20–25 cm above ground and divided into 5 sections for sampling (base, center and tip of the pseudo-stem and lamina and petioles of the leaves), all 15 samples from each portion were dried, ground and incubated by duplicate within 2 Jersey-Reyna ruminally fistulated cows with results analyzed using the Marquadt algorithm. Values ranging from 18.38% to 47.43% were found for the percentage of soluble fraction, from 33.45% to 45.76% for the degradable fraction, from 1.65%/h to 7.51%/h for the degradation speed and from 64.14% to 82.86% for the potentially degradable percentage, depending on the plant part. According to the obtained results, pseudo-stems have better degradability values than leaves and may be used as a feed source on cattle diets where the material is available.

**Table 1 (Abstr. W199).** Ruminal degradability constants on a *Musa* sp. fodder bank

Plant section	Soluble fraction, %	Degradable fraction, %	Degradation speed, %/h	Potentially degradable fraction, %
Pseudo-stem base	47.43 <sup>a</sup>	35.43 <sup>b</sup>	0.07518 <sup>a</sup>	82.86 <sup>a</sup>
Pseudo-stem center	42.51 <sup>a</sup>	34.13 <sup>b</sup>	0.06512 <sup>ab</sup>	76.64 <sup>a</sup>
Pseudo-stem tip	44.77 <sup>a</sup>	33.45 <sup>b</sup>	0.05824 <sup>abc</sup>	78.22 <sup>a</sup>
Petiole	33.34 <sup>b</sup>	38.85 <sup>ab</sup>	0.06689 <sup>ab</sup>	72.19 <sup>b</sup>
Lamina	18.38 <sup>c</sup>	45.76 <sup>ab</sup>	0.01653 <sup>d</sup>	64.14 <sup>c</sup>

<sup>a-c</sup>Indicate statistical differences ( $P < 0.05$ ) within a column.

**Key Words:** degradability, *Musa*, fodder bank

**W200 Purple prairie clover (*Dalea purpurea* Vent) reduces fecal shedding of *Escherichia coli* in pastured cattle.** Yuxi Wang<sup>\*1</sup>, Long Jin<sup>1,2</sup>, Alan Iwaasa<sup>3</sup>, Yuanheng Li<sup>3,5</sup>, Zhong Xu<sup>1</sup>, Mike Schellenberg<sup>3</sup>, Xiuli Liu<sup>1,6</sup>, Tim McAllister<sup>1</sup>, and Kim Stanford<sup>4</sup>, <sup>1</sup>Lethbridge Research Centre, Agriculture and Agri-Food Canada, Lethbridge, AB, Canada, <sup>2</sup>Department of Animal Science, Northeast Agricultural University, Harbin, China, <sup>3</sup>Semiarid Prairie Agricultural Research Centre, Agriculture & Agri-Food Canada, Swift Current, SK, Canada, <sup>4</sup>Alberta Agriculture and Rural Development, Lethbridge Agriculture Centre, Lethbridge, AB, Canada, <sup>5</sup>Institute

of Grassland Research, Chinese Academy of Agricultural Sciences, Hohhot, China, <sup>6</sup>Inner Mongolia Academy of Agricultural and Animal Husbandry Sciences, Hohhot, China.

A 3-year (2009–2011) grazing study was conducted to assess the effects of purple prairie clover (PPC, *Dalea purpurea* Vent) on fecal shedding of total *Escherichia coli* in cattle. Three pasture types were used in the experiment; bromegrass (Check), mixed cool season grasses with PPC (Simple) and mixed cool and warm grasses with PPC (Complex). Pastures were rotationally grazed (5, 10 and 10 cattle for Check, Simple and Complex) during a summer and fall grazing period. Purple prairie clover was grazed in summer at the vegetative/early flower stage and at the flower/early seed stage during the fall. Weekly and biweekly fecal samples were collected from each animal for enumeration of *E. coli* and chemical analyses. Forage samples were collected throughout the season for analysis. Condensed tannins (CT) were only detected in Simple and Complex pastures that contained PPC, with concentrations being higher in the fall than in the summer. Fecal counts of *E. coli* in cattle grazing Simple and Complex pastures linearly decreased ( $P < 0.05$ ) over summer to fall in all 3 years, an outcome not observed in cattle grazing the Check pasture. Across the 3 grazing seasons, fecal *E. coli* was lower ( $P < 0.05$ ) in cattle grazing Simple and Complex pastures than Check during the fall. During the fall, feces collected from cattle grazing the Check pasture had higher ( $P < 0.05$ ) pH, N, NH<sub>3</sub>-N, total volatile fatty acids (VFA) and branched-chain VFA, but a lower ( $P < 0.05$ ) acetate: propionate ratio than that collected from cattle grazing Simple or Complex pastures. In a second experiment, 2 strains of *E. coli* were cultured in M9 medium containing 25 to 200 µg/mL of PPC CT. Growth of *E. coli* was linearly ( $P < 0.01$ ) reduced by increasing levels of PPC CT. The scanning electron micrographs showed electron dense filamentous material associated with the outer membrane of *E. coli* cells exposed to CT. Incorporation of PPC into forage reduced the fecal shedding of *E. coli* from grazing cattle, likely due to the anti-*E. coli* properties of PPC CT.

**Key Words:** grazing pasture, purple prairie clover, *Escherichia coli*

**W201 Yield, chemical composition, and efficiency of use of nitrogen by Marandu grass.** Tadeu Silva de Oliveira<sup>\*1</sup>, Ricardo Augusto Mendonça Vieira<sup>1</sup>, Alberto Magno Fernandes<sup>1</sup>, Danielle Ferreira Baffa<sup>2</sup>, and José Carlos Pereira<sup>2</sup>, <sup>1</sup>Universidade Estadual do Norte Fluminense-Darcy Ribeiro, Campos dos Goytacazes, Rio de Janeiro, Brazil, <sup>2</sup>Universidade Federal de Viçosa, Campus Universitário, Viçosa, Minas Gerais, Brazil.

The objective of this study was to evaluate the effect of 5 nitrogen levels on the yield and quality characteristics and the use efficiency of *Brachiaria brizantha* 'Marandu' grass. The soil, classified as a Red-Yellow Acrisol, was sampled at a depth of 0 to 0.20 m, for analyses. Soil analyses results were follows: pH in water = 5.6; CEC (T) = 8.17; H + Al = 4.21; sum of bases = 3.96; Ca = 2.8; Mg = 1.1 cmolc dm<sup>-3</sup>; P = 66.6 mg dm<sup>-3</sup>; K = 123 mg dm<sup>-3</sup>; and 1.49 dag kg<sup>-1</sup> organic matter. The area was prepared by plowing followed by disking two times and then fertilized with 300 kg/ha of N-P-K formulation (8-28-16). The *Brachiaria brizantha* 'Marandu' was sown at a rate of 3 kg/ha of pure viable seeds in a row spacing of 0.45m. Because of the uniformity of the area, we adopted a completely randomized design with four replicates. The treatments consisted of four levels of nitrogen (0, 60, 120, 180 and 240 kg/ha). Samples of the material were collected to analyze productivity variables, concentrations of neutral detergent fiber, crude protein and neutral detergent insoluble nitrogen, and in vitro dry matter digestibility. The nitrogen use efficiency, recovery of the applied nitrogen (RAN) and agronomic efficiency of the applied nitrogen (AE) were calculated. The

statistical analyses involved the application of procedure PROC MIXED. To select the regression models the criteria utilized was Akaike's information criterion. Dry mass production increased ( $P < 0.001$ ) by 1,624.67 (kg/ha) as the nitrogen doses were increased. The doses of nitrogen affected ( $P < 0.05$ ) the concentrations of neutral detergent fiber (from 294.6 to 381.4 g/kg, in the leaf), crude protein (from 86.1 to 99.6 g/kg, in the leaf) and neutral detergent insoluble protein (from 402.9 to 396.2 g/kg CP, in the leaf). Nitrogen use efficiency increased, whereas RAN and AE were not affected ( $P > 0.05$ ) by the nitrogen doses. In conclusion, nitrogen fertilization promotes improvement in productivity and chemical composition of Marandu grass, also improving the efficiency with which the grass utilizes the nitrogen.

**Key Words:** nitrogen use efficiency, nutritional characteristics, urea

#### **W202 Effect of ensiling on kernel processing score in whole-plant corn silage harvested with varied processors and settings.**

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Two experiments were conducted to evaluate the impact of (1) ensiling on kernel processing score (KPS; % of starch passing through 4.75 mm sieve) of whole-plant corn silage (WPCS) of varied processors and settings; and (2) extended ensiling times on KPS of corn shreddage. In Experiment 1, samples from 2 self-propelled forage harvesters (SPFH) were collected at harvest. One SPFH was fitted with a conventional-type processor with greater (32% vs. 21%) roll-speed differential than normal. Samples collected were from 3 theoretical lengths of cut (TLOC; 19, 22 and 26 mm; 3-mm roll-gap setting) in 2 hybrid types (leafy and dual-purpose). The other SPFH was fitted with an intermeshing-disc processor and samples were from 3 TLOC (17, 19 and 26 mm) at 2 roll-gap settings (1.5 and 2.5 mm) in one hybrid. Samples for each processor and settings collected at harvest (12 samples total) were divided into 2 piles. The first pile was unfermented and analyzed for KPS in duplicate. The second pile was vacuum-sealed in 3 plastic bags and ensiled for 30 d. After fermentation, the 3 bags were composited and analyzed for KPS in duplicate. Data were analyzed using Proc Mixed of SAS with the fixed effect of fermentation and the random effect of sample. In Experiment 2, WPCS was harvested using a SPFH fitted with a shreddage processor with cross-grooved rolls set at 2.5 mm roll-gap and 32% roll speed differential and the SPFH set at 26 mm of TLOC. Samples were collected at harvest and vacuum-sealed in plastic bags and ensiled for 0, 30, and 120 d in triplicate. Samples from each bag were analyzed for KPS in duplicate. Data were analyzed using Proc Mixed of SAS with the fixed effect of ensiling time and the random effect of bag. In Experiment 1, ensiled samples had 10%-units greater ( $P = 0.01$ ) KPS than the unfermented samples (60.1% vs. 50.2%, respectively). In experiment 2, WPCS ensiled for 120 d tended ( $P = 0.07$ ) to have 7%-units greater KPS than 0 d (67.2% vs. 60.3%, respectively), but not 30 d (63.6%). These findings suggest that fermentation increases KPS.

**Key Words:** corn silage, ensiling time, corn silage processing score

#### **W203 Morphological responses and chemical composition of Tanzania grass subjected to two pasture-management strategies.**

Alberto Magno Fernandes\*<sup>1</sup>, Ricardo Augusto Mendonça Vieira<sup>1</sup>, Tadeu Silva de Oliveira<sup>1</sup>, and Fermino Deresz<sup>2</sup>, <sup>1</sup>Universidade Estadual do Norte Fluminense, Campos dos Goytacazes, Rio de Janeiro, Brazil, <sup>2</sup>EMBRAPA-Gado de Leite, Juiz de Fora, Minas Gerais, Brazil.

The objective of this study was to evaluate the morphogenetic traits and chemical composition of Tanzania grass using a fixed resting period (FR) or according to 95% light interception (LI, by accuparLP-80) for managing Tanzania-grass pastures. As the LI of the herbage mass reached 95%, the animals were allowed to graze each paddock for 3 d; and (2) 30 d defoliation interval with a 3 d grazing period (FR). Experimental unit consisted of 11 paddocks grazed by 5 Holstein-Gir cows allowing for 2 replicates per defoliation treatment in the first year with the same areas assigned to the same treatment the following year grazed by 4 Holstein-Gir cows per experimental unit. Six samples per paddock were taken on 2 crossed transect lines. Sward height, forage mass, and morphological components and their chemical composition were evaluated. The fixed effects were treatments (LI or FR), year (one or 2), and the interaction, whereas the random effect was area within treatment and the model was fitted as a mixed model with yearly repeated measures (using SAS). The interaction was significant only for senescent material ( $P = 0.026$ ). The treatments did not affect pasture mass, height, and the proportion of stems in the pre- ( $P = 0.455, 0.715, 0.457$ , respectively) and post-grazing ( $P = 0.199, 0.160, 0.457$ , respectively) conditions. However, the proportion of leaves and crude protein content of the herbage mass were greater in LI than FR paddocks ( $P = 0.031$ , and 0.018, respectively). The concentration of other nutrients were not affected by treatments ( $P > 0.05$ ). Therefore, LI might affect at least some quality traits of the paddock.

**Key Words:** grass, grazing system, sward height

#### **W204 How does the chemical additive calcium oxide affect the in vitro growth of lactic acid bacteria and yeast? R. A. de Paula, O. G. Pereira\*, T. C. da Silva, K. G. Ribeiro, and H. C. Mantovani, Universidade Federal de Vicosa, Vicosa, Minas Gerais, Brazil.**

Sugarcane has a high concentration of sucrose, which is metabolized by yeasts during ensiling producing high concentrations of ethanol. Calcium oxide (CaO) has been used as a silage additive resulting in a reduction of yeasts, less ethanol and improved DM recovery. The objective of this study was to evaluate the effect of CaO on the in vitro growth of lactic acid bacteria (LAB) and yeast in an aqueous-extract of sugarcane. The aqueous-extract of sugarcane was made by blending fresh sugarcane and distilled water in the proportion 5:1 for 5 min. The extract was filtered through layers of cheesecloth and sterilized at 121°C for 15 min. The microorganisms evaluated were one yeast (*Saccharomyces cerevisiae*), and 3 LAB (*Lactobacillus brevis*, *L. plantarum*, and *Pediococcus pentosaceus*, isolated from alfalfa silage). The LAB and the yeast (initial population of 6 log cfu/mL) were grown in the sugarcane extract with levels of CaO: 0 (control), 0.5, 1, 1.5, and 2% (fresh basis) and incubated at 37 and 30°C for 48 h, respectively. Two tests were conducted, one with no pH adjustment and another adjusting the initial pH to 6 ± 0.13. The pH was evaluated before and after the incubation. In the first trial, the final pH of the control was 5.62 whereas the pH of the extracts containing CaO were higher than 11.6 for all microorganisms. In addition, there was inhibition of growth of all microorganisms at the concentration of 0.5% CaO. In the second test, the numbers of *L. brevis* and *P. pentosaceus* decreased from 7.94 and 7.82 log cfu/mL to 4.97 and 5.98 log cfu/mL with adding 1% CaO and no growth was observed for all LAB at 1.5% CaO, except for *L. brevis* (3.75 log cfu/mL at 1.5% CaO). Numbers of *S. cerevisiae* remained constant in all concentrations of CaO (average 7.82 log cfu/mL). The results indicate that the initial rise in the pH caused by the addition of CaO delays the fermentation process and thus controls the growth of yeasts and ethanol production.

In addition, other microorganisms other than those in the present study are responsible for lowering the pH in the early stages of fermentation.

**Key Words:** lactic acid bacteria, sugarcane silage, *Saccharomyces cerevisiae*

**W205 Application of *Pediococcus pentosaceus*, *Pichia anomala*, and chitinase to high moisture alfalfa hay at baling: Effects on chemical composition and conservation characteristics.** Long Jin<sup>1</sup>, Lysiane Dunière<sup>1</sup>, Joseph P. Lynch<sup>1</sup>, Eric Chevaux<sup>2</sup>, Tim A. McAllister<sup>1</sup>, John Baah<sup>3</sup>, and Yuxi Wang\*<sup>1</sup>, <sup>1</sup>*Agriculture and Agri-Food Canada, Lethbridge, AB, Canada*, <sup>2</sup>*Lallemand animal nutrition, Blagnac, France*, <sup>3</sup>*Best Environmental Technologies Inc., Edmonton, AB, Canada*.

The need to attempt to bale alfalfa hay at higher than optimum moisture level has increased in recent years due to unpredictable weather conditions driven by climate change. This often results in heating of the forage and a reduction in its nutritive quality. Although organic acids such as propionic acid (PA) are generally effective in preventing fungal growth in moist forage, the high rate of application and field handling costs coupled with environmental concerns makes hay producers reluctant to use them. The objective of this study was to assess the effects of applying *Pediococcus pentosaceus* (PED) and *Pichia anomala* (PIC) alone or in combinations with chitinase (CH) at baling on conservation characteristics of high moisture alfalfa hay. Alfalfa was harvested at the mid-bloom stage, wilted in the field to 73–77% DM (HMH) and baled without or with PED, PIC, PED+PIC, PED+CH, PIC+CH, and PA. Control (HMH), PED+CH, PIC+CH and PA and alfalfa hay baled at >85% DM (NMH) treatments were repeated over 3 yrs while other treatments were examined only within a single year. Interior temperature was recorded and samples were taken 90 d after baling for chemical and microbial assessment. Averaged over 3 yr, HMH had higher ( $P < 0.001$ ) temperature degree days (>30°C, HDD) than NMH. The HDD of PED+CH was lower ( $P < 0.001$ ) than that of other treatments in yr 1 and 3, but the max temperature achieved was lower ( $P < 0.05$ ) for PED+CH as compared with other treatments only in yr 3. The NDF and NH<sub>3</sub>-N contents were lower ( $P < 0.001$ ) for PED+CH and PIC+CH, but WSC was higher ( $P < 0.001$ ) for PED+CH than that of HMH and PA in yr 1. Mold and yeast counts did not differ among treatments, but CH+PED and CH+PIC had lower ( $P < 0.05$ ) total culturable bacteria than other treatments in yr 1. The results indicated that *P. pentosaceus* and *P. anomala* combined with chitinase applied at baling have the potential to improve the conservation of HMH alfalfa by preventing spontaneously heating.

**Key Words:** high moisture alfalfa hay, additives, spontaneous heating

**W206 Nitrogenous compounds and fermentation characteristics of king grass-cassava tops silages.** Tyrone Clavero\*, *Universidad del Zulia, Maracaibo, Estado Zulia, Venezuela*.

To increase nitrogenous compounds and improve fermentation quality of King grass (*Pennisetum purpureum* × *Pennisetum typhoides*) silage, ensiling with *Manihot esculenta* was tested. The treatments for silage making were: 100% king grass (KG), 25% cassava (C):75% KG, 50% C:50% KG, 75% C:25% KG and 100% C. Fresh plant materials were chopped to 1 cm length, mixed according to treatments, ensiled in laboratory silos and stored at 25°C for 60 d. After opening silos, dry matter (DM), pH, total nitrogen content (TN), protein nitrogen (PN), soluble nitrogen (SN), PN/TN, ammonia nitrogen (AN), neutral detergent fiber

nitrogen of total nitrogen (NDF/TN), and acid detergent fiber nitrogen of total nitrogen (NADF/TN) were determined. The data were analyzed according to a completely randomized design with 3 replications, significance among mixing levels was determined by Tukey test. Silage DM increased ( $P < 0.01$ ) with the inclusion of cassava in the mixtures, reaching the highest values (25.95%) with 75%. The introduction of cassava significantly ( $P < 0.05$ ) decreased pH levels of the silages, showing the lowest value (3.88) with 100% cassava. The TN, PN, SN, and PN/TN contents increased linearly ( $P < 0.05$ ) as the percentage of cassava increased in the mixtures. Proportion of neutral and acid detergent nitrogen (NDF and NADF) to total nitrogen were not affected by treatments ( $P > 0.05$ ). A small amount of AN was detected in silage, however, there were not significant differences ( $P > 0.05$ ) between 50 and 100% mixing levels of cassava. In this study, the inclusion of cassava at the rate of 25% or more, increased nitrogenous compounds and improved silage fermentation.

**Key Words:** silage mixture, cassava, nitrogenous compound

**W207 Relationship between grinding energy and chemical composition and NDF digestibility in forages.** E. Prinsloo<sup>1</sup>, C. Anelich<sup>1</sup>, E. Raffrenato\*<sup>2</sup>, W. A. van Niekerk<sup>1</sup>, and L. J. Erasmus<sup>1</sup>, <sup>1</sup>*Department of Animal & Wildlife Sciences, University of Pretoria, Pretoria, South Africa*, <sup>2</sup>*Department of Animal Sciences, Stellenbosch University, Stellenbosch, South Africa*.

Objectives of our work were to assess the relationship between chemical components and forage fragility, when measured as grinding energy (GrE), and between fragility and digestibility of forages. In Experiment 1 28 forages were analyzed for DM, total N, starch, NDF, ADF, ADL, 24 h ivNDFd, total phenols (TP) and non-tannic phenols (NTP). Initial particle size (IPS) was determined after pre-grinding the samples with a 2 cm fitted screen knife mill. The GrE was measured using an ultra-centrifugal and a hammer mill, fitted with a 1 mm screen and final particle size (FPS) was determined. Difference between the 2 mills was determined using a *t*-test for 2 dependent samples. Multiple regressions were used to best predict observed GrE measurements from the set of parameters obtained. Energy measurements ranged from 34.9 to 356 J/g with the ultra-centrifugal mill consistently using less energy than the hammer mill ( $P < 0.001$ ). The independent variables selected were able to explain 77 to 89% of the GrE and IPS consistently accounted for most of the variation, showing the difficulty of having a homogenous IPS. Forage resistance to grinding did not depend on the fiber quantity alone, but on the different quality and interaction of its components. In Experiment 2 GrE was measured on 34 forages, including corn silages, alfalfa and eragrostis hays. In vitro NDF digestibility was obtained from 0 to 240 h. Correlations between GrE and NDFd at various time points and NDF rates of digestion (kd) were calculated and the best predictors among fiber, its components and digestibility values were selected. The results showed high negative correlations of NDFd and kd with GrE ( $P < 0.001$ ). In particular 18 and 24 h NDFd had the highest correlation values (−0.65 and −0.77) across samples and species, showing the important relationship between fragility and fiber digestibility. The IPS, FPS, cellulose (ADF-ADL) and kd explained up to 60% of the variation. Grinding energy can be used to rank forages to account for different fragility, but IPS represents a limitation. Forages with similar peNDF may then be adjusted to account for different GrE and fragility.

**Key Words:** forage fragility, grinding energy, particle size



**W208 The effect of a bacteria inoculant on fermentation and aerobic stability of a brown-midrib and conventional corn silage.**

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Objective of this study was to test the effect of a forage inoculant (Bonsilage corn, Schaumann Inc., Mendota Heights, MN) containing *Lactobacillus buchneri*, (DSM16774) and *Lactobacillus plantarum*, (DSM12837) on fermentation and aerobic stability of corn silage. Brown-midrib and conventional corn (33.8% and 37.0% DM, respectively) were ensiled untreated or treated with inoculant at  $5 \times 10^5$  cfu/g of fresh forage in bags. Data were analyzed by proc GLM (SAS v.9.4) as a  $2 \times 2$  factorial with inoculant and corn silage as main effects. Ninety days after ensiling, the silages showed a similar response to the inoculant, except that BMR silage had a higher content of acetic acid and ethanol than conventional corn silage. The treated corn silages had less lactic acid compared with untreated silages and a greater concentration of acetic acid. The pH of treated silages was higher than untreated silages because of the decreased lactic acid and higher content of acetic acid. Untreated corn silages increased significantly in temperature after 95h of exposure to air, while treated silages did not increase in temperature during 168h of monitoring. Inoculating corn silage with  $5 \times 10^5$  cfu/g of the bacteria inoculant resulted in a more hetero fermentation and markedly improved the aerobic stability of corn silage.

**Table 1 (Abstr. W208).** The chemical composition and aerobic stability of corn silage<sup>1</sup> after 90 d of ensiling

Item	BMR		CS		P-value	
	Control	Treated	Control	Treated	Inoculant	Silage
DM, % as fed	32.8	31.2	37.5	36.2	0.92	<0.001
pH	3.80	4.23	3.80	4.20	<0.001	0.37
Lactic acid, % of DM	4.75	1.04	4.76	1.18	<0.001	0.57
Acetic acid, % of DM	1.69	3.81	1.58	3.17	<0.001	0.01
Total acids, % of DM	6.44	4.88	6.34	4.35	<0.001	0.12
Ethanol, % of DM	0.88	0.81	0.39	0.56	0.37	<0.001
L/A Ratio	2.82	0.27	3.02	0.37	<0.001	0.11
Aerobic stability <sup>2</sup>	98 h	ND	95 h	ND		

<sup>1</sup>BMR = brown-midrib; CS = conventional corn silage; treated with  $5 \times 10^5$  cfu/g of inoculant.

<sup>2</sup>Hours before temperature of silage increased 2° above ambient temperature.

**Key Words:** inoculant, buchneri, silage

**W209 Nutritional evaluation of leaves, twigs, and fruits of *Gmelina arborea* as feed resource for ruminants.** Ronke Y. Aderinboye\*, Olukayode O. Showunmi, Chryst F. I. Onwuka, and Victoria O. A. Ojo, Federal University of Agriculture, Abeokuta, Ogun, Nigeria.

Browse species have played major roles as feed alternatives particularly when grasses are of low nutritive quality serving as sources of crude protein in the tropics. Although, the leaves of some browses have been harvested for small ruminant feeding, the pods or fruits have not been extensively utilized. In this study, the nutritional value of the leaves, twigs and fruits of *Gmelina arborea* was evaluated through determination of their chemical composition, in vitro gas production and digestibility in a completely randomized design. Five hundred grams (n = 6) each of fresh *Gmelina* leaves, twigs and fruits was harvested from the different tree stands and oven-dried to constant weight for dry matter determination. Dried samples were used for chemical analysis and in

vitro studies. In vitro digestibility of leaves, twigs and fruits and their combinations were evaluated. Statistical analyses was done using one-way ANOVA procedure and means separated using Duncan's multiple range test. Results revealed that *Gmelina* leaves had higher dry matter (DM) and crude protein (CP) contents (39.07%DM; 15.54%CP) than the twigs (35.26%DM; 10.63%CP) and fruits (34.36%DM; 10.42%CP). Leaves and twigs were higher in calcium and phosphorus content than the fruits. Cell wall fractions were higher in the twigs than the leaves and fruits. The fruits had the highest nitrogen free extract content, higher ( $P < 0.05$ ) in vitro net gas production, dry matter degradability (63.4%) and short chain fatty acid content. The gas produced from degradable fraction; b was lower in twigs relative to the leaves and fruits while rate of degradation; c was similar for the leaves, twigs and fruits. The combination of leaves, twigs and fruits showed better nutritive value than sole twigs. Chemical composition of *Gmelina* leaves, twigs and fruits suggests them as a nutritive feed resources for ruminants. The leaves and fruits can serve as potential sources of crude protein and fermentable carbohydrate while the twigs can be a good source of fiber.

**Key Words:** *Gmelina*, aerial part, ruminant

**W210 Comparison of Ankom filter bag types for the determination of acid and neutral detergent fibers.** Chris D. Teutsch, Brian T. Campbell\*, and W. Mac Tilson, Virginia Tech, Blacksburg, VA.

In the mid-1990s Ankom Technology developed a filter bag system for fiber determination. This system reduced labor and costs and simplified the operation. More recently, a new filter bag (F-58) was developed. This bag possesses reduced pore size and has been directed toward end users who utilize a finer grind size to support near infrared spectroscopy (NIRS). Little work has compared the older (F-57) and newer (F-58) bags. The primary objective of this study was to compare F-57 and F-58 filter bags for determination of ADF and NDF. Four forage species were collected, dried, and sequentially ground to pass through a 2 and 1 mm screen. These species included orchardgrass (*Dactylis glomerata* L.), bermudagrass (*Cynodon dactylon* L.), white clover (*Trifolium repens* L.), and sericea lespedeza (*Lespedeza cuneata* L.). Acid and NDF were determined using the Ankom 200 Fiber Analyzer. Trial x treatment interactions occurred for ADF ( $P < 0.01$ ). Therefore data are presented by trial. Within trials, significant forage x filter bag interactions occurred ( $P < 0.01$ ). In general, the use of the F-58 bag decreased ADF concentrations in all forages except bermudagrass ( $P < 0.01$ ) and had no effect on NDF concentrations ( $P > 0.05$ ). However, the impact on ADF concentrations was not consistent across forage species. Bermudagrass ADF concentrations did not differ between bags for either trial and orchardgrass differed in Trial I only where the % ADF was decreased from 38 to 35% for F-57 and F-58 bags respectively ( $P < 0.01$ ). At the current time our data indicate that the inconsistent performance across forage species and the increased cost does not justify the use of the F-58 bags.

**Key Words:** fiber, forage, detergent

**W211 Effect of corn planting density on yield and nutritional quality of corn silage when planted after ryegrass harvested for silage.** Gonzalo Ferreira\*<sup>1</sup>, Paul Hammock<sup>2</sup>, Mary Hammock<sup>2</sup>, Issac Hammock<sup>2</sup>, and Nathan Hammock<sup>2</sup>, <sup>1</sup>Department of Dairy Science, Virginia Polytechnic Institute and State University, Blacksburg, VA, <sup>2</sup>Hammock Dairy Inc., Museville, VA.

The objective of this study was to determine the effect of planting density on yield and nutritional composition of corn silage in a  $2 \times$ -crop

**Table 1 (Abstr. W211).** Effect of corn planting density (plants/ha × 1000) on yield and nutritional quality of corn silage

	Nonirrigated				Irrigated				SEM	<i>P</i> <		
	55	70	85	100	55	70	85	100		I	D	I×D
DM yield, kg/ha × 1000	18.9	18.2	19.2	21.5	18.9	20.5	20.1	21.6	0.13	0.44	0.23	0.80
Plant dry weight, g/plant	340	280	250	220	343	337	265	228	16	0.17	0.01	0.33
DM, %	27.7	27.4	27.7	26.7	27.8	27.7	27.6	25.5	0.53	0.10	0.36	0.46
CP, %	9.9	10.1	9.6	9.8	10.3	10.6	10.6	10.1	0.20	0.03	0.22	0.41
NDF, %	38.8	38.6	40.1	39.7	35.2	34.6	37.2	38.3	1.11	0.04	0.12	0.68
ADF, %	22.9	23.7	25.2	25.2	22.8	22.7	23.4	24.7	0.81	0.09	0.17	0.83
ADL, %	3.1	2.9	2.8	3.2	2.7	2.5	2.7	3.0	0.19	0.14	0.23	0.73
Starch, %	29.2	31.2	30.1	31.3	30.3	31.2	32.1	30.6	1.29	0.56	0.68	0.75
IVNDFD, %	55.1	55.5	56.0	53.9	55.1	55.9	56.3	54.3	0.78	0.60	0.08	0.99

rotation system. The study was performed at an 800-cow dairy farm located in south Virginia. Corn was planted in experimental plots within 2 cornfields, one of which was irrigated with a central-pivot irrigating system. Planting densities were 55, 70, 85, and 100 seeds/ha (×1000) in 4 replicates per cornfield (2 fields × 4 densities × 4 replicates = 32 plots). Plots were 12 25-m long rows separated by 76 cm. The irrigated cornfield was watered with 100 mm of water before tasseling. At the early-dent stage of maturity, 10 plants from each plot were cut (15 cm above ground), weighed, chopped, mixed thoroughly, and ensiled in bags for 60 d. Nutritional composition was performed by wet chemistry. The statistical model included the effects of irrigation (I), density (D),

the 2-way interaction (I×D), block, and field-by-block interaction. No interactions between irrigation and planting density were observed for any of the variables of interest. Dry matter yield was not affected by planting density. Increasing planting density reduced plant biomass. Planting density did not affect the concentrations of CP, NDF, ADF, ADL, and starch, but tended to affect IVNDFD quadratically. In conclusion, under a double-crop rotation system increasing planting density of corn does not increase DM yield and also does not affect the nutritional composition of corn silage when harvested at an early stage of maturity.

**Key Words:** plant density, corn silage, nutritional quality

## Growth and Development II

**W212 Sodium butyrate induces adipocytic differentiation of porcine mesenchymal stem cells.** Benedetta Tugnoli<sup>1</sup>, Chiara Bernardini<sup>1</sup>, Monica Forni<sup>1</sup>, Andrea Piva<sup>1</sup>, Chad H. Stahl<sup>2</sup>, and Ester Grilli<sup>\*1</sup>, <sup>1</sup>DIMEVET, University of Bologna, Ozzano Emilia, Bologna, Italy, <sup>2</sup>Laboratory of Developmental Nutrition, College of Agriculture and Life Sciences, North Carolina State University, Raleigh, NC.

Sodium butyrate (SB) has been shown to affect the differentiation of mesenchymal stem cells (MSC) through the activation of different transcriptional pathways. The aim of this study was to investigate the effects of SB on the proliferation and differentiation potential of porcine bone marrow-derived MSC. Third passage MSC were verified by flow cytometry to be > 95% CD105<sup>+</sup>, CD90<sup>+</sup>, CD44<sup>+</sup>, CD45<sup>-</sup>, and CD34<sup>-</sup>. Cells were cultured in either a low glucose DMEM+10% FBS (BM), BM + 2.5 mM SB (SB2.5) or BM + 5 mM SB (SB5) in a randomized complete block design. Data were analyzed with 1-way ANOVA followed by linear and quadratic contrasts and the treatments had 6 independent replicates (n = 6). Cell proliferation was significantly decreased by both SB2.5 and SB5 after 48h (-55% on average,  $P = 0.001$ ) and 72h (-63% on average,  $P = 0.001$ ). To assess the impact of SB on spontaneous differentiation, MSC were cultured for 27 d, with complete media change every 3 d. At 3 d, SB treated cells showed changes in morphology compared with controls, from spindle-shaped fibroblast-like to larger sail-shaped cells with intracellular shiny droplets. At 27d, cells were stained for osteocytic (Alizarin Red), chondrocytic (Alcian Blue) and adipocytic (Oil-Red-O) differentiation. No terminal differentiation was detected in MSC cultured in the BM alone, while accumulated lipids were clearly stained by Oil-Red-O in MSC cultured in the presence of SB. The phenotypic changes observed from 3 to 27 d were supported by a change in the pattern of gene expression, analyzed by semiquantitative real-time PCR. At 3 d, both SB2.5 and SB5 increased mRNA expression of peroxisome proliferator-activated receptor gamma (PPAR $\gamma$ ;  $P = 0.05$ , linear effect) and decreased osteocalcin (OC;  $P = 0.02$ , quadratic) and aggrecan ( $P = 0.04$ , quadratic) mRNA levels compared with untreated cells. At 27d, there was an increase in PPAR $\gamma$  mRNA level ( $P = 0.02$ , linear) and decreased OC and aggrecan ( $P = 0.02$  and  $P = 0.002$ , respectively, both linear and quadratic) with SB treatment. To conclude, our data suggest that SB promotes the differentiation of porcine bone marrow-derived MSC toward an adipocytic lineage.

**Key Words:** sodium butyrate, mesenchymal stem cells, pig

**W214 Effect of butyrate on inflammatory and oxidative gene markers in porcine IPEC-J2 intestinal epithelial cells.** Hui Yan\* and Kolapo Ajuwon, Purdue University, West Lafayette, IN.

The small intestine is important for nutrient digestion and absorption as well as immune function. Newborns are sensitive to pathogen and diet-induced inflammation, which influences development of intestinal function. Butyrate has been shown to possess immune modulatory effect in various cells and tissues. But its effect on LPS-induced inflammation in the porcine IPEC-J2 intestinal cell model are unclear. Therefore, we investigated the effects of butyrate on expression of cytokines and metabolic gene markers in this cell line. A randomized complete block design was used. Main effect was the different concentrations of butyrate and replicate was used as the blocking factor. There were at least 6 replicates per treatment. Data were analyzed using PROC GLM. Gene expression was determined by RT-PCR. High concentration of butyrate (1mM) significantly increased ( $P < 0.05$ ) LPS-induced TNF $\alpha$ , IL-6, IL-8

and MCP1 expression, and this indicated that this concentration was harmful to the cells. However, lower concentrations of butyrate (10  $\mu$ M to 100  $\mu$ M) significantly decreased ( $P < 0.05$ ) LPS-induced MCP-1 and TLR4 expression (100  $\mu$ M concentration) and IL-1 $\beta$  (50  $\mu$ M) compared with control. Butyrate treatment also significantly increased expression of metabolic genes such as ACO (100  $\mu$ M) and PPAR $\alpha$  (10 to 100  $\mu$ M), indicating that butyrate increases energy expenditure. LPS treatment significantly reduced ACO expression and eliminated the effect of butyrate on ACO and PPAR $\alpha$  expression. Taken together, butyrate exhibited limited anti-inflammatory effect in IPEC-J2 cells, but strongly enhanced oxidative capacity through induction of oxidative genes.

**Key Words:** IPEC-J2, butyrate, inflammation.

**W215 MicroRNA exert a role in the process of arginine promoting rat mammary gland development.** Lianmin Chen<sup>\*1</sup>, Liangyu Hu<sup>1</sup>, Mengzhi Wang<sup>1</sup>, J. J. Loo<sup>2</sup>, Hongrong Wang<sup>1</sup>, and Lihuai Yu<sup>1</sup>, <sup>1</sup>College of animal science and technology, Yangzhou University, Yangzhou, Jiangsu Province, China, <sup>2</sup>University of Illinois, Urbana, IL.

Except for its use during milk protein synthesis, extra arginine (Arg) might exert other unknown biological or metabolic functions in the mammary gland. Previous work focused on the effect of Arg on bovine mammary epithelial cell casein production showed that 556.00 mg/L Arg (2  $\times$  the basal level) in the medium elicited the greatest stimulation of casein and mTOR-related genes. Whether Arg regulates cell development via effects on microRNA (miR) is unclear. The specific objective was to determine if miR expression in rat mammary tissue are altered by feeding 2  $\times$  Arg. Twelve pregnant littermate Wistar rats were randomly divided into 2 groups and experimental diets designed according to the AIN-93G purified diets. The Arg concentration was 12.8 g/kg in the Arg group while in the Control was 6.4 g/kg. A total of 21.62 g/kg glutamic acid was added to the Control group to keep diets iso-nitrogenous. At 17 d postpartum rats were killed and the mammary tissue harvested. Histological changes were measured by paraffin section, and mammary acinar area measured using digital imaging system software (MC30). Statistical analysis was carried out via *t*-test using SPSS16.0. Differentially expressed miR were measured using Solexa miR-Seq and verified by RT-qPCR. The miR target genes were analyzed using SBC prediction wizard (TargetScan, miRanda, PicTar, MirTarget2, PITA supported). Gene functions were analyzed via DAVID 6.7 and GO analysis. The mammary acinar area was significantly greater ( $P < 0.001$ ) due to Arg (452.71  $\mu$ m<sup>2</sup>) compared with the Control (388.68  $\mu$ m<sup>2</sup>). A total of 8 miR had greater ( $P < 0.05$ , fold-change > 2) expression in Arg compared with the Control. Among those upregulated, miR-1-3p plays a role in the control of cellular component synthesis; miR-133a-3p and miR-133a-5p play a role in the regulation of cell developmental process; miR-133b-3p exerts control of transporter activity and enzyme activity; and miR-206-3p has an important role in the regulation of cellular processes. There was no clear cellular function prediction for the upregulated miR such as miR-149-5p, miR-1b and miR-486. Results suggest dietary Arg might promote mammary tissue development through altering miR expression. The exact regulatory mechanisms need further investigation.

**Key Words:** micro RNA, arginine, development of mammary



**W216 Poor maternal nutrition decreases longissimus dorsi cross sectional area of fetal offspring at d 45 of gestation.** Joseline S. Raja\*, Sambhu M. Pillai, Amanda K. Jones, Maria L. Hoffman, Katelyn K. McFadden, Kristen E. Govoni, Steven A. Zinn, and Sarah A. Reed, *Department of Animal Science, University of Connecticut, Storrs, CT.*

Poor maternal nutrition during gestation results in long-term postnatal changes in muscle morphometrics, muscle mass and fiber number, and intramuscular adipose deposition of the offspring. Thus, we hypothesized that under- and over-feeding ewes during gestation would inhibit fetal muscle development. To test this hypothesis, 82 pregnant Western Whiteface ewes were individually housed and fed 100%, 60%, or 140% of NRC requirements for TDN beginning at d 30.2 ± 0.2 of gestation. Offspring (CON, RES, and OVER, respectively), were necropsied at d 45, 90, and 135 of gestation and within 24 h of birth. At d 45 of gestation (n = 7 ewes per diet), the triceps brachii (TB), semitendinosus (STN), and longissimus (LM) muscles were weighed. The LM were frozen and cryosectioned to determine muscle fiber cross sectional area (CSA) and the number of muscle fibers per  $\mu\text{m}^2$ . Muscle weight was expressed as percent of fetal weight and data were analyzed using the MIXED procedure of SAS, with maternal diet as the main effect. Fetal weight tended to be less in OVER compared with CON ( $P = 0.07$ ; CON: 11.0 ± 0.6 g, RES: 10.1 ± 0.5 g, OVER: 9.2 ± 0.5 g). No differences in the weight of TB, STN, or LM due to maternal diet were detected ( $P \geq 0.53$ ). The CSA of LM (n = 6 fetuses per treatment [2 fetuses per ewe]) from OVER and RES were smaller compared with CON ( $P \leq 0.008$ ; CON: 287.1 ± 3.2  $\mu\text{m}^2$ ; RES: 223.4 ± 2.9  $\mu\text{m}^2$ ; OVER: 254.9 ± 3.2  $\mu\text{m}^2$ ). However, the number of fibers per  $\mu\text{m}^2$  of LM was not different due to maternal diet ( $P = 0.47$ ). Smaller muscle fiber CSA could result from inadequate protein accretion, decreased fusion of myogenic precursor cells, or both. Similar fiber numbers suggest increased interfibrillar space, which may lead to increased connective tissue formation. In conclusion, poor maternal nutrition during early gestation alters the formation of muscle fibers as early as d45, potentially resulting in decreased muscle mass at birth.

**Key Words:** muscle, maternal nutrition

**W217 The effects of maternal under- and over-feeding on muscle development of lambs as determined by RNA-Seq analysis.** Maria L. Hoffman\*, Kristen N. Peck, Jill L. Wegryzn, Sarah A. Reed, Steven A. Zinn, and Kristen E. Govoni, *University of Connecticut, Storrs, CT.*

We previously reported that both maternal under- and over-feeding altered muscle cross sectional area and lipid content in offspring. Therefore, we hypothesized that poor maternal nutrition would alter key pathways and novel genes involved in muscle development of lambs. Ewes (n = 3/treatment) were fed 100% (CON), 60% (RES), or 140% (OVER) of NRC requirements for TDN and one twin lamb per ewe was euthanized within 24 h of birth and muscle tissue was collected for RNA-Seq analysis. RNA was prepared into cDNA libraries and sequenced using the Ion Torrent Proton. Data were aligned to the *Bos taurus* (Btau\_4.61) and *Ovis aries* (Oar\_V.3.1) reference genomes using Tophat and expression evaluated with Cufflinks. Differential gene expression was determined using the Benjamini-Hochberg multiple testing correction ( $q \leq 0.05$ ). Using the ovine and bovine references, 10 and 35 differentially expressed genes were identified, respectively. The differentially expressed genes are involved in metabolic regulation, hypertrophy, nutrient uptake, and protein turnover. Using the ovine reference, compared with CON, expression of *myogenic factor 6*, and *myostatin* increased 1.7 and 2.0-fold in OVER ( $q \leq 0.05$ ). *BTG family member 2*, *ankyrin repeat domain 1*, and *phosphoserine phosphatase*

expression decreased 1.5, 2.5, and 3.1-fold, respectively in OVER ( $q \leq 0.05$ ). Expression of *FHJ murine osteosarcoma viral oncogene homolog (FOS)* was reduced 1.6-fold in RES compared with CON ( $q = 0.05$ ). Using the bovine annotation, compared with CON, *arrestin domain containing 2* and *AT rich interactive domain 5B* expression were 3.5 and 2.9-fold greater, respectively in OVER ( $q = 0.05$ ). Expression of *PPAR $\gamma$  coactivator 1  $\alpha$* , *regulator of G-protein signaling 16*, *thrombomodulin*, and *jun B proto-oncogene* were reduced  $\geq 2$ -fold ( $q \leq 0.05$ ) in OVER. *Tripartit motif containing 63*, *FOS* and *suppressor of cytokine signal 3* expression were reduced 1.4, 1.6, and 2.8-fold, respectively in RES ( $q \leq 0.05$ ). In conclusion, although both under- and over-feeding had similar effects on offspring muscle development, based on gene expression, the mechanisms through which this occurs appear to be different.

**Key Words:** transcriptome, muscle, sheep

**W218 Muscle fiber hypertrophy is associated with increased expression of key transcriptional and epigenome regulatory genes.** Yue Lu, Jennifer S. Bradley, Sarah R. McCoski, Adam J. Geiger, R. Michael Akers, Alan D. Ealy, and Sally E. Johnson\*, *Virginia Polytechnic Institute and State University, Blacksburg, VA.*

Early neonatal nutrition affects the rate and extent of muscle growth and fiber hypertrophy in calves. The objective of the experiment was to examine the effect of caloric intake on LM, infraspinatus (INF) and semitendinosus (ST) fiber cross-sectional area (CSA), fiber type and muscle gene expression in neonatal heifers. Newborn Holstein heifers were assigned to a high nutritional plane (HNP; 28% CP, 25% fat, n = 5) or low nutritional plane (LNP; 20% CP, 20% fat, n = 5) milk replacer diet for 8 wks. Calves were pair-fed equivalent amounts of grain (25% CP, 4% fat) beginning at wk 5. Caloric intake supported ADG of 220 g/d and 771 g/d for the LNP and HNP heifers, respectively. Heifers were euthanized at 8 wks of age and samples of LM, INF and ST were collected. Gene expression was measured by RT-qPCR. Data were analyzed by ANOVA for the main effect of diet, muscle and their interaction with post-hoc Tukey separation of means. Plane of nutrition differentially affected muscle fiber hypertrophy with HNP LM and ST fiber cross-sectional area (CSA) larger ( $P < 0.05$ ) than LNP. No differences in CSA were noted between HNP and LNP INF. The smaller LNP LM and LNP ST sizes were reflected by greater ( $P < 0.05$ ) numbers of fibers/mm<sup>2</sup>. *Pax7* expression is 38% greater ( $P < 0.05$ ) in HNP LM than LNP LM and *BTG2*, *E2F6* and *myogenin* mRNA amounts are 60% greater ( $P < 0.05$ ) suggesting that muscle stem and progenitor cells are active and supplying the growing fiber with myonuclei. Messenger levels of *IGF-1* are greater ( $P < 0.05$ ) in HNP LM than LNP LM further supporting increased fiber hypertrophy. No differences ( $P > 0.05$ ) were observed in the protein degradation enzyme genes, *FOXO3*, *atrogin* or *MuRF*, indicating that protein accretion is due to elevated protein synthesis. Diet can serve as an epigenetic modifier of phenotype. Calves maintained on the LNP diet expressed lower ( $P < 0.05$ ) amounts of the histone deacetylase genes, *HDAC1* and *HDAC3*, and the lysine-specific demethylase, *KDM2A*, in the LM than their HNP counterparts. Altered expression of epigenome modifiers provides evidence for diet-induced changes in the muscle fiber chromatin architecture that may contribute to long-term metabolic responses.

**Key Words:** skeletal muscle, hypertrophy, epigenome

### W219 Myogenic regulatory factors are increased in bovine satellite cells by polyamines and their precursor amino acid ornithine.

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Skeletal muscle is unique in its growth and developmental characteristics in that normal hypertrophy is carried out through activation and fusion of satellite cells (SC) surrounding individual muscle fibers. These SC are responsive to signals from their extracellular environment especially growth factors and nutrients. Polyamines (PA) are a class of small, positively charged molecules that favorably affect growing cells through somewhat undefined physiological mechanisms. Polyamines exist in mammalian species primarily as ornithine derivatives. Abnormalities in polyamine metabolism are associated with irregular development in mammals emphasizing their critical role as regulators of growth. To test the direct effect of PA on skeletal muscle development, primary bovine SC cultures were differentiated in media containing either methionine (10 mM control 1), ornithine (10 mM), putrescine (5 mM), spermine (0.5 mM), or no supplement (control 2). SC were differentiated as a monoculture (MC) as well as in coculture (CC) alongside preadipocytes isolated from bovine intramuscular fat depots. This coculture provided a more realistic environment of that which would surround skeletal muscle and impact SC activity. Following treatment, SC were isolated from CC using laser micro-dissection technique. SC populations were analyzed for temporal expression of the myogenic regulatory factors (MRF) *MyoD*, *Myf5*, and *Myf4* (myogenin) to identify differentiating cells along with the genes *Pax7* and *Spry1* representative of quiescent cells. Protein isolation and Western Blot analyses were also performed to measure protein expression in a temporal manner. Exposure of SC's to PA in CC resulted in upregulation of *MyoD* ( $P = 0.05$ ), *Myf5* ( $P = 0.02$ ), and *Myf4* ( $P = 0.09$ ) and attenuation of *Pax7* ( $P = 0.10$ ) and *Spry1* ( $P = 0.07$ ). Statistics generated in SAS (Cary, NC) using PROC MIXED procedure. Treatment was included as a fixed effect. These results suggest that ornithine metabolism and polyamine metabolites can affect bovine skeletal muscle myogenesis, and may therefore be promising as candidates for natural means of promoting growth of lean tissue in cattle.

**Key Words:** polyamine, satellite cell, myogenic regulatory factor

### W220 Muscle gene expression patterns in finishing steers supplemented with dietary Amaize (*Aspergillus oryzae* extract).

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We evaluated the effect of dietary *Aspergillus oryzae* extract on gene expression profiles in the *Longissimus lumborum* from finishing steers. Cross-bred (Simmental × Angus) yearling steers were randomly assigned to one of 2 groups (n = 10/treatment). From receiving until d21, starter and step-up diets were fed to acclimate steers to a traditional mid-west finishing diet. Steers were then fed *ad libitum* to meet or exceed NRC requirements until slaughter (d140): basal diet with or without 5 g/hd/d of *A. oryzae* extract (Amaize, Alltech Inc., Nicholasville, KY) containing  $\alpha$ -amylase (AMZ). On d90, *Longissimus lumborum* tissue was collected for gene expression analysis. Serum was collected at d 40, 90 and 140 for analysis of metabolites (BHBA, glucose, insulin, urea). Data from blood metabolites was analyzed using a mixed model. Gene expression was profiled using the Affymetrix Bovine Gene 1.0 ST Array. Performance did not differ between treatments. Hierarchical clustering indicated a treatment effect ( $P < 0.05$ ) of the AMZ-supplemented group compared with the control. A total of 1148 genes were differentially affected ( $P$

$< 0.05$ ; 430 upregulated; 718 downregulated) between treatments. The genes affected, enriched ( $P < 0.05$ ) and activated several pathways, including IGF-1 signaling, insulin receptor signaling, valine degradation, and VDR/RXR activation. The pathways activated are involved in the regulation of muscle development and growth. Blood metabolites indicated greater levels of BHBA, urea and insulin ( $P < 0.05$ ) at d40 in AMZ-supplemented steers. Only insulin remained at a greater concentration in the AMZ group than the control throughout the experiment. In conclusion, AMZ supplementation in the finishing diet affects muscle gene expression and insulin metabolisms, potentially causing a positive effect for the development of skeletal muscle in finishing steers.

**Key Words:** amylase, muscle, cattle

### W221 Investigation of effects of maternal nutrition intensification and fetal sex on development of skeletal muscle of bovine fetuses.

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This study aimed to evaluate the effects of maternal feeding level (MN) and fetal sex (FS) on skeletal muscle development of bovine fetuses at different stages of gestation (SG). Fourty-4 multiparous, dry Holstein × Gyr cows with average initial body weight of  $480 \pm 10$  kg were fed either restricted feeding at 1.15% of body weight (CO, n = 24) or *ad libitum* (ON, n = 20) with the same diet (93% corn silage and 7% concentrate; 111 g/kg of CP and 674 g/kg of TDN). Eleven cows of each dietary treatment were slaughtered at 139, 199, 241 and 268 d of gestation. Fetuses were necropsied to evaluate the development of skeletal muscle. Data were analyzed by MIXED procedure of SAS considering the fixed effects of MN, FS and SG ( $2 \times 2 \times 4$  factorial). Modifications in gene expression of skeletal muscle of fetuses were observed in function of MN and FS despite the lack of effect of MN ( $P = 0.330$ ) and FS ( $P = 0.518$ ) on fetal weight. The muscle mRNA expression of myogenic markers  $\beta$ -catenin and MyoD was greater in male than in female fetuses, as well the expression of all adipogenic markers evaluated (Zfp423, C/EBP $\alpha$  and PPAR $\gamma$ ), 3 of the 4 fibrogenic markers evaluated (Collagen I, Collagen III and Fibronectin) and the number of myocytes in muscle. Marginal effects of MN were observed on mRNA expression as well on the phenotypic indicators of myogenesis, adipogenesis and fibrogenesis. At the mid-gestation (139 DG)  $\beta$ -Catenin, Zfp423 and PPAR $\gamma$  expression and myocytes number were greater in ON than in CO fetuses and in males than in females, but these differences were not observed at subsequent SG. Fat content of fetal muscle was not affected by MN and FS. Almost all myogenic, adipogenic and fibrogenic markers were less expressed in late gestation than in mid-gestation, however collagen deposition, fat and crude protein content of fetal muscle were greater at late gestation than in mid-gestation. The MN changed gene expression of myogenic, adipogenic and fibrogenic markers at mid-gestation (greater in ON than in CO) but some compensatory gene expression made the effect of MN not significant in late gestation.

**Key Words:** adipogenesis, fetal programming, fibrogenesis

### W222 High-energy diet reduced myogenic gene expression of Hanwoo steers fed to three different endpoints.

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High energy diets have been used for enhancing marbling fat in high quality beef cattle. However, this has been associated with reduced yield grade when the steers were fed for long-term. The aim of this experiment was to determine the effect of additional 3% TDN (77%) diet compared with control diet (74%) on the level of adipogenic and myogenic gene expressions at 3 different endpoints. A 2 × 3 factorial arrangement (High, Control, and 26, 28, 30 mo endpoints) was used to feed 48 Hanwoo steers. Four steers were fed in same pen and 12 pens were used for treatment. Longissimus dorsi (L.D.) muscle was collected within 10 min of harvest for analysis of PPAR $\alpha$ , SCD, GLUT4, MHC1, MHC2X, and myogenin mRNA abundance. Real-Time RT-PCR was used to measure the quantity of respective mRNA relative to a ribosomal protein subunit 9 (RPS9) mRNA. Data were analyzed as a completely randomized design using the MIXED model, each treatment performed in triplicate. Difference between control and treatments were determined using the LSD procedure. Overall ADG did not differ between high-energy diet or control diet ( $P > 0.05$ ). ADG tend to increase in high-energy diet at 30 mo endpoint compared with control diet ( $P = 0.09$ ). Marbling score and carcass weight were greater at 30-mo endpoint than at other endpoints ( $P < 0.05$ ). Yield grade was not different among endpoints ( $P > 0.05$ ). Percentage of protein in L.D. muscle tend to decrease at 30 mo endpoints ( $P = 0.09$ ). Real-time quantitative PCR revealed that the mRNA content of MHC2X in muscle from high-energy diet cattle decreased (diet × endpoint,  $P < 0.05$ ) as compared with the control group. There was no interaction for MHC1 gene expression among 3 endpoints ( $P > 0.05$ ). These data indicated that high-energy diet decreased relative mRNA level of MHC2X on long-fattening periods. These fast-glycolytic genes may affect muscle protein composition in Hanwoo L.D. muscle.

**Key Words:** Hanwoo, myogenic, gene expression

#### **W223 $\alpha$ -Solanine induces myogenesis of bovine satellite cells isolated from semimembranosus and longissimus muscle tissue.**

K. Y. Chung<sup>1</sup>, J. K. Kim<sup>\*2</sup>, H. S. Kim<sup>1</sup>, E. M. Lee<sup>1</sup>, H. J. Kim<sup>1</sup>, S. S. Chang<sup>1</sup>, and H. S. Kang<sup>1</sup>, <sup>1</sup>Hanwoo Research Institute, NIAS, Pyeongchang, Korea, <sup>2</sup>Animal and Food Science, Texas Tech University, Lubbock, TX.

Solanine is a glycoalkaloid compound found in leaves, fruit and tubers of potato, tomato, and eggplant. Solanine poisoning was reported in various animal models such as mice, rabbit, and chicken. However, it is used as a treatment for human asthma. Some medicine used for treating human asthma has been used to induce myogenesis of bovine skeletal muscle. We hypothesized that  $\alpha$ -solanine may affect myogenesis of bovine satellite cells isolated from different muscle depots. Bovine satellite cells were pronase-liberated from semimembranosus (SM) and longissimus dorsi (LD) muscle tissues of 3 newborn Hanwoo calves. Bovine SM and LD satellite cells were incubated with dulbecco modified eagle medium (DMEM) with 10% fetal bovine serum for proliferation and induced differentiation with DMEM with 3% horse serum. Bovine satellite cells were treated with various levels of  $\alpha$ -solanine (control, 0.001, 0.01, 0.1, 1, and 10  $\mu$ M). mRNA abundance for myosin heavy chain 1 (MHC1), MHC2X, glucose transporter 4 (GLUT4), myogenin, G-coupled protein receptor 43 (GPR43), and  $\beta$ 2-adrenergic receptor ( $\beta$ 2-

AR) were measured by real-time quantitative PCR. Data were analyzed as a completely randomized design using the MIXED model, each treatment performed in triplicate. Means were considered different at  $P < 0.05$ . Relative MHC2X mRNA abundance was greater in both, SM and LD satellite cells with 1  $\mu$ M solanine treatments compared with the control ( $P < 0.05$ ). Relative level of MHC2X and GLUT4 were decreased in 10  $\mu$ M solanine treatments ( $P < 0.05$ ). Relative level of MHC2X and  $\beta$ 2-AR were greater in SM satellite cell compared with LD satellite cells ( $P < 0.05$ ). There was no tissue × dose interaction among MHC1 mRNA concentration ( $P > 0.05$ ). These result indicated that  $\alpha$ -solanine have a dose-dependent effect on MHC2X mRNA but did not affect to MHC1 mRNA in bovine satellite cell.

**Key Words:**  $\alpha$ -solanine, Hanwoo, semimembranosus

#### **W224 Role of epidermal growth factor receptor and erbB2 in trenbolone acetate mediated increases in bovine satellite cell proliferation and protein synthesis and decreases in protein degradation.**

Kara J. Thornton\*, Ernest Kamanga-Sollo, Michael E. White, and William R. Dayton, University of Minnesota, St. Paul, MN.

Trenbolone acetate, TBA, has been shown to increase proliferation and protein synthesis rates and decrease protein degradation rate in bovine satellite cell (BSC) cultures. Although we have previously shown that the androgen receptor is involved in these TBA effects, our current data show that 2 receptors from the erbB family, epidermal growth factor receptor (EGFR) and erbB2, also play a role in the effects of TBA on BSC proliferation, protein synthesis and protein degradation in BSC cultures. We have assessed the effects of treating BSC with AG1478, a specific EGFR tyrosine kinase inhibitor, and/or AG879, a specific erbB2 tyrosine kinase inhibitor, on the ability of TBA to affect proliferation rate (<sup>3</sup>H-Thymidine incorporation) in proliferating BSC cultures and to affect protein synthesis rate (<sup>3</sup>H-phenylalanine incorporation/mg protein) and percent protein degradation in fused BSC cultures. Statistics were done using Proc MIXED in SAS; the model included treatment as a fixed effect and experiment, BSC number and experiment replicate as random effects. Treatment with AG1478, AG879 or a combination of AG1478/AG879 significantly ( $P < 0.05$ ) suppressed TBA-induced increases in proliferation. Treatment with AG1478 significantly ( $P < 0.05$ ) suppressed specific TBA-induced increases in protein synthesis. Treatment with AG879 or a combination of AG879/1478 also decreased TBA-induced increases in protein synthesis ( $P < 0.05$ ) although the effects of AG879 or AG879/1478 may be non-specific as there was also a significant ( $P < 0.05$ ) decrease in protein synthesis rate in control cells (not treated with TBA) receiving these treatments. While treatment with either AG1478 or AG879 alone had no effect on the ability of TBA to reduce protein degradation rate in fused BSC cultures, treatment with a combination of AG879/1478 significantly ( $P < 0.05$ ) suppressed TBA-induced decreases in protein degradation rates. In summary our results show that receptors from the Erb family are necessary to support TBA-induced effects on proliferation, protein synthesis and protein degradation in BSC cultures.

**Key Words:** bovine, satellite cell, EGFR



## Nonruminant Nutrition: Energy and fiber

**W225 Fitting and validating prediction equations of metabolizable energy of meat and bone meal for pigs.** R. A. Castilho<sup>1</sup>, P. C. Pozza<sup>2</sup>, N. T. E. Oliveira<sup>3</sup>, C. P. Sangali<sup>2</sup>, C. N. Langer<sup>4</sup>, and R. V. Nunes<sup>3</sup>, <sup>1</sup>*Safedns Nutrição Animal, Cascavel, Paraná, Brazil*, <sup>2</sup>*Universidade Estadual de Maringá, Maringá, Paraná, Brazil*, <sup>3</sup>*Universidade Estadual do Oeste do Paraná, Mchal Candido Rondon, Paraná, Brazil*, <sup>4</sup>*Swine Production System, Quatro Pontes, Paraná, Brazil*.

The aim of this study was to fit and validate models to predict the metabolizable energy (ME) of meat and bone meal (MBM) for pigs. Thirty-two barrows (26.75 ± 1.45 kg) were allotted in a randomized blocks design with 8 treatments (7 MBM that replaced 20% the basal diet) and 4 replicates, to determine the ME. All MBM samples were analyzed for dry matter (DM), crude protein (CP), ether extract (EE), gross energy (GE), crude fiber (CF), mineral matter (MM), calcium (Ca) and phosphorus (P). The multiple linear regression models were adjusted using GE, CP, EE, MM, Ca and P as regressors (DM basis) using the ordinary least square method. To validate prediction equations, a database was compiled containing 48 pairs of observed and predicted ME. The 48 observed ME values were compiled from the literature and classified according to the scientific origin, resulting in 15 Brazilian data and 33 from international literature. The validity was initially assessed by adjusting the linear regression of 1st degree of the observed ME in function of the estimated ME, using the ordinary least squares. This procedure was adopted for the databases from the Brazilian, international and mixed literature. The validation of the estimated equations, as predictors of the linear ratio of ME, was checked by fitting a linear model of 1st degree of the predicted values ( $y = b_0 + b_1x_i$ ) of ME by the equations initially estimated in function of the observed values. The ME values of MBM ranged from 1645 to 2645 kcal/kg. The prediction equations  $ME_1 = -4233.58 + 0.4134GE + 72P + 89.62MM - 159.06Ca$  ( $R^2 = 0.90$ );  $ME_2 = 2087.49 + 0.3446GE + 31.82MM - 189.18Ca$  ( $R^2 = 0.87$ );  $ME_3 = 2140.13 + 0.3845GE - 112.33Ca$  ( $R^2 = 0.86$ );  $ME_4 = -346.58 + 0.656GE$  ( $R^2 = 0.80$ ); and  $ME_5 = 3221.27 + 178.96EE - 76.55MM$  ( $R^2 = 0.82$ ) were effective in predicting the ME of Brazilian MBM. However, there was no validation when using data obtained from international researches. In conclusion, the equations that efficiently estimates the ME of MBM for pigs in Brazilian conditions are  $ME_1 = -4233.58 + 0.4134GE + 72CP + 89.62MM - 159.06Ca$  and  $ME_2 = 2087.49 + 0.3446GE + 31.82EE - 189.18Ca$ .

**Key Words:** chemical composition, feedstuff, model

**W226 Effects of different net energy concentrations of diets on the growth performance of growing or finishing pigs housed individually.** Gang Il Lee<sup>1</sup>, Kwang-sik Kim<sup>2</sup>, Jun Cheol Park<sup>2</sup>, and Dong Yong Kil<sup>1</sup>, <sup>1</sup>*Chung-Ang University, Anseong-si, Gyeonggi-do, Republic of Korea*, <sup>2</sup>*Rural Development Administration, Cheonan-si, Chungnam, Republic of Korea*.

This experiment was conducted to investigate the effects of different net energy (NE) concentrations of diets on the growth performance of growing or finishing pigs housed individually. In experiment 1 (growing pigs), a total of 60 growing pigs (Landrace × Yorkshire × Duroc; initial BW = 34.3 ± 3.7 kg) were allotted to 5 dietary treatments with 12 replicate pens and 1 pig per pen in a 35-d feeding trial. In experiment 2 (finishing pigs), a total of 60 finishing pigs (Landrace × Yorkshire × Duroc; initial BW = 73.7 ± 5.6 kg) were allotted to 5 dietary treatments

with 12 replicate pens and 1 pig per pen in a 28-d feeding trial. Within each experiment, a corn-soybean meal-based basal diet (91.75%) was prepared to meet all nutrient requirements for growing or finishing pigs (NRC, 2012). The remaining portion (8.25%) was added to the basal diet by different inclusion ratio of soybean oil and starch to create 5 NE concentrations (2,318, 2,390, 2,462, 2,533, or 2,605 kcal NE/kg) of diets for growing or finishing pigs. The NE and amino acids concentrations in diets were calculated based on the values from NRC (2012). Pigs were allowed ad libitum access to feed and water in both experiments. Statistical model included dietary treatment as a fixed effect. Orthogonal polynomial contrast test was performed to determine linear and quadratic effects of NE concentrations of diets. Results indicated that the final BW, ADG, ADFI, and feed efficiency (G:F) of growing pigs were not affected by dietary treatments. Similarly, final BW, ADG, and ADFI of finishing pigs were not affected by dietary treatments. However, the G:F of finishing pigs was improved (linear,  $P < 0.01$ ) with increasing NE concentrations in diets (0.326, 0.331, 0.323, 0.352, and 0.349 for 2,318, 2,390, 2,462, 2,533, or 2,605 kcal NE/kg, respectively). In conclusion, increasing NE concentrations in diets from 2,318 to 2,605 kcal/kg have no beneficial effects on the growth performance of growing pigs, but improve feed efficiency of finishing pigs.

**Key Words:** growth performance, net energy, pig

**W227 Effects of dietary metabolizable energy levels on performance and energetic metabolism of broiler chickens.** M. P. F. Teixeira, N. C. Baiao, L. J. C. Lara, M. A. Pompeu\*, L. F. P. Pereira, C. W. R. Gondim, K. R. Soares, and W. L. S. Climaco, *Universidade Federal de Minas Gerais, Belo Horizonte, MG, Brazil*.

Recent research has demonstrated a reduction in dietary heat increment and improvements in production performance from supplementing lipids as an energy source. This performance improvement can be attributed to increased availability of nutrients from feed ingredients, in addition to improved energy efficiency by increasing net energy (NE) of diet. The present study was conducted to evaluate the effects of increasing metabolizable energy (ME) levels in the diet on performance parameters, nutrient digestibility and energy balance of 19 to 41 d of age broilers kept in thermoneutral conditions. A total of 336 male broiler chickens were evaluated in this study, distributed in a completely randomized design with 4 treatments and 6 replicates with 14 broilers each. The treatments consisted of 4 diets containing 19% CP and increasing ME levels (2,980, 3,080, 3,180 and 3,280 kcal/kg), which were obtained by addition of soybean oil. The data were submitted to ANOVA and the means were compared by the Tukey test ( $\alpha = 0.05$ ). Feed intake was not affected by ME levels; although the best results for weight gain (2.3 kg) and feed conversion ratio (1.53 kg/kg) were obtained in broilers fed with diets containing 3,280 kcal/kg ( $P < 0.01$ ). The viability rate and feed cost per kg of live weight did not differ between treatments, but production efficiency ratio increased linearly as ME was increased ( $P < 0.01$ ). The ME levels affected the EE digestibility, broilers fed with more energetic diets (3,180 and 3,280 kcal/kg) achieved better EE digestibility (79.8% and 81.4%, respectively;  $P < 0.01$ ). Heat production and increment was not affected by ME levels. The dietary ME levels had a linear effect over NE ( $P < 0.03$ ), but ME conversion into NE remained constant between diets. In conclusion, ME levels has influence on performance and energetic metabolism of broiler chickens under thermoneutral condition.

**Key Words:** heat increment, net energy, thermal comfort

**W228 Effects of dietary metabolizable energy levels on performance and energetic metabolism of broiler chickens under cyclic heat stress condition.** M. P. F. Teixeira, N. C. Baiao, L. J. C. Lara, M. A. Pompeu\*, L. F. P. Pereira, K. R. Soares, A. F. Silva, J. F. V. Braga, and A. R. C. Abreu, *Universidade Federal de Minas Gerais, Belo Horizonte, MG, Brazil.*

Improving broiler performance during heat stress has been the aim of many researches, and one of most common approach is to increase dietary metabolizable energy (ME), which may increase nutrients availability and improve energy efficiency by both reducing heat increment (HI) and increasing ME conversion into net energy (k). This study aimed to evaluate the effects of increasing ME levels on performance, nutrient digestibility and energy balance of 19 to 41 d old broilers kept under cyclic heat stress (16 h of  $23 \pm 2^\circ\text{C}$  and 8 h of  $32 \pm 2^\circ\text{C}$ ;  $60 \pm 2\%$  RH). A total of 336 male broilers were used, distributed in a completely randomized design with 4 treatments and 6 replicates of 14 birds each. Treatments consisted of 4 diets containing 19% CP and 4 ME levels (2,980, 3,080, 3,180 and 3,280 kcal/kg), which were obtained by the addition of soybean oil. Data were submitted to ANOVA and the means were compared by Tukey and SNK test for performance and energetic metabolism, respectively ( $\alpha = 0.05$ ). Weight gain, feed intake, viability and productive efficiency ratio were not influenced by ME levels ( $P > 0.05$ ). However, broilers presented a better feed conversion ratio when fed with 3,280 kcal/kg, followed by 3,180 kcal/kg ( $P = 0.003$ ). Moreover, the lowest feed cost per kg of live weight was observed with 2,980 kcal/kg, followed by broilers that consumed 3,080 kcal/kg of feed ( $P = 0.028$ ). The digestibility of CP and EE increased linearly with ME increase in diet ( $P < 0.001$ ). The consumption of ME adjusted to zero nitrogen balance was not affected by diet treatment ( $P > 0.05$ ), but a higher heat production was observed in broilers that received diets with the lowest ME level ( $P = 0.012$ ). HI was higher in broilers fed with 2,980 kcal/kg diet ( $93.9 \text{ kcal/kg}^{0.75}/\text{day}$ ) ( $P = 0.016$ ). The dietary ME levels had a linear effect over net energy ( $P < 0.021$ ), but k remained constant between diets ( $P > 0.05$ ). We conclude that ME level influences on performance and energetic metabolism.

**Key Words:** cyclic heat stress, heat production, net energy

**W229 Determination of apparent digestibility of fibrous sources for finishing pigs.** Bernardo Berenchein\*<sup>1</sup>, Evelyn Brito<sup>1</sup>, Adibe Abdalla<sup>2</sup>, Helder Louvandini<sup>2</sup>, Dineshkumar Danashekar<sup>2</sup>, Adibe Abdalla Filho<sup>2</sup>, Paulo Lima<sup>2</sup>, and Patrícia Righetto<sup>2</sup>, <sup>1</sup>*Laboratory of Studies and Researches of Production and Nutrition of Poultry and Swine; Federal University of Amazonas, Parintins, AM, Brazil,* <sup>2</sup>*Center of Nuclear Energy in Agriculture, Piracicaba, SP, Brazil.*

Many studies evidenced that the possibility of using forages and other grasses as feedstuffs in swine production. However, there is a need to study the potential fibrous feedstuffs in swine production in terms of identification, quantification and evaluation of interactions between physiological and associative effects on digestibility and animal performance. Hence, the objective of the present study was to evaluate the apparent digestibility of Tifton-hay, Citric Pulp and Soybean hulls in finishing pig diets. Twenty crossbred barrows ( $70 \pm 1.95 \text{ kg}$ ) were distributed in a randomized block design with 4 treatments ( $n = 5$ ) fed 20% replacement of fibrous diets on basal diet, Group I- 100% basal diet; Group II- 80% basal diet +20% Tifton hay; Group III- 80% basal diet +20% Citric pulp and Group IV- 80% basal diet +20% Soybean hulls, in individual metabolism cages. Apparent digestibility of Dry Matter and nutrients was performed by total feces and urine collection method for pigs, which animals were housed in metabolism cages for 12 d (7 d for acclimation and 5 d for collection). Feeds and feces were

analyzed for dry matter (DM), crude energy (CE), crude protein (CP), neutral detergent fiber (NDF) and acid detergent fiber (ADF). Our result shows that Tifton hay had 2.425 kcal/kg of apparent digestible energy and 3.36% of apparent digestible protein. In addition, apparent digestibility of NDF and ADF of Tifton hay were 28.47 and 22.63% respectively. However, the results for Citric pulp were 2.850 kcal/kg of apparent digestible energy, 1.47% of apparent digestible protein and also apparent digestibility of NDF and ADF, were 8.14 and 7.85%. Soybeans hulls showed 2.250 kcal/kg of apparent digestible energy, 7.85% of apparent digestible protein and apparent digestibility of NDF and ADF were 21.7 and 26.39%, respectively. Therefore, our present study concluded that fibrous sources were used like feedstuffs in finishing pig diets. Moreover, it confirms that fibrous feeds will be both nutritionally and economically beneficial to swine production. The authors thank for the support of CNPq and FAPEAM.

**Key Words:** citric pulp, soybean hulls, Tifton hay

**W230 Feeding diets containing low-protein or rapidly fermentable carbohydrate to weanling pigs does not affect growth performance.** Vivian V. Almeida\*<sup>1</sup>, Amoracyr J. C. Nuñez<sup>1</sup>, Patrícia V. A. Alvarenga<sup>2</sup>, Fabrício R. Castelin<sup>2</sup>, Ysenia V. Silva-Guillen<sup>2</sup>, and Maria Cristina Thomaz<sup>2</sup>, <sup>1</sup>*Department of Animal Sciences, Purdue University, West Lafayette, Indiana,* <sup>2</sup>*Department of Animal Science, São Paulo State University, Jaboticabal, São Paulo, Brazil.*

The objective in this study was to evaluate the effects of dietary CP and dried citrus pulp (DCP, rapidly fermentable carbohydrate source) contents on growth performance of weanling pigs. A total of 108 barrows weaned at 21 d of age were blocked by initial BW ( $5.82 \pm 0.96 \text{ kg}$ ) and randomly assigned to 4 treatments with 9 replicate pens per treatment and 3 pigs per pen. Treatments were arranged in a  $2 \times 2$  factorial, with 2 CP contents (HCP and LCP; high- and low-CP diets, respectively) and 2 DCP contents (0 and 7.5%, as-fed basis). The HCP treatments consisted of feeding 20 and 21% CP diets (as-fed basis) throughout the pre-starter I (1 to 14 d) and pre-starter II (15 to 28 d) phases, respectively. For the LCP treatments, CP contents were 16 and 17% for the pre-starter I and II diets, respectively. The AA contents in the diets were balanced by supplementation with crystalline AA, such as L-Lys, DL-Met, L-Thr, L-Trp, L-Val, and L-Ile to maintain an ideal AA pattern. Ideal AA ratios (standardized ileal digestible basis) were: 100% Lys; 28% Met; 63% Thr; 18% Trp; 69% Val; and 55% Ile. Individual pig BW and pen feed disappearance were recorded weekly to determine ADG, ADFI, and G:F. Data were analyzed as repeated measures using the MIXED procedure of SAS, with pen and block as the random effects and CP, DCP, day, and 2-way and 3-way interactions as the fixed effects. There were no CP x DCP interactions for BW, ADG, ADFI, and G:F; thus, the main effects are discussed. Growth performance was not affected by the dietary DCP contents. Even though BW and ADFI did not differ between dietary CP contents during the 28-d feeding period, pigs fed the LCP-diet showed decreased ADG ( $P = 0.03$ ) and G:F ( $P = 0.02$ ) from d 21 to 28 post-weaning compared with those fed the HCP-diet. However, overall ADG and G:F were not affected by reducing the dietary CP content. In conclusion, feeding diets containing 7.5% DCP to weanling pigs does not affect growth performance. Moreover, dietary CP content in pig pre-starter diets may be reduced by 4 percentage units when combined with AA supplementation without depressing overall growth performance.

**Key Words:** amino acid, fiber, pig

**W231 Effects of dietary protein and rapidly fermentable carbohydrate contents on cecal microbial fermentation profile of weanling pigs.** Vivian V. Almeida\*<sup>1</sup>, Amoracyr J. C. Nuñez<sup>1</sup>, Patrícia V. A. Alvarenga<sup>2</sup>, Fabrício R. Castellini<sup>2</sup>, Ysenia V. Silva-Guillen<sup>2</sup>, and Maria Cristina Thomaz<sup>2</sup>, <sup>1</sup>Department of Animal Sciences, Purdue University, West Lafayette, Indiana, <sup>2</sup>Department of Animal Science, São Paulo State University, Jaboticabal, São Paulo, Brazil.

A total of 108 barrows weaned at 21 d ( $5.82 \pm 0.96$  kg initial BW) were used to determine the effects of dietary CP and dried citrus pulp (DCP; rapidly fermentable carbohydrate source) contents on VFA and ammonia concentrations in the cecal digesta of weanling pigs. Pigs were blocked by initial BW and randomly assigned to 1 of 4 treatments with 9 replicate pens per treatment and 3 pigs per pen in a randomized complete block design. Treatments were arranged in a  $2 \times 2$  factorial, with 2 CP contents (HCP and LCP; high- and low-CP diets, respectively) and 2 DCP contents (0 and 7.5%, as-fed basis). The HCP treatments consisted of feeding 20 and 21% CP diets (as-fed basis) throughout the pre-starter I (1 to 14 d) and pre-starter II (15 to 28 d) phases, respectively. For the LCP treatments, CP contents were 16 and 17% for the pre-starter I and II diets, respectively. Crystalline AA were added to the HCP and LCP diets to maintain an ideal AA pattern. Cecal digesta samples were collected from 1 pig per pen on d 7 and 28 postweaning for determinations of VFA profile by gas chromatography and ammonia concentration by colorimetry. Data were analyzed as repeated measures using the MIXED procedure of SAS, with pen and block as the random effects and CP, DCP, day, and 2-way and 3-way interactions as the fixed effects. Pigs fed diets containing 7.5% DCP tended ( $P = 0.06$ ) to have increased acetate concentration and decreased ( $P < 0.01$ ) propionate, isovalerate, and valerate concentrations in the cecum. Cecal butyrate, isobutyrate, and total VFA concentrations were not affected by the treatments. There was a DCP  $\times$  day interaction ( $P < 0.05$ ) for isovalerate concentration, in which dietary inclusion of 7.5% DCP decreased ( $P < 0.01$ ) isovalerate concentration only at 28 d postweaning. There was a CP  $\times$  DCP interaction for ammonia concentration ( $P < 0.01$ ), in which adding 7.5% DCP to the diet tended ( $P = 0.06$ ) to decrease cecal ammonia concentration only for pigs fed the LCP diet. In conclusion, low-protein AA-supplemented diets with 7.5% DCP inclusion may reduce proteolytic fermentation in the cecum of weanling pigs.

**Key Words:** microbial metabolite, protein, piglet

**W232 Effect of feeding wheat- or barley-based diets with high- or low- nutrient density on diet nutrient digestibility and growth performance in weaned pigs.** X. Zhou\*<sup>1</sup>, M. G. Young<sup>2</sup>, M. L. Swift<sup>1,3</sup>, E. Beltranena<sup>1,4</sup>, and R. T. Zijlstra<sup>1</sup>, <sup>1</sup>University of Alberta, Edmonton, AB, Canada, <sup>2</sup>Gowans Feed Consulting, Wainwright, AB, Canada, <sup>3</sup>Alberta Agriculture and Rural Development, Lethbridge, AB, Canada, <sup>4</sup>Alberta Agriculture and Rural Development, Edmonton, AB, Canada.

Barley grain is a commonly fed energy feedstuff for grower-finisher pigs in western Canada. However, its feeding value for weaned pigs is poorly characterized, especially by modern feed formulation method using NE values and standardized ileal digestible (SID) AA coefficients for feedstuffs. Formulating lower nutrient dense diets may reduce feed cost, provided weaned pigs can maintain growth performance by increasing ADFI to meet requirements. To compare the feeding value of wheat- and barley-based diets with high- or low- nutrient density, 4 diets based on 64% wheat or 68% barley grain providing either 2.4 or 2.3 Mcal NE/kg were fed in a  $2 \times 2$  factorial arrangement to 208 weaned pigs (9.0 kg) housed in 13 pens per diet. Diets were formulated to 4.5

g SID Lys/Mcal NE and were fed for 21 d. Feed added, remaining, and pig BW were measured weekly to calculate pen ADFI, ADG, and G:F. Freshly-voided feces were collected on d 19 and 20 to measure diet apparent total-tract digestibility (ATTD) of DM, GE, and CP and diet DE values. No interactions between cereal source and nutrient density were observed. Compared with wheat-based diets, feeding barley-based diets reduced ( $P < 0.01$ ) the ATTD of DM, GE, and CP by 2.7, 3.0, and 4.4%, respectively, and reduced ( $P < 0.01$ ) measured diet DE value by 0.05 Mcal/kg. Compared with high-nutrient density diets, feeding low-nutrient density diets reduced ( $P < 0.01$ ) ATTD of DM, GE, and CP by 2.6, 1.8, and 2.3%, respectively, while measured diet DE value was not affected. Overall, feeding barley-based diets increased ( $P < 0.01$ ) ADG by 41 g and G:F by 0.04 g:g compared with feeding wheat-based diets, but ADFI was not affected. Diet nutrient density did not affect ADFI, ADG, or G:F. In conclusion, feeding barley grain instead of wheat as cereal energy source for weaned pigs may increase growth performance. Growth performance of weaned pigs was not affected by reducing dietary NE by 0.1 Mcal/kg when SID Lys/NE was maintained. Feeding barley- instead of wheat-based diets provided advantages for the feeding of weaned pigs.

**Key Words:** barley, energy, weaned pig

**W233 Graded concentrations of dietary wheat bran reduce ileal and total tract digestibility of nutrients but increase hindgut digestibility of dry matter and organic matter in pigs.** A. R. Son\*, W. B. Kwon, and B. G. Kim, Konkuk University, Seoul, Republic of Korea.

The objective was to determine the effects of graded concentrations of wheat bran (WB) on apparent ileal (AID), apparent total-tract (ATTD), and hind gut digestibility of nutrients and to test the effects of time for collecting fecal grab samples on nutrient digestibility in pigs. Six barrows (initial mean BW of  $70.7 \pm 5.7$  kg) surgically fitted with a T-cannula in the distal ileum were used. A replicated  $3 \times 3$  Latin square design was used with 3 diets, 3 periods, and 6 pigs. A basal diet was prepared to mainly contain wheat, soybean meal, and cornstarch. Two additional diets were formulated to contain 20 or 40% of WB at the expense of cornstarch. Each experimental period consisted of a 7-d adaptation period and a 4-d collection period. After the adaptation period, fecal samples were collected on d 8 and 11 using a grab sampling method, and ileal digesta samples were collected on d 9 and 10. To compare differences between nutrient digestibility calculated based on time points for fecal grab sampling, before or after the collection period for ileal digesta, a paired *t*-test was used. The AID of DM (81.8, 71.4, and 58.4%), OM (84.9, 75.0, and 62.4%), CP (78.2, 77.5, and 68.9%), and ADF (49.7, 45.4, and 36.1%) linearly decreased ( $P < 0.05$ ) with increasing inclusion rate of WB from 0 to 40%. The ATTD of DM (91.8, 84.7, and 76.0%), OM (93.9, 87.2, and 78.8%), CP (89.7, 88.5, and 84.1%), and ADF (68.6, 53.4, and 39.1%) linearly decreased ( $P < 0.01$ ) as the inclusion rate of WB increased. Hindgut digestibility of DM (9.6, 13.4, and 17.6%) and OM (8.7, 12.2, and 16.4%) linearly increased ( $P < 0.05$ ), and that of NDF (30.5, 15.2, and 10.3%) and Ca (19.9, 18.2, and 10.1%) linearly decreased ( $P < 0.05$ ) with increasing inclusion rate of WB. However, there were no differences in ATTD of nutrients between time points for feces grab sampling. In conclusion, the inclusion of WB reduced AID and ATTD of nutrients but increased hindgut digestibility of DM and OM, and there were no difference in the ATTD between the fecal collection time points of before and after ileal digesta collection in pigs.

**Key Words:** digestibility, swine, wheat bran



**W234 Up to 30% insoluble dietary fiber reduces carcass fat content of heavy pigs.** Daniela Junqueira Rodrigues<sup>1</sup>, Maria Cristina Thomaz<sup>1</sup>, Urbano dos Santos Ruiz<sup>2</sup>, Maryane Sespere Oliveira\*<sup>1</sup>, Everton Daniel<sup>1</sup>, Fabrício Rogério Castelini<sup>1</sup>, Ysenia Victoria da Silva Guillen<sup>1</sup>, Vivian Vezzoni Almeida<sup>1</sup>, and Sarah Sgavioli<sup>1</sup>, <sup>1</sup>Universidade Estadual Paulista, Jaboticabal, Sao Paulo, Brazil, <sup>2</sup>Universidade de São Paulo, Piracicaba, Sao Paulo Brazil.

The slaughter of heavy pigs (120 to 130kg BW) provides relatively bigger carcasses and a higher amount of meat, but has the disadvantage of a higher fat deposition, given that animals have higher food intake while protein deposition remains the same. Qualitative feed restriction is a technique that provides caloric intake control, diluting the diet energy with a fibrous ingredient. Our aim was to test the effects of diets containing increasing concentrations of insoluble fiber as part of a qualitative feed restriction program on carcass traits. Thirty 2 barrows (commercial crossbred; initial BW 78.53 ± 11.02kg), were assigned to 4 dietary treatments (ad libitum) with 8 replicate pens per treatments and one animal per pen in a randomized block design: control (corn and soybean meal based; 3230 kcal/kg calculated ME, 13.9% CP, 15.5% insoluble fiber) and 3 diets with increasing concentrations of insoluble fibers (20, 25 and 30%; from purified cellulose), and consequently decreased concentrations of calculated ME. The slaughtered happened when BW means were 133.30 ± 11.59kg. The yields were calculated based on the final BW, the backfat thickness, loin eye and fat area were measured at last ribs, in the insertion of the last thoracic vertebra to the first lumbar. Data were analyzed by ANOVA using GLM procedure of SAS, polynomial regression were used to determine the effect of insoluble fiber. We observed linear reduction ( $P < 0.05$ ) to backfat thickness and fat area, moreover, linear increase to meat:fat ratio. There were no differences in final BW, yield, lean meat, loin eye area and ham yield. In conclusion, using up to 30% of insoluble fiber in diets of heavy pigs as part of a qualitative feed restriction program reduces carcass fat without altering other carcass traits.

**Table 1 (Abstr. W234).** Carcass traits from swine fed different levels of insoluble fiber

Item	Experimental diets (% IF)				SEM	Effect of IF
	15.5	20	25	30		
Final BW, kg	136.18	132.38	131.83	130.97	2.23	—
Yield, %	80.83	80.54	79.72	79.14	0.76	—
Lean meat, %	55.53	54.53	56.11	57.39	0.75	—
Backfat thickness, mm	33.97	31.62	32.16	26.78	0.90	Linear
Loin eye area, cm <sup>2</sup>	44.14	39.14	43.14	45.43	1.36	—
Fat area, cm <sup>2</sup>	28.86	33.86	23.29	23.14	1.48	Linear
Meat:Fat	1.63	1.16	1.97	2.14	0.12	Linear
Ham yield, %	29.22	27.93	30.75	29.04	0.55	—

**Key Words:** backfat thickness, cellulose, feed restriction

**W235 Effect of dietary inclusion of insoluble fiber from sugar cane on meat quality of finishing swine.** Maryane Sespere Oliveira\*, Maria Cristina Thomaz, Marco Monteiro Lima, Fabrício Faleiros Castro, Patricia Versuti Arantes Alvarenga, Manuela Vantini Marujo, and Daniela Junqueira Rodrigues, *Universidade Estadual Paulista, Jaboticabal, Sao Paulo, Brazil.*

The objective of this study was to evaluate the effects of insoluble fiber, extracted from sugar cane, in finishing swine diets on meat quality. Fifty barrows were blocked by initial BW (79.33 ± 2.20 kg) and randomly assigned to one of 5 treatments with 10 replicate pens per treatment and

one animal per pen. Treatments consisted of corn/soybean meal-based diets formulated to contain 0, 5, 10, 15 to 20% insoluble fiber and the levels of calculated ME (3230, 3069, 2905, 2745 and 2586 kcal/kg) were decreased according to increased dietary fiber. Pigs were slaughtered when BW means were around 130 kg. Initial pH (pH45) in *longissimus dorsi* (LD) was measured at the last rib position 45 min after slaughter and final pH (pHu) was measured 24 h after slaughter. The left LD muscle was removed, and sections of about 10 cm thickness were cut from the anterior end for meat quality analysis, such as, color, drip loss, oxidative stability, shear force, the concentration of cholesterol and lipids. The color of LD was measured with a Minolta Chromameter. The oxidative stability was assessed by TBARS. The shear force required to shear each block of muscle was determined by means of a Stable Micro systems texture analyzer fitted with Volodkevitch jaws. Data were analyzed by ANOVA using the MIXED procedure of SAS, diet was the fixed effect and replicate was the random effect. Polynomial contrasts were used to determine the effect of insoluble fiber inclusion. The drip loss, color parameters *L* (lightness), *a* (redness), *b* (yellowness) of LD and muscle pH were unaffected by dietary fiber treatments. But linear responses in Shear force (2.00, 1.74, 1.75, 2.00 and 1.87 ± 0.12,  $P < 0.05$ ) and quadratic responses in Cholesterol (27.35, 25.24, 28.59, 27.00 and 24.51 ± 2.12,  $P = 0.02$ ) were observed as amount of insoluble fiber increased in the diet. Insoluble fiber level had no effect on oxidative stability and lipids of LD. In conclusion, increasing insoluble fiber in the diet may soften meat and reduce cholesterol in the swine meat, without altering other meat qualities.

**Key Words:** insoluble fiber, meat quality, pig

**W236 Mannans and glucans in diets for weanling pigs and their effects on the diarrhea incidence and pH of the digestive tract.** Patricia V. A. Alvarenga, Maria C. Thomaz, Marco M. Lima\*, Daniela J. Rodrigues, Manuela V. Marujo, Fabrício F. Castro, and Maryane S. F. Oliveira, *Univesidade Estadual Paulista, Jaboticabal, Sao Paulo, Brazil.*

An experiment was conducted to evaluate the effects of diets containing mannans and glucans on diarrhea incidence and pH of the digestive tract organs of weanling pigs. Ninety-six piglets weaned at 24 d (7.7 ± 1.76 kg initial BW) were blocked by initial BW and randomly assigned to 4 treatments with 8 replicate pens per treatment and 3 pigs per pen in a randomized complete block design. Pigs had ad libitum access to feed and water over a 42 d study. The treatments were: PC (positive control diet: 40ppm of colistine sulfate); NC (negative control diet: free of antimicrobial); NC+CA (NC + Compound A: 10% MOS and 18% β-glucan); NC+CB (NC + Compound B: 18% MOS and 20% β-glucan). The inclusion of Compound A was 10 kg/ton and of Compound B was 2 kg/ton. The diarrhea incidence was measured 2 times a day, every day until the 21 d of experiment, using subjective scores according to the consistency and appearance of feces (1 = well-formed, 2 = soft, and 3 = diarrhea). One animal per pen was slaughtered on d 14 of experiment. The pH of the organs contents were measured using a pH meter. Data were analyzed by ANOVA and Tukey's test (5%), using the GLM procedure of SAS. Higher incidence of diarrhea and also less incidence of well-formed and soft feces were observed for NC ( $P < 0.01$ ). The pH of the stomach content was lower for NC+CB and PC ( $P < 0.05$ ), and the pH of the small intestine was lower for NC+CA compared with PC ( $P < 0.05$ ), but did not differed from NC and NC+CB. In conclusion, feeding diet with no additives results in higher incidence of diarrhea, also treatments NC+CB and PC are able to reduce the pH of the stomach.

**Table 1 (Abstr. W236).** Diarrhea incidence and pH of the digestive tract organs

Item	Treatments				SEM	P-value
	NC	NC+CA	NC+CB	PC		
pH						
Stomach	3.309 <sup>b</sup>	3.278 <sup>b</sup>	2.615 <sup>a</sup>	2.591 <sup>a</sup>	0.1994	<0.05
Small intestine	5.366 <sup>ab</sup>	4.900 <sup>a</sup>	5.310 <sup>ab</sup>	5.763 <sup>b</sup>	0.1988	<0.05
Cecum	5.504	5.230	4.931	5.023	0.1867	>0.05
Diarrhea incidence						
Well-formed	6.870 <sup>b</sup>	17.375 <sup>a</sup>	15.000 <sup>a</sup>	15.250 <sup>a</sup>	1.9483	<0.01
Soft	6.000 <sup>b</sup>	10.750 <sup>a</sup>	11.500 <sup>a</sup>	15.000 <sup>a</sup>	1.5889	<0.01
Diarrhea	25.000 <sup>b</sup>	9.250 <sup>a</sup>	11.375 <sup>a</sup>	7.625 <sup>a</sup>	1.6219	<0.01

**Key Words:** glucan, mannan, diarrhea

**W237 Influence of hydrolysable tannin extract supplementation on performance of growing-finishing pig.** Rubén Aguirre<sup>1</sup>, Javier A. Romo<sup>1</sup>, Rubén Barajas\*<sup>1</sup>, Juan M. Romo<sup>1</sup>, and Héctor R. Güémez<sup>1,2</sup>, <sup>1</sup>FMVZ-Universidad Autónoma de Sinaloa, Culiacán, Sinaloa, México, <sup>2</sup>Granja Porcina La Huerta, Culiacán, Sinaloa, México.

Seventy-two pigs (36 males and 36 females) with  $26.98 \pm 4.26$  kg of initial weight and 70 d old were used in a 90 d experiment to evaluate the influence of hydrolysable tannin extract supplementation on performance of growing-finishing pig. Animals were weighed, allotted by initial

weight, and in groups of 6 (3 males and 3 females) they were placed in 12 paved floor pens ( $7 \times 1.5$  m). Pen constituted the experimental unit. In a randomized complete block design, pens were assigned to treatments as follows: 1) Growing and finishing diets formulated with corn and soybean meal (Control); or 2) Control plus addition of 0.2% of hydrolysable tannin extract (HT). The hydrolysable tannin extract was provided as SilvaFeed NutriP (Pronutrient Developer, Mexico). Results were analyzed by ANOVA for a randomized complete block design. During the first 49 d that constituted the growing phase (d 1–49), HT supplementation augmented ( $P = 0.04$ ) body weight (66.11 vs. 60.18 kg) and improved average daily gain ( $P = 0.04$ ) by 18% (0.802 vs. 0.680 kg). Addition of HT increased ( $P < 0.01$ ) average daily feed intake (0.805 vs. 1.675 kg), but the feed intake as proportion of mean body weight was similar between treatments (3.84 vs. 3.89%). The feed efficiency (Gain/feed ratio) was not affected by treatments (0.410 vs. 0.447). During the finishing phase (d 50 to d 90), HT supplementation increased ( $P = 0.02$ ) feed intake (2.44 vs. 2.08 kg) without effect on ADG and feed efficiency. In complete 90 d experiment (joint growing and finishing phases), HT supplementation tended ( $P = 0.06$ ) to improve final weight (97.17 vs. 88.17 kg) and average daily gain (0.782 vs. 0.681 kg); increased ( $P < 0.01$ ) feed intake (2.093 vs. 1.858 kg), without effect on feed efficiency. It is concluded that 0.2% hydrolysable tannin supplementation during the growing phase improves performance of pig, but its addition during the finishing phase does not represent any advantage.

**Key Words:** growth performance, hydrolyzable tannin, pig

## Physiology and Endocrinology: Male reproduction, deers, and poultry

**W238 The effect of moringa oleifera leaf meal on follicle stimulating hormone, luteinizing hormone and testosterone of wad goat bucks serum.** Abimbola O. Ladokun\*, Khadeejah Kareem-Ibrahim, Bukola D. Adenaike, Oladimeji M. Abioja, and John A. Abiona, *Federal University of Agriculture, Abeokuta, Ogun, Nigeria.*

Moringa as a plant has been attributed with possessing anti-oxidative properties and lowering of blood cholesterol. It has also been reported to be involved as an endocrine system blocker (ESB). This study investigated the effect of moringa leaf meal supplemented in the feed of West African dwarf (WAD) goat bucks on their serum FSH, LH and testosterone. Sixteen matured WAD bucks were used; they were assigned into 4 groups with 2 replicates and 2 bucks per replicate. They were semi-intensively managed, allowed to graze and their concentrate supplemented with blended moringa leaf at 0g/kg concentrate, 100, 200, and 300 g/kg respectively for 8 weeks. Blood was sampled 3 times (Start, 4th week and end of 8th week) and serum harvested for analyses using EIA for testosterone and ELISA kits and reader for FSH and LH. Data were analyzed using one-way ANOVA of SAS, and significant ( $P = 0.05$ ) means were separated using the Duncan test in SAS. Concentrations of testosterone were similar across the groups (5.65–5.95ng/L). At the 4th and 8th week, the concentrations though greater than start, they were also similar across the groups indicating that moringa did not influence testosterone concentration in WAD bucks. Concentrations of FSH were also similar at the start (4.85–5.0), but there were decreases in concentrations at the 4th and 8th week as compared with start. It is noteworthy that for bucks supplemented with 200g/kg moringa, there was a significant ( $P = 0.05$ ) decrease in concentration at 4th and 8th week (5.0–4.40–4.30ng/L). The average serum LH at start was similar among groups (3.75–4.10ng/L). At the 4th and 8th week, treatment did not affect concentrations of LH. Results indicate that moringa leaf at the level used in this study did not elicit any response indicative of disrupting the concentration of testosterone, FSH and LH in WAD bucks.

**Key Words:** moringa, goat buck, hormone

**W239 Proteomic analysis of testicular proteins between yak and its sterile hybrid by iTRAQ labeling mass spectrometry.** Wei Fu<sup>1</sup>, Wenlin Bai<sup>2</sup>, Lin Huang<sup>1</sup>, Wenjing Liu<sup>3</sup>, Caixia Li<sup>1</sup>, Suyu Jin<sup>1</sup>, Xiang Qiu<sup>1</sup>, Liang Ren<sup>1</sup>, and Yucai Zheng\*<sup>1</sup>, <sup>1</sup>Southwest University for Nationalities, Chengdu, Sichuan Province, China, <sup>2</sup>Shenyang Agricultural University, Shenyang, Liaoning Province, China, <sup>3</sup>Southwest University of Science and Technology, Mianyang, Sichuan Province, China.

The objective of the present study aimed at identifying differential expression proteins in testes of adult yak (*Bos grunniens*) and its sterile hybrid (cattle-yak), to provide basic data for elucidating the molecular mechanisms of the male sterility of cattle-yak. Cattle-yak is a hybrid of male cattle (*Bos taurus*) with female yak, exhibiting higher body size, milk and meat yields than those of yak, but F<sub>1</sub> to F<sub>3</sub> male sterility, the mechanism is still unknown. In the present study, total proteins were extracted from the testes of adult yaks (n = 6) and cattle-yaks (n = 4), respectively, followed by analysis of the testicular protein profiles (pooled samples for each group) by isobaric tags for relative and absolute quantitation (iTRAQ) technique. Proteins with a fold change of >1.2 and  $P < 0.05$  (*t*-test) were considered to be significantly differentially expressed. Hypergeometric test was used in Gene Ontology analysis. A total of 4464 proteins were identified, of which 714 proteins

were differentially expressed, including 432 downregulated and 282 upregulated proteins in the testis of cattle-yak compared with that of yak. Interestingly, approximately 30% of the top 50 downregulated proteins in cattle-yak testis are testis-specific or highly expressed in testis, and most likely exert crucial effects on spermatogenesis. Gene Ontology analysis of the differentially expressed proteins demonstrated that 19 major protein categories belong to carbohydrate metabolism and protein folding were highly enriched in the Biological process category, implying the important roles of the 2 processes in spermatogenesis. Further analysis of Biological process category revealed 43 proteins associated with reproductive process, and most of which were downregulated in the testis of cattle-yak. In addition, fatty acid oxidation metabolism was found to be more active in cattle-yak testis than in yak testis. Based on the testicular protein profiles, protein functions and protein-protein interaction analysis, we propose that the abnormal expression of several downregulated proteins of testis-specificity or with high abundance in testis, such as YBX2, PIWIL1, TDRD1, and BAG6, might be closely associated with the sterility of male cattle-yak.

**Key Words:** yak, hybrid sterility, proteomics

**W240 Effect of androstenone level on the boar testis transcriptome.** Dianelys Gonzalez-Pena\*, Robmay Garcia, and Robert V. Knox, and Sandra L. Rodriguez-Zas, *University of Illinois at Urbana-Champaign, Urbana, IL.*

Androstenone is the most abundant steroid produced in boar testis and share a precursor (pregnenolone) with testosterone, the principal male sex hormone needed for sperm production in the testis and its maturation on the epididymis. Androstenone is a pheromone of non-castrate males present in urine, sweat, saliva, and fatty tissues responsible for boar taint, which includes unpleasant odor and meat flavor. The objective of this study was to understand the differences in the transcriptome between high (greater than 1.0 µg/g) and low (less than 0.5 µg/g) androstenone boars. Testis mRNA from 10 Duroc crossbred boars was profiled. Single-end reads were mapped to the *Sus scrofa* reference genome (UCSC susScr3) using Tophat v2.0.12. In total, 4,969 transcripts from 4,744 genes were tested and 69 transcripts from 72 genes were differentially expressed between the high and low androstenone groups using Cufflink v2.2.1 (False Discovery Rate adjusted P-value <0.05). Among these, steroidogenic acute regulatory protein (STAR), cytochrome P450 family 19 subfamily A polypeptide 1 (CYP19A1), and sulfotransferase family cytosolic 2A dehydroepiandrosterone-preferring member 1 (SULT2A1) were overexpressed, while macrophage receptor with collagenous structure (MARCO) and anti-Mullerian hormone receptor type II (AMHR2) were underexpressed in the high relative to low androstenone groups. CYP19A1 encodes enzymes that participate in the synthesis of cholesterol, steroids, and other lipids and STAR encodes a protein critical in steroid hormone synthesis by enhancing the metabolism of cholesterol into pregnenolone the precursor of testosterone and androstenone. The overexpression of SULT2A1 in high relative to low androstenone groups is consistent with the role of this enzyme in the sulfo-conjugating of androstenone. Functional analysis of the differentially expressed genes using DAVID identified 2 category clusters (enrichment score >2) that include hormone metabolism biological processes and regulation, steroid and lipid biosynthetic and metabolism. These functional categories suggest that practices to reduce androstenone should aim to



minimize transcriptome changes that can negatively impact testosterone and boar fertility.

**Key Words:** transcriptome, androstenone, testis

**W241 Gonadal sperm characteristics of growing boars fed varying levels of raw or fermented cottonseed cake.** Mayowa Mojeed Oguntunde\* and Olajide Abraham Amao, *Ladoke Akintola University of Technology, Ogbomoso, Oyo, Nigeria.*

Cottonseed cake (CSC) is a veritable source of protein, energy and fiber for a variety of livestock species. However, it contains a deleterious polyphenolic compound, gossypol that restricts its use to ruminant feeding. Fermentation is one of the biotechnological options for reducing toxicity of many feed ingredients. This study was conducted to determine the effect of different levels of raw or fermented CSC on sperm characteristics of growing boars. Thirty (30) cross-bred (Landrace × Large White) with average weight of 7 kg, weanling boars (n = 6 per treatment) were randomly assigned to 5 dietary treatments arranged in a 2 × 2 factorial with a control within a completely randomized design (CRD). The CSC was included in diets at 0 (control), 10 and 20% (raw or fermented). Animals were fed for 12 weeks after which they were killed at average weight of 17.1 kg and the reproductive organs removed and processed for sperm characteristics. Sperm concentration was significantly ( $P < 0.05$ ) higher for the control ( $48.50 \times 10^6/\text{mL}$ ) than other treatments. Sperm motility for the raw CSC (68.55%) was similar to that of the control (68.01%). Boars that were fed 10% fermented CSC recorded lowest sperm motility (56.05%). Normal sperm was highest (71.34%) ( $P < 0.05$ ) for the control while the lowest value was obtained at 10% fermented CSC (52.76%). Gonadal sperm reserve (GSR) was the highest ( $P < 0.05$ ) for the control group ( $1.928 \times 10^9$ ), while the lowest value was observed at 20% fermented CSC ( $0.610 \times 10^9$ ). Boars fed raw CSC, irrespective of level, had higher GSR than those fed fermented CSC. Daily sperm production (DSP) was significantly ( $P < 0.05$ ) higher for the control than 10% and 20% fermented CSC groups. Daily sperm production/gram (DSP/g) was significantly ( $P < 0.05$ ) higher for 10% raw CSC ( $40.44 \times 10^6$ ) compared with other treatments. This study shows that growing boars could tolerate raw CSC up to 10% in diets on a short-term basis without adverse effects on the gonadal sperm characteristics.

**Key Words:** boar, cottonseed cake, fermentation and sperm characteristics

**W242 GameteGuard treatment improves post-thaw sperm quality and pregnancy per insemination in dairy cows.** Lisa Herickhoff\*<sup>1</sup>, Allison Lindsey<sup>1</sup>, Amanda Fritts<sup>1</sup>, and Patrick Burns<sup>2</sup>, <sup>1</sup>Membrane Protective Technologies Inc., Fort Collins, CO, <sup>2</sup>School of Biological Sciences, University of Northern Colorado, Greeley, CO.

GameteGuard is a blend of plant extracts containing antioxidants and membrane stabilizers. Previous studies have shown improvements in post-thaw sperm health with GameteGuard addition to the extender, and therefore it was hypothesized that the addition of GameteGuard to sperm extender would result in increased pregnancy rates per AI in lactating cows. Sperm from 2 ABS Global Inc. production Holstein bulls was extended in egg yolk citrate or egg yolk citrate plus GameteGuard to  $30 \times 10^6$  sperm/mK ( $15 \times 10^6$  sperm/dose) in a split ejaculate study. Sperm was packaged and frozen according to industry standards. The effectiveness of GameteGuard on pregnancy was assessed by first postpartum insemination of 208 Holstein cows. Over 6 mo, cows were cleared for

insemination by the farm veterinarian then 2 farm personnel determined estrus status using visual cues such as disturbed tail chalk. The cows were inseminated within 12 h by one trained farm employee. Cows were inseminated with either Bull 1 or 2, treated or control sperm, where treatment was blind to farm personnel. Bull and treatment combinations were dictated by a calendar day. For example, all cows to be bred on May 15 would use bull 1, red straw (control) while May 16 would use bull 1, blue straw (treated). Cows having postpartum problems and those with reproductive issues, such as ovarian cysts, were excluded from the study. Thirty-five to 45 d post-insemination pregnancy was assessed using transrectal ultrasonography and data were analyzed using Chi-squared. GameteGuard improved pregnancy per AI by 31% (from 38 to 50%;  $P < 0.05$ ; ~100 cows/treatment) across both bulls. Importantly, of the 124 animals bred within 83 d of calving, GameteGuard improved pregnancy per AI 36.1%.

**Key Words:** artificial insemination, sperm quality, pregnancy per insemination

**W243 Evaluation of factors affecting pregnancy rate to AI in pen-raised white-tailed deer.** Kyle J. Stutts\*, Jessica L. Leatherwood, Christopher R. Stewart, Mark J. Anderson, Marcy M. Beverly, and Stanley F. Kelley, *Sam Houston State University, Huntsville, TX.*

Factors known to influence pregnancy rate in livestock species were evaluated in white-tailed deer. Pen-raised white-tailed does (n = 53) ranging in age from 1.5 yr to 6.5 yr were utilized to evaluate the effects of BCS, BW, disposition, and serum cortisol concentration on fawning rate to AI. All does received a CIDR for 14 d and an injection (i.m.) of 300 IU PMSG at CIDR removal. Laparoscopic insemination was conducted at approximately 60 h following CIDR removal. Prior to insemination, does were restrained in a drop-floor chute and were administered sedative. At this time, BW was collected and disposition scores ranging from 1 to 5 (1 = docile and 5 = aggressive) were assigned by independent observers to evaluate deer behavior as an indicator of stress while restrained in the chute. Immediately following insemination, BCS was evaluated and blood samples were obtained via jugular venipuncture to determine cortisol concentration by RIA analysis. Previous research indicated a moderately positive relationship ( $r = 0.30$ ) between disposition score and serum cortisol concentration. Does were placed with cover bucks one week post AI. During fawning season, DNA samples were collected from all fawns to identify the sire of each fawn and determine fawning rate to AI. Data were analyzed using the LOGISTIC procedure of SAS. Pregnancy rate to AI was 67.9% and final pregnancy rate was 94.0% following natural service. Mean BCS was  $3.02 \pm 0.52$  (range of 2 to 4), and mean disposition score was  $2.16 \pm 1.21$  (range of 1 to 4). There was no significant effect of BCS ( $P = 0.59$ ), BW ( $P = 0.19$ ), cortisol concentration ( $P = 0.30$ ), or disposition score ( $P = 0.64$ ) on pregnancy rate to AI. Results of this study indicate that body condition and BW may not have as great of an influence on pregnancy rate in white-tailed deer as observed in livestock species. These data also indicate that the stress response to being restrained may not influence pregnancy rate to AI in white-tailed deer. Given the small sample size of this study, further research is needed to fully elucidate the influence of these factors on pregnancy rate to AI in white-tailed does.

**Key Words:** white-tailed deer, artificial insemination, disposition

**W244 Blood profiles of Ross 308 broiler and indigenous Venda chickens aged 42 and 90 days fed a similar diet.** Monnye Mabelebele\*, David Norris, Jones Ng'ambi, and John Alabi, *University of Limpopo, Polokwane, Limpopo, South Africa.*

A study was conducted to assess blood profiles of Ross 308 broiler and indigenous Venda chickens. A total of 120 Ross 308 broiler and Venda chickens were raised for a period of 90 d on a commercial grower diet containing 20% crude protein and 13 MJ ME/kg DM. Ten chickens per breed were randomly selected at 42 and 90 d of age and 3 mL of blood samples from both chicken breeds were collected from the jugular vein into 2 test tubes per bird to determine the blood profiles. A *t*-test was used to test the significance of differences between breeds ( $P < 0.05$ ). Differences among hematological parameters of Ross-308 and Venda chickens at 42 d of age were not significant ( $P > 0.05$ ) except for platelets ( $\times 10^3/\mu\text{L}$ ) for Ross 308 broiler ( $59.2 \pm 10.13$ ) and Venda chickens ( $24.0 \pm 10.00$ ); serum creatinine ( $\mu\text{mol/L}$ ) for Ross 308 broiler ( $17.5 \pm 50.05$ ) and Venda chickens ( $9.0 \pm 5.02$ ) and total protein (g/L)

of Venda ( $25.5 \pm 3.20$ ) and Ross 308 broiler chickens ( $20.0 \pm 3.12$ ). At 90 d of age, white blood cells ( $734.4 \pm 18.97 \times 10^3 \text{ cell}/\mu\text{L}$ ), red blood cells ( $2.3 \pm 0.15; \times 10^6 \text{ cell}/\mu\text{L}$ ), mean corpuscular volume ( $121.1 \pm 4.10 \text{ fL}$ ), serum calcium ( $2.6 \pm 0.01 \text{ mmol/L}$ ), creatinine ( $10.7 \pm 0.76 \mu\text{mol/L}$ ), cholesterol ( $3.9 \pm 0.12 \text{ mmol/L}$ ) and albumin ( $9.5 \pm 0.23 \text{ g/L}$ ) were higher for Ross 308 broiler chickens. However, platelets ( $\times 10^3/\mu\text{L}$ ) were higher in Venda ( $14.4 \pm 3.29$ ) than in Ross 308 broiler ( $12.2 \pm 3.00$ ) chickens. In the adult stage, broiler chickens have better hematological properties than indigenous chickens. Further investigation on immunological parameters of these breeds needs to be conducted.

**Key Words:** hematology, chicken, disease

# Physiology and Endocrinology: Metabolism, health, and physiological processes

**W245 Transition period concentrations of nonesterified fatty acids and  $\beta$ -hydroxybutyrate in dairy cows are not well correlated.** Maris M. McCarthy<sup>\*1</sup>, Sabine Mann<sup>2</sup>, Daryl V. Nydam<sup>2</sup>, Thomas R. Overton<sup>1</sup>, and Jessica A. A. McArt<sup>2</sup>, <sup>1</sup>*Department of Animal Science, Cornell University, Ithaca, NY*, <sup>2</sup>*Department of Population Medicine and Diagnostic Sciences, Cornell University, Ithaca, NY*.

The objective was to use longitudinal data of blood nonesterified fatty acids (NEFA) and  $\beta$ -hydroxybutyrate (BHBA) concentrations to describe the relationship between NEFA and BHBA in dairy cows during the periparturient period. Blood NEFA and BHBA concentration data collected from d -21 prepartum to 21 postpartum for 269 multiparous Holstein cows were recruited from 4 different studies carried out within our research groups. Of the 269 cows enrolled in the data set, 117 cows (43.5%) had at least 1 postpartum hyperketonemic event (BHBA  $\geq$  1.2 mmol/L), and 202 cows (75.1%) had at least 1 event of elevated postpartum NEFA ( $\geq$ 700  $\mu$ Eq/L) between 3 and 21 d in milk. Area under the curve (AUC) was used to investigate relationships between metabolites over time. Overall, Pearson correlation relationships between transition period NEFA and BHBA AUC were poor. There was a negative correlation between postpartum NEFA AUC and prepartum BHBA AUC, although the correlation coefficient was low ( $r = -0.26$ ). A positive correlation existed between postpartum NEFA AUC and postpartum BHBA AUC; however, the correlation coefficient was low ( $r = 0.26$ ), reinforcing a poor relationship between these metabolites during the periparturient period. Large variation was found between the day of maximum NEFA concentration within the first 21 d in milk and day of maximum BHBA concentration for the same time period. The mean and median days of maximum NEFA concentration were 6.8 and 6 d, respectively, whereas the mean and median days of maximum BHBA concentration were 9.6 and 8 d, respectively; however the range in days for both the mean and median day of maximum concentrations was very large. Overall, our data set indicates a poor relationship between blood concentrations of NEFA and BHBA during the periparturient period of dairy cows, suggesting that elevated concentrations of one should not be extrapolated to suggest elevated concentrations of the other metabolite.

**Key Words:** transition period, nonesterified fatty acids,  $\beta$ -hydroxybutyrate

**W246 Associations of circulating haptoglobin with performance and metabolism in dairy cows during early lactation.** Maris M. McCarthy<sup>\*</sup>, Takashi Yasui, and Thomas R. Overton, *Department of Animal Science, Cornell University, Ithaca, NY*.

The objective of the current study was to clarify associations between the severity of systemic inflammation during the early postpartum period with performance and energy metabolism. Cows were assigned to categorical quartiles (Q; Q1 = 0.18–0.59, Q2 = 0.60–1.14, Q3 = 1.15–2.05, and Q4 = 2.06–2.50 g haptoglobin/L) based on the highest plasma haptoglobin concentration measured during wk 1 postpartum. Linear and quadratic contrasts were tested for all measurements, and data analyzed over time were subjected to repeated-measures ANOVA using PROC MIXED of SAS (v 9.3) and the REPEATED statement. There was a quadratic relationship ( $P = 0.01$ ) on prepartum net energy for lactation (NE<sub>L</sub>) intake such that cows in Q3 had lower prepartum NE<sub>L</sub> intake compared with cows in the other Q (Q1 = 21.1, Q2 = 19.2,

Q3 = 16.6, Q4 = 19.6 Mcal/d). There was a quadratic relationship ( $P = 0.02$ ) on postpartum NE<sub>L</sub> intake such that cows in Q3 also had lower postpartum NE<sub>L</sub> intakes compared with cows in the other Q (Q1 = 36.3, Q2 = 34.6, Q3 = 29.8, Q4 = 33.7 Mcal/d). There was also a quadratic relationship ( $P = 0.05$ ) with postpartum milk yield (Q1 = 42.6, Q2 = 40.1, Q3 = 34.2, Q4 = 37.4 kg/d), with similar relationships with 3.5% fat-corrected milk ( $P = 0.02$ ) and energy-corrected milk ( $P = 0.03$ ). There was a linear tendency ( $P = 0.06$ ) for cows with increasing inflammation to have higher plasma glucose concentrations compared with cows with low inflammation. There was a similar linear relationship ( $P = 0.04$ ) for plasma insulin and cows in Q3 and Q4 had higher postpartum insulin compared with cows in Q1 and Q2. There was a linear relationship ( $P = 0.04$ ) for plasma NEFA and cows with increasing inflammation had lower NEFA (Q1 = 498, Q2 = 456, Q3 = 430, Q4 = 407  $\mu$ Eq/L). There was a quadratic relationship of inflammation ( $P = 0.02$ ) with liver triglyceride content at d 7 postpartum and cows in Q3 had the lowest triglyceride content. There was also a quadratic relationship ( $P = 0.02$ ) for liver glycogen and cows in Q3 had elevated liver glycogen content on d 7 postpartum compared with cows in other Q. Overall, cows with elevated haptoglobin in the first week after calving had reduced milk yields and alterations in metabolism; however, cows that have high early lactation haptoglobin ( $>1.14$  g/L) had a diverse range of production responses and further investigation is warranted.

**Key Words:** transition period, haptoglobin, metabolism

**W247 Intestinal permeability and incidence of diarrhea in Holstein calves.** Gemma Araujo<sup>1</sup>, Cristina Yunta<sup>1</sup>, Marta Terré<sup>1</sup>, Alessandro Mereu<sup>2</sup>, Ignacio Ipharraguerre<sup>2</sup>, and Alex Bach<sup>\*3,1</sup>, <sup>1</sup>*Department of Ruminant Production, IRTA (Institut de Recerca i Tecnologia Agroalimentàries), Caldes de Montbui, Spain*, <sup>2</sup>*Lucta S.A., Montornès del Vallès, Spain*, <sup>3</sup>*ICREA (Institut Catalana de Recerca i Estudis Avançats), Barcelona, Spain*.

Seventy-six newborn Holstein calves (44.4  $\pm$  6.15 kg BW) were involved in this study from birth until 21 d of age. Within 2 h after birth, calves received 4 L of maternal colostrum via an esophageal tube. The following 3 meals consisted of 2 L of late colostrum (or transition milk). After that, calves were fed 1.5 L of milk replacer (22.9% CP, 20.1% fat) twice daily. Calves were considered diarrheic when showed fecal scores  $\geq$ 3 for 3 consecutive days. Then, data from a random subset of 30 calves (45.9  $\pm$  5.47 kg BW), 15 that never had diarrhea and 15 that had diarrhea were used to assess potential associations between intestinal permeability and incidence of diarrhea. On 0, 7, 14 and 21 d of life, intestinal permeability of calves was measured by dosing 2 markers (lactulose and D-mannitol) and assessing their concentration in serum by ultra-high performance liquid chromatography-mass spectrometry. Plasma IgG was measured at birth and at 6 and 24 h after first colostrum intake and efficiency of IgG absorption calculated. Plasma and colostrum IgG contents were determined by radioimmunoassay and bacterial load in colostrum samples by colony counting. Data were analyzed with a mixed-effects model for repeated measures. All diarrhea incidences occurred between 7 to 14 d of study. Overall colostrum quality was good, with an IgG content  $>50$  mg/mL but total bacterial load was high ( $>100,000$  cfu/mL). However, there were no differences in these 2 parameters between colostrums consumed by calves that did and those that did not incur diarrhea. Also, efficiency of IgG absorption was similar for all calves ( $\sim$ 16%).



However, intestinal permeability was increased in diarrheic compared with healthy calves. Diarrheic calves had greater ( $P < 0.01$ ) lactulose serum concentrations ( $15.3 \pm 0.37 \mu\text{g/mL}$ ) than healthy calves ( $8.4 \pm 0.37 \mu\text{g/mL}$ ). Furthermore, diarrheic calves tended ( $P = 0.06$ ) to have a greater lactulose to D-mannitol ratio ( $1.26 \pm 0.16$ ) since birth until 21 d of life than healthy calves ( $0.81 \pm 0.16$ ). In conclusion, calves correctly immunized that develop diarrhea may be predisposed to suffer scours due to altered intestinal permeability right from birth.

**Key Words:** colostrum, intestinal integrity, scours

**W248 Effects of realimentation on umbilical blood flow, fetal and placental measurements, and birth weight in nutrient-restricted pregnant ewes.** Manuel Vasquez\*, Kendall Swanson, and Kimberley Vonnahme, *North Dakota State University, Fargo, ND.*

Nutritional restriction (60% of total nutritional requirement) from d 50 to 130 applied in nulliparous ewes has shown to reduce umbilical blood flow (UBF; Lemley et al., 2012; AJP 302:R454-R467). We hypothesized that during restriction, UBF and fetal and placental measurements would be less than in adequately fed ewes, but upon realimentation, ewes would have similar UBF as ewes that were never restricted. Second parity Dorset ewes were assigned either to an adequate nutrition group (CON,  $n = 7$ ) or a restricted (60% of CON) group (RES,  $n = 8$ ), from d 50 to 90 of gestation. On d 90, all ewes were fed 100% of nutritional requirements according to body weight. Ewe body weight and conceptus measurements via ultrasonography were recorded every 10 d from d 50 to 130 of gestation. Every 10 d, length and width from 10 random placentomes were averaged and then multiplied to obtain placental area. Fetal biparietal and abdominal lengths were recorded. Doppler mode was used to obtain UBF, pulsatility index (PI), and resistance index (RI). At birth, lambs and placental measurements were obtained. The data were analyzed using the Proc Mixed procedure of SAS. Treatment and day were treated as fixed effects, ewe as random. By d 70, RES ewes were lighter ( $P < 0.01$ ), and remained lighter than CON ewes throughout the experiment. While there were no treatment by day interactions or main effects of treatment ( $P > 0.13$ ) for any measurements obtained by ultrasonography, there were some interesting observations. On d 80, UBF and placental area were decreased, and PI, RI, and biparietal distance increased in RES vs CON ewes ( $P \leq 0.05$ ; means separation of unprotected F test). On d 90, before the realimentation, all ultrasound measurements were similar. After realimentation, there was no effect of treatment on any of the ultrasound measurements. At birth, lambs and placental measurements were similar ( $P > 0.43$ ). Perhaps the increased resistance indices and smaller placental size on d 80 were a trigger to the dam to enhance UBF to the growing fetus. Further studies are needed to determine the impact of maternal age and parity in the face of nutrient restriction on UBF.

**Key Words:** ewe, pregnancy, blood flow

**W249 Changes in insulin-like growth factor I and II profiles following anti-bPL antibodies infusions in six long-term-cannulated bovine fetuses at late gestation.** Andrea Alvarez-Oxiley\*<sup>1</sup>, Noelita Melo de Sousa<sup>2</sup>, Jean L. Hornick<sup>2</sup>, Kamal Touati<sup>2</sup>, and Jean F. Beckers<sup>2</sup>, <sup>1</sup>Facultad de Agronomía, Universidad de la República, Montevideo, Montevideo, Uruguay, <sup>2</sup>Faculty of Veterinary Medicine, University of Liege, Liege, Liege, Belgium.

Studies were conducted to evaluate the changes in plasma concentrations of bovine fetal placental lactogen (bPL), insulin-like growth factor (IGF-I) and IGF-II following anti-bPL antibody infusion in fetal

circulation. Abdominal aorta of bovine fetuses ( $n = 6$ ) were chronically cannulated on the medial tarsal artery and infusion of rabbit anti-bPL IgG was performed during late gestation. From the first infusion, blood samples were collected from the fetus on a daily basis. Fetuses remained cannulated during the last 10 to 95 d of gestation. The time-series of hormonal data (bPL, IGF-I and IGF-II) were statistically analyzed for repeated measures as a split plot in time with number of anti-bPL infusions (one vs. more than one), dose of anti-bPL infused L (low, 4, 6, 8 mL) vs. H (high, 20 mL) and days after infusions (d 1 to d 7) as the fixed effects using the Mixed procedure of SAS (1998). IGF-I concentrations tended to reach a minimal concentration on d 3 after infusions ( $15.5 \pm 3.4 \text{ ng/mL}$ ;  $P = 0.056$ ). No changes were observed on IGF-II concentrations. The mean values of bPL on d 3 after infusion showed a significant increase. When low doses of anti bPL IgG were administered, no peak of binding in fetal serum was observed. When higher or repeated doses of anti bPL IgG were injected, a significant binding rate of the tracer (B/T) was recorded in the fetal serum during 1, 2 or several days. This high B/T can be due to circulating free anti bPL IgG; these rabbit IgG can remain in solution during the step of separation of free and bound fraction of the RIA and thus can bind part of the tracer and create an artifactual overestimation of the circulating bPL. However generally, concentrations of circulating bPL after anti bPL IgG injection were decreased justifying a transitory decrease in IGF-I concentrations. These data showed that following anti-bPL infusion, a transient decrease in IGF-I but not in IGF-II is observed. Our work suggests that PL somatogenic activities at the maternal-fetal interface are mediated through the IGF-I rather than IGF-II.

**Key Words:** IGF, bovine placental lactogen, fetus cannulation

**W250 Effects of different feeding intensities during the first weeks of rearing on the metabolic status and on the circulating concentrations of adiponectin in dairy calves until 110 days of age.** Julia Kesser<sup>1</sup>, Miriam Hill<sup>1,2</sup>, Christian Koch<sup>2</sup>, Marion Piechotta<sup>3</sup>, Jürgen Rehage<sup>3</sup>, Klaus Eder<sup>4</sup>, Hassan Sadri<sup>1</sup>, Ute Müller<sup>1</sup>, and Helga Sauerwein\*<sup>1</sup>, <sup>1</sup>Physiology and Hygiene Group, Institute for Animal Science, University of Bonn, Bonn, Germany, <sup>2</sup>Lehr- und Versuchsanstalt Neumühle, Münchweiler an der Alsenz, Germany, <sup>3</sup>Clinic for Cattle, University of Veterinary Medicine Hannover, Hannover, Germany, <sup>4</sup>Animal Nutrition, University of Giessen, Gießen, Germany.

Dairy calves are commonly reared at restrictive levels of feeding, but greater allowances of feed intake may yield beneficial effects for both animal welfare and later milk production. We aimed to test the hypothesis that different feeding levels in early life will continue to affect the circulating concentrations of metabolically relevant hormones beyond the time of differential feeding. After receiving colostrum for the first 3 d of life, 57 German Holstein calves were randomly allocated to 3 groups fed either restrictively (r) with milk replacer (MR) (group MRr: 130 g MR/L, 6 L/d,  $n = 20$ ), or MR ad libitum (al) (MRal: 160 g MR/L,  $n = 17$ ) or whole milk (MI) al (MIal,  $n = 20$ ). All calves received colostrum from their dams for the first 3 d of life, and were then fed according to their group regimen from d 1 – 27 (phase (P) 1). Thereafter all calves were fed according to the MRr plan and were gradually weaned from d 56 – 70 (P2). Calves were further observed for the subsequent P3, i.e., d 71 – 109. All calves had free access to hay, water and concentrate and received a TMR in P3. Blood samples were collected on d 0 (before colostrum feeding), and on 10 other d covering P1-P3 to assess the concentrations of glucose, NEFA, leptin and adiponectin (bovine specific ELISA). Data were analyzed using the linear mixed model (SPSS). Differences ( $P < 0.05$ ) between the groups were largely limited to the

time of differential feeding (P1): glucose was greater in Mlal than in MR fed groups (glucose: 1.1-fold), insulin was greater in MRal and Mlal than in MRr (2.3-fold), whereas NEFA were lower in MRal than in MRr (0.65-fold). Leptin was not different between the groups at any time. Adiponectin concentrations did not differ between groups in P1, but tended ( $P < 0.1$ ) to lower values in MRr as compared with MRal and Mlal in P2. In P3 the MRal group had higher values ( $P < 0.05$ ) than MRr and Mlal, thus supporting continued effects of the differential feeding. In view of the insulin-sensitizing effects of adiponectin, feeding intensity in early life might thus affect insulin sensitivity at older ages.

**Key Words:** adiponectin, dairy calves, insulin sensitivity

**W251 Mitochondrial DNA copy numbers in blood cells during early and late lactation in dairy cows.** Lilian Laubenthal, Michael Hölker, Karl-Heinz Südekum, Helga Sauerwein, and Susanne Häussler\*, *University of Bonn, Institute of Animal Science, Bonn, Germany.*

During the transition period most high-yielding dairy cows suffer from negative energy balance due to decreased energy intake and increased energy demands required for milk synthesis. Mitochondria are the main sites of energy metabolism in mammalian cells and their number varies depending on age, sex, organ, and physiological or pathological conditions. Mitochondria exhibit their own genome, the mitochondrial DNA (mtDNA), and its copy numbers reflect the abundance of mitochondria within a cell. In the course of lactation, environmental, physiological, and energetic conditions alter. We hypothesized that these changes may influence the number of mtDNA/cell in dairy cows and thus investigated the number of mtDNA copies in blood during early and late lactation. German Holstein cows ( $n = 21$ ; BCS:  $3.0 \pm 0.1$ ) were fed according to their requirements. Estimated total energy requirements were calculated by adding the requirements for maintenance and milk production. Blood samples from the jugular vein were collected 3 and 35 wk postpartum. Genomic DNA was extracted from whole blood using a commercially available kit. Based on the amplification of the 12S rRNA (mtDNA target gene) and the  $\beta$ -globin (reference gene) to normalize the DNA content in each sample, mtDNA copy number was assessed by a multiplex qPCR. Data (mean  $\pm$  SEM) were analyzed by the pairwise Student's *t*-test (SPSS 22). In early lactation the number of mtDNA ( $87.4 \pm 15.5$  copies/cell) was about twice as much as in late lactation ( $46.0 \pm 3.7$ ;  $P = 0.008$ ). In early lactation energy demands are increased compared with late lactation, as indicated by the estimated total energy requirements for wk 3 ( $132 \pm 8.26$  MJ NE<sub>L</sub>) and wk 35 ( $103 \pm 5.77$  MJ NE<sub>L</sub>). The greater energy demands in early lactation were accompanied by elevated mtDNA copy numbers in peripheral blood when compared with late lactation. The observed changes meet the expectation that the metabolic load during early lactation requires more mitochondria. Peripheral blood thus forms a suitable matrix to assess the cellular content of mitochondria and the cellular energetic status of dairy cows when tissues are not accessible.

**Key Words:** mtDNA copy number, dairy cow, lactation

**W252 Mitochondrial DNA copy number in liver, mammary gland, and adipose tissue of early lactating dairy cows.** Lilian Laubenthal, Michael Hölker, Karl-Heinz Südekum, Helga Sauerwein, and Susanne Häussler\*, *University of Bonn, Institute of Animal Science, Bonn, Germany.*

With the onset of lactation, energy requirements rapidly increase in high-yielding dairy cows. To adapt to lactation, energy metabolism

needs to be regulated and coordinated among the key organs, namely adipose tissue (AT), liver, and mammary gland. Mitochondria are the main site for energy production in mammalian cells and their number depends on the energy demand and physiological state of each individual. Mitochondria have their own DNA and therefore the abundance of mitochondria in a cell is reflected by the copy number of mitochondrial DNA (mtDNA). Age-related differences of mtDNA are known for mice and humans, in which mtDNA varies between different tissues. However, little is known about mtDNA copy numbers in dairy cows; we thus aimed to provide an overview of mtDNA copy numbers in liver, subcutaneous (sc) AT and mammary gland of lactating dairy cows. Lactating German Holstein cows ( $n = 21$ ; BCS:  $3.0 \pm 0.1$ ) were fed according to their requirements. Liver, mammary gland and subcutaneous (sc) AT from the tailhead region were sampled during early lactation (3 wks postpartum), in which the estimated total energy requirement was  $132 \pm 8.26$  MJ NE<sub>L</sub>. Biopsies were immediately snap frozen after sampling. Genomic DNA was extracted using commercially available kits and the number of mtDNA copies/cell was quantified by a multiplex qPCR, targeting the 12S rRNA gene and using  $\beta$ -globin as reference gene. Tissue-specific differences were examined by Student's *t*-test (SPSS 22). Data are presented as means  $\pm$  SEM. The number of mtDNA copies/cell in liver ( $360 \pm 22.3$  copies/cell) was 7.7-fold and 5.3-fold higher ( $P < 0.001$ ) than in scAT ( $46.7 \pm 2.32$  copies/cell) and mammary gland ( $68.3 \pm 4.66$ ), respectively. Moreover, mammary gland contained 1.5-fold more mtDNA copies/cell than scAT ( $P = 0.001$ ). The differences in mtDNA content observed between the organs investigated herein are presumably reflecting their metabolic activity during the first weeks of lactation with liver playing a key role.

**Key Words:** mtDNA copy number, dairy cow, liver

**W253 Lipolysis induces adipose tissue macrophage infiltration in lactating dairy cows.** G. Andres Contreras\*<sup>1</sup>, Kyan Thelen<sup>2</sup>, Courtney L. Preseault<sup>2</sup>, Sarah E. Schmidt<sup>2</sup>, and Adam L. Lock<sup>2</sup>, <sup>1</sup>*Department of Large Animal Clinical Sciences, Michigan State University, East Lansing, MI*, <sup>2</sup>*Department of Animal Science, Michigan State University, East Lansing, MI.*

Excessive rates of lipolysis during periods of negative energy balance (NEB) are associated with increased susceptibility to disease. Lipolysis increases adipose tissue macrophage (ATM) populations. Depending on their phenotype, ATMs modify inflammatory processes and alter adipose metabolic functions. Classically activated ATMs (M1) are pro-inflammatory while alternatively activated ATMs (M2) promote inflammation resolution. The objective of this study was to evaluate changes in ATM trafficking and phenotype in healthy cows during feed restriction-induced NEB. Lactating multiparous dairy cows (DIM 119–210) were fed a common diet to meet nutrient requirements during a 14d preliminary period (d1 to 14) and then randomly assigned to one of 2 feeding protocols: ad libitum (AL;  $n = 6$ ) or feed-restricted (FR;  $n = 7$ ). Caloric intake was reduced in FR cows for 4 d (d15 to 18) to achieve a targeted NEB of  $-15$  Mcal/d. Omental and subcutaneous adipose tissue samples were collected to harvest stromal vascular cells (SVC) on d11 and d18. Data were analyzed in a mixed model with treatment and day as fixed effects and cow as a random effect. FR cows reached a NEB of  $-13.5 \pm 1.9$  Mcal/d inducing a lipolytic response (NEFA on d18: FR =  $0.52$  mEq/L; AL =  $0.17$  mEq/L;  $P < 0.01$ ), while AL animals remained in positive energy balance ( $3.2 \pm 2.2$  Mcal/d). Flow cytometry analysis revealed that at d18, FR increased the infiltration ratio of CD68<sup>+</sup>, a specific ATM surface marker, in omental SVC (FR =  $2.85 \pm 0.39$ ; AL =  $1.25 \pm 0.35$ ;  $P = 0.01$ ), while the expression ratio of CD14, an M1 marker, remained unaltered ( $P = 0.86$ ). Adipose tissue from FR cows

exhibited an increased expression of the macrophage-related gene SIRPA ( $P=0.01$ ), but no change in M1 genes CCL2 and TNF $\alpha$  (both  $P>0.49$ ). Additionally, compared with AL, FR upregulated the expression of M2 specific genes IL10 and ARG1 (both  $P<0.01$ ). This finding contrasts with the predominately M1 phenotype observed previously in ATMs from clinically diseased cows. These results provide evidence for an active role of ATMs during NEB in ruminants and emphasize changes in their inflammatory phenotype during lipolytic periods.

**Key Words:** lipolysis, adipose tissue macrophages, negative energy balance

**W254 Longitudinal characterization of the gene expression of key components of the mTOR signaling and ubiquitin proteasome system in skeletal muscle of dairy cows during the periparturient period and subsequent lactation.** Yi Yang<sup>1</sup>, Helga Sauerwein<sup>\*1</sup>, Sven Dänicke<sup>2</sup>, Jürgen Rehage<sup>3</sup>, and Hassan Sadri<sup>1</sup>, <sup>1</sup>Institute of Animal Science, Physiology and Hygiene Group, University of Bonn, Bonn, North Rhine-Westphalia, Germany, <sup>2</sup>Institute of Animal Nutrition, Friedrich-Loeffler-Institute (FLI), Braunschweig, Lower Saxony, Germany, <sup>3</sup>Clinic for Cattle, University for Veterinary Medicine, Foundation, Hannover, Lower Saxony, Germany.

At the onset of lactation when voluntary feed intake is insufficient to cover the total needs for maintenance and lactation, dairy cows need to mobilize body reserves. Besides fat, skeletal muscle protein is also degraded. Muscle is the main labile source of amino acids (AA) and may thus partition AA to protein syntheses in other organs, mainly the mammary gland, for gluconeogenesis and for generating ATP. The mammalian target of rapamycin (mTOR) and ubiquitin-proteasome system (UPS) are considered as the major regulators of protein synthesis and protein degradation, respectively. We hypothesized that the transcript abundance of key components of mTOR signaling and of 2 major muscle-specific E3 ubiquitin ligases, MuRF1 (muscle RING-finger protein-1) and atrogin-1 in skeletal muscle will change throughout late pregnancy and the subsequent lactation period. From 14 German Holstein cows, muscle tissue (*M. semitendinosus*) were obtained for biopsy on d -21, 1, 21, 70, 105, 182, 196, 224, and 252 relative to calving. The target mRNAs were quantified by qPCR. Data were analyzed by the MIXED procedure of SAS and results are reported as LSM with P-values adjusted by the method of Tukey-Kramer. The mRNA abundance of mTOR increased ( $P<0.05$ ) from d -21 to d 1, followed by a decline toward pre-partum values by d 180 and then increased thereafter. A 6-fold increase in eukaryotic initiation factor 4E-binding protein mRNA was observed from d -21 to d 1, and then a gradual decrease until d 105 with relatively stable values thereafter. The encoding ribosomal protein S6 kinase mRNA decreased ( $P<0.0001$ ) during lactation. The abundance of MuRF1 mRNA increased (3.6-fold) from d -21 to d 1, declined to nearly pre-partum values by d 105 and then remained unchanged. The mRNA abundance of atrogin-1 followed almost a similar trend as that of MuRF1; a 2.2 fold increase was noted from d -21 to d 1, and then a decrease until d 21 and unchanged values thereafter. In conclusion, our data show that key components of mTOR and UPS are upregulated at the level of the mRNA on d 1, suggesting a greater rate of protein turnover in muscle around calving.

**Key Words:** skeletal muscle, mTOR, ubiquitin-proteasome system

**W255 Comparison of fractional gluconeogenesis estimates in sheep determined with D<sub>2</sub>O administered via vein or rumen and by intravenous infusion of <sup>13</sup>C<sub>6</sub>-glucose.** Cornelia C. Metges<sup>\*1</sup>, Solvig Görö<sup>1</sup>, Gürbüz Das<sup>1</sup>, Umang Agarwal<sup>2</sup>, and Brian J. Bequette<sup>2</sup>,

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In ruminants, the main precursor for GNG is ruminal propionate which fundamentally differs from gluconeogenic precursors of monogastrics. The D<sub>2</sub>O method has been used to measure fractional gluconeogenesis (GNG) in humans. We aimed to determine whether the route of D<sub>2</sub>O administration (rumen (IR) or jugular vein (IV)) affects D enrichment of rumen fluid, ruminal propionate and estimates of fractional GNG. In addition, we compared these GNG estimates with estimates derived from <sup>13</sup>C<sub>6</sub>-glucose infusion. Four sheep (23.5 ± 1 kg BW), equipped with a rumen fistula and a jugular vein catheter, were fed a pelleted ration (35 g/kg BW and d; 9 MJ ME/d) at 2-h intervals. Water was offered ad lib. Sheep were given 2 boli of 7 g D<sub>2</sub>O (99.2 atom% (AP) D/kg BW) at 0800 and 1200 h either IR (method 1) or IV (method 2), or received continuous IV infusion of <sup>13</sup>C<sub>6</sub>-glucose (0.15 g/h) for 10 h (method 3) in a crossover design with 1 wk separating tracer administrations. Ruminant D enrichments were measured by isotope ratio mass spectrometry whereas ruminal propionate and plasma glucose D enrichment was measured by GC-MS. Fractional GNG was calculated from the ratio of D enrichment at C-5 of glucose (labeling via GNG only) to that at C-2 (labeling during GNG and glycogenolysis). The <sup>13</sup>C-mass isotopomer enrichment of plasma glucose was determined by GC-MS and GNG was calculated. Statistical comparison of GNG estimates was made with repeated measures ANOVA using PROC MIXED of SAS. Rumen fluid D enrichment attained a plateau 6 h after the first bolus (IR: 1.51; IV: 1.43 APE;  $P>0.1$ ). The D enrichment of ruminal propionate as a precursor for GNG showed faster labeling with the IR route (3 h:  $P<0.10$ ; 6 h:  $P<0.05$ ). However, the plateau enrichments of rumen fluid D and propionate D did not differ ( $P>0.10$ ). GNG estimates derived from IR and IV routes of D<sub>2</sub>O administration did not differ ( $P=0.83$ ) which resulted in an overall GNG estimate of 58.8%. In contrast, GNG estimate derived from <sup>13</sup>C<sub>6</sub> glucose dilution was at 72.8%, which did not differ from the other 2 methods ( $P>0.13$ ). Thus all 3 methods yield similar estimates of GNG in ruminants.

**Key Words:** ruminants, gluconeogenesis, stable isotope-labeled tracer

**W256 Propionate and cyclic AMP induced bovine PCK1 gene transcription is concurrently mediated by CRE and HNF4 $\alpha$  binding elements.** Qian Zhang, Stephanie L. Koser, and Shawn S. Donkin<sup>\*</sup>, Purdue University, West Lafayette, IN.

Cytosolic phosphoenolpyruvate carboxykinase (PCK1), a key gluconeogenic enzyme, is controlled at the transcriptional level. Our objective was to determine regulatory elements within the bovine PCK1 promoter that control transcription in response to cyclic AMP (cAMP), glucocorticoids, and propionate (PROP). Putative DNA promoter transcription factor protein binding sequences were identified for cAMP response element (CRE) at -94 to -87 and for Hepatic Nuclear Factor 4 $\alpha$  (HNF4 $\alpha$ ) at +68 to +72 (HNF4 $\alpha$ 1) and -1078 to -1074 (HNF4 $\alpha$ 2). To test control of transcription, the wild-type (WT) bovine PCK1 promoter (-1238 to +221) was ligated to a luciferase reporter gene and transfected into H4IIE cells followed by incubation with 2.5 mM PROP, 1 mM cAMP (cAMP), 5  $\mu$ M dexamethasone (DEX) or their combinations. The functionality of CRE, HNF4 $\alpha$ 1, and HNF4 $\alpha$ 2 cis-regulatory elements was determined using deletion mutations of the core transcription factor binding regions within the PCK1 promoter DNA. The deletion mutations tested were HNF4 $\alpha$ 1<sup>-</sup>; HNF4 $\alpha$ 2<sup>-</sup>; CRE<sup>-</sup>; HNF4 $\alpha$ 1<sup>-</sup>/HNF4 $\alpha$ 2<sup>-</sup>; CRE<sup>-</sup>/HNF4 $\alpha$ 1<sup>-</sup>; CRE<sup>-</sup>/HNF4 $\alpha$ 2<sup>-</sup>; and CRE<sup>-</sup>/HNF4 $\alpha$ 1<sup>-</sup>/HNF4 $\alpha$ 2<sup>-</sup>. H4IIE cells were transfected with the promoter-reporter constructs and exposed to treat-



ments for 23 h. Luciferase activity was measured in the cell lysates as a direct proxy for bovine PCK1 promoter activity. Within each construct, treatment effect was expressed as the fold change of luciferase activity relative to base media control (n = 3 cell preparations). Analyses of variance of the data were performed using the Proc Mixed procedure of SAS 9.3. Exposure to cAMP, DEX, cAMP+DEX, PROP, cAMP+PROP, cAMP+DEX+PROP induced ( $P < 0.05$ ) expression of the WT promoter relative to no addition controls by 2.0, 2.3, 3.9, 6.0, 7.3,  $14.4 \pm 1.4$  x respectively. A similar pattern was observed for each single mutant bovine PCK1 promoter. Responses to cAMP, DEX, PROP and their combinations were abolished for mutations lacking both HNF4 $\alpha$ 1 and CRE binding sites indicating that these elements act synergistically to control bovine PCK1 transcription.

**Key Words:** promoter, transcription factor binding site

### **W257 Hepatic mRNA expression of genes related to somatotropic axis and metabolism of dairy cows treated with recombinant bovine somatotropin during the periparturient period.**

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Objectives were to determine the effects recombinant bovine somatotropin (rbST) treatment during the peripartum period on hepatic mRNA expression of genes related to inflammation and immune response. Holstein cows were assigned randomly to receive no treatment (control; n = 10), 87.5 mg (rbST87.5; n = 12), or 125 mg (rbST125; n = 10) of rbST every 7 d from -21 to 21 d relative to calving. Liver biopsies were collected -21, -7, and 7 d relative to calving. Twenty 4 genes were assessed by direct molecular counts using NanoString technology. Continuous data were analyzed by ANOVA. Gene expression on d -21 was used as a covariate for analyses of mRNA expression on d -7. No differences in mRNA expression were observed among treatments on d -21 for all the genes except SOCS3, which had lower ( $P \leq 0.05$ ) mRNA expression in control cows compared with rbST87.5 cows. On d -7, expression of mRNA for ANGPTL4 and SCARB1 was higher ( $P \leq 0.05$ ) in rbST87.5 and rbST125 cows than control. Cows in the rbST87.5 treatment had ( $P \leq 0.05$ ) higher mRNA expression for HP, ICAM1, SOCS2 and XBP1 on d -7 than control cows. Control cows had ( $P \leq 0.05$ ) higher mRNA expression for HIF1A than rbST125 cows on d -7. On d 7, control cows had ( $P \leq 0.05$ ) higher mRNA expression for CXCL1, IL1RN, MYD88, NFKBIA, and SOCS3 compared with rbST87.5 and rbST125 cows. Control cows had ( $P \leq 0.05$ ) higher mRNA expression for ICAM1 and XBP1 than rbST125 cows and had higher mRNA expression for HIF1A than rbST87.5 cows. On the other hand, expression of mRNA for NR3C1 and SOCS2 was ( $P \leq 0.05$ ) lower in control cows than rbST125 and rbST87.5 cows, respectively. Treatment did not affect hepatic expression of the genes CEBPD, JUN, M-CSF1, NFKB1, PPARGC1A, STAT5B, TLR2, TNF, TNFRSF1 and TNFRSF5. The gene G-CSF was not detected. Weekly treatment of periparturient cows with rbST regulates liver mRNA expression of genes related to inflammation and immune response during the prepartum and postpartum periods. Increased postpartum mRNA expression of inflammatory and immune responses genes in control cows might be a consequence of increased incidence of postpartum diseases.

**Key Words:** periparturient cow, recombinant bovine somatotropin, hepatic gene expression

### **W258 A direct method is not as effective as an indirect method for determination of fatty acids from bovine placental tissue.**

Patricia A. Dutra<sup>1,2</sup>, Mohanathas Gobikrushanth\*<sup>2</sup>, Reza Salehi<sup>2</sup>, Ana Ruiz-Sanchez<sup>2</sup>, Marcos G. Colazo<sup>3</sup>, and Divakar J. Ambrose<sup>3,2</sup>, <sup>1</sup>Departamento de Zootecnia, Universidade Federal da Bahia, Salvador, Bahia, Brazil, <sup>2</sup>Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, Alberta, Canada, <sup>3</sup>Live-stock Research Branch, Alberta Agriculture and Rural Development, Edmonton, Alberta, Canada.

Quantification of fatty acids (FA) in reproductive tissues including the placenta is important to understand the influence of FA on reproductive function. Two methods of FA determination (direct and indirect) are commonly used, but their comparative efficacy to quantify FA in bovine tissue is not known. Therefore, we compared the efficacy of the 2 methods of FA determination in bovine placental tissue. Placenta (fetal cotyledon) samples from 13 dairy cows were collected within 5 h of calving, before placental release, snap frozen and stored. Tissues (0.05 g) were assigned in duplicate to either direct methylation (samples directly subjected to methylation process with no extraction step [direct method]) or indirect methylation (samples first subjected to FA extraction and then methylated [indirect method]). The indirect method was approximately 6 h long that involved an 18-step procedure, whereas the direct method took 3 h and had only 10 steps. Briefly, frozen tissue samples were pulverized under liquid nitrogen and methylated either directly, without extraction, or indirectly, after extraction. The FA methyl esters were then injected for gas chromatographic analysis. Fatty acid data were analyzed using the Mixed procedure of SAS. Forty-five different FA were identified from the placental tissue, of which 32 FA were significantly higher in the indirect method. Moreover, the unsaturated-long chain FA of our interest, i.e., oleic, linoleic,  $\alpha$ -linolenic, eicosapentaenoic and docosahexaenoic acid were significantly higher ( $P < 0.0001$ ) in the indirect method than in the direct method (0.42, 0.08, 0.008, 0.02 and 0.05 vs. 0.30, 0.06, 0.005, 0.01 and 0.04 mg per 50 mg sample, respectively). In addition, the total amount of FA, saturated FA, polyunsaturated FA, omega-3 and -6 FA were also higher ( $P < 0.0001$ ) with the indirect method than the direct method. Even though, the direct method is shorter and faster, it reduced the amount of FA extracted. We conclude that the indirect method has higher efficacy and should be preferred over the direct method for extraction of FA from placental tissue.

**Key Words:** extraction, methylation, bovine placenta

### **W259 The effect of prepartum diets supplemented with oilseeds on maternal and newborn calf plasma fatty acid profile.**

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Long-chain polyunsaturated fatty acids (PUFA) have important roles during pregnancy, both in the dam and the fetus. However, limited information is available regarding the transfer of specific PUFA from dam to fetus in cattle. Our objective was to examine the effects of oilseed (oilseed vs. no oilseed) and type of oilseed (canola vs. sunflower) supplementation during late gestation on the fatty acid (FA) profile of maternal and newborn calf plasma. Pregnant Holsteins were assigned to 1 of 3 diets containing 8% rolled sunflower (SUN, high in linoleic acid; 8 cows) or canola seed (CAN, high in oleic acid; 7 cows) on dry matter (DM) basis, or no oilseed (CON; 7 cows), for the last  $35 \pm 2$  d of gestation. Blood samples were collected within 3 h after calving from dam and newborn calf to determine FA profile. Data were analyzed using the Mixed procedure of SAS. The proportion of total saturated FA (TSFA) was higher in CON ( $46.8 \pm 3.7$ ) fed cows than in those fed

oilseeds ( $37.5 \pm 2.7$ ). Feeding SUN increased the proportion of total PUFA and linoleic acid ( $47.8 \pm 2.5$  and  $45.5 \pm 2.9$ ) relative to those fed CAN ( $39.5 \pm 2.7$  and  $36.0 \pm 2.9$ , respectively). Moreover, SUN fed cows had higher TPUFA:TSFA ratio ( $1.29 \pm 0.06$ ) than those fed CAN ( $0.99 \pm 0.06$ ) or CON ( $0.88 \pm 0.06$ ). However, oilseed or type of oilseed supplementation during prepartum period did not affect FA profile of calves. Correlation analysis in CON treatment indicated that calf and dam TSFA were positively associated ( $R = 0.79$ ,  $P = 0.03$ ). Total monounsaturated FA (TMUFA,  $R = -0.66$ ,  $P = 0.10$ ) and oleic acid (OLA,  $R = -0.68$ ,  $P = 0.08$ ) in calf were negatively correlated with dam TSFA in CON treatment. Calf OLA ( $R = 0.68$ ,  $P = 0.05$ ) and TMUFA ( $R = 0.67$ ,  $P = 0.06$ ) in SUN treatment had positive association with dam total n-6. There was no relationship between dam and calf FA profile in CAN treatment. In summary, oilseed and type of oilseed supplemented during late gestation affected maternal FA profile but not that of newborn calf. Moreover, oilseed supplementation and the type of oilseed supplemented during prepartum period affected the association between maternal and newborn calf FA.

**Key Words:** sunflower seed, canola seed, fatty acid profile

#### **W260 Examining peripheral activity of catechol-O-methyltransferase (COMT) in Holstein cows following artificial insemination.**

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The objective was to determine peripheral activity of COMT in pregnant versus non-pregnant lactating Holstein cows. Cows ( $n = 22$ ) were synchronized using the Ovsynch plus CIDR protocol and bred via artificial insemination on d 0. Cows were retrospectively classified as pregnant ( $n = 4$ ) or open ( $n = 14$ ) based on rectal palpation at d 35 post-insemination. Moreover, cows were classified as rebred ( $n = 4$ ) if they showed signs of estrus and were re-inseminated before rectal palpation. Blood samples were collected on d 0, 4, 8, 12, 16, 20, and 24 post-insemination. Plasma was separated by layering 3 mL of blood onto 3 mL of Ficoll and centrifuged at  $400 \times g$  at room temperature for 30 min. One mL of erythrocytes were collected and stored at  $-80^\circ\text{C}$ . Peripheral activity of COMT was determined by incubating erythrocyte cell homogenates with 2 mM *s*-adenosyl methionine, 3 mM cysteine, 10 mM  $\text{MgCl}_2$ , and 1 mM of the COMT specific substrate 6-7-dihydroxycoumarin. Samples and substrates were incubated at  $37^\circ\text{C}$  for 30 min and enzyme reactions were stopped by adding 0.5 M HCl, 10%  $\text{NaNO}_2$ , 10%  $\text{NaMoO}_4$ , and 1 M NaOH to each sample. Lastly, the disappearance of 6-7-dihydroxycoumarin was determined by measuring the amount of light absorbed at 510 nm using a Spectra Max Plus plate reader. Activity of COMT was analyzed using repeated measures ANOVA of the MIXED procedure of SAS and the model statement included day, pregnancy status, and their respective interaction. Activity of COMT was increased ( $P < 0.01$ ) on d 16 post-insemination compared with all other days in cows classified as open or pregnant. Activity of COMT was not different ( $P = 0.87$ ) throughout the sampling period in cows classified as rebred. Moreover, activity of COMT was increased ( $P < 0.01$ ) on d 16 in pregnant cows versus rebred cows. Therefore, peripheral activity of COMT in lactating Holstein cows, which is involved in catechol-estrogen metabolism, may be altered by pregnancy status as well as days post-insemination.

**Key Words:** catechol-O-methyltransferase, erythrocytes, pregnancy

**W261 IGF-1 concentrations during early pregnancy in suckled Nellore beef cows.** Rogerio F. G. Peres\*<sup>1</sup>, Ky G. Pohler<sup>3</sup>, Hugo B. Graff<sup>2</sup>, Adnan D. P. Rodrigues<sup>1</sup>, Michael F. Smith<sup>3</sup>, Duane H. Keisler<sup>3</sup>, and Jose L. M. Vasconcelos<sup>1</sup>, <sup>1</sup>*Departamento de Produção Animal, Faculdade de Medicina Veterinária e Zootecnia-UNESP, Botucatu, São Paulo, Brazil*, <sup>2</sup>*Agropecuária Fazenda Brasil, Barra do Garças, Mato Grosso, Brazil*, <sup>3</sup>*Department of Animal Sciences, University of Missouri, Columbia, MO.*

The objective of this experiment was to evaluate temporal changes in serum IGF-1 concentrations in Nellore postpartum cows and its effect on the pregnancy rates to TAI. Cows ( $n = 1208$ ) from 2 farms with different grass quality (Farm1 = high quality forage;  $n = 931$ , BCS  $2.81 \pm 0.01$ ; Farm2 = low quality forage;  $n = 277$ , BCS  $2.97 \pm 0.03$ ) with  $53 \pm 6.2$  d postpartum were supplemented with mineral premix (110 g/cow/day) throughout the study. Cows underwent an estrous synchronization protocol with TAI. Blood was sampled on D-11 (first day of protocol), D0 (TAI), and D30 (pregnancy diagnosis) and analyzed for IGF-1 concentrations (all cows). Concentrations of IGF-1 on Days -11, 0 and 30 were analyzed using PROC MIXED including farm, BCS, pregnancy status on D30 and cow within pasture as a random effect. PROC GLM was used to evaluate IGF-1 concentrations effect on pregnancy rates. The pregnancies to TAI were 48.2% (449/931) and 45.5% (126/277) for Farm1 and 2, respectively. IGF-1 concentrations were greater ( $P < 0.05$ ) in all days in Farm1 compared with Farm2 ( $P < 0.05$ ). IGF-1 concentrations decreased during the study on Farm1 (D-11:  $145.0 \pm 2.4a$ , D0:  $125.2 \pm 2.9b$  and D30:  $98.6 \pm 1.8c$  ng/mL,  $P < 0.05$ ). On farm 2 the IGF-1 concentrations were greater on D-11 compared with D0 and D30 (D-11:  $73.0 \pm 1.5a$ , D0:  $65.1 \pm 1.5b$ , D30:  $63.0 \pm 1.4b$  ng/mL,  $P < 0.05$ ). There was a linear negative association of IGF-1 concentrations on pregnancy rates ( $P < 0.05$ ). There was no difference in IGF-1 concentrations on D-11 and D0 between pregnant and open cows. Cows in Farm1 that became pregnant ( $96.0 \pm 1.9a$  ng/mL) had less IGF-1 concentrations on D30 compared with nonpregnant cows ( $102.4 \pm 1.9b$  ng/mL). In Farm 2 pregnant ( $60.0 \pm 1.7b$  ng/mL) cows tended ( $P < 0.06$ ) to have less IGF-1 concentration than nonpregnant cows ( $63.7 \pm 1.5b$  ng/mL). These data show that Nellore cows have a decrease in IGF-1 concentrations within 30 d of the first insemination and pregnant cows had less IGF-1 concentration on D30 compared with nonpregnant cows. Although the farm with better quality forage had cows with increased IGF-1 concentrations compared with cows in the farm with low quality forage, in both farms pregnant cows had a decrease in IGF-1 after TAI. Further research is needed to understand mechanisms by which less IGF-1 is associated with pregnancy in beef cows. FAPESP Project #2014/03209-0.

**W262 Circulating anti-Müllerian hormone (AMH) in Holstein and Jersey breeds, at different physiological states and in dam-daughter pairs.** E. O. S. Batista\*<sup>1,2</sup>, C. Collar<sup>1</sup>, N. Silva-Del-Rio<sup>1</sup>, P. D. Carvalho<sup>4</sup>, J. P. Verstegen<sup>3</sup>, P. S. Baruselli<sup>2</sup>, M. C. Wiltbank<sup>4</sup>, and A. H. Souza<sup>1</sup>, <sup>1</sup>*University of California, Tulare, CA*, <sup>2</sup>*University of Sao Paulo, Sao Paulo, SP, Brazil*, <sup>3</sup>*Mofa Global, International Center for Biotechnology, Wisconsin, WI*, <sup>4</sup>*University of Wisconsin, Wisconsin, WI.*

Our aim was to investigate circulating levels of the anti-Müllerian hormone (AMH) in Holstein and Jersey breeds at different physiological states. A second objective was to study possible associations between circulating AMH in dam-daughter pairs. Mature cows and heifers were located in 2 commercial facilities in Central-California. Blood sample for AMH analysis was taken from Holstein (Mature cows = 141; pregnant Heifers = 408) and Jersey (Mature cows = 148; pregnant Heifers =

123) breeds and measured with the MofA-Global bovine fertility assay. Data were analyzed by the GLIMMIX and CORR procedures of SAS. Circulating AMH differed ( $P < 0.01$ ) across cattle breeds (Holstein cows = 243.9 pg/mL; Holstein heifers = 237.8 pg/mL vs Jersey cows = 312.2 pg/mL; Jersey heifers = 334.4 pg/mL). Despite of cattle breed, circulating AMH seem to be increased in cows with greater days in milk ( $P = 0.01$ ); but was not influenced by level of milk production in lactating cows ( $P = 0.92$ ) or even parity number ( $P = 0.68$ ). In virgin heifers, stage of pregnancy did not influence circulating AMH ( $P > 0.10$ ) in both Holstein and Jersey breeds. Interestingly, there was a significant although somewhat low correlation of circulating AMH in dam-daughter pairs

(Holsteins: n-pairs = 116,  $r = 0.18$ ,  $P < 0.01$ ; Jerseys: n-pairs = 106,  $r = 0.22$ ,  $P < 0.01$ ). In conclusion, AMH results are specific within major dairy breeds and some important nuisance variables may need to be taken into account when interpreting AMH results. The significant correlation between circulating AMH in dams and their respective daughters might allow for selection of this important heritable trait.

**Key Words:** anti-Müllerian hormone (AMH), dairy breed, heritability



## Production, Management and the Environment III

**W263 Technical and economic performance on beef cattle in the Livestock Low Carbon Integrated Project-ICV.** Fabiano Alvim Barbosa\*<sup>1</sup>, Vando Telles Oliveira<sup>2</sup>, Filipe Lage Bicalho<sup>3</sup>, Luciano Bastos Lopes<sup>4</sup>, Juliana Mergh Leão<sup>1</sup>, and Lucas Luz Emerick<sup>1</sup>, <sup>1</sup>Universidade Federal de Minas Gerais, UFMG, Belo Horizonte, Minas Gerais, Brazil, <sup>2</sup>Instituto Centro de Vida, ICV, Alta Floresta, Mato Grosso, Brazil, <sup>3</sup>Soluções Integradas ao Agronegócio, SIGA, Alta Floresta, Mato Grosso, Brazil, <sup>4</sup>Empresa Brasileira de Pesquisa Agropecuária, Embrapa, Sinop, Mato Grosso, Brazil.

Economic efficiency and productivity of 6 farms on cattle production systems were evaluated in Mato Grosso State, Amazon biome, during January 2013 and December 2014, part of the Livestock Low Carbon Integrated Project-ICV. Production and economic data were collected using control software (*Prodap Professional GP*) and analyzed by electronic spreadsheets (Microsoft Excel). The systems was developed exclusively on a grazing system with a base herd comprised of Zebu (Nelore) and crossbred *Bos taurus* × *Bos indicus* cattle in 4 farms. The total of pasture was 6,560 ha (ha) and 14,260 cattle. The average of pasture was 547 ha/farm and 1,188 cattle/farm. Introduction of technologies related to good practices agricultural of fertilization of pasture, feeding supplementation, breeding program, animal health practices, associated with technical-administrative management were evaluated. The zootechnical and economic indexes were compared by Duncan and Tukey test with significance level of  $P < 0.05$ . There was no statistical difference ( $P > 0.05$ ) between zootechnical indexes, stocking rates was 2.17 animals/ha, 755 kg live weight/ha, 256.73 kg body weight produced/ha and 0.87% of mortality rate. There was no statistical difference ( $P > 0.05$ ) between economic indexes, total operating costs was US\$0.68/kg, net margin of US\$165.09/ha. The net present values (NPV) and internal rate of return (IRR) were higher in 2014 than 2013 ( $P < 0.05$ ), due to increased financial resources for the technologies in 2013. NPV was US\$74.92 and US\$208.68/ha in 2013 and 2014, respectively ( $P < 0.05$ ), IRR was 1.43 and 3.49% per month in 2013 and 2014, respectively ( $P < 0.05$ ). The economic results indicated that the activity of beef cattle in Amazon biome can be sustainable use good agricultural practices.

**Key Words:** costs, profitability, sustainability

**W264 Lactation and immune responses of lactating dairy cows vary with different environmental stressors.** Ricardo O. Rodrigues\*<sup>1</sup>, Ann L. Kenny<sup>1</sup>, Matthew R. Waldron<sup>1,2</sup>, and Thomas B. McFadden<sup>1</sup>, <sup>1</sup>Division of Animal Sciences, University of Missouri, Columbia, MO, <sup>2</sup>Nutrition Professionals Inc., Chilton, WI.

Meta-analysis was used to evaluate the response of dairy cows to different stressors. Holstein cows ( $n = 24$ ;  $2.8 \pm 0.7$  parity;  $153 \pm 25$  DIM) were housed in free-stalls for 14d (P1), then moved to environmentally controlled tie-stall rooms at thermoneutral temperature for 7d (P2; constant 20°C), then exposed to 14d of programmed 12h cyclical heat stress (HS; mean THI range  $72.5 \pm 0.1$  to  $81.3 \pm 0.5$ ; P3), and finally, moved back to free-stalls for 7d (P4). Response of animals to housing (P1 vs. P2), HS (P2 vs. P3), and recovery from HS (P3 vs. P4; effect confounded with housing) were evaluated. For analyses, each variable within each period was averaged over the 3 median days. Data were analyzed using Proc MIXED of SAS; the original treatments were random effects and period was a repeated effect. For each comparison, data from the previous period were used to covariate-adjust for carryover effects. Moving cows from free- to tie-stalls did not affect BW, but reduced ( $P < 0.01$ )

milk yield and DMI by 5 and 10%, respectively. Feed efficiency (FE) and ECM FE improved 6% ( $P < 0.03$ ) but milk components declined ( $P < 0.05$ ) from free- to tie-stalls, except for fat and SCS, which increased ( $P < 0.05$ ). Plasma NEFA decreased ( $P < 0.01$ ) and BHBA increased ( $P < 0.03$ ) in tie-stalls. Immune measurements were minimally affected. Respiration rate (RR) increased ( $P < 0.06$ ) 10% in cows housed in tie-stalls, but rectal temperature (RT) was only 0.2°C higher ( $P < 0.01$ ). Heat stress negatively affected ( $P < 0.01$ ) BW, milk yield and DMI (10, 28 and 28% decline, respectively) but did not affect FE, ECM FE or 3.5% FCM FE. Immune response was negatively affected ( $P < 0.05$ ) during HS. Rectal temperature and RR increased ( $P < 0.01$ ) 1.5°C and 65%, respectively, in P3 compared with P2. During recovery from HS (P4), milk yield and DMI were higher ( $P < 0.03$ ), FE and ECM FE were lower ( $P < 0.01$ ), and BW did not change from P3. Although recovery from HS markedly reduced RT and RR (1.7°C and 42%, respectively;  $P < 0.01$ ), milk composition and most immune responses did not change from P3 to P4. We conclude that different stressors elicit different responses in performance variables and immune function.

**Key Words:** adaptation, heat stress, housing

**W265 A descriptive analysis of how dairy cows convert feed into food in the United States.** Juan M. Tricarico\*, *Innovation Center for U.S. Dairy, Rosemont, IL.*

The net contributions dairy cows make to the food system in the United States are not necessarily well understood by consumers. Estimates of nutrient conversion efficiency are sometimes used to describe these contributions but are often poorly documented or based on dubious assumptions. The main objectives of this study were to 1) define coefficients to calculate human-edible fractions of major dairy feed ingredients used in the United States, and 2) estimate the share of the dairy ration that is human-edible on a national level using these coefficients. The analysis was performed on a national average dairy ration computed from 350 farm surveys used in the carbon footprint life cycle assessment for fluid milk (available at <http://www.lcacommons.gov/>). The national average ration includes weighed rations for calves, open heifers, bred heifers, first calf heifers, springers, lactating cows, and dry cows, and accounts for forage grazed during the year. The national average ration includes 33 ingredients and contains 53% forage and 47% concentrate (DM basis). Food, fuel, and fiber industry by-products (14 ingredients) account for 19% of dairy feed DM. Eight major crops account for 80% of dairy feed DM (corn 42%, alfalfa 22%, wheat 3.1%, soybean 3.0%, canola 1.8%, sorghum 1.7%, barley 1.4%, and cottonseed 1.4%). Two coefficients were calculated to estimate human-edible fractions of each ingredient. The first coefficient was calculated as 1 minus NDF content. The non-NDF fraction was considered human-edible if it does not contain toxic compounds, and ingredients containing more than 30% NDF were excluded. The second coefficient was calculated by multiplying the first coefficient by the proportion of total ingredient production currently demanded by the US food industry. This coefficient incorporates current consumer demand, preferences and eating habits. The amount of human-edible dairy feed is either 20 or 0.9% of ration DM when using coefficients 1 or 2, respectively. Dairy cows make a net positive contribution to food supply in the United States by converting significant amounts of otherwise unusable plant matter in feed into food.

**Key Words:** dairy, human-edible, food security

**W266 Study of ethyl-2-nitropropionate, ethyl nitroacetate, nitroethane, and 2-nitroethanol as alternatives to reduce ruminal methane production.** Pedro A. Ochoa\*<sup>1</sup>, Agustín Corral<sup>1</sup>, Michael Hume<sup>2</sup>, Oscar Ruiz<sup>1</sup>, Claudio Arzola<sup>1</sup>, and Robin C. Anderson<sup>2</sup>, <sup>1</sup>*Facultad de Zootecnia y Ecología. Universidad Autónoma de Chihuahua, Chihuahua, Chihuahua, México,* <sup>2</sup>*U.S. Department of Agriculture, College Station, TX.*

Methane (CH<sub>4</sub>) is a greenhouse gas, as well as an energy loss in ruminant animals. Much research has been done aimed to reduce CH<sub>4</sub> emission from cattle industry. Nitrocompounds such as nitroethane (NE), 2-nitroethanol (2NEOH), and 3-nitropropionic acid (3NPA), have been shown to possess the ability to inhibit methane production on in vitro conditions. However, the potential of ethyl-2-nitropropionate (E2NPA) and ethyl nitroacetate (ENA) to reduce methane production have never been evaluated. We evaluated the ability of ENA, E2NPA, 2NEOH, and NE to reduce total gas, CH<sub>4</sub> production and microbial diversity changes in a consecutive batch culture. Mixed populations of ruminal bacteria were incubated in tubes 18 × 150 mm containing a basal medium with 0.2 g of ground alfalfa and a H<sub>2</sub>/CO<sub>2</sub> (1:1) gas phase. Tubes were supplemented with water (control) or the corresponding nitrocompound to reach a 12 mM concentration and incubated at 39°C. Total gas production was determined after 24 h. Microbial diversity was determined by DGGE. Gas composition was determined by gas chromatography. Data were analyzed using PROC GLM of SAS by a complete randomized design considering each nitrocompound as sole effect. According to our results, the use of nitrocompounds as supplement had no significant effect on total gas production, except for nitroethane ( $P \leq 0.000$ ; Table 1). In the other way, methane production was decreased ( $P \leq 0.0001$ ) by all nitrocompounds compared with the controls. In addition, each nitrocompound does not showed difference on its capacity of reducing methane emissions ( $P \leq 0.05$ ). The DGGE indicates that all nitrocompounds change the microbial diversity. Results of the study also reveal that the capacity of ENA and E2NPA to decrease methane production is very similar to the 2NPOH and NE. There is some evidence that these nitrocompounds could be supplemented to reduce methanogenesis.

**Table 1 (Abstr. W266).**

Treatment (nitrocompound)	Total gas volume production (mL)	Reduction (%)	Methane (μM/mL)	Reduction (%)
Control	19.00	—	43.77	—
Ethyl nitroacetate	18.10	4.73	1.41	96.80
2-Nitropropionate	18.79	1.10	1.06	97.70
2-Nitroethanol	18.36	3.36	1.17	97.33
Nitroethane	15.40	18.94	1.83	95.86

**Key Words:** nitrocompounds, methane, ruminant

**W267 Use of udder skin temperature as a heat stress indicator in lactating dairy cattle.** Kristen M. Perano\* and Kifle G. Gebremedhin, *Cornell University, Ithaca, NY.*

The objective of this research was to test whether the udder skin temperature of a cow is useful as a heat stress indicator. Heat stress leads to an increase in respiration rate, skin temperature, and rectal temperature, followed by a decrease in milk production and feed consumption. Rectal temperature is a reliable indicator of heat stress, but measuring rectal temperature may require restraint of the animal. Skin temperature is considered a less reliable indicator of heat stress, but skin temperature measurements are fast and non-invasive. For this research, 8 primiparous, mid-lactation Holstein cows producing  $34.4 \pm 3.7$  kg/d of milk

were housed in 2 identical climate-controlled rooms with tie stalls and exposed to moderate heat stress (THI  $79.5 \pm 1.2$ ) for 8 h per day. Cows were given 1 week to adjust to the facility, then data was collected for 5 weeks on milk production, feed consumption, rectal temperature, respiration rate, udder skin temperature, and body surface temperature in 3 unshaved locations (neck, back and side). Cows were milked twice daily and fed ad libitum. Rectal temperature was measured twice each day before heat stress conditions and after 8 h of heat stress. Respiration rate, udder skin temperature, and body surface temperatures were measured before heat stress and every 2 h during heat stress. Four experimental cows were randomly assigned to be cooled with conductive cooling by circulating chilled water through modified DCC waterbeds, and these cows experienced less heat stress than control cows. For the final week of the experiment, experimental and control cows were switched. Correlations among daily milk production and feed consumption, rectal temperature at the end of heat stress, and the daily average under heat stress of respiration rate, udder skin temperature, and body surface temperature were analyzed in JMP. Correlation with milk production was strongest for rectal temperature (0.67), followed by feed consumption (0.58), udder skin temperature (0.53), respiration rate (0.38), and body surface temperature (0.23). Thus, udder skin temperature may be useful as a fast, non-invasive measurement of heat stress.

**Key Words:** heat stress, skin temperature, milk production

**W268 Maternal heat stress affects calf passive immunity: Effects on intestinal cell apoptosis.** Bahroz M. S. Ahmed\*<sup>1</sup>, Ana Paula A. Monteiro<sup>1</sup>, Umair Younas<sup>1</sup>, Turkey O. Asar<sup>1</sup>, J-D. Liu<sup>2</sup>, Joyce Hayen<sup>1</sup>, Sha Tao<sup>2</sup>, and Geoffrey E. Dahl<sup>1</sup>, <sup>1</sup>*University of Florida, Gainesville, FL,* <sup>2</sup>*University of Georgia, Tifton, GA.*

Heat stress (HT) in utero not only induces fetal growth retardation but also influences postnatal performance of the offspring, such as immune function and metabolic adaptation. The objective was to examine the cellular mechanism of altered passive immunity in neonatal bull calves after in utero heat stress during late gestation. Specifically, by examination of the rate of apoptosis of intestinal cells early in life, as that has been shown to influence gut closure. Cows were dried off 60 d before expected calving and randomly assigned to one of 2 treatments: HT or cooling (CL). During the dry period, all cows were housed in a freestall barn, the pen for CL cows was equipped with active cooling including water soakers and fans whereas the pen for HT cows had no soakers. Heat stress was moderate compared with other studies, as HT cows had only 0.1°C increase in rectal temperature and 8 breath/min increase in respiration rate compared with CL cows. After birth all bull calves were immediately separated from their dams and weighed. Bull calves (n = 30) were killed at birth without colostrum feeding (5/trt) and 1 and 2 d of age (DOA, following colostrum feeding, 5/trt). Colostrum (3.8 L) was fed within 2 h after birth to bulls slaughtered on 1 and 2 DOA. After slaughter, the intestine was removed, weighed, and dissected into duodenal, jejunal and ileal segments, and tissue samples from each section were fixed in 4% neutral formalin and then transferred to 70% ethanol for immunohistochemistry. Apoptosis was measured in the jejunum using TUNEL labeling, counting 6 different sections per slide per calf. Small intestine weights did not differ (HT:  $1376 \pm 45$ ; CL:  $1309 \pm 45$  g;  $P = 0.30$ ). In both groups, apoptosis reactive cell counts decreased with time after birth (HT: 0DOA: 58.8; 1DOA: 21.6; 2DOA: 41.4; CL: 0DOA: 88.6; 1DOA: 58.0; 2DOA: 53.8; SEM = 16.7;  $P = 0.05$ ). However, apoptotic counts in CL bulls were higher than HT bulls ( $66.8 \pm 9.6$  vs.  $40.6 \pm 9.6$ , respectively;  $P = 0.10$ ). The results indicate that

jejunal cell apoptosis progresses with time after birth, but in utero HT reduces the apoptotic rate.

**Key Words:** heat stress, bull, apoptosis

#### **W269 Methane emissions from Holstein cows in tropical**

**environment.** Camila S. Cunha<sup>\*1</sup>, Marcos I. Marcondes<sup>1</sup>, Cristina M. Veloso<sup>1</sup>, Maria I. B. Brandão<sup>1</sup>, Elves C. Godinho Jr<sup>1</sup>, Marcelo M. D. Castro<sup>1</sup>, Aline S. Trece<sup>1</sup>, Luisa F. L. Salazar<sup>2</sup>, Erick Iglesias<sup>1</sup>, Otávio H. G. B. D. Siqueira<sup>1</sup>, Diego Zanetti<sup>1</sup>, and Tadeu E. Silva<sup>1</sup>, <sup>1</sup>*Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil*, <sup>2</sup>*Universidad de Antioquia, Medellín, Colômbia*.

The objective was to quantify enteric methane (CH<sub>4</sub>) emissions from Holstein cows of 2 levels of milk production in tropical environment and compare data observed in this study to data obtained from the equations proposed by the Intergovernmental Panel in Climate Change (IPCC). The study was carried out at the Universidade Federal de Viçosa, Brazil. High milk production cows (29 ± 0.89 L/d, n = 6) and low milk production cows (10 ± 0.34 L/d, n = 5) were used in the assays. Each assay had the first 5 d for cows' adaptation plus the number of days required to have 5 d of gases samples of each animal. Cows were held in free stall barns. High production cows diet were constituted, in dry matter (DM) basis, by 48.5% of corn silage, 1.5% of Coast-cross (*Cynodon* sp.) hay and 50% of concentrate based on corn and soybean meal. The diet of low production cows were in DM basis, 80% of corn silage and 20% of concentrate based on corn, soybean meal and wheat meal. Food and refusals were sampled to dry matter ingestion (DMI) analyses. Average feed intake observed to each group of cows were, respectively, 20.14 e 13.13 kg of DM/day. The sulfur hexafluoride (SF<sub>6</sub>) tracer methodology was used to obtain the observed methane emissions. IPCC Tier 2 equations were used to obtain the calculated methane emissions. A *t*-test were used for test the differences between the observed and the calculated data. Methane emissions observed from high production cows were 115.67 g CH<sub>4</sub>/day, 5.74 g CH<sub>4</sub>/kg of DMI and 3.94 g CH<sub>4</sub>/L of milk and from low production cows were 174.68 g CH<sub>4</sub>/day, 13.30 g CH<sub>4</sub>/kg of DMI and 17.22 g CH<sub>4</sub>/L of milk. When using the IPCC equations, methane emissions for the 2 groups of cows in g of CH<sub>4</sub>/day were 403.08 and 261.15, respectively. There are significant differences (*P* < 0.01) between the observed and the calculated methane emissions data for the 2 groups of cows. It was concluded that the equations for estimating methane emissions from IPCC are not representative for high and low production cows in tropical environment.

**Key Words:** greenhouse gas, lactation, sulfur hexafluoride

#### **W270 Effects of condensed tannin extract supplementation on beef cattle performance and nitrogen balance: I. Growing phase.**

Landon G. Canterbury<sup>\*</sup>, Lee-Anne J. Walter, Brandon M. Koch, David G. Lust, and Eric A. Bailey, *West Texas A&M University, Canyon, TX*.

Condensed tannins (CT), a polyphenolic of various forages, reportedly binds with dietary protein in the rumen and may affect site of nitrogen excretion in beef cattle. To evaluate the effect of CT on N excretion, a commercially available condensed tannin extract was top-dressed on a cereal grain-based diet at 3 levels (0, 1, or 2% of diet, DM basis). Nutrient digestibility and nitrogen balance were measured in British-cross steers (n = 18; BW = 374 ± 34 kg) offered ad libitum access to the basal diet (15.6% CP). Due to a limited number of metabolism crates, steers were randomly assigned to 3 groups first and then assigned to treatments within group. Steers were offered treatments for 10 d (7 d in

individual stalls and 3 d in metabolism crates), followed by 4 d for total fecal and urine collections in metabolism crates. No group by treatment interactions was detected (*P* ≥ 0.18) among the response variables. Provision of CT did not affect (*P* ≥ 0.56) OM intake or apparent total-tract OM digestion. Similarly, NDF intake and apparent total-tract NDF digestion were not different (*P* ≥ 0.52) among treatments. Nitrogen intake was not affected (*P* = 0.58) by inclusion of CT in the diet, but fecal N output increased (*P* = 0.02) at 2% CT inclusion compared with control. However, there was no difference (*P* = 0.36) in urine N output among treatments. Nitrogen retention was lesser (*P* = 0.03) with 2% CT than 0 or 1% CT. Proportion of total N excreted in urine decreased (*P* = 0.03) with CT supplementation at 1 or 2% in the diet. Similarly the proportion of total N excreted in feces increased (*P* = 0.03) with 1 or 2% CT inclusion. Under the conditions of this experiment, site of N excretion was shifted away from urine and toward feces when CT was included in a complete diet fed to beef cattle. Further work is needed to investigate possible effects of CT on N emissions from concentrated animal feeding operations.

**Key Words:** condensed tannin, digestibility, nitrogen balance

#### **W271 Tannin extract supplementation on gas production in feces of receiving bull-calves.**

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The addition of condensed tannins to feces of beef cattle decreases the amount of in vitro gas production; but it is not clear if the same effect occurs when tannin extracts are fed to cattle. In this research, 30 Brahman bull-calves (230 ± 12.7 kg) were used to evaluate the influence of tannin extract supplementation on gas production in feces of receiving bull-calves. Fecal samples were taken from each bull-calf during 3 consecutive days, 40 g aliquots were placed in 600 mL plastic flasks, and 40 g of distilled water were added, flask were closed, and connected with a plastic pipe toward a 250 mL graduate glass flask inverted in a water bath. Flasks were incubated at 37°C during 24 h, and displaced water was recorded as the in vitro gas production. Other feces aliquots of 20 g were used to DM determination. Before receiving the treatments, the values of in vitro gas production (BGP) were computed as mL gas/g of feces DM basis. Bull-calves were placed in 6 dirt-floor pens, using a completely randomized design experiment, 3 treatments were randomly assigned: 1) corn silage based-diet (15.2% CP; 1.36 Mcal NEM/kg DM) without extra additives (Control); 2) Control plus 0.6% of condensed tannin extract (CT); and 3) Control plus 0.6% of hydrolysable tannin extract (HT). After 28 d on treatment diets, fecal samples were taken during 3 continuous days again, and after treatment fecal gas production (AGP) was measured as previously described. Each bull-calf was considered as the experimental unit. Results were analyzed by ANOVA for a completely randomized design. The BGP values were similar between treatments (*P* = 0.53) with a mean of 4.7 ± 0.45 mL/g of feces DM. The AGP values were not affected by treatments (*P* = 0.54) with a mean of 5.9 ± 0.49 mL/g of feces DM. Results indicate that when both, hydrolysable or condensed tannin are supplemented in the diet; they have no influence on the amount of gas produced in the feces of receiving bull-calves.

**Key Words:** feces, gas production, tannin



**W272 Composting of dairy manure and grape vine prunings as a tool to reduce both industries' environmental impact.** Mario E. de Haro-Marti<sup>\*1</sup>, Mireille Chahine<sup>2</sup>, Ariel Agenbroad<sup>3</sup>, and Tony McCammon<sup>2</sup>, <sup>1</sup>University of Idaho, Gooding, ID, <sup>2</sup>University of Idaho, Twin Falls, ID, <sup>3</sup>University of Idaho, Caldwell, ID.

Composting of dairy manures is an acceptable and greatly used technique, but in most cases dairy manures don't have the proper carbon to nitrogen ratio (C:N) for composting without the loss of nitrogen as ammonia during the composting process. At the same time, the grape industry uses burning as the most widespread technique to dispose of carbon rich annual prunings. The objectives of this project were to demonstrate the effects of increasing the C:N content of dairy manures using grape vine prunings and to showcase 3 different composting techniques. Manure from an open lot dairy was mixed with ground grape vine prunings from an organic vineyard. The C:N ratios of the initial composting mix were adjusted to >40:1 to meet organic standards. Because grape prunings had a low C:N (79:1), sawdust from a horse stable was added to reach the desired C:N level. Three composting systems were showcased, mechanically turned windrows, passively aerated, and forced aerated. Three replications were built for each system. Control windrows consisted of 3 replications of mechanically turned windrows using only dairy manure and some straw to simulate regular dairy operations (C:N 23:1). A significant nitrogen loss reduction (4.33 kg/t, equivalent to an 85% reduction on total nitrogen loss) was observed in compost windrows with carbon-enhanced mixes (0.72 kg/t,  $P < 0.05$ ) as compared with just dairy manure mix (5.03 kg/t,  $P < 0.05$ ). Compounds' concentrations (nutrients) that usually limit compost application rates (e.g., P, K, salts) were also reduced in the final product when grape prunings were added to the initial mix. The reduction in these components can allow an increase in field application rates of compost before reaching maximum concentrations of limiting nutrients. The project demonstrated the feasibility of using composting as a Best Management Practice to reduce or eliminate the annual burning of grape prunings, reducing carbon emissions from the grape industry. It also demonstrated the reduction in ammonia emissions that can be achieved by the dairy industry when mixing wastes rich in C:N.

**Key Words:** composting, manure, grapes

**W273 Effect of floating islands on parlor wastewater multi-stage treatment system effectiveness.** Vinicius R. Moreira<sup>\*1</sup>, Brian D. LeBlanc<sup>2</sup>, Eric Achberger<sup>3</sup>, and Laura Zeringue<sup>1</sup>, <sup>1</sup>LSU AgCenter Southeast Research Station, Franklinton, LA, <sup>2</sup>LSU AgCenter School of Plant, Environmental and Soil Sciences, Baton Rouge, LA, <sup>3</sup>LSU AgCenter, Baton Rouge, LA.

Most manure stored in grazing dairy farms is wastewater collected from the milking parlor and adjacencies. Anaerobic lagoons are designed to treat this liquid waste by reducing solids and oxygen demand, but may release methane, ammonia, odors, and pathogens in the process. Treatment intensification may be a more efficient and economical long-term solution for small grazing dairies. LSU AgCenter Dairy Waste and Nutrient Management Team have been evaluating technologies and practices to improve wastewater treatment using the Southeast Research Station Dairy Wastewater Treatment Evaluation System (DWTES). The system is a replicated set of anaerobic lagoon, aerobic lagoon and constructed wetlands. The objective of this study was to evaluate the effect of artificial floating islands on DWTES effluent quality. Biohaven Floating Islands (Martin Ecosystems, Baton Rouge, LA) were deployed in an anaerobic lagoon (186-m<sup>2</sup> floating islands) and an aerobic lagoon (93 m<sup>2</sup> floating islands). Wastewater was sampled from every stage, approximately every 3 mo for 17 mo. Samples were analyzed for tem-

perature, pH, chlorophyll-A, chemical oxygen demand (COD), total solids, total dissolved solids, total suspended solids (TSS), ammonia-N, nitrate-N, nitrite-N, total Kjeldahl nitrogen (TKN), total phosphorus, anions, dissolved oxygen concentrations, total coliforms and *E. coli*. Total solids (931 mg/L), COD (711 mg/L), TKN (72 mg/L) and sulfate (14 mg/L) concentrations in raw wastewater decreased by 50% or more in the system's effluent. *Escherichia coli* counts were reduced from 6.5 log MPN/100 mL in raw wastewater to  $3 \pm 0.41$  log MPN/100 mL in wetland effluents. Floating islands improved treatment effluent for TSS (66 vs.  $49 \pm 18$  mg/L), COD (212 vs.  $168 \pm 16$  mg/L), and TKN (27 vs.  $21 \pm 2$  mg/L). Ammonia-N as a proportion of TKN and nitrate-N ( $3.79$  vs.  $4.73 \pm 1.1$  mg/L) increased in the replicate containing floating islands. Floating islands appeared to effectively improve dairy parlor wastewater treatment in a multi-stage treatment system.

**Key Words:** dairy wastewater, floating island, lagoon

**W274 Cross-species intake responses to temperature stress.** Robin R. White<sup>\*1,2</sup> and Mark D. Hanigan<sup>1</sup>, <sup>1</sup>Department of Dairy Science, Virginia Tech, Blacksburg, VA, <sup>2</sup>National Animal Nutrition Program, University of Kentucky, Lexington, KY.

The objectives of this study were to compare and model feed intake responses to temperature across species and to assess opportunities to use cross-species (CS) data to parameterize models when species-specific (SS) data were limited. Literature searches were conducted to identify studies reporting intake during climate-stress treatments compared with 1 or more thermoneutral treatments. The resulting data set comprised 614 treatment means for 108 studies on livestock responses to heat or cold stress. An ANOVA was conducted on the CS data set to identify the effects of species, temperature and species by temperature interactions on intake as a fraction of thermoneutral intake (FFI). Four models were derived from the CS data set and SS root mean squared prediction error (RMSPE) and concordance correlation coefficients (CCC) of these models were compared with models of the same form derived from SS data sets. Models used explanatory variables for (1) duration of exposure; (2) mean temperature; (3) low and high temperatures; and (4) difference between low and high temperatures and duration of exposure. An additional model accounting for temperature and stage of production was derived from the SS data. ANOVA demonstrated that the species by temperature interaction did not have a significant effect on FFI ( $P = 0.162$ ). Across species, intake decreased with temperature. Notably, all species demonstrated a constant decrease in intake across the thermoneutral zone indicating the previous assumption of constant intake during thermoneutrality may be incorrect. The CS-derived models had marginally lower RMSPE and higher CCC when compared with those derived from the SS data sets. The model fit with production data had the lowest RMSPE and highest CCC within the study. When compared over areas with notable knowledge gaps, using CS models often had reduced RMSPE and improved CCC when compared with SS models. Although fitting models based on SS data allows for incorporating unique covariates, like level of production, fitting responses based on CS data can help to improve model estimates when knowledge gaps exist.

**Key Words:** modeling, multi-species, heat stress

**W275 Comparative feedlot response of Angus-cross and Brahman bull-calves to pen-shade under hot weather conditions.** Ruben Barajas<sup>\*1</sup>, Billy J. Cervantes<sup>2</sup>, Alejandro Camacho<sup>1</sup>, Leopoldo R. Flores<sup>1</sup>, Juan J. Lomeli<sup>1</sup>, and Javier A. Romo<sup>1</sup>, <sup>1</sup>FMVZ-Universidad Autónoma de Sinaloa, Culiacán, Sinaloa, México, <sup>2</sup>Ganadera Los Migueles, S.A. de C.V., Culiacán, Sinaloa, México.

Pen-shade improves performance of beef cattle under hot weather. Brahman cattle pose better capability than Angus-cross cattle to confront hot environmental conditions. There are few comparative studies evaluating pen-shade benefits across breed's types. This research was performed to determine the comparative feedlot response of Angus-cross (AC) and Brahman (BR) bull-calves to pen shade under hot weather conditions. In a completely randomized design experiment with a 2 × 2 factorial arrangement, 30 AC and 30 BR bulls-calves (233 ± 24.9 kg) were grouped by breed type and placed in 2 types of pens: (1) Without shade (NO SHADE); or (2) Shaded (SHADE). Shade was provided by a ceiling of 6 × 4 m of metal sheeting positioned 3.7 m over soil level. Bull-calves were fed with a corn silage-based diet (15% CP 1.35 Mcal NEm/kg DM), served at 1700 h to minimize digestion heat load impact. Length experiment was 98 d, and pen was the experimental unit. Results were analyzed by covariance, and initial body weight was used as the associated co-variable. Across experiment, climatic conditions were air temperature 36.4 ± 4.04°C (range 27.6 to 49.5°C), relative humidity 47.9 ± 11.87% (range 24.7 to 82.5%), and THI 85.7 ± 3.16 (range 79.1 to 96.4). Interactions breed × pen shade ( $P < 0.05$ ) were observed on final body weight, average daily gain, and feed efficiency. Angus-cross SHADE bull-calves were heavier ( $P < 0.01$ ) as compared with AC NO SHADE (369.9 vs. 350.3 kg), while SHADE had no influence ( $P > 0.10$ ) on final weight of BR. The Angus cross in SHADE gain 16% more wt ( $P < 0.01$ ) than Angus cross allotted in NO SHADE (1.40 vs. 1.20 kg/d), but ADG was similar ( $P > 0.10$ ) for BR both in SHADE and NO SHADE. AC SHADE bull-calves had a 12% better feed/gain ratio ( $P = 0.03$ ) than AC NO SHADE (0.18 vs. 0.16 kg gain/kg DMI). Results suggest that feedlot response to pen shade vary with breed type, and become important as the proportion of Brahman breed decreases in genetic composition of beef cattle under hot climate conditions.

**Key Words:** bull-calves, feedlot performance, pen shade

#### **W276 A multi-objective diet optimization to reduce land, water, and greenhouse gas emissions from US dairy production.**

Robin R. White\* and Mark D. Hanigan, *Department of Dairy Science, Virginia Tech, Blacksburg, VA.*

The objectives of this study were to construct an optimization model to identify opportunities to reduce land use, water use and greenhouse gas (GHG) emissions within dairy production systems and to assess how improved energy and protein use efficiency could affect opportunities to reduce environmental impact (EI) of dairy production systems. Non-linear programming was used to adjust monthly diets fed to 10 cattle groups to minimize EI associated with an average United States (US) dairy production system. System boundaries extended from the inputs to the cropping system to the dairy farm gate. The effects of improved biological efficiencies were modeled as a 15% decrease in the allometric energy constant used to calculate maintenance energy requirements or as a 15% decrease in maintenance metabolizable protein requirements. Least-cost optimization was used as a baseline. A total of 28 scenarios were simulated which varied in objective, biological efficiency and allowable cost increase. Objectives included minimizing land, water, greenhouse gases, or all EI metrics. Allowable cost increases ranged from 1% to 20%. Baseline land use (1.22 m<sup>2</sup>/kg milk), water use (14.6 m<sup>3</sup>/kg) and GHG emissions (1.45 kg CO<sub>2</sub>/kg) agreed well with established values for US dairy production. At cost increases between 1% and 20% above baseline, EI metrics could be simultaneously reduced by 5.9 to 16.6%. Improving energy or protein efficiency greatly improved opportunity to reduce EI. When both energy and protein efficiency were improved by 15%, EI reductions ranged from 14.6 to 21.1%. Opportunity to reduce EI varied greatly with allowable cost increase and

diminishing environmental returns to cost were apparent. The cost of reducing environmental impact by 15% was decreased by 87.7% when energy and protein efficiency improved compared with the national average production efficiency scenario. Improving energy and protein use efficiency of dairy cattle represents a promising way to reduce EI without sacrificing profitability.

**Key Words:** feed efficiency, dairy, environmental impact

#### **W277 Effect of temperature humidity index patterns on fertility, postpartum disease and culling risk in New York dairy farms.**

Benjamin D. Scott\* and Julio O. Giordano, *Department of Animal Science, Cornell University, Ithaca, NY.*

Objectives were to use within-barn measurements of temperature-humidity index (THI) to evaluate its effect on (1) Pregnancies per AI (P/AI) relative to THI at or near AI; (2) postpartum disease incidence rates (DZ30); and (3) exit from the herd (E30) relative to THI near parturition. Temperature and humidity measurements were collected hourly on 5 farms using HOBOWare data loggers. Hourly measurements were averaged daily. The effect of THI on P/AI was evaluated using a THI threshold of 72 on the day of AI (THI1) or by 3-d (THI3) and 7-d (THI7) rolling averages (day of AI and prior). The DZ30 and E30 were analyzed by THI on the day of parturition (THIC), THI 7 d before and after parturition (THI14), 7 d before parturition (PreTHI), and the day of parturition plus 6 d after (PostTHI). The DZ30 and E30 were analyzed by a 68 THI threshold and represent a 30-d risk period. DZ30 represented a minimum of 1 reported event of: displaced abomasum, milk fever, metritis, retained placenta, ketosis, or mastitis. Pregnancies per AI were lower ( $P < 0.01$ ) for AI occurring at ≥72 THI threshold using all 3 THI approximations. Mean P/AI were 38.7% (n = 9,334) vs 32.5% (n = 1,140) for THI1 <72 or ≥72, 34.8% (n = 9,291) vs 30.3% (n = 1,074) for THI3 <72 or ≥72, and 38.8% (n = 9,392) vs 28.9% (n = 748) for THI7 <72 or ≥72. The DZ30 for THI ≥68 vs THI <68 tended to be different for THIC ( $P = 0.08$ ), was similar for PostTHI ( $P = 0.15$ ) but it was greater for THI14 ( $P = 0.01$ ) and PreTHI ( $P = 0.02$ ). Mean (95% CI) DZ30 was 26.3% (24.3–28.4%) vs 30.3% (27.9–32.9%) for THI14 <68 (n = 2,381) or ≥68 (n = 1,922) and was 26.3% (24.3–28.5%) vs 30.2% (27.8–32.7%) for PreTHI <68 (n = 2,388) or ≥68 (n = 1,905). Relative risk (RR) of DZ30 (95% CI) was greater than 1 for THI14 ≥68:THI14 <68 (1.15, 1.03–1.29) and PreTHI ≥68:PreTHI <68 (1.15, 1.02–1.29). Thus, attributable risk (AR) of THI14 ≥68 to DZ30 is 13.2% and of PreTHI ≥68 is 12.9%. Rates of E30 were similar ( $P > 0.10$ ) for all THI measurements using a 68-unit threshold. We conclude that THI before insemination reduces P/AI, that THI around parturition affects 30-d postpartum disease risk with prepartum THI having the greatest effect. Also, THI around parturition does not strongly affect postpartum 30-d exit probabilities.

**Key Words:** heat stress, disease risk, transition

#### **W278 Methane and carbon dioxide emissions from manure of dairy cows fed regular or brown midrib corn silage-based diets.**

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The objective of this study was to examine the effects of feeding dairy cows with regular corn (RCS) or brown midrib (BMCS) corn silage on CH<sub>4</sub> and CO<sub>2</sub> emissions from manure storage. For this purpose, 8 lactating cows fed diets containing 59% of RCS or BMCS were used in a block design. Total manure (feces and urine) collection from each

cow was performed on 2 consecutive days. Manure was mixed (1:1) with an inoculum from a bioreactor and stored (20°C) under anaerobic conditions in glass bottles (6 replicates/cow) for 17 weeks. Quantity of gas produced was measured daily and sampled to determine gas composition, while manure was sampled weekly to measure volatile fatty acid (VFA) and NH<sub>3</sub> concentrations. The MIXED procedure of SAS was used to determine the effects of corn silage cultivar on measured parameters and significance was declared at  $P \leq 0.05$ . Cows fed BMCS-based diets excreted more ( $P < 0.01$ ) manure (86 vs 64 kg/d, respectively) and organic matter (8.6 vs 6.5 kg/d, respectively) than cows fed RCS-based diets. Excretion of N averaged 370 g/d and was not affected ( $P = 0.16$ ) by the dietary treatment. Compared with manure from cows fed RCS-based diets, manure from cows fed BMCS-based diets emitted more ( $P \leq 0.01$ ) CH<sub>4</sub> (173 vs 146 L/kg organic matter [OM]) and CO<sub>2</sub> (148 vs 118 L/kg OM) during storage period. Total VFA concentration tended to higher ( $P = 0.08$ ) in manure from cows fed BMCS compared with cows fed RCS, indicating more extensive degradation of OM during storage of manure when cows were fed BMCS vs. RCS. Losses of OM from manure of cows fed BMCS based-diets was higher ( $P < 0.01$ ) than losses from cows fed RCS-based diets (38 vs. 31%). In conclusion, feeding BMCS-based diets to dairy cows can increase daily volatile solid excretion and CH<sub>4</sub> and CO<sub>2</sub> emissions (per kg volatile solids) compared with feeding RCS-based diets.

**Key Words:** manure, CH<sub>4</sub> emission, brown midrib corn silage

**W279 Determination of climatologically suitable places in Turkey for feedlot cattle production by using comprehensive climate index model.** Hayati Koknaroglu<sup>1</sup>, John A. Harrington Jr<sup>2</sup>, and Terry L. Mader<sup>3</sup>, <sup>1</sup>Suleyman Demirel University, Isparta, Turkey, <sup>2</sup>Kansas State University, Manhattan, KS, <sup>3</sup>University Nebraska, Lincoln, NE.

The objective of this study was to determine climatologically suitable places to raise feedlot cattle in Turkey. Daily average temperature, relative humidity and wind speed values for 15 locations (Antalya, Balikesir, Corum, Diyarbakir, Edirne, Elazig, Erzincan, Erzurum, Eskisehir, Isparta, Izmir, Kayseri, Konya, Sivas and Van), spanning last 30 years, were obtained. Daily solar radiation values could not be obtained from the weather stations and were calculated based on a formula that takes hemisphere, latitude and day of the year into account. The comprehensive climate index (CCI) model that enables one to quantify beef cattle performance based on environmental conditions (temperature, relative humidity, wind speed, solar radiation) at any time in the year was used to predict dry matter intake (DMI), average daily gain and feed efficiency of feedlot cattle. Because mostly dairy breed calves are placed into the feedlot in Turkey, Holstein option in CCI was chosen to calculate maintenance energy requirement of cattle. Based on the previous feedlot feeding studies conducted in Turkey, it was assumed that calves would be placed on feed at 250 kg and be marketed at 520 kg, diet would have 2600 kcal/kg metabolic energy and would have dry matter intake of 2.31% of the body weight. Results comparing the 15 locations indicate that cattle raised in Antalya, known to be hottest place and Erzurum, known to be coldest place, had the lowest and highest DMI, respectively ( $P < 0.05$ ). Environmental conditions in summer lowered cattle DMI in hotter locations and winter conditions increased DMI in colder locations ( $P < 0.05$ ). Feedlot cattle raised in hotter and colder portion of Turkey had lower average daily gain than other places having a milder climate ( $P < 0.05$ ). In general, cattle raised in hotter climate had better feed efficiency than those raised in cold climate ( $P < 0.05$ ).

**Key Words:** comprehensive climate index, feedlot, performance

**W280 Methane prediction equations for beef cattle fed high forage diet.** Paul Escobar-Bahamondes<sup>1,2</sup>, Masahito Oba<sup>1</sup>, and Karen A. Beauchemin<sup>2</sup>, <sup>1</sup>University of Alberta, Edmonton, AB, Canada, <sup>2</sup>Agriculture and Agri-Food Canada, Lethbridge, AB, Canada.

The study aim was to improve the prediction of CH<sub>4</sub> emissions from beef cattle by developing equations specific for high forage diets ( $\geq 40\%$  DM basis). Treatment means from 38 beef studies published between 2000 and 2014 with dietary forage  $\geq 40\%$  DM were compiled into a database. Criteria for inclusion in the database were intake, diet composition and enteric CH<sub>4</sub> production. Principal component analysis detected relevant variables associated with CH<sub>4</sub>. Because of the limited size of the original database ( $n = 123$ ), a Monte Carlo technique was used to resample 1,000 times the data from each study to create a new virtual data set. Outliers were excluded by Mahalanobis distance in both the original database and virtual data set. Using the original database, forward stepwise multiple regression was used to obtain prediction equations. The random effect of study was included in the analysis using the Mixed procedure and 'leave-one-out' cross validation was used to internally validate the equations. Using the virtual data set ( $n = 100,305$ ), equations were developed using forward stepwise multiple regression and K-fold cross validation ( $n = 10$ ). Model performance was evaluated as observed-predicted values using concordance correlation ( $r_c$ ) and root mean square prediction error (RMSPE, g/d). Statistical analysis was performed using JMP v11. Using the original database, the best-fit equation was: CH<sub>4</sub> (g/d) = 71.5(±11.45) + 0.12(±0.03) × BW (kg) + 0.10(±0.01) × DMI<sup>3</sup> (kg/d) - 244.8(±56.44) × fat<sup>3</sup> (kg/d) with  $P < 0.0001$ ,  $r_c$ : 0.72 and RMSPE: 39.1; where BW = body weight; DMI = dry matter intake. Using the Monte Carlo data set, the best-fit equation was: CH<sub>4</sub> (g/d) = 25.9(±0.54) + 0.13(±0.001) × BW (kg) + 145.4 (±1.31) × fat (kg/d) + 10.3(±0.16) × (NDF - ADF)<sup>2</sup> (kg/d) + 0.1(±0.00) × DMI<sup>3</sup> (kg/d) - 27.4 (±0.20) × (starch/NDF) with  $P < 0.0001$ ,  $r_c$ : 0.81 and RMSPE: 36.4 where NDF = neutral detergent fiber and ADF = acid detergent fiber. Monte Carlo data set equation improved prediction accuracy compared with the original database equation, but extensive feed analysis is required to use the equation. Both equations specifically developed for beef cattle fed forage diets may increase the accuracy of predicting CH<sub>4</sub> production.

**Key Words:** methane, beef cattle, equation

**W281 Mathematical models to predict phosphorus output in manure and milk from lactating dairy cows.** G. Alvarez<sup>1,2</sup>, J. A. D. R.N. Appuhamy<sup>2</sup>, and E. Kebreab<sup>2</sup>, <sup>1</sup>Universidad Autonoma de San Luis Potosi, San Luis Potosi, Mexico, <sup>2</sup>Department of Animal Science, University of California, Davis, CA.

Accurate manure and milk phosphorus (P) output estimates are important for monitoring P released to the environment and assist in quantifying P balance in lactating dairy cows. The objectives of the study were (1) to develop empirical models, and (2) evaluate extant models for estimating P output from lactating dairy cows. A meta-regression analysis was conducted using 191 fecal (Pf), 83 urine (Pu), and 110 milk (Pm) P output measurements from 39 studies. Dry matter intake (kg/cow/d), diet composition, milk yield and composition and DIM were used as predictor variables. The Pu was highly variable with a mean of  $0.34 \pm 0.027$  g/cow/d. Two models; with and without DMI data were developed for each Pf and Pm (g/cow/d, Table 1). The DMI-based extant model by Weiss and Wyatt (2004) was the most successful in predicting manure P output [root mean square prediction error (RMSPE) as a percentage of average observed value = 19.6%]. Models developed in this study with and without DMI predicted Pf with RMSPE of 18.6 and 24.0%, respectively. The Pm predictions with and without DMI were related to



RMSPE <17.0% of the average measured Pm. Our models also predicted total P output [Pf + Pm + Pu] from lactating dairy cows with good accuracy (RMSPE = 11.8 – 17.3%). The models can be used to improve our understanding of P utilization and excretion in dairy production systems.

**Table 1 (Abstr. W281).** Models developed in this study

Equation <sup>1</sup>	RMSPE (%)
With DMI data	
$Pf = -10.7(4.06) + 0.69(0.03) \times \text{Pintake} + 0.56(0.24) \times \text{CP} + 0.68(0.17) \times \text{GE} - 0.50(0.10) \times \text{Milk}$	18.6
$Pm = 19.1(7.28) + 1.25(0.17) \times \text{DMI} + 0.20(0.06) \times \text{Milk} - 6.20(2.21) \times \text{MProt} - 0.02(0.01) \times \text{DIM}$	13.9
Without DMI data	
$Pf = -10.2(7.43) + 151.0(10.6) \times P + 0.96(0.22) \times \text{GE} - 0.52(0.22) \times \text{ADF}$	24.0
$Pm = 36.6(8.46) + 0.45(0.06) \times \text{Milk} - 0.03(0.01) \times \text{DIM} - 5.20(2.7) \times \text{MProt}$	16.9

<sup>1</sup>Pintake = P intake (g/cow/d), GE = dietary gross energy (MJ/kg DM), Milk = milk yield (kg/cow/d), MProt = milk protein (%), CP, P and ADF (% of dietary DM).

**Key Words:** mathematical model, phosphorus excretion, dairy cow

**W282 Enteric methane mitigation and evaluation ruminal parameters of cattle fed cottonseed and vitamin E.** Ricardo Galbiatti Sandoval Nogueira\*, Flavio Perna Jr., Eduardo Cuellar Orlandi Cassiano, Lizbeth Collazo Paucar, Mariane Cheschin Ernandes, Diana Carolina Zapata Vasquez, Adrielle Matias Ferrinho, Romulo Germano de Resende, Felipe Bispo Mendonça, Renata Gardennalli, Angélica Simone Cravo Pereira, and Paulo Henrique Mazza Rodrigues, *University of São Paulo, Pirassununga, São Paulo, Brazil.*

Objective this study was verify the enteric methane emissions and evaluate ruminal parameters caused by the inclusion of 30% of cottonseed (diet with 8,32% ether extract) and 0,4% vitamin E (500 IU) in the diet of cattle. Six cannulated cows were distributed in a replicate 3x3 Latin square. Treatments were (1) control: basal diet; 2) CS: basal diet plus 30% cottonseed and 3) VitE: basal diet plus 30% cottonseed plus 0,4% vitamin E. Ruminal fermentation ex situ (micro-rumen) technique, in which, a sample (10 mL solid and 20 mL liquid) of rumen content was collected via cannula and put inside a sealed glass flask and incubated for 30 min at 39°C, starting samples removed the vials analyzed for the enteric methane emissions, total short chain fatty acids production (SCFA) and energy loss on methane compared with other products of rumen fermentation (REL). Results were compared through orthogonal contrast, where contrast 1: CS and VitE vs control; contrast 2: CS vs VitE. Data analyzed for SAS 9.3 and was considered 5% significance level. Results are shown in Table 1. Cottonseed inclusion reduced methane emissions. Production of propionate was similar between treatments, in average 82.76 g kg d<sup>-1</sup>. Cottonseed inclusion reduced the acetate, butyrate, SCFA and acetate propionate ratio (C2:C3). REL was similar between treatments, in average 29.45%. Include 30% of cottonseed in cattle diet reduces rumen fermentation, resulting in lower enteric methane emissions and total short chain fatty acids production. Vitamin E does not affect ruminal fermentation products.

*Contd.*

**Table 1 (Abstr. W282).** Effect of including cottonseed and vitamin E on the production of SCFA, methane, and REL

Variable	Control	CS	VitE	SEM	C1	C2
Acetic, g kg d <sup>-1</sup>	210.1	143.9	133.9	11.8	0.0028	0.7091
Propionic, g kg d <sup>-1</sup>	80.94	85.35	82.02	5.51	0.8017	0.7910
Butyric, g kg d <sup>-1</sup>	90.72	51.36	44.30	4.22	0.0001	0.3883
SCFA total, g kg d <sup>-1</sup>	381.8	280.6	260.2	19.6	0.0045	0.6411
C2:C3	3.607	2.193	2.413	0.17	0.0097	0.6139
Methane, g kg d <sup>-1</sup>	43.79	26.40	24.13	1.42	0.0001	0.3584
REL	31.31	29.67	27.35	1.36	0.4623	0.5980

C1 = CS and VitE vs. control; C2 = CS vs. VitE.

**Key Words:** lipids, energy loss on methane, short-chain fatty acids

**W283 The effect of temperature, pH, total solids and type of shape of goat manure for biogas production.** Bruno Biagioli\*<sup>1</sup>, Kleber T. Resende<sup>1</sup>, Izabelle A. M. A. Teixeira<sup>1</sup>, Normand St-Pierre<sup>2</sup>, Carla J. Härter<sup>1</sup>, and Márcia H. M. R. Fernandes<sup>1</sup>, <sup>1</sup>Univ. Estadual Paulista, Department of Animal Sciences, Jaboticabal, SP, Brazil, <sup>2</sup>The Ohio State University, Department of Animal Sciences, Columbus, OH.

The aim of this study was to obtain the best combination of factors that could be easily manipulated by men to optimize the production of biogas. To evaluate the anaerobic digestion of goat manure, we used 24 digesters distributed in factorial design 3 × 3 × 3 × 2, and considered the following factors: temperature (20.1°C, 21.5°C, 30.2°C, 33.7°C and 34.7°C), pH (6.8, 6.84, 7.01, 7.15, 7.22), total solids (2.6%, 3.5%, 4.5%, 6.8%, 6.9%), and type of shape (whole goat manure and broken goat manure) in a central composite design. Data were analyzed as a mixed model with fixed effect of temperature, pH, TS, shape of manure, and their interactions and the random effect of day and digester, using the PROC MIXED procedure of SAS (version 9.0). The shape of manure did not affect biogas production. The isolated effect of temperature and pH in the biogas production was not significant whereas only the highest level of total solids resulted in greater biogas production by day (0.11 m<sup>3</sup> ± 0.02; P = 0.0008). We also observed interaction among temperature, pH, and total solids (5.36 m<sup>3</sup> ± 0.12 P = 0.0020) on the biogas production. Additionally, the percentage of methane in the biogas was not affected by the amount of total solids, pH, and temperature in the biodigester. The best volume of biogas production was obtained when the following values were applied: 34.7 (temperature), 6.8 (pH) and 7.0 (total solids). Our results reveal that even under low pH and low temperature, biogas production increases as a function of increasing total solids of goat manure, however none of the factors affected the proportion of methane in the biogas. The fact that we detected the best biogas yield using 6.8% total solids is extremely important, because a greater supply of substrate requires fewer amounts of water and time, it is less labor-intensive, while methane production is greater than that found using 5% total solids. Even though water returns in the biofertilizer form, if less water is required through biodigestion it would represent a significant water saving.

**Key Words:** biodigester, methane, caprine

**W284 Idle cattle, water buffalo, and swine consume 44% of global feed resources.** J. R. Knapp\*<sup>1</sup> and R. A. Cady<sup>2</sup>, <sup>1</sup>Fox Hollow Consulting LLC, Columbus, OH, <sup>2</sup>Elanco Animal Health, Greenfield, IN.

The objectives of this study were to estimate the number of idle cattle, water buffalo, and swine globally and evaluate their impact on feed and water resource utilization as part of the larger Food Forward Sustainability Project. The term “idle” is defined as an animal that is not growing, pregnant, lactating, held for breeding, or used for draft. Two data sources were used: 1) USDA Foreign Ag Service data that includes cattle and swine inventories for 19 countries, and 2) FAO data that includes inventories of cattle, water buffalo, and swine for 16 sub-continental regions. A gamma distribution was used to estimate survival and the number of non-breeding animals alive after 4, 5, and 1 years for cattle, water buffalo, and swine, respectively. Non-breeding stocks were adjusted for imports and exports. Turnover age was calculated as stock numbers divided by slaughter numbers and represents the average lifespan of an animal in the non-breeding population in steady state conditions. Feed consumption estimates were cross-validated using maintenance energy requirements and found to be in reasonable agreement. Turnover ages ranged from 2.6 to 14.7 years for cattle, 3.0 to 8.5 years for water buffalo, and 4.8 to 21.1 mo for swine across regions. It was estimated that  $46.6 \pm 0.6\%$  of 1.45 billion cattle,  $61.1 \pm 0.5\%$  of 190 million buffalo, and  $9.8 \pm 0.3\%$  of 946 million swine in the world in 2010 were mature, non-breeding animals. With cattle and water buffalo, a portion of these animals may be maintained for draft purposes and would not be completely idle. At this time, no data are available to reliably estimate the draft use of cattle and water buffalo. Also, in some societies, mature animals may be maintained as a form of personal wealth, defense against times of food scarcity, or both. Idle and draft cattle, water buffalo, and swine consumed an estimated 1068, 132, and 62 million metric tons of feed (DM basis) or 44% of total feed required, and 2840, 364, and 154 billion liters of water, respectively, in 2010. Reducing the number of idle cattle, water buffalo, and swine is one of several viable options to increase system-wide production efficiency and decrease the total amount of resources required to produce animal-based foods.

**Key Words:** sustainability, food supply, resource utilization

**W285 Crop and grazing land requirements to meet consumer demand for animal products in 2050.** J. R. Knapp\*<sup>1</sup> and R. A. Cady<sup>2</sup>, <sup>1</sup>*Fox Hollow Consulting LLC, Columbus, OH*, <sup>2</sup>*Elanco Animal Health, Greenfield, IN*.

As part of the larger Food Forward Sustainability Project, we have estimated the quantity of feed required to produce animal products and meet global consumer demands in 2050 under different production scenarios using population-based models (Table). The objectives of this study were 1) to determine how much crops and crop residues were available globally in 2010 in support of animal feed production and might be available in 2050 under reasonable estimates of increasing crop yields, 2) compare them to feed requirements, and 3) to evaluate the impact on land requirements for feed production. Data from USDA National Agricultural Statistics Service and FAO were utilized to estimate yields and utilization of major grain and oilseed crops. Proportions of crops used in feed, food, seed, and other uses were assumed to be the same in 2050 as in 2010. Crop residues were estimated from crop yields and represent the maximum potentially available feed, but do not account for use in bedding, soil amendment, etc. While production and utilization of grain, oilseed, and byproducts for feed in 2010 appears to be lower than feed requirements (Table), it is likely underestimated due to under-reporting of grain byproducts by FAO and neither data source fully accounting for secondary byproducts or animal protein byproducts. These results indicate that continued innovation supporting sustainable intensification in livestock and poultry agriculture and increasing crop yields can produce adequate amounts of food and feed

in 2050 without increasing crop lands (Table). Also, increasing crop yields have the potential to provide more crop residues for feeding ruminant livestock that could increase the efficiency of feed use and reduce pressure on grazing lands (Table). Without innovation in animal and plant agriculture, 36 to 58% more land would be required in 2050 to produce food and feed.

**Table 1 (Asbtr. W285).** Feed requirements and annual production (million metric tonnes as is, 85-90% dry matter)

	2010	2050 Continued innovation	2050 Frozen productivity
<b>Feed requirements</b>			
Grain & byproducts	1,111	1,331	1,872
Oilseed & byproducts	320	396	557
Crop residues & forage	3,146	3,245	4,545
<b>Feed production &amp; utilization</b>			
Grains & by-products	878	1,403	
Oilseeds & byproducts	261	367	
Crop residues	2,075	2,905	

**Key Words:** sustainability, food supply, animal protein

**W286 Climatic factors associated with abortion occurrences in Japanese commercial pig herds.** Ryosuke Iida, Satomi Tani\*, and Yuzo Koketsu, *Meiji University, Kawasaki, Kanagawa, Japan*.

Our objectives were to determine climatic and production factors associated with abortions in female pigs in commercial herds and to compare the reproductive performances and culling patterns between aborting and nonaborting reserviced female pigs. We analyzed 309,427 service records of female pigs entered into 100 herds located in humid subtropical or continental climate zones. Climate data were obtained from 21 weather stations located close to the studied herds. Mean daily average temperatures (Tavg) for the 21-d pre-mating period for each female pig were coordinated with that female’s reproductive data. Generalized linear models were conducted for whether or not a female pig aborting. Abortion risk per service ( $\pm$ SEM) was  $0.7 \pm 0.06\%$ , and mean daily average temperature (range) was  $15.0^\circ\text{C}$  ( $-11.0$  to  $32.7^\circ\text{C}$ ). Risk factors associated with an increased abortion risk per service were higher parity, having more stillborn piglets, higher Tavg and reservicing ( $P < 0.05$ ). Also, abortion risks in parity 1 and parity 2–5 sows linearly increased by 0.3 and 0.1%, respectively, as Tavg increased from 20 to  $30^\circ\text{C}$  ( $P < 0.05$ ), but there were no such associations in parity 0 and or parity 6 or higher female pigs ( $P \geq 0.37$ ). Abortions reserviced female pigs had 0.4 fewer pigs born alive than nonaborting reserviced female pigs ( $P < 0.05$ ). Also, 64.6% of all aborting female pigs were culled for reproductive failure, compared with only 23.4% of nonaborting females. In conclusion, producers should closely monitor female pigs at high risk of aborting and apply more advanced cooling systems.

**Key Words:** abortion, heat stress, incidence rate

**W287 Estimated feed and water requirements to meet global 2050 demand for animal proteins.** J. R. Knapp\*<sup>1</sup> and R. A. Cady<sup>2</sup>, <sup>1</sup>*Fox Hollow Consulting LLC, Columbus, OH*, <sup>2</sup>*Elanco Animal Health, Greenfield, IN*.

FAO has published predictions for 2050 global milk and meat consumption. As part of the larger Food Forward Sustainability Project, the objective of this study was to estimate the quantities of feed and water

required to produce the FAO forecasted quantities of animal products under 2 scenarios and independent of advances in crop production. In the first scenario, innovation continues, and in the second, development and application of technologies are limited, freezing productivity at 2010 levels. Models capturing animal population dynamics were formulated with biological constraints and used to predict the resources needed for egg, dairy, and meat production from chickens, swine, cattle, and water buffalo. Global consumption of chicken meat, pork, and beef is expected to increase 134, 51, 43%, respectively, based on increases in population and per capita consumption. For swine and beef, options exist with current technology and management that if applied more broadly could increase production efficiency and maintain or reduce resource utilization by these species' production systems relative to 2010 levels. The impact of increased dairy consumption would increase feed and water utilization by 10% with continued innovation. In swine, beef, and dairy, it is biologically possible to increase total production while decreasing animal numbers. In contrast, production of chicken meat cannot be increased to meet the very large projected demand without significant increases in both animal numbers and carcass size. Likewise, increasing egg production to meet future demands will require modest increases in eggs per hen and number of hens. Innovation and broader adoption of existing technology have strong potential to mitigate the impacts of increased animal production on feed and water utilization while meeting future demands for animal products, thus minimizing the expansion of animal numbers as has happened historically.

**Table 1 (Abstr. W287).**

	2010	2050 Cont. Innovation	2050 Frozen Productivity
Human population (billion)	6.9	9.4	9.4
Animal product consumption (kg×yr <sup>-1</sup> ×person <sup>-1</sup> )	148.4	170.7	170.7
Total animal product consumption (billion MT)	1.02	1.60	1.60
Feed (as is, billion MT)	4.6	5.0	7.0
Water (trillion L)	15.8	17.3	24.9

**Key Words:** sustainability, food supply, animal protein

**W288 Climatic factors associated with reproductive performance in English Berkshire pigs raised in a subtropical climate region of Japan.** Shiho Usui, Satomi Tani\*, and Yuzo Koketsu, *Meiji University, Kawasaki, Kanagawa, Japan.*

Our objective was to clarify the reproductive characteristics of English Berkshire female pigs in a humid subtropical zone. We examined interactions between 2 breeds (English Berkshire and crossbred females) and climatic factors for reproductive performance. We analyzed 63,227 first-service records of 11,992 females in 12 herds. Climate data were obtained from 4 weather stations located close to the studied herds. Mean daily maximum temperatures (Tmax) and daily average relative humidity (ARH) for different periods around servicing and farrowing of each female were coordinated with that female's reproductive performance data. Multilevel mixed-effects models were applied to the data. There were 2-way interactions between the breed and either Tmax or ARH for weaning-to-first-mating interval (WMI), total born and farrowing rate ( $P < 0.05$ ). The WMI in Berkshire sows increased by 0.10 (SE: 0.008) days as Tmax increased by each degree Celsius ( $P < 0.05$ ), whereas in crossbred sows it only increased by 0.01 (0.005) days ( $P < 0.05$ ). The WMI in crossbred sows also increased by 0.03 (0.007) days as ARH increased by one percent ( $P < 0.05$ ). However, there was no such association in Berkshire sows ( $P = 0.37$ ). In Berkshire females total born decreased by 0.02 (0.003) pigs for each degree Celsius increase in Tmax ( $P < 0.05$ ), whereas that in crossbred females decreased by 0.03 (0.002) pigs ( $P < 0.05$ ). With regard to farrowing rates, the odds ratios in Berkshire and crossbred females were 0.973 (95% confidence intervals: 0.967–0.979) and 0.986 (0.981–0.991) for Tmax, respectively. Therefore, we recommend producers applying advanced cooling systems for Berkshire females as well as crossbred females.

**Key Words:** Berkshire, climatic factor, reproductive performance



## Ruminant Nutrition: Beef III

### W289 Effect of increasing levels of alfalfa hay on ruminal fermentation in growing Simmental heifers fed high-concentrate diets.

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Eight rumen cannulated Simmental heifers (BW = 281.4 ± 18.5 kg) were randomly assigned to 1 of 4 experimental treatments to determine the effects of increasing levels of alfalfa hay on ruminal fermentation. Treatments tested were: a) total mixed ration with 10% barley straw as forage source (10BS), b) total mixed ration with 13% alfalfa hay as forage source (13AH), c) total mixed ration with 16% alfalfa hay as forage source (16AH) and d) total mixed ration with 19% alfalfa hay as forage source (19AH). Forages were coarsely chopped before their incorporation in total mixed ration. Diets were offered on an ad libitum basis, and formulated to be isocaloric (2.83 Mcal ME/kg DM) and iso-nitrogenous (14% CP on DM basis). After 2 weeks of diet adaptation, ruminal samples were taken immediately before feeding and at 4, 8, 12, 16, and 24 h after feeding on d1, d4 and d7 of the sampling week. Differences were analyzed by using the MIXED procedure of SAS. The model contained the fixed effects of treatment and the random effect of heifer. Day and hour were used as repeated measure. Intake of DM recorded during the sampling week was affected by treatment, being higher in heifers fed 16AH and 19AH than in heifers fed 10BS and 13AH (8.9 and 8 kg/d on average, respectively;  $P < 0.001$ ). Heifers fed 19AH had a higher mean ruminal pH compared with the other treatments (6.74 vs. 6.55 on average;  $P = 0.02$ ) and a higher total area under the pH curve ( $P = 0.01$ ). Concentration of NH<sub>3</sub>-N (1.8 mg N/100mL) and total VFA (84 mM) were not affected by treatment ( $P > 0.10$ ). Heifers fed 10BS presented the lowest proportion of acetate ( $P = 0.05$ ) and the highest proportion of propionate ( $P = 0.04$ ). Molar proportions of butyrate, valerate, isobutyrate and isovalerate were not affected by treatment ( $P > 0.10$ ). Results indicate that the inclusion of alfalfa hay at 19% of total DM in a high-concentrate diet fed to growing heifers did not affect total VFA and NH<sub>3</sub>-N concentration, increased acetate and decreased propionate proportion, and increased mean ruminal pH, which can reduce the risk of ruminal acidosis.

**Key Words:** alfalfa hay, beef cattle, high-concentrate diet

### W290 Protein requirements of Nellore and Angus young bulls.

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We used 48 animals with initial body weight (BW) of 380.2 ± 4.73 kg to determine protein requirements of Nellore and Angus young bulls by comparative slaughter technique. Four animals of each breed were slaughtered at beginning of the experiment (baseline animals). The remainder were housed in individual stalls, where 8 animals of each breed were fed ad libitum with a whole corn shell (WCS) diet (85% WCS and 15% of a pellet based on soybean meal) or a silage/concentrate (SC) diet (30% silage and 70% concentrate based on corn and soybean meal).

Other 4 animals of each breed were fed with the SC diet at 50% of the dry matter intake adjusted for metabolic BW of animals that received the SC diet ad libitum. Intake was measured daily and a metabolism trial was conducted with total collection of feces and urine. These data were used to estimate the crude and metabolizable protein intake (CPI and MPI, respectively). At 90 d of growth the cattle were slaughtered. The net requirement of protein for maintenance (NP<sub>m</sub>, g/kg EBW<sup>0.75</sup>/d) was assumed to be the intercept of the linear regression of the retained protein (RP) on CPI and the slope of the linear regression of RP on MPI was assumed to be the efficiency of protein utilization for maintenance (k). The NP<sub>m</sub> was divided by the k to obtain the metabolizable requirement of protein for maintenance (MP<sub>m</sub>, g/kg EBW<sup>0.75</sup>/d). The net requirement of protein for growth (NP<sub>g</sub>) was calculated by NP<sub>g</sub> (g/kg EBG/day) = a + b × RE, where EBG is the empty body gain and RE is the retained energy. The data were analyzed using the GLM procedures of SAS adopting significance level of 0.05. We did not observe an effect ( $P > 0.05$ ) of breeds on NP<sub>m</sub> (1.01 g/kg EBW<sup>0.75</sup>/d) and k (38%). MP<sub>m</sub> was 2.68 g/kg EBW<sup>0.75</sup>/d. In addition, breed did not significantly affect ( $P > 0.05$ ) on NP<sub>g</sub> (NP<sub>g</sub> g/day = EBG × [286 - 8.23 × (ER/EBG)]). Funded by FAPEMIG, INCT-CA and CNPq.

**Key Words:** maintenance, growth

### W291 Intake and digestibility of Nellore steers fed different sources of forage in diets with crude glycerin in feedlot.

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Nine ruminally cannulated Nellore steers (300.0 ± 30kg of BW and 18 ± 2 mo of age) were used in a 3x3 Latin Square experimental design to evaluate the effect of different sources of forage in diets with crude glycerin (80.64% of glycerol) on DM, CP, NDF and NFC intake and digestibility. The treatments were different sources of forage (fixed 15% of NDF from forage; fNDF): corn silage, sugar cane and sugar cane bagasse, in diets with 10% (DM) of crude glycerin. For this study intake, refusals and total feces collection were recorded for 3 d, experimental periods were 17 d (14 d for adaptation, 3 d for intake and total digestibility). Data were analyzed as a triple Latin square design with 3 treatments and 3 animals in 3 simultaneous triplicates using the PROC MIXED procedure of SAS. The least squares means were generated and compared ( $P < 0.05$ ) using Tukey's test. There was no difference on DM and CP intake among diets. On the other hand, the intake of NDF increased ( $P < 0.05$ ) in animals fed corn silage which did not differ from animal fed with sugar cane bagasse. Furthermore, animals fed with sugar cane bagasse decreased NFC intake ( $P < 0.05$ ). Animals fed with corn silage increased DM and NDF digestibility compared with the other diets ( $P < 0.05$ ). Additionally, an increase on CP digestibility was observed in animals feed with sugar cane bagasse, which not differed from animals fed with corn silage ( $P < 0.05$ ). NFC digestibility increased in animals fed with sugar cane ( $P < 0.05$ ). Different sources of forage included in 15% of fNDF in diets with crude glycerin (10% DM) altered intake and digestibility.

**Key Words:** biofuel, corn silage, sugar cane

**W292 Effect of glycerol supplementation and ambient temperature on growth and metabolic and immunological responses in Korean cattle steers.** Hyeok Joong Kang, Min yu Piao, Hyun Jin Kim, In Kyu Lee, Min Jeong Gu, Cheol Hee Yun, and Myunggi Baik\*, *Seoul National University, Seoul, Republic of Korea.*

This study was performed to evaluate whether glycerol supplementation and ambient temperature affects growth and blood metabolic and immunological parameters in beef cattle. Twenty Korean cattle steers with average 14.4 mo of age and 405 kg of body weight (BW) were used. Animals were divided into conventional control diet group and 2% glycerol supplementation group. Steers were allowed a concentrate diet with the amount of 1.5% of BW and a tall fescue with the amount of 0.75% of BW for 8 weeks. Experimental period 1 (P1) was 4 weeks from July 28 to August 26, and period 2 (P2) 4 weeks from August 27 to September 26. Blood was collected 4 times at July 28, August 11, August 27, and September 26. Maximum temperature-humidity index (THI: 75.8) of August (P1) was higher ( $P < 0.001$ ) than that (69.7) of September (P2), although maximum ambient temperature (27.8°C) of P1 was numerically higher than that (26.3°C) of P2 without statistical significance. Glycerol supplementation did not affect both concentrate and forage intake, average daily gain (ADG), and feed/gain ratio at P1 and P2. ADG was lower ( $P = 0.03$ ) at P2 (0.74 kg/d) compared with P1 (0.89 kg/d). Feed/gain ratio was improved ( $P < 0.001$ ) at P2 compared with P1. Glycerol supplementation did not affect blood concentrations of glucose, triglyceride (TG), cholesterol, high-density lipoprotein (HDL), low density lipoprotein (LDL), nonesterified fatty acid, and albumin. Blood concentrations of TG, cholesterol, HDL, LDL, glucose, and albumin were lowest ( $P < 0.05$ ) at late July compared with other times (August and late September). Glycerol supplementation decreased blood CD8+ T cell population at late July and mid-August. However, it did not affect other blood immune cell populations. Blood granulocyte to lymphocyte ratio was highest at August 27. Monocyte and B cell populations were lowest ( $P < 0.05$ ) at August 11. In conclusion, glycerol supplementation did not affect growth and blood metabolic parameters. Feed efficiency was lower at higher THI August compared with September. Some blood metabolic and immunological parameters were influenced by ambient temperature.

**Key Words:** beef cattle, ambient temperature, glycerol supplementation

**W293 Comparison of fatty acid profiles and volatile compounds among quality grades and their association with carcass and sensory traits in loin and rump of Korean cattle steer.** Min Yu Piao, Hyun Jin Kim, Cheorun Jo, Hyun Joo Kim, and Myunggi Baik\*, *Department of Agricultural Biotechnology, College of Agriculture and Life Science, Seoul National University, Seoul, Republic of Korea.*

This study was performed to compare fatty acid contents and volatile compounds among quality grades (QG) and to understand association between QG and above parameters in loin and rump of Korean cattle steers. Loin and rump samples were obtained from 48 Korean cattle steers with each of 4 quality grades (QG 1++, 1+, 1, and 2; average 32 mo of age). In loin, oleic acid (C18:1n9) contents were highest ( $P < 0.05$ ) in QG 1++ and lowest in QG 2. Linoleic acid (C18:2n6), eicosapentaenoic acid (C20:5: EPA), and polyunsaturated acid (PUFA) contents were lowest ( $P < 0.05$ ) in QG 1++ and highest in QG 2. Palmitic acid, stearic acid, oleic acid, SFA, and MUFA contents were higher ( $P < 0.05$ ) in loin than in rump. In loin, major fatty acid contents per meat, including palmitic acid (C16:0), oleic acid and saturated fatty acid (SFA) and USFA, were highest ( $P < 0.05$ ) in QG 1++ and lowest in QG 2. Marbling score (MS), QG, and overall acceptability of sensory evaluation were

positively correlated ( $r > 0.47$ ,  $P < 0.05$ ) with oleic acid contents of fat. In contrast, MS, QG, tenderness, juiciness, flavor and overall acceptability were inversely correlated ( $r < -0.62$ ,  $P < 0.05$ ) with linoleic acid, EPA, and PUFA contents. MS and QG were positively correlated ( $r > 0.59$ ,  $P < 0.001$ ) with most of fatty acid contents per meat, including palmitic acid, stearic acid, oleic acid, SFA and USFA contents. Total 20 volatile compounds, including aldehydes and hydrocarbons, were detected in loin and rump by GC/MS analysis. Few hydrocarbon contents, including n-pentane, n-hexane, and 2-butene, were higher in QGs 1++ and 1+ than in QGs 1 and 2. Other volatile compounds did not show significant patterns among QGs. MS and QG were positively correlated ( $r > 0.67$ ,  $P < 0.05$ ) with 2-butene, n-pentane, chloroform, and n-hexane contents. Our results revealed that MS and QG are linked with fatty acid contents such as oleic acid in loin. Among 20 volatile compounds detected, only few compounds were associated with MS and QG.

**Key Words:** Korean cattle steers, quality grade, fatty acid

**W294 Effects of essential oils and exogenous enzymes on in vitro ruminal fermentation.** Fabiola A. Lino<sup>1</sup>, Lidiamar L. R. Vieira<sup>1</sup>, Andrea M. Mobiglia<sup>1</sup>, Débora G. Sousa<sup>1</sup>, Fernando R. Camilo<sup>1</sup>, José Tiago Neves Neto<sup>1</sup>, Tiago S. Acedo<sup>2</sup>, Cristina S. Cortinhas<sup>2</sup>, João Ricardo R. Dórea<sup>2</sup>, Luis Fernando M. Tamassia<sup>2</sup>, and Juliano J. R. Fernandes<sup>\*1</sup>, <sup>1</sup>Universidade Federal de Goiás, Goiânia, Goiás, Brazil, <sup>2</sup>DSM Nutritional Products, São Paulo, São Paulo, Brazil.

The objective of this study was to evaluate the inclusion of a blend of essential oils and exogenous enzymes on in vitro ruminal fermentation (dual flow continuous culture fermenters). Ten fermenters with dual continuous flow and they each have 1.4 L capacity. The dilution rate of the liquid and solid content has maintained 10% and 5%.h<sup>-1</sup>, respectively. The design was randomized complete block with 4 replicates. Two experimental periods, each one has 8 d, were considered a block. The ruminal fluid was collected from 5 Nelore steers with ruminal cannula. The experimental period has 8 d with a 5-d adaptation period and 3-d collection period. The diet composition on dry matter (DM) was 8.5% sugarcane bagasse, 82.5% corn, 5% soybean meal and 4% mineral mixture. Each fermenter received daily 100g of the diet (on DM) with additives includes, as follows: T1–300mg of monensin (0.012%, monensin from Tortuga); T2–essential oil (0.008%, Crina Ruminants); T3–blend of essential oil (0.008%, Crina Ruminants) and 300 mg of monensin (0.0024%, monensin from Tortuga); T4–blend of essential oil (0.008%, Crina Ruminants) and exogenous amylase (0.05%, Ronozyme Rumistar); T5– blend of essential oil (0.008%, Crina Ruminants), exogenous amylase (0.05%, Ronozyme Rumistar) and exogenous protease (0.08%, Ronozyme Proact). In this study were evaluated short-chain fatty acids (SCFAs) and the apparent digestibility of DM and crude protein (CP). The data were analyzed by R software and the probability of 5% was considered as statistical difference. There were no statistical differences for all variable studied. The data demonstrate that the use of essential oils and exogenous enzymes and their associations must be studied further because they showed the same fermentation parameters than the monensin treatment. Thus, these additives represent a good alternative to monensin.

*Contd.*

**Table 1 (Abstr. W294).** Production and ratio of SCFA, DM and CP digestibility on in vitro fermentation of diet with high concentrate level and different kind of additives

	Treatments					P-value
	T1	T2	T3	T4	T5	
Total SCFA, mM	149.99	143.44	142.03	147.91	152.10	0.801
SCFA %						
Acetate	50.3	50.7	51.3	53.3	48.4	0.885
Propionate	34.4	27.2	26.8	27.4	34.1	0.465
Butyrate	9.5	17.4	17.0	14.7	12.4	0.176
Valerate	3.21	1.99	1.99	2.00	2.58	0.311
Acetate: propionate	1.57	1.94	1.95	1.90	1.54	0.575
Apparent digestibility (%)						
DM	41.90	43.51	47.96	48.12	43.65	0.614
CP	35.05	34.39	39.69	38.76	39.69	0.664

**Key Words:** amylase, essential oil, protease

**W295 Effect of steam-flaking on in situ degradability and PDI values of maize, wheat and rice.** Fei Wang\*, Yunlong Huo, and Qingxiang Meng, *China Agricultural University, Beijing, China.*

The objective of this study was to investigate effects of cereal type and steam-flaking processing on nutritive values of maize, wheat and rice (with whole hulls). Intact and steam-flaked grains of maize, wheat and rice were measured for in situ degradability using nylon bag method and predicted for protein digested in the small intestine (PDI) using the method of French Protein System. Data obtained were subjected to ANOVA as a 2 × 3 factorial design with 3 replicates by the GLM procedure of SAS (Version 9.0; SAS Institute, Cary), and a Duncan significant difference test was used to determine the differences among means. Significance was declared at  $P \leq 0.05$ . Three ruminally cannulated Limousin steers averaging 400kg of body weight were used. Steers were fed a TMR diet twice daily. The results showed that the rumen effective degradation rate of dry matter (DM), organic matter (OM), starch and crude protein (CP) of the grains were: wheat (80.91%, 81.34%, 87.68% and 81.34%) > rice (71.10%, 72.84%, 84.79% and 72.84%) > maize (65.80%, 63.99%, 74.60% and 63.99%). The rumen degradation rate from fast to slow were: rice > wheat > maize. DM, OM, Starch and CP effective degradability of rice were increased by steam-flaking (SF) processing. SF processing also increased DM, OM, Starch and CP degradation rates of maize and wheat. However, DM, OM, Starch and CP degradation rates of rice were decreased by SF processing. Rumen nondegradable nitrogen from feed (PDIA) and PDI of wheat (50.07 and 99.97g/kg DM) were significantly higher ( $P < 0.01$ ) than maize (34.83 and 55.03 g/kg DM) and rice (23.16 and 46.27 g/kg DM). PDI of maize was reduced ( $P < 0.01$ ) by SF processing, while no significant differences were observed between PDI values of intact and steam-flaked wheat or rice. In summary, rumen effective degradation rate and PDI of wheat are higher than maize and rice. Steam-flaking can significantly improve the rumen effective degradability of rice and decrease the PDI value of maize, but have no significant effect on PDI values of wheat or rice.

**Key Words:** steam-flaking, in situ, protein digested in the small intestine (PDI) DI

**W296 Effects of conventional dietary adaptation over periods of 6, 9, 14, and 21 days on feedlot performance and carcass**

**traits of Nellore cattle.** Daniela Dutra Estevam<sup>1</sup>, Danilo Domingues Millen<sup>2</sup>, Ismael de Castro Pereira<sup>1</sup>, Ramon Argentin Rizzieri<sup>1</sup>, Gabriel Fernandes Melo<sup>1</sup>, Alexandre Perdigão<sup>1</sup>, Cyntia Ludovico Martins<sup>1</sup>, and Mario De Beni Arrigoni<sup>1</sup>, <sup>1</sup>São Paulo State University (UNESP), Botucatu, São Paulo, Brazil, <sup>2</sup>São Paulo State University (UNESP), Dracena, São Paulo, Brazil.

This study, conducted at the São Paulo State University feedlot, Botucatu Campus, Brazil, was designed to determine the effects of adaptation periods of 6, 9, 14 and 21 d on feedlot performance and carcass traits of Nellore cattle. Cattle were fed for 88-d regardless of adaptation period adopted. The experiment was designed as a completely randomized block, replicated 6 times, in which ninety-six 20-mo-old yearling Nellore bulls (391.1 ± 30.9 kg) were fed in 24 pens (4 animals/pen) according to the different adaptation periods adopted: 6, 9, 14, and 21 d. The adaptation program consisted of ad libitum feeding of 3 diets over adaptation periods with concentrate level increasing from 70% to 86% of diet DM. Each of the adaptation diets containing 70.0%, 75.0%, and 80.5% concentrate were fed for 2-d, 3-d and 7-d to cattle adapted for 6-d, 9-d and 21-d; respectively. Also, the adaptation diets containing 70.0%, 75.0%, and 80.5% concentrate were fed for 4-d, 5-d and 5-d, respectively, to cattle adapted for 14-d. The finishing diet contained: 71.5% cracked corn grain, 14.0% sugarcane bagasse, 10.5% peanut meal, 2.5% supplement, 1.0% urea, and 0.5% limestone (DM basis). Cattle were fed ad libitum twice daily throughout the study, and feed offerings and refusals were weighed daily. Orthogonal contrasts were used to evaluate linear, quadratic, and cubic relationship between adaptation periods and the dependent variable. The use of different adaptation periods did not affect ( $P > 0.10$ ) DMI, either expressed in kg or as % of BW. However, as the adaptation period lasted longer, final BW (6-d = 512.0 kg; 9-d = 521.9 kg; 14-d = 527.6 kg; 21-d = 514.8 kg), ADG (6-d = 1.37 kg; 9-d = 1.48 kg; 14-d = 1.56 kg; 21-d = 1.41 kg), G:F ratio (6-d = 0.142; 9-d = 0.153; 14-d = 0.153; 21-d = 0.146), and HCW (6-d = 277.3 kg; 9-d = 283.3 kg; 14-d = 287.8 kg; 21-d = 283.3 kg) were affected ( $P < 0.05$ ) quadratically. Moreover, dressing percentage increased linearly ( $P = 0.05$ ), as the adaptation period lasted longer (6-d = 54.13%; 9-d = 54.27%; 14-d = 54.55%; 21-d = 55.04%). Thus, yearling Nellore bulls should be adapted for 14 d. Grant provided by São Paulo State Foundation (FAPESP), São Paulo, São Paulo, Brazil.

**Key Words:** adaptation, Nellore

**W297 The effect of crude glycerin concentration on fiber digestion in beef calves.** Robert G. Bondurant\*, Jana Harding, Melissa Jolly-Breithaupt, James C. MacDonald, Andrea R. McCain, and Samodha C. Fernando, *University of Nebraska-Lincoln, Lincoln, NE.*

Ruminally and duodenally cannulated steers (n = 7; 363 kg) were utilized in a 7 × 4 Latin rectangle to determine the effects of crude glycerin (GLY) on total-tract digestibility, rate and extent of fiber digestibility an rumen fermentation parameters. Crude glycerin replaced soybean hulls (SH) at 0, 4, 8, or 12% of diet DM. Basal diets consisted of 50% wheat straw (WS), SH, 4% supplement, and soybean meal, to maintain a consistent CP level. Fecal samples, rumen and duodenal fluid were collected 1 h before feeding and at 2, 5, and 8 h post feeding. In situ bags containing ground WS or SH were incubated in the rumen for 0, 6, 12, 16, 24, 48, and 96 h to determine NDF digestion rates. Both DM intake and OM intake, decreased quadratically ( $P = 0.04$ ) as GLY increased in the diet, with lowest DM intake and OM intake at 4% GLY inclusion. As GLY inclusion increased from 0 to 12% diet DM, NDF intake linearly decreased ( $P < 0.01$ ) (5.86 and 5.27kg, respectively). Glycerin inclusion level had no effect on total-tract DM or OM digestibility ( $P \geq 0.66$ ). As



GLY inclusion increased from 0 to 12%, total-tract NDF digestibility decreased linearly ( $P = 0.02$ ) from 64.78% to 53.76%, respectively. The inclusion of GLY had no effect on in situ rate of NDF digestibility ( $P \geq 0.27$ ) for WS or SH. Molar acetate proportion decreased linearly ( $P < 0.01$ ) as time post-feeding progressed as well as GLY increased. Molar propionate concentrations increased ( $P < 0.01$ ) as GLY inclusion increased and time to feeding increased from -1 to 8 h. Butyrate concentration increased ( $P < 0.01$ ) at 2, 5, and 8 h post feeding for 8 and 12% GLY, while concentrations were unchanged for 0 and 4% GLY. Subsequently, acetate to propionate ratio decreased ( $P < 0.01$ ) as GLY level increased from 0 to 12% and time from feeding increased from -1 to 8 h. Although the inclusion of GLY in forage-based diets has a negative impact on NDF digestibility, a decrease in acetate to propionate ratio could result in improved G:F.

**Key Words:** glycerin, fiber, volatile fatty acid

### W298 Effect of supplementing feedlot cattle with live *Saccharomyces cerevisiae* on feed intake and rumen parameters.

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Our objective was to evaluate the effect of supplementation with live yeast (*Saccharomyces cerevisiae* CNCM I-1077) on feed intake and rumen parameters of beef cattle. Eight rumen cannulated steers with 24 mo and weighting 450 kg were used in a duplicated 4x4 Latin square design. Two levels of concentrate (60 or 80% of DM) and 2 treatments (with or without yeast supplementation) were applied, having sugarcane silage as the roughage source. The product was given in capsules, via rumen cannula, to achieve  $8 \times 10^9$  cfu of yeast per animal. Animals were housed individually in tie-stalls with free access to water, and fed ad libitum allowing for 5 to 10% of orts. Feed was offered twice daily and orts weighed daily to measure DMI. Each period lasted for 28d to allow for complete washout of the previous treatment. The last 2 d of each treatment period were used for rumen fluid sample collection for pH and short chain fatty acids (SCFA) measurements. Feeding diets with 80% concentrate increased feed intake (9.4 vs. 8.6 kg  $\pm$  0.4,  $P = 0.05$ ); however, yeast supplementation had no effect on feed intake. Feeding a greater concentrate diet decreased mean rumen pH (6.52 vs. 6.69  $\pm$  0.07,  $P < 0.01$ ), and live yeast supplementation prevented the drop in rumen pH. Feeding the greater concentrate diet increased rumen total concentration of SCFA (43 vs. 36 mM  $\pm$  3,  $P = 0.03$ ), but only without yeast supplementation. Supplementation with live yeast increased the molar proportion of acetate (63.2 vs. 62.4%  $\pm$  0.8,  $P = 0.03$ ), and of valerate (1.23 vs. 1.18%  $\pm$  0.06,  $P = 0.06$ ) in the rumen, and decreased the concentration of butyrate (10.6 vs. 11.4%  $\pm$  0.5,  $P = 0.03$ ). Yeast supplementation promoted changes in rumen fermentation that are associated with better maintenance of a higher pH, and a higher proportion of acetate in the rumen. However, these changes did not translate into a detectable increase of feed intake.

**Key Words:** short chain fatty acids, sugarcane silage, yeast

**W299 Effect of adding virginiamycin in combination with different doses of monensin on feedlot performance.** Raul Lizarraga<sup>1</sup>, Juan Pablo Russi<sup>1</sup>, Luis Casares<sup>2</sup>, Milton Gorocica<sup>2</sup>, and Alejandro Relling<sup>\*1,3</sup>, <sup>1</sup>Fac Cs Veterinarias, UNLP, Argentina, <sup>2</sup>Phibro Animal Health, Argentina, <sup>3</sup>IGEVET, CCT La Plata, CONICET, Argentina.

The objective of this experiment was to evaluate the effect of the addition of virginiamycin (Vm) in combination with different doses of monensin (Mn) on feedlot performance. Two-hundred 40 crossbred steers (189  $\pm$  3 kg) were used in an 80 d experiment. Steers were blocked by BW and randomly distributed in 24 pens, 9 animals each. Steers were fed a finishing diet (90% concentrate) based on whole-shelled corn and milo silage as roughage source. Three dietary treatments were evaluated: T1, basal diet supplemented with 180 mg Mn/hd/d; T2, basal diet supplemented with 200 mg Vm and 180 mg Mn hd/d; and T3, 200 mg Vm and 120 mg Mn hd/d. Body weight was measured on d 0, 40 and 80. Dry matter intake was measured weekly and G:F was estimated for the entire feeding period. Prior to harvest, on d 80, an ultrasound was performed to measure back fat (BF) and ribeye area (REA). Hot carcass weight was measured at harvest. Data were analyzed using a complete randomized block design with block as random effect and treatment as fixed effect. Fisher's protected test was used for mean separation. Pen was used as the experimental unit. Hot carcass weight and G:F were greater and DMI lower in the T2 group (all  $P < 0.05$ , Table 1). Back-fat tended to be greater in T2 ( $P = 0.09$ ), BW on d 40 and 80, and REA were numerically greater in T2 but not statistically different among treatments. Feedlot performance (ADG, G:F) of T3 was numerically greater than that of T1, but differences were not significant (all  $P > 0.10$ ). In conclusion the addition of 200 mg Vm to a higher dose of Mn improved feedlot performance (HCW and G:F).

**Table 1 (Absr. W299).** Feedlot performance and carcass characteristics in cattle supplemented with Vm in diets with two levels of Mn

	T1	T2	T3	P-value
BW d 0, kg	189.4	188.9	188.9	0.80
BW d 40, kg	278.3	282.8	279.6	0.44
BW d 80, kg	302.6	305.8	303.1	0.64
ADG, kg/d	1.42	1.46	1.42	0.73
DMI, kg/d	9.32 <sup>a</sup>	8.59 <sup>b</sup>	9.19 <sup>a</sup>	<0.01
G:F	0.137 <sup>a</sup>	0.150 <sup>b</sup>	0.137 <sup>a</sup>	0.05
Carcass weight, kg	173.5 <sup>c</sup>	178.0 <sup>d</sup>	171.2 <sup>c</sup>	0.05
Ribeye area, cm <sup>2</sup>	52.1	52.4	52.1	0.95
Back fat, cm	0.49	0.53	0.50	0.09

<sup>ab</sup>Values with different superscripts are statistically different at  $P < 0.05$ .

<sup>cd</sup>Values with different superscripts are statistically different at  $P < 0.10$ .

**Key Words:** beef cattle, virginiamycin, monensin

### W300 Evaluation of rumen microbiota in sugarcane silage based diet with different sources of nonfibrous carbohydrates. J.

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With the increasing adoption of sugarcane silage in ruminant feed, it becomes essential to understand the effects of supplementation with different levels and sources of non-fiber carbohydrates (NFC) on ruminal microorganisms. The aim of this study was to characterize the population change of cellulolytic and amylolytic rumen bacteria, caused by the increase of concentrate, and by the use of different sources of NFC in diets with sugarcane silage. Six Nellore beef cattle, castrated, and cannulated in the rumen, were used in a 3 0215 3 Latin square design. The experimental diets were formulated with 2 levels of concentrate: 60% or 80%, and the roughage used was sugarcane silage. Within each level of concentrate inclusion, 3 different sources of NFC were used: steam flaked corn (SFC), pelleted citrus pulp (PCP), or ground corn (GC). SFC and PCP were included in the diet in partial replacement of 70% of GC.

At d 14 of each period, samples of rumen contents were collected for DNA extraction and subsequent analysis of the relative quantification of rumen microorganisms by real time PCR. The increase of concentrate in the diet resulted in a decrease in population of *Fibrobacter succinogenes* ( $P < 0.01$ ) and *Streptococcus bovis* ( $P < 0.01$ ), and in an increased of *Ruminococcus flavefaciens* ( $P = 0.05$ ) and *Megasphaera elsdenii* population ( $P = 0.04$ ), without changing the *Ruminococcus albus* population ( $P = 0.63$ ). The partial replacement of GC by PCP resulted in an increase of *S. bovis* population ( $P = 0.01$ ) and in a reduction of *R. flavefaciens* ( $P < 0.01$ ), without changing *F. succinogenes* ( $P = 0.64$ ). Moreover, the replacement of GC by SFC increased the population of *M. elsdenii* ( $P = 0.03$ ) and reduced *R. albus* ( $P < 0.01$ ). There was a significant Diet\*NFC interaction only for *M. elsdenii* ( $P = 0.02$ ), where SFC increase *M. elsdenii* population only at the 80% concentrate diet. In conclusion, substituting GC for PCP resulted in an increase only in population of *S. bovis*, while substituting GC for SFC resulted in a decrease in population of *R. albus*, and in an increase of *M. elsdenii*, in diets with sugarcane silage as the roughage source.

**Key Words:** beef cattle, rumen microorganism, qPCR

**W301 Effects of rotating antibiotic and ionophore feed additives on enteric methane and volatile fatty acid production of steers consuming a high forage diet.** Whitney Crossland<sup>\*1</sup>, Luis Tedeschi<sup>1</sup>, Todd Callaway<sup>2</sup>, Mike Miller<sup>1</sup>, Brandon Smith<sup>1</sup>, and Matt Cravey<sup>3</sup>, <sup>1</sup>Texas A&M University, College Station, TX, <sup>2</sup>USDA-ARS Southern Plain Region, College Station, TX, <sup>3</sup>Huvepharma Inc., Amarillo, TX.

Feed additives such as ionophores and antibiotics have been shown to decrease ruminal methanogenesis, but evidence as a long-term means of mitigation is lacking. In the present study, we proposed a rotation of feed additives as an alternative to reduce methane (CH<sub>4</sub>) production and to increase animal responses. Rumen-cannulated steers ( $n = 12$ ) were fed a basal high forage diet at 2% of BW (DM) for 13 wk in a Calan gate facility for individual DMI measurement. Steers were randomly assigned to 1 of 6 treatments: (1) control (C) containing the basal forage diet and no additive, (2) bambermycin (B) = C + 20 mg B/hd/d, (3) monensin (M) = C + 200 mg M/hd/d, (4) B7M = rotating B and M treatments weekly, (5) B14M = rotating B and M treatments every 14 d, and (6) B21M = rotating B and M treatments every 21 d. Steers were blocked by weight in a RCB design with repeated measures each wk. Performance data and rumen fluid were collected weekly for in vitro analysis ( $n = 13$ ) and results were interpreted on organic matter intake (OMI) basis. Treatments did not affect ADG. Potential activity of CH<sub>4</sub> (PAM) was greatest for M-fed steers and least for B21M-fed steers (0.219 vs 0.172 mM/kg OMI, respectively;  $P < 0.05$ ). Additionally, PAM of the B21M-fed steers was most consistent of all treatments. Total VFA concentration differed ( $P < 0.05$ ), being greatest for M- and B14M-fed steers (3.46 and 3.47 mM/Kg OMI) and lowest for the B7M treatment (2.87 mM/Kg OMI), but were not different from other treatments. The PAM differed over time for all treatments decreasing toward wk 6 then increasing toward wk 12. Wk also affected total VFA peaking at wk 3 followed by a significant depression in wk 4 (4.02 vs 2.86 mM/Kg OMI;  $P < 0.05$ ). There is an evidence to suggest that weekly rotation of B and M feed additives may not provide additional benefit at either the ruminal or environmental level when compared with continuous feeding of single feed additives. However, a 21-d rotation may combine desirable animal performance, decreased CH<sub>4</sub> emissions, and provide a novel practical approach for industry feeding protocol.

**Key Words:** feed additive, CH<sub>4</sub>, VFA

**W302 Flint corn processing methods and dietary concentrations of roughage NDF for finishing cattle: 1. Intake and digestibility of nutrients.** Antonio Humberto Fleury de Melo<sup>1</sup>, Murillo Alves Porto Meschiatti<sup>1</sup>, João Meneghel de Moraes<sup>1</sup>, Camila Delveaux Araujo Batalha<sup>1</sup>, Lucas Jado Chagas<sup>1</sup>, Débora de Carvalho Basto<sup>1</sup>, Jonas de Souza<sup>1</sup>, Nayana Carla Gonçalves Barbosa<sup>2</sup>, and Flávio Augusto Portela Santos<sup>\*1</sup>, <sup>1</sup>University of São Paulo, Piracicaba, São Paulo, Brazil, <sup>2</sup>Federal University of Goiás, Jataí, Goiás, Brazil.

The objective of this trial was to evaluate intake and total-tract apparent digestibility (AD) of nutrients in finishing diets containing flint corn (70 – 75% vitreous endosperm) either ground (particle size: 2.70 mm) or steam-flaked (bulk density: 360 g/L), combined with dietary contents of 4, 7, 10 or 13% (DM basis) NDF from sugarcane bagasse. Sixteen Nellore bulls (BW = 484.46 ± 7.47 kg) fitted with rumen cannula, were fed diets containing 76–86% corn and randomly allocated to 16 pens for 2 periods (14 d for adaptation and 6 d for collection) in a 2 × 4 factorial arrangement. Total feces collection was carried out for 4 d. During the collection period the amount of diet offered was restricted to 85% of previous ad libitum intake. The data were analyzed using a PROC MIXED model with period and animal as random effect. Linear and quadratic effects were tested for dietary concentrations of roughage NDF. There were no interactions between corn processing and dietary concentrations of sugarcane bagasse NDF. Steam flaking of flint corn tended ( $P = 0.07$ ) to decrease DMI (7.76 vs. 7.28 kg.d<sup>-1</sup>), it decreased ( $P < 0.01$ ) AD of NDF (50.73 vs. 33.12%) and of crude protein (75.05 vs. 71.93%), but it increased ( $P < 0.01$ ) AD of DM (73.45 vs. 77.38%) of TCHO (75.19 vs. 81.93%), of NFC (84.1 vs. 95.16%) and of starch (95.95 vs. 99.36%). Steam flaking increased ( $P < 0.01$ ) dietary TDN concentration (80.26% vs. 84.22%) but had no effect ( $P = 0.73$ ) on TDN intake (6.22 vs. 6.15 kg.d<sup>-1</sup>). Increasing roughage NDF content of the diets caused a quadratic response for DMI (6.95, 7.90, 7.80 and 7.41 kg.d<sup>-1</sup>, respectively) and nutrient intakes and a linear decrease for AD of DM (78.80; 75.95; 75.90 and 72.80% respectively), of TCHO (81.55, 78.76, 78.86 and 75.07% respectively) and had no effect ( $P = 0.40, 0.11, 0.94$  and  $0.29$ , respectively) on AD of NDF, NFC, CP, EE and starch. In conclusion, steam flaking increases energy value of flint corn and feeding around 7% of sugarcane bagasse NDF in finishing diets optimizes energy intake of Nellore bulls.

**Key Words:** feedlot, steam-flaked corn, roughage NDF

**W303 Effect of excess MP supplementation from corn gluten meal or soybean meal on plasma amino acid concentrations of beef cows consuming low quality forage.** Taylor C. Geppert<sup>\*1</sup>, Allison M. Meyer<sup>2</sup>, and Patrick J. Gunn<sup>1</sup>, <sup>1</sup>Department of Animal Science, Iowa State University, Ames, IA, <sup>2</sup>Division of Animal Sciences, University of Missouri, Columbia, MO.

The objective was to determine the effects of excess MP supplementation from feedstuffs differing in rumen degradability of protein on plasma AA concentrations. As part of a larger study, we hypothesized excess MP supplementation from feedstuffs differing in rumen degradability would alter reproductive performance of beef cows around the time of ovulation, potentially due to changing circulating AA concentrations. Non-pregnant, nonlactating mature beef cows ( $n = 18$ ) were allotted by age, BW and BCS to ad libitum corn stalks and 1 of 2 isocaloric, isonitrogenous dietary supplements consisting primarily of either (1) corn gluten meal (GLM) or (2) soybean meal (SBM), supplemented at 150% of NRC MP requirements. Cows were individually supplemented once daily for 58 d and coccygeal blood samples were taken on d 47 for AA analysis. Data were analyzed using the MIXED procedures of SAS.

On d 47, total AA, essential AA, nonessential AA, glycogenic AA and branched-chain AA were not different ( $P \geq 0.21$ ) between treatments. However, total ketogenic AA tended to be greater ( $P = 0.07$ ) in excess GLM than excess SBM supplemented cows. When expressed as a percent of total AA, arginine, lysine, threonine, tryptophan and valine were lesser ( $P \leq 0.05$ ) in GLM than SBM supplemented cows. However as a percent of total AA, leucine, methionine and phenylalanine were greater ( $P \leq 0.03$ ) in GLM than SBM cows. In addition, as a percent of total AA asparagine and proline were greater ( $P < 0.05$ ) in cows supplemented with GLM than SBM. As a percent of essential AA, arginine, lysine, valine, tryptophan and threonine were lesser ( $P \leq 0.04$ ), and methionine, phenylalanine and leucine were greater ( $P \leq 0.03$ ) in GLM than SBM. These data illustrate that supplementing MP at 150% of NRC requirements from feedstuffs differing in rumen degradability may change the profile of circulating plasma AA around the time of ovulation in beef cows consuming low quality forage. However, the effects of this change in AA profile on reproductive function of beef cows are still largely undetermined and warrant further investigation.

**Key Words:** amino acid, corn stalk, protein

**W304 Flint corn processing methods and dietary concentrations of roughage NDF for finishing cattle: 2. Rumen fermentation, N metabolism, and ingestive behavior.** Antonio Humberto Fleury de Melo<sup>1</sup>, Murillo Alves Porto Meschiatti<sup>1</sup>, Camila Delveaux Araujo Batalha<sup>1</sup>, Jonas de Souza<sup>1</sup>, João Meneghel de Moraes<sup>1</sup>, Débora de Carvalho Basto<sup>1</sup>, Lucas Jado Chagas<sup>1</sup>, Nayana Carla Gonçalves Barbosa<sup>2</sup>, and Flávio Augusto Portela Santos<sup>\*1</sup>, <sup>1</sup>University of São Paulo, Piracicaba, São Paulo, Brazil, <sup>2</sup>Federal University of Goiás, Jataí, Goiás, Brazil.

The objective of this trial was to evaluate rumen fermentation, N metabolism and ingestive behavior of cattle fed diets containing flint corn (70 – 75% vitreous endosperm) either ground at 2.70 mm or steam-flaked at 360 g/L, combined with dietary contents of 4, 7, 10 or 13% (DM basis) NDF from sugarcane bagasse. Sixteen Nellore bulls (484.46 ± 7.47 kg) fitted with rumen cannula, were fed diets containing 76–86% corn and randomly allocated to 16 pens for 2 periods (14d adaptation; 6d collection) in a 2 × 4 factorial arrangement. During the collection period the amount of diet offered once a day was restricted to 85% of previous ad libitum intake. Rumen samples were collected on d 16 at 0, 2, 4, 6, 8, 10, 12, 16 and 24 h post feeding. Blood samples and urine spot samples were collected on d 15, 4 h post feeding. Microbial synthesis was estimated based on the purine derivative method. The data were analyzed using a PROC MIXED model with period and animal as random effect. Linear and quadratic effects were tested for dietary concentrations of roughage NDF. No interactions were observed between corn processing and NDF level. Steam-flaking increased ( $P < 0.01$ ) rumen propionate and decreased ( $P < 0.01$ ) butyrate and N-NH<sub>3</sub> concentrations. Steam-flaking decreased ( $P < 0.01$ ) N ingestion, blood urea nitrogen (BUN), N urine and total N excretion and increased ( $P = 0.03$ ) microbial efficiency. Increasing dietary sugarcane bagasse NDF tended to increase linearly ( $P = 0.10$ ) rumen pH and decreased linearly ( $P = 0.05$ ) rumen propionate and ( $P = 0.05$ ) total VFA. Increasing bagasse NDF caused a quadratic effect ( $P < 0.01$ ) on N ingestion and absorption. Rumination and mastication time were increased linearly ( $P < 0.01$ ) as roughage NDF was increased. Blood glucose and BUN were not affected ( $P = 0.53$  and 0.54, respectively) by bagasse NDF level. In conclusion, steam-flaking of flint corn improves energetics and N metabolism of beef cattle. Feeding increasing levels of bagasse

NDF stimulates rumination but it decreases the efficiency of energy production in the rumen.

**Key Words:** feedlot, steam-flaked corn, roughage NDF

**W305 Carcass traits of Nellore cows finished in different times of high intake supplementation in pasture.** Flavio Dutra de Resende<sup>\*1,2</sup>, Ana Paula Reiff Janini<sup>2</sup>, Ivanna Moraes de Oliveira<sup>1</sup>, Aline Domingues Moreira<sup>2</sup>, Fernanda Diamantino dos Santos<sup>3</sup>, Paloma Helena Gonçalves<sup>3</sup>, Michele Aparecida Prado Alves<sup>3</sup>, and Gustavo Rezende Siqueira<sup>1,2</sup>, <sup>1</sup>Agência Paulista de Tecnologia dos Agronegócios, Colina, São Paulo, Brazil, <sup>2</sup>UNESP, Jaboticabal, São Paulo, Brazil, <sup>3</sup>UNIFEB, Barretos, São Paulo, Brazil.

This project aimed to evaluate the carcass traits of Nellore cull cows finished in different times of high intake supplementation in *Brachiaria brizantha* ‘Marandu’ pasture, in dry season. It is assumed that the high intake supplementation in pasture could improve the carcass traits of these animals. The experiment was performed from 09/2014 to 11/2014, assessing 4 times of finishing with high intake supplementation – 2% of BW – in pasture (0, 21, 42 and 63 d). Several 60 Nellore cull cows were used and kept, before the beginning of the study, in system extensive production, pasture plus mineral salt ad libitum. The cull cows were distributed randomly (experimental design in randomized blocks) on 20 experimental units (paddock), composed by 3 animals/paddock and 5 paddocks/supplementation time. At zero time, the cows were conducted directly to slaughter and, at the end of feeding times 21, 42 and 63 d, the remaining cull cows were slaughtered for evaluation of carcass traits. The supplementation time did not alter the carcass length (136.43 cm;  $P = 0.90$ ), carcass depth (45.59 cm;  $P = 0.53$ ) and size of round (87.03 cm;  $P = 0.79$ ) from cull cows. It was a linear increase ( $P < 0.01$ ) on weight and carcass yield, *longissimus* muscle area (LM area) and subcutaneous fat thickness (12th-rib fat). Cull cows finished with 63 d of supplementation at pasture presented increased weight from 203.7 kg to 257.8 kg and the LM area became from 56.9 cm<sup>2</sup> to 70.8 cm<sup>2</sup>, regarding the slaughtered cull cows at d 0. At the same period, it was registered an increase of 3.59 percentage points in carcass yield, initially of 51.5%; as well in 12th-rib fat, passing from 2.02 mm to 6.15 mm. Therefore, the increase in supplementation time applied in finishing of Nellore cull cows allows an improvement the carcass traits.

**Key Words:** carcass, loin eye area, subcutaneous fat thickness

**W306 Sugar cane straw replacing whole corn on growth and intake of feedlot finishing bulls.** Barbara J. M. Lemos<sup>\*1</sup>, Flavio G. F. Castro<sup>2</sup>, Bruno P. C. Mendonça<sup>2</sup>, Dheividu B. Fernandes<sup>2</sup>, and Juliano J. R. Fernandes<sup>1</sup>, <sup>1</sup>Universidade Federal de Goiás, Goiania, Goiás, Brazil, <sup>2</sup>AgroCria, Goiania, Goiás, Brazil.

The effects of increasing amounts of pelleted sugar cane straw (SCS) replacing corn in finishing diets (85% whole corn + 15% pelleted concentrate) of feedlot young bulls were evaluated during a 103 d period. Nellore bulls (n = 80; 337 ± 31 kg of BW) were allotted to 20 outdoor pens (4 bulls per pen) and assigned to a complete randomized block design with 5 blocks (initial BW) and 4 dietary treatments (Table 1): Control (whole corn), SCS3%, SCS6%, SCS9%. Bulls were adapted over 21 d by decreasing inclusion of SCS on dietary DM (d 1 to 7, 30% of SCS; d 8 to 14, 20% of SCS; d 15 to 21, 10% of SCS). The DMI increased linearly up to 23%, as kg/d and 22% as % of BW ( $P = 0.01$ ), 19% on DIP intake ( $P = 0.03$ ) and 39% on peNDF intake ( $P < 0.01$ ) with increased addition of SCS. No effects were observed in NE intake ( $P > 0.05$ ). The Final BW and ADG tended to increase linearly ( $P = 0.10$ ,



BW;  $P = 0.07$ , ADG) with increasing amounts of SCS. In conclusion, SCS is a possible peNDF source replacing whole corn. There were evidences of growth and nutrients intake improvement.

**Table 1 (Abstr. W306).** Composition of finishing diets with increasing amounts of pelleted sugar cane straw (SCS; composition of 92% DM, 6.9% ash, 1.8% CP, 0.5% EE, 69% NDF, 13% peNDF, 37% ADF) replacing corn (3, 6, and 9% of dietary DM) and effects on growth and intake of Nellore bulls fed 85% whole corn + 15% pelleted concentrate

Item	Diet				SEM	P-value	
	Control	SCS3%	SCS6%	SCS9%		Lin	Quad
Diet composition, % of DM							
CP	14.6	14.3	14.1	13.8	—	—	—
DIP	8.1	7.9	7.8	7.7	—	—	—
NDF	14.1	15.8	17.6	19.3	—	—	—
peNDF	5.4	5.6	5.9	6.1	—	—	—
NEm, Mcal/kg	2.10	2.03	1.97	1.90	—	—	—
NEg, Mcal/kg	1.44	1.40	1.35	1.31	—	—	—
Growth							
Initial BW, kg	340.1	338.3	336.5	336.5	1.50	0.95	0.21
Final BW, kg	490.3	499.8	498.6	500.8	6.00	0.10	0.24
ADG, kg/d	1.459	1.567	1.574	1.595	0.052	0.07	0.34
Intake							
DM, kg/d	8.59	9.40	10.05	10.57	0.499	0.01	0.71
DM, % of BW	2.07	2.25	2.41	2.53	0.113	0.01	0.84
NEm+g, Mcal/d							
DIP, kg/d	0.684	0.745	0.784	0.813	0.039	0.03	0.69
peNDF, kg/d	0.461	0.530	0.626	0.641	0.032	<0.01	0.41
G:F	0.170	0.168	0.157	0.153	0.007	0.08	0.91

**Key Words:** beef cattle, byproduct, fiber

**W307 Impact of a molasses-based liquid feed supplement on the diet selection behavior and growth of fattening calves.** Lisa J. Gordon and Trevor J. DeVries\*, *Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada.*

The objective of this study was to assess the impact of a molasses-based liquid feed (LF) supplement on the feed sorting behavior and growth of young, fattening cattle fed a high-grain diet. Twenty-four male Holstein calves,  $90.2 \pm 2.6$  d old and weighing  $137.5 \pm 16.9$  kg, split into groups of 4, were exposed to each of 2 treatments in a crossover design with 35-d treatment periods. Treatments were (1) control diet (76.0% high-moisture corn, 19.0% protein supplement, 5.0% alfalfa/grass haylage), and (2) LF diet (68.4% corn, 17.1% protein supplement; 9.0% molasses-based LF, and 4.5% alfalfa/grass haylage). Diets were designed to support 1.5 kg/d ADG. Data were collected for the final 3 wk of each treatment period. Feed intakes were recorded daily and calves were weighed 2x/wk. Feed samples of fresh feed and refusals were collected 3x/wk for particle size analysis. The particle size separator had 3 screens (19, 8, and 1.18 mm) and a bottom pan, resulting in 4 fractions (long, medium, short, fine). Sorting was calculated as the actual intake of each fraction expressed as a % of its predicted intake. Data were summarized by pen and week and analyzed in a repeated measures general linear mixed model. Calves tended ( $SE = 4.8$ ;  $P = 0.08$ ) to sort for long particles on the control diet (110.5%) and did not sort these particles on the LF diet (96.8%). Sorting for medium particles ( $102.6 \pm 0.6\%$ ) was similar ( $P = 0.9$ ) across diets. Calves sorted against short particles on

the LF diet (97.5%;  $SE = 0.5$ ;  $P = 0.04$ ), but did not sort this fraction on the control diet (99.4%). Calves sorted against fine particles ( $79.3 \pm 4.0\%$ ), to a similar extent ( $P = 0.2$ ), on both diets. DMI was similar across diets (6.1 kg/d;  $SE = 0.1$ ;  $P = 0.9$ ), but day-to-day variability in DMI was higher (0.5 vs 0.4 kg/d;  $SE = 0.02$ ;  $P = 0.04$ ) when calves were fed the control compared with the LF diet. Calves on both diets had similar ADG (1.6 kg/d;  $SE = 0.04$ ;  $P = 0.8$ ) as well as within-pen variability in ADG (0.4 kg/d;  $SE = 0.05$ ;  $P = 0.7$ ). Feed conversion was also similar between control and LF diets (4.3 vs 3.9 kg DM/kg gain;  $SE = 0.3$ ;  $P = 0.4$ ). The results suggest that despite promoting more consistency in DMI, addition of a molasses-based LF to a high-grain, finishing calf diet did not affect calf growth.

**Key Words:** molasses, fattening calf, sorting

**W308 Feedlot performance and carcass traits of Nellore and ½ Angus x Nellore cattle adapted either for 9 or 14 days.**

Daniel Hideki Mariano Watanabe\*<sup>1</sup>, Murillo Ceola Stefano Pereira<sup>2</sup>, Ana Carolina Janssen Pinto<sup>1</sup>, Wilson Inácio Silva Filho<sup>1</sup>, Gustavo Perina Bertoldi<sup>1</sup>, André Luiz Nagatani Rigueiro<sup>1</sup>, Anderson Augusto Santos<sup>1</sup>, Paulo César da Silva Santos<sup>1</sup>, Luis Felipe Rebeschini Oliveira<sup>1</sup>, Pedro Fernando Santi<sup>1</sup>, João Victor Tino Dellaqua<sup>1</sup>, Mário De Beni Arrigoni<sup>2</sup>, and Danilo Domingues Millen<sup>1</sup>, <sup>1</sup>São Paulo State University (UNESP), Dracena, São Paulo, Brazil, <sup>2</sup>São Paulo State University (UNESP), Botucatu, São Paulo, Brazil.

This study, conducted at the São Paulo State University feedlot, Dracena Campus, Brazil, was designed to evaluate the length of adaptation period to high concentrate diets on overall feedlot performance and carcass traits of Nellore (NE) and 1/2 Angus x Nellore (AN) cattle. The experiment was designed as a completely randomized block with  $2 \times 2$  factorial arrangement, replicated 6 times (3 animals/pen), in which seventy-two 22-mo-old yearling bulls [36 NE, and 36 AN] were fed in 24 pens for 89 d according to the treatments: NE adapted for 9-d; NE adapted for 14-d, AN adapted for 9-d, and AN adapted for 14-d. Average initial BW was  $319.2 \pm 18.5$  kg, and  $307.9 \pm 29.5$  kg for NE and AN, respectively. The adaptation program consisted of ad libitum feeding of 3 diets over adaptation periods with concentrate level increasing from 62% to 86% of diet DM. Each of the adaptation diets containing 62%, 70%, and 78% concentrate was fed for 3-d to cattle adapted for 9-d. For cattle adapted for 14-d, the adaptation diets containing 62%, 70%, and 78% concentrate were fed for 5-d, 4-d and 5-d, respectively. The finishing diet contained: 66.5% cracked corn grain, 14.0% sugarcane bagasse, 16.0% cottonseed meal, 1.5% supplement, 1.2% urea, and 0.8% limestone (DM basis). Cattle were fed ad libitum twice daily throughout the study, and feed offerings and refusals were weighed daily. No significant ( $P > 0.10$ ) adaptation length main effects were observed for any of the feedlot performance and carcass traits variables evaluated. However, significant ( $P < 0.01$ ) biotype main effects were observed, in which AN yearling bulls had greater final BW (466.6 kg vs. 425.8 kg), ADG (1.71 kg vs. 1.27 kg), DMI expressed in kg (11.8 kg vs. 9.5 kg), and DMI expressed as % of BW (3.07% vs. 2.53%), improved G:F ratio (0.146 vs. 0.134), and heavier HCW (243.6 kg vs. 229.0 kg). No significant ( $P = 0.74$ ) biotype main effect was observed for dressing percentage (AN = 53.5%; NE = 53.2%). Thus, AN yearling bulls performed better than NE yearling bulls regardless of length of the adaptation period adopted. Cattle should be adapted for 14-d, because longer adaptation periods are safer. Grant provided by São Paulo State Foundation (FAPESP), São Paulo, São Paulo, Brazil.

**Key Words:** adaptation, biotype

**W309 Rumen morphometrics of Nellore and ½ Nellore x Angus cattle adapted either for 9 or 14 days to high-concentrate diets.** André Luiz Nagatani Rigueiro\*<sup>1</sup>, Daniel Hideki Mariano Watanabe<sup>1</sup>, Murillo Ceola Stefano Pereira<sup>2</sup>, Wilson Inácio Silva Filho<sup>1</sup>, Gustavo Perina Bertoldi<sup>1</sup>, Ana Carolina Janssen Pinto<sup>1</sup>, Anderson Augusto Santos<sup>1</sup>, Mariana Squizatti<sup>1</sup>, Daniela Dutra Estevam<sup>2</sup>, Lais Aquino Tomaz<sup>1</sup>, Osvaldo Alex Souza<sup>1</sup>, and Danilo Domingues Millen<sup>1</sup>, <sup>1</sup>São Paulo State University (UNESP), Dracena, São Paulo, Brazil, <sup>2</sup>São Paulo State University (UNESP), Botucatu, São Paulo, Brazil.

This study, conducted at the São Paulo State University feedlot, Dracena Campus, Brazil, was designed to evaluate the length of the adaptation period to high concentrate diets on rumen morphometrics and rumenitis of Nellore (NE) and 1/2 Angus × Nellore (AN) cattle. The experiment was designed as a completely randomized block with 2 × 2 factorial arrangement, replicated 6 times (3 animals/pen), in which seventy-two 22-mo-old yearling bulls [36 NE (319.2 ± 18.5 kg), and 36 AN (307.9 ± 29.5 kg)] were fed in 24 pens for 89 d according to the treatments: NE adapted for 9-d; NE adapted for 14-d, AN adapted for 9-d, and AN adapted for 14-d. The adaptation program consisted of ad libitum feeding of 3 diets over adaptation periods with concentrate level increasing from 62% to 86% of diet DM. Each of the adaptation diets containing 62%, 70%, and 78% concentrate was fed for 3-d to cattle adapted for 9-d. For cattle adapted for 14-d, the adaptation diets containing 62%, 70%, and 78% concentrate were fed for 5-d, 4-d and 5-d, respectively. The finishing diet contained: 66.5% cracked corn grain, 14.0% sugarcane bagasse, 16.0% cottonseed meal, 1.5% supplement, 1.2% urea, and 0.8% limestone (DM basis). At harvest, rumenitis incidence (RUM) was determined, on the entire washed rumen, using a scale of 0 (no lesions noted) to 10 (severe ulcerative RUM). Likewise, a 1-cm<sup>2</sup> fragment of each rumen was collect from cranial sac. The number of papillae per cm<sup>2</sup> of rumen wall (NOP) was determined, as well as the mean papillae area (MPA). The rumen wall absorptive surface area in cm<sup>2</sup> was calculated as follows: 1 + (NOP × MPA) – (NOP × 0.002). No significant ( $P > 0.10$ ) biotypes and adaptation length main effects were observed for any of the rumen morphometrics variables evaluated. However, a significant ( $P < 0.01$ ) biotype main effect was observed for RUM, where NE yearling bulls had greater RUM scores than AN yearling bulls (2.05 vs. 1.35). No significant ( $P = 0.15$ ) adaptation length main effect was observed for RUM. Thus, NE yearling bulls are more sensitive to rumen epithelium damage, regardless of length of the adaptation period adopted. Grant provided by São Paulo State Foundation (FAPESP), São Paulo, São Paulo, Brazil.

**Key Words:** adaptation, rumenitis

**W310 Relationships of feedlot performance and rumen morphometrics of Nellore cattle differing in phenotypic residual feed intake.** Murillo Ceola Stefano Pereira<sup>2</sup>, Gustavo Durante Cruz<sup>3</sup>, Mario De Beni Arrigoni<sup>2</sup>, Juliana Silva<sup>1</sup>, Tássia Veluma Barbosa Carrara<sup>2</sup>, and Danilo Domingues Millen\*<sup>1</sup>, <sup>1</sup>São Paulo State University (UNESP), Dracena, São Paulo, Brazil, <sup>2</sup>São Paulo State University (UNESP), Botucatu, São Paulo, Brazil, <sup>3</sup>Cargill Animal Nutrition, Elk River, MN.

The objective of this study was to examine the relationship of feedlot performance, DMI variation and rumen morphometrics of Nellore cattle classified by residual feed intake (RFI). A study was conducted, in 2 consecutive years, using individual pens at São Paulo State University feedlot, Dracena campus, Brazil. At year 1, forty-eight 20-mo-old Nellore bulls (358.2 ± 19.4 kg) were fed for 94 d. At year 2, sixty 20-mo-old Nellore bulls (402.5 ± 33.0 kg) were fed for 84-d. All Nellore bulls

were categorized as high RFI (RFI > 0.5 SD above the mean, n = 25), medium RFI (RFI ± 0.5 SD from the mean, n = 56) and low RFI (RFI < 0.5 SD below the mean, n = 27). The finishing diet, in both years, contained: 71.5% cracked corn grain, 16.0% sugarcane bagasse, 7.7% soybean meal, 3.5% supplement, and 1.3% urea (DM basis). Variables were analyzed by a linear model with year and RFI groups as the main effects, with interactions. Animals classified as low RFI had lower ( $P = 0.01$ ) initial BW (high = 382.5 kg; medium = 378.1 kg; low = 371.8 kg), daily DMI in kg (high = 10.1 kg; medium = 9.5 kg; low = 8.2 kg), daily DMI expressed as % of BW (high = 2.32%; medium = 2.18%; low = 1.92%), and improved ( $P = 0.01$ ) G:F ratio (high = 0.112; medium = 0.131; low = 0.143). However, no significant ( $P > 0.10$ ) RFI effect was observed for ADG (high = 1.14 kg; medium = 1.25 kg; low = 1.16 kg), final BW (high = 484.0 kg; medium = 489.5 kg; low = 474.7 kg) and HCW (high = 266.7 kg; medium = 262.7 kg; low = 256.1 kg). Moreover, animals categorized as high RFI had lower ( $P = 0.02$ ) DMI variation (high = 12.28%; medium = 15.14%; low = 16.24%). No significant ( $P > 0.10$ ) RFI effect was observed for rumenitis (high = 1.4; medium 1.4; low = 1.1), number of papillae (high = 45.6; medium = 43.6; low = 43.1), and absorptive surface area (high = 25.4 cm<sup>2</sup>; medium = 26.3 cm<sup>2</sup>; low = 22.7 cm<sup>2</sup>). Nonetheless, Nellore bulls ranked as low RFI presented smaller ( $P = 0.07$ ) mean papillae area (high = 0.55 cm<sup>2</sup>; medium = 0.60 cm<sup>2</sup>; low = 0.50 cm<sup>2</sup>). Selection of low RFI Nellore bulls is feasible because it does not negatively affect feedlot performance, HCW, and rumen characteristics.

**Key Words:** papillae, Nellore

**W311 Effects of crude glycerin on feed intake and apparent total-tract digestibility of finishing diets in crossbred heifers.** E. H. C. B. van Cleef\*<sup>1,3</sup>, S. Uwituze<sup>2</sup>, C. A. Gilis<sup>2</sup>, C. L. Van Bibber-Krueger<sup>2</sup>, K. A. Miller<sup>2</sup>, C. C. Aperce<sup>2</sup>, and J. S. Drouillard<sup>2</sup>, <sup>1</sup>São Paulo State University, Jaboticabal, São Paulo, Brazil, <sup>2</sup>Kansas State University, Manhattan, KS, <sup>3</sup>FAPESP, São Paulo, Brazil.

Expansion of the biodiesel industry worldwide has increased availability of crude glycerin, a major by-product of biodiesel production from animal and vegetable oils. Our objective was to evaluate DMI and apparent total-tract digestibilities of DM, OM, and NDF in crossbred heifers fed finishing diets in which 15% crude glycerin was used to replace a portion of dry-rolled corn. Twenty-four crossbred heifers (334.4 ± 0.9 kg initial BW) were randomly assigned to 12 pens and allowed ad libitum access to isonitrogenous finishing diets containing 10% corn silage and 90% concentrate composed of dry-rolled corn, soybean hulls, corn gluten feed, soybean meal, mineral premix, and 0 (CON) or 15% crude glycerin (GLY). Glycerin contained 81.5% glycerol, 13.3% water, 6.3% ash, and <0.02% methanol. Animals were fed chromic oxide (0.1% of diet DM), and fecal grab samples were collected 3 times daily (8, 16, and 24 h after feeding) on d 7, 21, and 35; composited by animal; dried; and ground through a 1-mm screen. Chromic oxide contents of feces and diets were determined by atomic absorption and used to calculate apparent total-tract digestibilities of DM, OM, and NDF. Weights of feed DM delivered and refused were recorded daily to calculate DMI. Data were analyzed as a completely randomized design with repeated measures using a mixed model with diet, time, and diet × time as fixed effects, and animal within diet × time as a random effect. There were no interactions between treatment and time of sampling ( $P > 0.10$ ), suggesting no adaptive response to dietary glycerin. Feed DMI was similar among treatments (10.93 and 10.92 kg/d for CON and GLY, respectively;  $P > 0.1$ ). Organic matter digestibility of GLY diet was greater than that of the CON diet ( $P = 0.04$ ), and DM digestibility followed a similar trend ( $P = 0.06$ ). Digestibility of NDF was unaffected

by glycerin addition ( $P > 0.10$ ). Crude glycerin can effectively replace dry-rolled corn in diets for beef heifers when fed at 15% of diet DM, improving OM digestion without adversely affecting NDF digestibility.

**Key Words:** apparent total-tract digestibility, by-product, glycerin

**W312 Impact of the nutritional plan in the growing phase on performance of Nellore cattle during the finishing phase.** Ivanna Moraes de Oliveira\*<sup>1</sup>, Matheus Henrique Moretti<sup>2</sup>, João Alexandrino Alves Neto<sup>3</sup>, Aline Domingues Moreira<sup>4</sup>, Rodolfo Maciel Fernandes<sup>4</sup>, João Marcos Beltrame Benatti<sup>5</sup>, Gustavo Rezende Siqueira<sup>1,4</sup>, and Flávio Dutra de Resende<sup>1,4</sup>, <sup>1</sup>Agência Paulista de Tecnologia dos Agronegócios, Colina, São Paulo, Brazil, <sup>2</sup>Agroceres Multimix, Rio Claro, São Paulo, Brazil, <sup>3</sup>Campos Rações e Minerais, Acreúna, Goiás, Brazil, <sup>4</sup>UNESP, Jaboticabal, São Paulo, Brazil, <sup>5</sup>Trouw Nutrition/Bellman, Mirassol, São Paulo, Brazil.

The effects of nutritional plans (NP) in the growing phase on the performance of Nellore cattle during the finishing phase were evaluated. Our hypothesis was that the NP in the growing phase affect the body weight gain rate (BWGR) in the finishing phase, and that there is an interaction between the NP. The evaluated NP were characterized by the combination of different types of supplementation in the dry season (post-weaning - phase I) e wet season (phase II) (both growing phase) and finishing stage (dry season - phase III). In phase I, the animals received protein supplementation (PS; 0.1% BW) or protein-energy supplementation (PES; 0.5% BW); in phase II, mineral salt (MS; ad libitum), or PES (0.5% BW); and in phase III, the animals under the different NP in the growing phase (PS/MS; PS/PES; PES/MS and PES/PES) received 1.5 or 2.0% BW of supplement. Animals were maintained on a Marandu-grass pasture at all phases. Animals were weighed after 16 h of solid-feed deprivation. The BWGR was calculated as  $(ADG \div \text{average BW of the animal} - \text{in each phase}) \times 100$ . The experimental design was completely randomized, with a 2 (supplements in phase I)  $\times$  2 (supplements in phase II)  $\times$  2 (supplements in phase III) factorial arrangement;  $n = 5$  (animals as replicate). The  $t$ -test was applied at 10% probability. No interaction was detected ( $P > 0.10$ ) between phases I, II, and III. In addition, the NP supplied in phase I did not influence ( $P > 0.10$ ) ADG, BWGR, or the final BW in phase III. The ADG during phase III was influenced only by the nutritional history of phase II. In phase III, the animals fed MS (1.02 kg) during phase II had a greater ( $P = 0.02$ ) ADG than those fed PES (0.92 kg). Likewise, the animals fed MS had a higher BWGR in phase III than those fed PES (0.27 vs. 0.21 kg/100 kg BW, respectively). In phase II, the animals fed PES gained 60.3 kg more than those which received MS. At the end of phase III, this difference became 47.2 kg, i.e., 78% of the gain obtained in phase II was maintained. The nutritional plan utilized during the growing phase, before finishing (wet season), alters the gain rate in the finishing. Trouw Nutrition/Bellman provided financial support, and FAPESP provided the fellowship grant.

**Key Words:** growth, historic nutritional, supplementation

**W313 Performance of grazing Nellore cattle finished with high-intake supplementation.** Flavio Dutra de Resende\*<sup>1,3</sup>, Beatriz Lima Vellini<sup>1</sup>, Ivanna Moraes de Oliveira<sup>1</sup>, João Marcos Beltrame Benatti<sup>2</sup>, Matheus Henrique Moretti<sup>2</sup>, Rodolfo Maciel Fernandes<sup>3</sup>, Aline Domingues Moreira<sup>3</sup>, and Gustavo Rezende Siqueira<sup>1,3</sup>, <sup>1</sup>Agência Paulista de Tecnologia dos Agronegócios, Colina, São Paulo, Brazil, <sup>2</sup>Trouw Nutrition/Bellman, Mirassol, São Paulo, Brazil, <sup>3</sup>UNESP, Jaboticabal, São Paulo, Brazil.

The effects of high intake supplementation on the performance of Nellore cattle finished on *Brachiaria brizantha* 'Marandu' pastures was evaluated. It was hypothesized that the performance Nellore cattle finished under high intake supplementation and with a high forage allowance is improved when corn is used as the source of energy, and that under conditions of low forage allowance, citrus pulp is the most suitable source of energy. Two forage allowances (FA; 2.5 and 4.5 kg DM kg<sup>-1</sup> BW) and 2 sources of energy (SE; corn and pulp included in the diet at 2% of the BW) were evaluated. In the experiment, were used 48 (48) uncastrated Nellore cattle at 30 mo of age, with an initial BW of 430.0  $\pm$  2.7 kg. The animals were blocked according to body weight and the data evaluated in randomized blocks in a 2  $\times$  2 factorial arrangement (FA and SE). The paddock was considered the experimental unit, with 3 replicates per treatment (4 animals/paddock). There was no interaction between SE and FA ( $P > 0.10$ ). None of the factors led to any differences ( $P > 0.10$ ) in carcass yield (58.8%), average daily gain (1.22 kg day<sup>-1</sup>) or gain yield (carcass gain/weight gain: 805.6 g kg<sup>-1</sup>). The higher FA resulted in an increase in BW (559.0 kg;  $P = 0.06$ ) as compared with the animals on lower FA (534.0 kg). However, no difference was detected for carcass gain (0.98 kg day<sup>-1</sup>;  $P = 0.28$ ), carcass weight (322.2 kg;  $P = 0.11$ ), subcutaneous fat thickness (SFT; 4.31 mm;  $P = 0.24$ ), or loin eye area (LEA; 78.4 cm<sup>2</sup>;  $P = 0.49$ ) for the animals under different FA. Corn and pulp did not alter the final BW (546.5 kg;  $P = 0.19$ ). Contrastingly, the carcass gain (1.02 kg day<sup>-1</sup> vs 0.93 kg day<sup>-1</sup>;  $P = 0.09$ ), the carcass weight (327.6 kg vs 316.8 kg;  $P = 0.09$ ), and the SFT (4.72 mm vs 3.89 mm;  $P = 0.18$ ) were greater in the animals fed corn as compared with pulp. Citrus pulp provided animals with a larger LEA ( $P < 0.01$ ) as compared with corn (81.0 cm<sup>2</sup> vs 75.8 cm<sup>2</sup>). It is recommended to use a forage allowance of 4.5 kg DM kg<sup>-1</sup> BW and corn as the source of energy. Nutreco/Bellman provided support.

**Key Words:** carcass, energy source, forage allowance

**W314 Different supplementation strategies to grazing beef cattle in tropical conditions.** Bruna B. Menezes<sup>2</sup>, Henrique J. Fernandes\*<sup>1</sup>, Maria G. Morais<sup>2</sup>, Marina N. Bonin<sup>3</sup>, Gelson L. D. Feijó<sup>3</sup>, Ednéia P. Rosa<sup>1</sup>, Lucy M. Surita<sup>1,3</sup>, Yasmin S. Falcão<sup>1</sup>, and Amanda A. Perestrelo<sup>1</sup>, <sup>1</sup>State University of Mato Grosso do Sul, Aquidauana, MS, Brazil, <sup>2</sup>Federal University of Mato Grosso do Sul, Campo Grande, MS, Brazil, <sup>3</sup>EMBRAPA Beef Cattle, Campo Grande, MS, Brazil.

The objective of this study was to evaluate the effect of different supplementation strategies on the average daily gain (ADG) and carcass characteristics of Nellore bulls grazing Piatã grass (*B. brizantha* cv. BRS Piatã) pastures from the weaning until a Brazilian commercial slaughter weight of 450 kg. Thirty 2 bulls (initial BW of 189  $\pm$  15.7 kg) from the State University of Mato Grosso do Sul, Aquidauana, Brazil, were randomly distributed in 4 treatments (8 animals/experimental units per treatment): mineral supplementation (Control), continuous concentrate supplementation (Continuous), concentrate supplementation from weaning until a BW of 320 kg (Growing), and concentrate supplementation from 320 kg until 450 kg BW (Finishing). Concentrate supplement used was Recrimax AC (until BW of 320 kg) and Lipomax (BW from 320 to 450 kg) (Real H, Campo Grande, MS, Brazil) and was supplied at 0.5% of body weight. Recrimax AC and Lipomax were formulated with corn, soybean meal, urea, molasses, rumen-protected fat (Lactoplus), salt (sodium chloride), dicalcium phosphate, virginiamycin, and a premix with methionine, lysine, minerals and vitamins A, D and E. These supplements had 75.6 and 85.1% OM, 24.3 and 18.6% CP, 1.34 and 2.3% EE, 7.1 and 12.3% NDF, 44.2 and 54.2% NFC, 68.3 and 77.6% TDN, respectively. Mineral supplement was supplied ad libitum.



To calculate ADG, animals were weighted (after 16 h of solid fasting) at the beginning of the trial and at the slaughter day. After slaughter, measurements of loin eye areas (LEA) and fat thickness at the 12th rib (FT) were taken. The effect of the concentrate supplementation, of the continuous or tactical (just in part of the animal's life) concentrate supplementation, and of the phase of the tactical supplementation were evaluated by the partition of the sum of squares of treatment in orthogonal contrasts. ADG was greater to supplemented animals and to continuous compared with tactical concentrate supplementation ( $P < 0.05$ ), and did not differ between phases of tactical concentrate supplementation (0.449, 0.611, 0.519 and 0.557 kg/d for Control, Continuous, Growing and Finishing treatments, respectively). Concentrate supplementation and continuous supplementation (when compared with tactical supplementation) also increased ( $P < 0.05$ ) LEA (64.6, 88.8, 72.0 and 77.6 cm<sup>2</sup> for Control, Continuous, Growing and Finishing, respectively). FT (1.59, 1.72, 2.05 and 1.85 mm for Control, Continuous, Growing and Finishing, respectively) was not affected ( $P > 0.05$ ) by the treatments. Acknowledgments to FUNDECT – Foundation to Support development of education, science and technology of the state of Mato Grosso do Sul, for financial support of the project.

**Key Words:** carcass, concentrate, Nelore

**W315 Effect of high-fiber concentrate diets on intake, performance, carcass traits and meat quality of feedlot Nelore heifers.** J. M. B. Ezequiel<sup>\*1</sup>, P. S. N. Oliveira<sup>1</sup>, R. L. Galati<sup>2</sup>, D. S. Ferreira<sup>1</sup>, V. C. Santos<sup>1</sup>, A. C. Homem Junior<sup>1</sup>, and E. H. C. B. van Cleef<sup>1</sup>, <sup>1</sup>São Paulo State University, Jaboticabal, São Paulo, Brazil, <sup>2</sup>Federal University of Mato Grosso, Cuiabá, Mato Grosso, Brazil.

The use of fiber-rich by-products, as corn germ meal, could prevent metabolic disorders in feedlot cattle fed high-concentrate diets, avoiding losses in performance. Therefore, the objective of this study was to evaluate high-concentrate diets with corn germ meal totally replacing corn grain on performance, carcass traits, and meat sensory characteristics of feedlot Nelore heifers. Twenty-eight heifers (200 ± 22.5 kg BW) were randomly assigned to 4 treatments in a 2 × 2 factorial arrangement: 2 concentrate levels (70 and 80%), and 2 energy sources (corn grain and corn germ meal). Corn silage was used as roughage, and concentrate was composed of soybean hulls, sunflower meal, mineral premix and corn or corn germ meal. Animals were gradually adapted to the finishing isoenergetic and isonitrogenous diets over a 6-wk period. Weights of feed delivered and refused were recorded daily and composited samples were analyzed for DM, CP and aNDF contents. At the end of 112-d feedlot period, heifers were transported for 100 km to a commercial abattoir. There was no interaction ( $P > 0.05$ ) of concentrate levels and energy sources for DM intake, which was not affected by treatments. The concentrate level of 80% and corn, allowed the greatest CP (1.17 kg/day) and aNDF (4.05 kg/day) intakes. Final BW ( $P < 0.05$ ) and ADG ( $P < 0.01$ ) were decreased when corn germ meal was used. Dressing percentage, HCW, LMA, as well as BF were unaffected by treatments. Carcass muscle and fat were greater for animals fed 80% concentrate diets ( $P < 0.05$ ). Preference and texture of *Longissimus* muscle were greater when animals were fed 80% concentrate diets with corn germ ( $P < 0.05$ ). In conclusion, high proportions of concentrate containing corn as energy source seems to provide best performance, but the total replacement of corn with corn germ meal in high-concentrate diets promote better sensory characteristics of meat from finishing Nelore heifers.

**Key Words:** by-product, cattle, corn germ meal

**W316 Use of virginiamycin in Nelore cattle fed mineral supplement or protein supplement.** Gustavo Rezende Siqueira<sup>\*1,3</sup>, Michele Aparecida Prado Alves<sup>2</sup>, Paloma Helena Gonçalves<sup>2</sup>, Ivanna Moraes de Oliveira<sup>1</sup>, Rodolfo Maciel Fernandes<sup>3</sup>, Guilherme Felipe Berti<sup>2</sup>, Fernanda Diamantino dos Santos<sup>2</sup>, and Flávio Dutra de Resende<sup>1,3</sup>, <sup>1</sup>Agência Paulista de Tecnologia dos Agronegócios, Colina, São Paulo, Brazil, <sup>2</sup>UNIFEB, Barretos, São Paulo, Brazil, <sup>3</sup>UNESP, Jaboticabal, São Paulo, Brazil.

The objective of this study was to evaluate the use of virginiamycin (VM) and the supplementation level on the performance of Nelore cattle in their post-weaning phase grazing on *Brachiaria brizantha* 'Marandu' during the wet season. We hypothesized that VM works on the gram-positive bacteria by inhibiting processes that result in loss of energy, modulating the ruminal fermentation and promoting an improvement in animal performance. The following supplements were utilized in this experiment: mineral (ad libitum) and protein (0.1% of BW), with or without inclusion of VM. The experiment was conducted from January to March 2014, divided into 3 25-d periods. Eighty (80) post-weaning uncastrated Nelore, with an average initial BW of 205.4 ± 21.9 were used in the experiment. Animals were weighed at the end of each period, after a solid food-deprivation period of 16 h. Data were analyzed in randomized blocks with repeated measures over time, in 2 × 2 factorial arrangement in which the factors were the supplements and lack or inclusion of VM. The paddock was considered the experimental unit, with 4 paddocks per treatment (5 animals/paddock). Means were compared by the *t*-test at 10% probability, and a trend at 15%. There was no interaction between VM and the supplementation level ( $P = 0.78$ ). It was considered that there was an upward trend of 0.06 kg in ADG for animals fed VM ( $P = 0.16$ ), which resulted in the increased final BW of the animals fed VM (269.5 vs 264.8;  $P = 0.09$ ). The protein supplement provided an additional gain of 0.09 kg/day as compared with the mineral supplement, which represented a 11% higher ADG ( $P = 0.03$ ) and an increase in BW (271.7 vs 262.6;  $P = 0.01$ ). The highest ADG ( $P < 0.01$ ) was observed in the first period (1.01 kg), decreasing by 48% in the second period, and increasing by 0.30 kg/day in the third period as compared with the second; this variation was due to the climatic conditions. The use of VM and protein supplement improves animal performance during the wet season. Acknowledgments: Paraíso Nutrição Animal.

**Key Words:** additive, supplements, wet season

**W317 Finishing strategies for grazing Nelore cattle in the wet season.** Gustavo Rezende Siqueira<sup>\*1,3</sup>, Paloma Helena Gonçalves<sup>2</sup>, Michele Aparecida Prado Alves<sup>2</sup>, Ivanna Moraes de Oliveira<sup>1</sup>, Rodolfo Maciel Fernandes<sup>3</sup>, Carlos Carvalho Marcolino<sup>2</sup>, Fernanda Diamantino dos Santos<sup>2</sup>, and Flavio Dutra de Resende<sup>1,3</sup>, <sup>1</sup>Agência Paulista de Tecnologia dos Agronegócios, Colina, São Paulo, Brazil, <sup>2</sup>UNIFEB, Barretos, São Paulo, Brazil, <sup>3</sup>UNESP, Jaboticabal, São Paulo, Brazil.

The effects of supplementation strategies on the performance of Nelore cattle finished on Marandu-grass pastures (wet season) were evaluated. It was expected that with the increased energy density at the final stage of finishing there would be improvement in animal performance. The experimental period (EP) was from 12/2013 to 05/2014 (five 28-d periods). Seventy-two uncastrated cattle with an average initial BW of 409.3 ± 17.8 kg were used in the experiment. The following nutritional plans were adopted: mineral supplement (MS; ad libitum), 140 d of the EP; energy supplement (ES; 0.7% of the BW), 140 d of the EP; MS/high supplementation on pasture (MHS; MS on the first 70 d of the EP and supplementation at 2% BW in the last 70 d of the EP). At the end of

each period, the animals were weighed (after 16 h of feed deprivation) to determine the average daily gain (ADG). To calculate the carcass gain and carcass gain percent (carcass gain/weight gain), 6 animals were slaughtered at the beginning of the EP (control slaughter). The experimental design was of randomized blocks, analyzed as a repeated measure over time, in which the experimental unit was the paddock (6 animals/paddock). There was interaction between periods and nutritional plans on ADG ( $P < 0.01$ ). In the first 2 periods, animals fed ES showed a 27% higher ADG ( $P < 0.01$ ) than the others. In the last 2 periods, however, MHS (1.83 kg/day) provided a greater ( $P < 0.01$ ) ADG as compared with ES (1.27 kg/day) and MS (0.92 kg/day). Carcass gain in the animals fed MHS was 56% and 16% higher ( $P < 0.01$ ) than in those receiving MS and ES, respectively. Nutritional plan MHS increased ( $P$

$< 0.01$ ) the carcass gain percent by 5% and 20% as compared with the animals fed MS and ES, respectively. Dressing percent and final carcass weight were higher ( $P < 0.01$ ) in the cattle that received MHS (58.7%; 355.2 kg), followed by those that received ES (56.8%; 333.0 kg) and then MS (52.8%; 281.9 kg). A greater uptake of nutrients, through increased supplementation during the final stage of finishing, improves the animal performance by increasing carcass gain percent, weight, and dressing percent. Acknowledgments: Trouw Nutrition/Bellman.

**Key Words:** high supplementation on pasture, nutritional plans, wet season

## Ruminant Nutrition: Dairy III

**W318 Milk yield and composition of Holstein cows fed increasing levels of amylolytic enzyme.** Caio Seiti Takiya\*<sup>1</sup>, Gustavo Delfino Calomeni<sup>1</sup>, Thiago Henrique Annibale Vendramini<sup>1</sup>, Thiago Henrique Silva<sup>1</sup>, Guilherme Gomes Silva<sup>1</sup>, Jessica Cristiane Bertoni<sup>1</sup>, Carlos Eduardo Cardoso Consentini<sup>1</sup>, Rodrigo Gardinal<sup>1</sup>, Jefferson Rodrigues Gandra<sup>2</sup>, José Esler Freitas Jr.<sup>3</sup>, and Francisco Palma Rennó<sup>1</sup>, <sup>1</sup>Departamento de Nutrição e Produção Animal da Universidade de São Paulo, Pirassununga, São Paulo, Brazil, <sup>2</sup>Faculdade de Ciências Agrárias da Universidade de Grande Dourados, Itahum, Mato Grosso do Sul, Brazil, <sup>3</sup>Departamento de Zootecnia da Universidade Federal da Bahia, Ondina, Bahia, Brazil.

The objective of this study was to evaluate the effects of increasing doses of amylolytic enzyme in diet of dairy cows on milk yield and composition. The enzyme used comes from *Aspergillus oryzae* extract enriched with  $\alpha$  amylase activity (Amaize, Alltech Inc., Nicholasville, KY). The extract is in powdered form and was offered hand mixed into concentrate. Twenty-four multiparous Holstein cows (average 120 DIM, 32 kg/d of milk yield and 580kg of live weight) were distributed in 4 × 4 Latin square design with 21-d periods and received the following treatments: (1) Control, without Amaize; (2) A150, diet containing 150 FAU/kg of Amaize (diet DM basis); (3) A300, diet containing 300 FAU/kg DM of Amaize (diet DM basis) diet; (4) A450, diet containing 450 FAU/kg of Amaize (diet DM basis). One FAU (fungal  $\alpha$ -amylase unit) is the amount of enzyme which breaks down 1g starch in 1 h based on 11–25 min at pH 4.7 and 30°C. Experimental diet was formulated to achieve 30% of starch content. Corn silage was used as forage source and diet contained 48:52 ratio of forage and grain mixture. Cows were mechanically milked twice daily, and samples were collected proportionally from each milking on d 15, 16 and 17 of each experimental period. Milk samples were analyzed for fat protein and lactose (Milkoscan; Foss Electric, Hillerod, Denmark). Data were subjected to ANOVA and regression using PROC MIXED of SAS (2001). No differences were observed in milk yield and composition (Table 1). Inclusion of exogenous amylolytic enzyme did not improve milk yield and composition of mid lactating cows.

**Table 1 (Abstr. W318)**

Item	Treatment <sup>1</sup>				SEM	P-value <sup>2</sup>	
	0	A150	A300	A450		L	Q
Milk yield, kg/d	32.81	32.72	32.34	32.40	1.37	0.275	0.829
3.5% FCM, kg/d	34.78	34.87	34.19	34.60	1.28	0.543	0.714
Fat, kg/d	1.26	1.28	1.24	1.26	0.05	0.594	0.917
Protein, kg/d	1.02	1.01	1.00	1.01	0.02	0.348	0.212
Fat, %	3.85	3.93	3.86	3.89	0.10	0.793	0.549
Protein, %	3.09	3.09	3.09	3.09	0.02	0.924	0.631

<sup>1</sup>0, 150, 300 or 450 FAU/kg (DM basis) of Amaize (Alltech Inc., Nicholasville, KY).

<sup>2</sup>Linear and quadratic effect.

**Key Words:** a-amylase, *Aspergillus oryzae*, starch

**W319 Biofortified milk: Selenium and vitamin E in cow's diet to improve nutritional components in milk.** Marcia S. V. Salles\*<sup>1</sup>, Arlindo Saran Netto<sup>2</sup>, Luiz C. Roma Junior<sup>1</sup>, Marcus A. Zanetti<sup>2</sup>, Karina Pfrimer<sup>3</sup>, and Fernando A. Salles<sup>1</sup>, <sup>1</sup>APTA, Ribeirão

Preto, SP, Brazil, <sup>2</sup>FZEA/USP, Pirassununga, SP, Brazil, <sup>3</sup>FMRP/USP, Ribeirão Preto, SP, Brazil.

Healthy nutrition is a preoccupation of most of the world's population, thus the importance of animal science studies to improve milk nutrient composition. The aim was to study the vitamin E and selenium with sunflower oil (SFO) added to the diet of lactating cows to improve the nutrient profile of milk. Twenty-eight cows were allocated in 4 treatments, as follows: C (control diet); O (4% of SFO in dry matter (DM) diet); A (3,5 mg/ kg DM of organic selenium + 2000 IU of vitamin E/ cow per day); OA (4% of SFO in DM diet + 3,5 mg/ kg DM of organic selenium + 2000 IU of vitamin E/cow per day). Cows were fed with 0.50 of concentrate, 0.42 of corn silage and 0.08 of coast-cross hay (DM). Blood and milk were taken in the last week of trial and analyzed for selenium and  $\alpha$ -tocopherol. Data were analyzed as a RCBD with a factorial treatment structure (GLM/SAS). The addition of selenium and vitamin E in the cow's diet increased selenium and  $\alpha$ -tocopherol serum (0.083, 0.085, 0.337 and 0.389  $\mu$ g/L,  $P < 0.0001$ , 0.022 SEM of selenium and 4.51, 6.26; 6.92 and 8.97 mg/dl,  $P = 0.0009$ , 0.66 SEM of  $\alpha$ -tocopherol for C, O, A and OA, respectively) and milk (0.011, 0.027, 0.235 and 0.358  $\mu$ g/L,  $P < 0.0001$ , 0.033 SEM of selenium and 2.27, 1.56; 3.08 and 2.89 mg/L,  $P = 0.0088$ , 0.36 SEM of  $\alpha$ -tocopherol for C, O, A and OA, respectively). SFO supplementation increased  $\alpha$ -tocopherol in cow's blood ( $P = 0.0097$ ). The antioxidants added to the diet of lactating cows improved the nutrient profile of milk. Financial support: FAPESP.

**Key Words:** nutrient, mineral, vitamin

**W320 Effect of fermented corn silage density and gas filled porosity on corn silage pH and fermentation end-products.** William L. Braman\*, John E. Kurtz, and Keith A. Bryan, *Chr. Hansen Inc., Milwaukee, WI.*

Increasing density (D) of ensiled corn forage by proper harvest dry matter, chop length and silo packing results in decreased presence of oxygen which enhances bacterial fermentation of corn silage (CS). This research measured the effect of CS D and calculated gas filled porosity (P) (Holmes and Muck, 2006) on selected fermentation characteristics. From December, 2012 to July, 2013, 46 dairy operations in the Midwest United States ranging in size from approximately 300 to 4,500 cows (62,500 total) participated in a study to evaluate the effect of silo density (kg/meter<sup>3</sup>) of ensiled CS on the pH, lactic acid (LA), total volatile fatty acids (VFA), ammonia (NH<sub>3</sub>), and ethanol (ETOH), all % of CS DM, of the corresponding corn silage. A minimum of at least 6 core samples from the face of CS bunkers or drive-over piles was consolidated and a composite sample sent to a commercial lab (Rock River Laboratories, Watertown, Wisconsin) for analysis using wet chemistry methods. Some dairies were sampled more than once as they changed sources of CS. The average D was 292.6 kg/meter<sup>3</sup> (range 174.6–350.8). The REG procedure of SAS was used to analyze the relationship between D/P and the dependent variables. There were negative relationships between D and pH ( $R^2 = 0.31$ ,  $P < 0.001$ ), NH<sub>3</sub> ( $R^2 = 0.25$ ,  $P < 0.001$ ), and ETOH ( $R^2 = 0.14$ ,  $P = 0.003$ ). There were positive relationships between D and LA ( $R^2 = 0.26$ ,  $P < 0.001$ ), VFA ( $R^2 = 0.17$ ,  $P < 0.05$ ). Gas filled P was calculated as an alternative measurement of CS D with 0.4 or lower P recommended for proper fermentation. The CS calculated P average was 0.32 (range 0.06–0.69). Regression analysis indicated a negative relationship between P and LA ( $R^2 = 0.23$ ,  $P < 0.001$ ) and VFA ( $R^2 =$



0.18,  $P = 0.001$ ). A positive relationship between P and pH ( $R^2 = 0.30$ ,  $P < 0.001$ ) was observed. These results suggest that increased CS bunker/pile D measured by silage face core sampling is strongly correlated with improved fermentation characteristics of low pH,  $\text{NH}_3$ , ETOH and high VFA and LA concentrations. Gas filled P is an alternative measurement that was correlated with the increased fermentation acids measured and a decreased pH. This work demonstrates the importance of high D on improving the fermentation quality of CS.

**Key Words:** silage, ensiling, density

**W321 Productive performance of dairy cows fed saturated and unsaturated fatty acids sources in the transition period and early lactation.** Gustavo D. Calomeni<sup>\*1</sup>, Rodrigo Gardina<sup>1</sup>, Jose Esler Freitas Junior<sup>2</sup>, Cybele E. Araújo<sup>1</sup>, Filipe Zanferari<sup>1</sup>, Caio S. Takiya<sup>1</sup>, Thiago H. A. Vendramini<sup>1</sup>, Victor C. Galvão<sup>1</sup>, and Francisco P. Renno<sup>1</sup>, <sup>1</sup>Department of Nutrition and Animal Production, School of Veterinary Medicine and Animal Science, University of São Paulo, Pirassununga, São Paulo, Brazil, <sup>2</sup>School of Veterinary Medicine, Federal University of Bahia, Ondina, Bahia, Brazil.

The objective of the present study was to evaluate milk yield and composition of dairy cows receiving saturated or unsaturated fatty acids (FA) sources during transition period and early lactation. Thirty Holstein cows were divided into 3 experimental groups in randomized complete design. The animals were randomly assigned to receive one of the following treatments: 1) control (C; without fat addition,  $n = 10$ ), basal diet containing (DM basis) 2.5 g/100g of ether extract (EE) in pre and postpartum period; 2) calcium salts of saturated FA (SAT; Magnapac - Tectron Ltd.,  $n = 10$ ), basal diet containing (DM basis) 4.5 g/100g of EE in pre-partum and 5.5 g/100g in postpartum; 3) whole raw soybeans (UNSAT;  $n = 10$ ), basal diet containing (DM basis) 4.5g/100g of EE in pre-partum and 5.5g/100g of EE in postpartum period. The animals were fed individually, and the experimental diets were fed since 35 d before the expected calving date, and provided until 90 d of lactation and were formulated to meet the nutritional requirements of each period (pre and postpartum). Milk yield was measured daily and samples for milk composition were collected weekly from the first to the 13th week of lactation. Data were analyzed using PROC MIXED of SAS 9.1, with the effect of diet, time and interaction as fixed effects, and animal as random effect. The data were analyzed by orthogonal contrasts (C vs. UNS + SAT and UNS vs. SAT). Fatty acids sources did not influence ( $P > 0.05$ ) milk yield, protein and lactose (kg/d and %). The average milk yields were 30.07, 28.86, and 30.85 kg for C, UNS and SAT, respectively. Unsaturated fatty acids source reduced the FCM (28.12 Kg/d vs 33.63 kg/d) ( $P = 0.029$ ), and milk fat (4.16% vs. 3.37%; 1.23 kg/d vs 0.95 kg/d) ( $P = 0.006$ ) ( $P = 0.004$ ) for UNS vs SAT contrast. The supplementation of dairy cows with fatty acids sources in the transition period and early lactation affected the FCM and milk fat yield and percent.

**Key Words:** milk yield, milk composition, fat source

**W322 Embryo production and oocyte quality of dairy cows fed saturated and unsaturated fatty acids in transition period and early lactation.** Gustavo D. Calomeni<sup>\*1</sup>, Rodrigo Gardinal<sup>1</sup>, Filipe Zanferari<sup>1</sup>, Caio S. Takiya<sup>1</sup>, Thiago H. A. Vendramini<sup>1</sup>, Jose Esler Freitas Junior<sup>2</sup>, Cybele E. Araújo<sup>1</sup>, Victor C. Galvão<sup>1</sup>, and Francisco P. Renno<sup>1</sup>, <sup>1</sup>Department of Nutrition and Animal Production, School of Veterinary Medicine and Animal Science, University of São Paulo, Pirassununga, São Paulo, Brazil, <sup>2</sup>School of Veterinary Medicine, Federal University of Bahia, Ondina, Bahia, Brazil.

The objective of the present study was to evaluate embryo production and oocyte quality of dairy cows receiving saturated or unsaturated fatty acids (FA) sources during transition period and early lactation. Thirty Holstein cows were divided into 3 experimental groups in randomized complete design. The animals were randomly assigned to receive one of the following treatments: 1) control (C; without fat addition,  $n = 10$ ), basal diet containing (DM basis) 2.5 g/100g of ether extract (EE) in pre and postpartum period; 2) whole raw soybeans (UNSAT;  $n = 10$ ), basal diet containing (DM basis) 4.5g/100g of EE in pre-partum and 5.5g/100g of EE in postpartum period; 3) calcium salts of saturated FA (SAT; MAGNAPAC - Tectron Ltd.,  $n = 10$ ), basal diet containing (DM basis) 4.5 g/100g of EE in pre-partum and 5.5 g/100g in postpartum. The experimental diets were fed from 35 d before the expected calving date, and provided until 90 DIM, formulated to meet the nutritional requirements of each period (pre- and postpartum). The procedure for ovum pick-up (OPU) was performed in 3 periods:  $30 \pm 7$  DIM (OPU1),  $60 \pm 7$  DIM (OPU2) and  $90 \pm 7$  DIM (OPU3). After OPU, the viable oocytes were classified in grade I, II and III. Only oocytes with grade I, II and III were submitted to in vitro fertilization (IVF). Data were analyzed using PROC MIXED of SAS 9.1, with the effect of diet, OPU and interaction as fixed effects, and animal as random effect. The data were analyzed by orthogonal contrasts (C vs. UNS + SAT and UNS vs. SAT). No effects ( $P > 0.05$ ) were observed for OPU, treatment and interaction (treatment\*OPU). No differences were observed among contrasts for total oocytes, viable oocytes, oocytes grade and total embryo produced. The saturated and unsaturated FA supplementation did not influence embryo production and oocyte quality in postpartum period.

**Key Words:** fat source, ovum pick-up, transition period

**W323 Effect of supplemental OmniGen AF and either a negative or positive DCAD prepartum on intake and blood and urine metabolites from dry off through 10 week postpartum.** J. K. Bernard<sup>\*1</sup>, Z. Wu<sup>4</sup>, D. J. Hurley<sup>2</sup>, A. L. Jones<sup>3</sup>, K. P. Zanzalari<sup>4</sup>, J. D. Chapman<sup>4</sup>, and D. J. McLean<sup>4</sup>, <sup>1</sup>Animal and Dairy Science, University of Georgia, Tifton, GA, <sup>2</sup>College of Veterinary Medicine, University of Georgia, Athens, GA, <sup>3</sup>College of Veterinary Medicine, University of Georgia, Tifton, GA, <sup>4</sup>Phibro Animal Health Corp., Quincy, IL.

Multiparous Holsteins (80) were enrolled 60 d before expected parturition through 70 DIM to evaluate the effects of feeding OmniGen AF (OG) and Animate (AN, Phibro Animal Health Corp., Quincy, IL) on DMI, blood and urine metabolites, and milk yield and composition. Cows were blocked by expected calving date and assigned randomly within block to one of 8 treatment combinations arranged as a  $2 \times 2 \times 2$  factorial to provide: 0 (OG-) or 57 g OM (OG+) beginning at dry off through parturition; 0 (POS) or 0.59 kg/d AN (NEG) beginning 28 d before predicted calving; and OG- or OG+ postpartum. Three cows were dropped from the trial due to reasons unrelated to treatments. During the far-off period, no differences were observed in DMI: 13.7 and 14.8 kg/d for OG- and OG+, respectively. Cows fed OG+ had higher ( $P < 0.05$ ) BUN and urinary pH and K concentrations compared with OG-. During the close up period, cows fed NEG had lower ( $P < 0.05$ ) DMI compared with POS; 13.9 and 15.7 kg/d for NEG and POS, respectively. Concentrations of K, Cl, ionized Ca, and ionized Mg were higher ( $P < 0.05$ ) for NEG (4.59, 104.05, 1.28, and 0.68) compared with POS (4.46 mEq/mL, 102.06 mEq/mL, 1.26 mmol/mL, and 0.66 mmol/mL, respectively). Immediately postcalving (0 - 2 DIM), cows fed NEG had lower concentrations of globulin, and urinary pH and Na concentrations but higher Cl ( $P < 0.05$ ): 2.93, 7.59, 31.51, and 55.99 compared with 3.17 g/dl, 7.95, 53.69 mEq/L, and 37.7 mEq/L for NEG and POS, respectively.

Aspartate aminotransferase was greater ( $P < 0.05$ ) for OG- compared with OG+: 127.16 and 106.21 U/L, respectively. No differences were observed in DMI or milk yield postpartum but cows fed OG+ had lower ( $P < 0.05$ ) SCC compared with OG-. Milk fat percentage was higher for cows fed OG+ postpartum compared with those fed OG+ prepartum only. No interactions were observed among OG and AN. Carryover effects of NEG fed prepartum on blood mineral concentrations were observed 0–2 DIM. Supplemental OG did not affect postpartum milk yield but decreased SCC consistent with previous research.

**Key Words:** Animate, blood metabolite, DCAD

**W324 Characterization of ingredient loading accuracy on commercial dairy farms in North America and Europe feeding total mixed rations.** Michael C. Barry\*, *AgModels LLC, Tully, NY.*

Successful implementation of precision feeding is predicated on the assumption that farms can accurately deliver the correct amounts and proportions of ingredients during the feed mixing process. Thirty-three commercial dairy herds of varying management styles across North America and Europe feeding total mixed rations (TMR) were surveyed with respect to ingredient loading accuracy. Across farms, 386,011 individual feed loading records were electronically recorded and compiled during the calendar year of 2014. Deviations from target load amounts were calculated as the difference between actual and the targeted amount of ingredient, both in absolute (kg) and relative (percent of target) terms. Absolute bullseyes were defined as those records with a deviation less than 5 kg from the target and averaged 24.1% across all observations, ranging from 4.3% to 73.9% between farms (median 19.8%). Relative bullseyes were defined as those records with a deviation less than 2% from the target and averaged 46.7% across all observations, ranging from 10.8% to 80.6% between farms (median 42.7%). Absolute deviation values that exceeded an absolute value greater than 62.5 kg from the target were designated as absolute outliers, and represented 11.85% of all values, ranging from 1.3 to 35.3% between farms (median 13.1%). Relative deviation values that differed more than 20% of target values were designated as relative outliers and represented 14.3% of all values and ranged from 2.7 to 40.3% between farms (median value of 15.1%). Several nonsensical values (those with actual values <0 kg) were identified and stratified by farm. 3.8% of all values were deemed as nonsensical and ranged from 0.7% to 10.2% between farms. Deviations were skewed toward positive values, indicating a tendency to overload ingredients. These results indicate a wide range of proficiency of feed loading across farms studied, and would suggest that there exists opportunities to assess and improve quality control procedures and the feed loading protocols on dairy farms feeding total mixed rations.

**Key Words:** total mixed ration, loading accuracy

**W325 Effects of hydroxy versus sulfate forms of trace minerals in milk replacer or starter on dairy calves through weaning.**

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Young dairy calves fed at a higher plane of nutrition may benefit from trace mineral (TM) sources with greater bioavailability, but few data are available for hydroxy forms of Zn, Cu, and Mn. The goal of this study was to evaluate differences in intake, growth, and health status of calves fed milk replacer and starter grain with different sources of TM. Male Holstein calves ( $n = 64$ ) < 1 wk old were transported from a commercial farm to the research facility. Calves were assigned to treat-

ments in a  $2 \times 2$  factorial arrangement of TM source in milk replacer and TM source in starter grain in a randomized complete block design. All calves were fed milk replacer (28% crude protein, 20% fat) at a fixed feeding rate [700 g/d of powder for wk 1, 950 g/d of powder for wk 2–6, and 450 g/d of powder for wk 7] and had ad libitum access to starter [22% CP] and water. Milk replacers were formulated to contain either sulfate (S) or hydroxy (H) TM (70, 10, and 50 mg/kg of Zn, Cu, and Mn, respectively). Starters formulated either with STM or HTM contained 150, 30, and 100 mg/kg of Zn, Cu, and Mn, respectively. All calves were weaned on d 49 and continued to have ad libitum access to water and starter until the end of the experiment at d 63. Body weights and measurements were taken on all calves on a weekly basis. Fecal and respiratory scores were monitored daily and any use of medications was recorded. Final body weight and average daily gain (overall mean of ADG = 0.91 kg/d) did not differ among treatments. Calves fed the HTM milk replacer plus STM starter had greater hip height ( $P = 0.03$ ) and hip height average daily gain ( $P = 0.007$ ). Starter intake was greater in calves fed HTM milk replacer than those fed STM milk replacer ( $P < 0.0001$ ). Calves fed the STM milk replacer had a greater frequency of elevated fecal scores in the first 3 wk of life (odds ratio STM to HTM = 1.80), although use of medication was not different among treatments. Results indicated that calves fed HTM milk replacer and starter had similar overall growth as calves fed STM but maintained a better health status throughout the pre-weaning phase.

**Key Words:** dairy calf, mineral, hydroxy

**W326 Effects of different forage supplement patterns on the growth of Holstein calves.** Zhaohai Wu, Shengli Li, and Zhijun Cao\*, *State Key Laboratory of Animal Nutrition, College of Animal Science and Technology, China Agricultural University, Beijing, Beijing, China.*

This study was conducted to investigate the effects of 2 forage sources supplemented from different time points on the growth performance of Holstein bull calves. Forty Holstein bull calves with similar body weights and birthdate were selected and randomly assigned to 5 treatments with 8 replicates each: control (CON) calves were fed starter feed only, and the other 4 treatments consisted of the same starter feed plus 2 forage sources started from different time points: chopped alfalfa hay started at the 15th day (AW) and the 3rd day (AD), chopped oat hay started at the 15th day (OW) and the 3rd day (OD). All calves were weaned at 56 d, and the study finished one week later. In the pre-weaning period, the starter feed intake was not significantly affected by forage supplement. Moreover, starter feed and total dry matter intake were increased in the first week of post-weaning by feeding forage from the 15th day and providing oat hay. In the pre-weaning period, the average daily gain of calves supplemented forage from the 15th day tended to be higher compared with that from the 3rd day ( $P = 0.079$ ). In the first week of post-weaning, the average daily gain of calves fed forage from the 15th day was significantly higher compared with that from the 3rd day ( $P = 0.045$ ) and CON calves ( $P = 0.032$ ). Meanwhile, the body length of calves pre-weaning improved by feeding oat hay ( $P = 0.071$ ), and the heart girth of calves fed forage from the 15th day tended to be greater than CON calves ( $P = 0.089$ ) in the first week post-weaning. The contents of LDH, AKP, GOT and GPT in the serum of calves were improved by supplementing oat hay and providing forage from the 15th day ( $P < 0.05$ ). The incidence of diarrhea in CON calves were numerically higher than the calves provided forage. In conclusion, forage supplementing did not adversely affect the growth and intake of calves in the pre-weaning period, and the growth performance of calves was improved in the first

week of post-weaning by providing forage from the milk-feeding period, especially offering oat hay from the 15th day.

**Key Words:** calves, oat hay, alfalfa hay

### **W327 Effects of acidified and pasteurized waste milk on calf**

**diarrhea occurrence.** J. Y. Ma<sup>\*1</sup>, X. X. Ren<sup>1</sup>, H. T. Shi<sup>1</sup>, G. Guo<sup>2</sup>, X. Z. Li<sup>2</sup>, and Z. J. Cao<sup>1</sup>, <sup>1</sup>State Key Laboratory of Animal Nutrition, China Agricultural University, Beijing, China, <sup>2</sup>Beijing Capital Agribusiness Group Co., Ltd., Beijing, China.

**Abstract:** Waste milk is unfit for human consumption because it might be harmful for human health. However, it may be acceptable for calf feeding if properly processed. This study was designed to investigate the effect of 2 different sterilization methods of waste milk on diarrhea occurrence of calves. Forty female Holstein calves were randomly assigned into one of 2 groups and fed either acidified (by formic acid) or pasteurized waste milk and defined as treatment group (TT) and control group (CT). The total number of bacteria in milk was detected and found higher in pasteurized milk than in acidified milk ( $3.6 \times 10^4$  vs.  $3.8 \times 10^3$  cfu/mL) on average. The birth weights of calves in 2 groups were similar (TT:  $43.38 \pm 4.100$ ; CT:  $43.83 \pm 4.854$ ;  $P > 0.05$ ). All calves were fed 4 L of colostrum within 1 h after birth and were subsequently fed milk 3 times per day at 0730, 1430, and 1930. Fecal samples were collected from all calves daily for 8 weeks from the third day after birth. Feces were scored by UW-Madison calf health scoring. Calf diarrhea occurrence was described by 3 indexes including diarrhea rate (DA), diarrhea frequency (DF) and fecal indexes (FI). The calculation equations are as bellow. Data were statistically analyzed using the Two-Sample *t*-test for means procedure of SAS (version 9.2, SAS Institute Inc., Cary, NC). Diarrhea rate = number of diarrhea calves/total number of calves  $\times$  100%; diarrhea frequency = ( $\sum$  number of diarrhea calves  $\times$  days in diarrhea)/total number of calves  $\times$  number of days on trial  $\times$  100%; fecal index = the sum of fecal score/(total number of calves). Results indicate that the overall calf diarrhea in TT group was lower compared with CT group. DF and FI of TT group were lower than those of CT group (DF:  $10.00 \pm 0.02$  vs.  $15.9 \pm 0.02\%$ ; FI:  $1.41 \pm 0.28$  vs.  $1.59 \pm 0.25$ ;  $P < 0.05$ ). DR values were not significantly different between two groups during the whole trial period of 8 weeks ( $P > 0.05$ ); however, DA, DF and FI within the first 6 weeks (d3 to d42) was lower in TT group than those in CT group (DR: 60 vs. 90%; DF:  $3.75 \pm 1.07$  vs.  $7.50 \pm 1.10\%$ ; FI:  $1.34 \pm 0.029$  vs.  $1.50 \pm 0.03$ ;  $P < 0.05$ ). We conclude that feeding waste milk acidified with formic acid can effectively reduce calf diarrhea occurrence compared with feeding pasteurized waste milk.

**Key Words:** acidification, pasteurization, calf diarrhea

### **W328 Investigating the impact of dietary changes on rumen microbial community during the transition period in Holstein dairy cows using high-throughput sequencing.**

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The transition period is a critical time in the life of a dairy cow and is associated with a dietary shift from a high-forage-based diet (HF) to a high-grain based (HG) diet. This study investigated the impact of such a dietary shift on the structure of the rumen bacterial population. Fourteen Holstein dairy cows were used in this experiment. Rumen digesta samples were obtained using an oral stomach tube 3 weeks before calving (HF) and 9 weeks postcalving (HG). Twenty-eight

samples were collected, and bacterial 16S rRNA genes were sequenced using Roche 454 pyrosequencing with titanium chemistry. Processing of sequences, performing  $\alpha$  and  $\beta$  diversities, and classifying sequences were conducted using MOTHUR. A total of 198,276 non-chimeric sequences were generated. Those sequences were assigned to 16,833 operational taxonomic units based on a 95% genetic similarity for each sample. Shannon and Inverse Simpson indices were calculated for each sample, and revealed an overall reduction ( $P < 0.05$ ) in the diversity of the rumen bacterial population when cows were shifted from the HF to the HG (from  $5.4 \pm 0.2$  to  $4.3 \pm 0.2$  and from  $153 \pm 26$  to  $47 \pm 26$ , respectively). UniFrac was used to test whether HF samples were different from HG samples. The normalized-weighted *P* test was  $< 0.001$ , indicating a significant difference between the 2 bacterial communities; and the calculated UniFrac metric value was 0.968, indicating that most of the branch length belonged to one community or the other. Sequences from HF were allocated mainly into the following phyla: Bacteroidetes ( $38 \pm 4\%$ ), an unclassified group ( $33 \pm 2\%$ ), Firmicutes ( $23 \pm 3\%$ ), and Proteobacteria ( $5 \pm 4\%$ ), whereas samples from HG were allocated mainly into the Proteobacteria ( $36 \pm 4\%$ ), Bacteroidetes ( $34 \pm 4\%$ ), Firmicutes ( $15 \pm 3\%$ ), and an unclassified group ( $14 \pm 2\%$ ). ANOVA with time as the fixed effect showed that Proteobacteria and the unclassified group were significantly different ( $P < 0.001$ ) between HF and HG, and Firmicutes had a tendency ( $P = 0.07$ ) to be different.

**Key Words:** dairy cow, transition period, 16S rRNA

### **W329 Metabolic profile and onset of puberty in dairy heifers fed reduced-fat distillers grains in replacement of forage.**

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Our objective was to determine the effect of increasing the inclusion rate of reduced-fat distillers dried grains (RFDDGS) in replacement of forage in limit-fed diets on the metabolic profile and onset of puberty in dairy heifers. A 16-wk randomized complete block design feeding study was conducted using 48 Holstein heifers ( $199 \pm 2$  d of age) with 3 treatments. Treatments were 30% RFDDGS with 68.5% grass hay (30DG), 40% RFDDGS with 58.5% grass hay (40DG), and 50% RFDDGS with 48.5% grass hay (50DG) on a DM basis. All diets also contained 1.5% mineral mix. Rations were limit-fed at 2.65, 2.50, and 2.35% of BW on a DM basis for 30DG, 40DG, and 50DG, respectively to have similar intakes of CP and energy among treatments. Jugular blood samples were collected on 2 d during wk 0, 4, 8, 12, and 16 for metabolite and metabolic hormone analysis. When heifers weighed 200 kg, coccygeal vein blood samples were taken twice per wk for progesterone analysis to estimate onset of puberty. Blood samples continued until cycling was confirmed via ultrasound for the presence of a corpus luteum. There were no interactions of treatment by wk for any of the metabolites and metabolic hormones measured. Glucose (76.3, 77.7, and 77.3 mg/dL; SEM = 1.60 for 30DG, 40DG, and 50DG, respectively) and leptin (4.42, 4.35, 4.59 ng/mL; SEM = 0.088) were similar ( $P > 0.05$ ) among treatments. There was a quadratic effect ( $P < 0.05$ ) for cholesterol (93.5, 89.2, and 97.1 mg/dL; SEM = 2.16), plasma urea nitrogen (17.8, 17.8, and 19.9 mg/dL; SEM = 0.35), and a quadratic tendency ( $P = 0.05$ ) for IGF-1 (102.7, 100.0, and 109.4 ng/mL; SEM = 3.59). Age at puberty (234.6, 244.3, and 235.5 d; SEM = 13.7) and BW at puberty (246.4, 261.3, and 254.0 kg; SEM = 24.9) were similar ( $P > 0.05$ ) among treatments. These results demonstrate that heifers can be limit-fed diets with greater inclusion rates of RFDDGS and maintain energy status without



accumulating excess adipose tissue as indicated by leptin. Treatments had no detrimental effects on age or BW at puberty; however, at this time no measures were made on reproductive performance post-trial.

**Key Words:** distillers grains, metabolic profile, dairy heifer

**W330 Gastrointestinal tract of healthy 1-week-old Jersey calves is well suited to digest, absorb, and incorporate nutrients into lean tissue even when fed a high plane of milk replacer.** Yu Liang\*<sup>1</sup>, Tyler L. Harris<sup>1</sup>, Jeff A. Carroll<sup>2</sup>, and Michael A. Ballou<sup>1</sup>,

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This study investigated the digestibilities of nutrients as well as nitrogen (N) retention of Jersey calves fed different planes of milk replacer nutrition. Twelve calves were blocked by BW at birth and randomly assigned to either a low (LPN) or high (HPN) plane of milk replacer nutrition. The LPN calves were fed 14.5 g DM/kg BW of a 20% protein and 20% fat milk replacer/d, and HPN calves 20 g DM/kg BW of a 28% protein and 20% fat milk replacer/d. Calves were fed twice daily at 0700 and 1900. All calves were fed 3 L of pooled colostrum within 6 h of birth and then were assigned treatments. Calves were given 1 d to adapt to treatments. The study was divided into two for the last 24 h of each period. Blood samples were collected at the beginning and end of each period and analyzed for plasma glucose and urea N concentrations. All data reported as HPN vs LPN, respectively. Fecal scores were greater (2.01 vs 1.52 ± 0.13;  $P = 0.004$ ) for HPN calves during both periods; however, there was no difference (30.9 vs 31.9 ± 0.6%;  $P \geq 0.253$ ) in the DM percentage of feces. The HPN calves had greater ADG over the entire study (0.211 vs -0.106 ± 0.6 kg/d;  $P < 0.001$ ). There were no differences ( $P \geq 0.239$ ) between treatments in either digestible or metabolizable energy efficiencies, which averaged 92.8 and 83.1%, respectively. There was a treatment × period interaction ( $P = 0.038$ ) on the percentage of intake N retained, whereas calves fed the HPN had a greater N retention during period 1 (88.0 vs. 78.7 ± 1.79%;  $P = 0.004$ ), but was not different from calves fed the LPN during period 2 (85.3 vs. 85.0 ± 1.79%;  $P = 0.904$ ). Therefore, these data indicate that healthy Jersey calves have the capability to digest and absorb the additional nutrients when fed a higher plane of nutrition during the first week of postnatal life. Further, the additional energy and amino N absorbed by calves fed the HPN were incorporated into lean tissue growth at a high efficiency.

**Key Words:** calf, digestibility, nutrition

**W331 Effects of glucose and propionate infusions on milk fat yield: A meta-analysis.** Sarah E. Schmidt\* and Adam L. Lock, Michigan State University, East Lansing, MI.

Milk fat depression (MFD) research has primarily focused on inhibitory effects of rumen biohydrogenation intermediates, particularly *trans*-10,*cis*-12 18:2 (CLA), on fat synthesis in the mammary gland. However, several studies have shown that directly increasing insulin-secretagogues through infusion also results in MFD. The objective of this meta-analysis was to evaluate the effects of glucose and propionate infusions on milk fat yield and energy balance. The analysis included 22 publications representing 27 glucose infusion treatments and 15 propionate infusion treatments. Trial-adjusted values were calculated in a linear mixed model with study included as a random variable and weighted by the inverse of the variance of daily milk yield. Trial-adjusted values for percent change in milk fat yield were regressed across levels of glucose (0.33 to 2.40 kg/d) and propionate (0.25 to 1.52 kg/d) infusion and lines

of best fit were applied ( $r^2 = 0.76$ ;  $r^2 = 0.65$ ). Change in milk fat yield ranged from 1.69% to -20.3% for glucose infusions and from 6.78% to -23.8% for propionate infusions. Increasing glucose and propionate infusion amount increased the magnitude of MFD ( $P < 0.01$ ;  $P < 0.01$ ). Cows in each glucose infusion study were categorized as having high (HF; milk fat >4.09%), moderate (MF; milk fat ≥3.58% and ≤4.09%), or low (LF; milk fat <3.58%) milk fat concentrations. Increasing glucose infusion amount decreased milk fat yield of HF cows at a greater rate compared with LF cows ( $P = 0.03$ ). MF cows also exhibited a greater response to glucose infusion compared with LF cows ( $P < 0.01$ ). Trial-adjusted values for change in energy balance were regressed on percent change in milk fat yield following glucose infusion and a line of best fit was applied ( $r^2 = 0.74$ ). Reducing milk fat yield from 1.69 to -20.3% through glucose infusion increased energy balance (-1.3 to 4.8 Mcal/d;  $P < 0.01$ ). While CLA has been shown to cause MFD, the effects of insulin-secretagogues on energy partitioning should also be considered in the development of a comprehensive model.

**Key Words:** energy balance, meta-analysis, milk fat depression

**W332 Effect of the starch level in diets with soybean or canola meal on the performance of lactating dairy cows.** Juan I. Sanchez-Durte\*<sup>1</sup>, Kenneth F. Kalscheur<sup>2</sup>, and David P. Casper<sup>1</sup>,

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This study was designed to test the impact of reducing corn grain starch with nonforage fiber sources in diets using either soybean meal or canola meal as the primary protein source. Sixteen Holstein cows were assigned to a 4 × 4 Latin square design with 4 periods of 28 d. Treatments were arranged as a 2 × 2 factorial with 2 protein sources [soybean meal (SBM) and canola meal (CM)] and 2 starch levels (21 and 27% of DM). Diets were formulated to contain 16.5% CP and the starch levels were achieved by replacing corn grain with soybean hulls and beet pulp. Protein source × starch interactions ( $P < 0.05$ ) were observed for DMI, feed efficiency (ECM/DMI), fat %, protein %, protein yield, and MUN. Cows fed the 27% starch diet consumed more DMI than cows fed the CM-21% diet. However, there was no interaction for milk yield, as cows fed 27% starch produced 2.5 kg/d more than cows fed 21% starch. Milk fat percentage was the least for cows fed CM-27% and greatest for cows fed SBM-27% and CM-21%. Milk protein percentage and yield was least for CM-21% compared with the other 3 diets. Milk urea nitrogen was least for cows fed CM-27% compared with the other 3 diets. Cows fed 27% starch produced 1.9 kg/d more energy-corrected milk (ECM) than cows fed 21% starch. Feed efficiency was the greatest for cows fed CM-21% and least for cows fed CM-27%. Overall, lower starch % in SBM or CM diets negatively affected DMI, milk yield, ECM, and milk protein percentage and yield, but not feed efficiency and milk fat percentage and yield. Increasing starch level in canola meal diets decreased milk fat percentage, but potentially improved protein balance because less MUN was observed.

*Contd.*

**Table 1 (Abstr. W332).**

Item	SBM		CM		SEM	<i>P</i> > <i>F</i> <sup>1</sup>
	21%	27%	21%	27%		
DMI, kg/d	26.2 <sup>ab</sup>	26.8 <sup>a</sup>	24.7 <sup>b</sup>	27.7 <sup>a</sup>	0.90	I
Milk, kg/d	36.5	38.5	36.6	39.7	1.30	S
ECM, kg/d	39.8	41.8	40.0	41.8	1.23	S
ECM/DMI	1.53 <sup>ab</sup>	1.57 <sup>ab</sup>	1.66 <sup>a</sup>	1.53 <sup>b</sup>	0.06	I
Fat, %	4.08 <sup>ab</sup>	4.14 <sup>a</sup>	4.21 <sup>a</sup>	3.86 <sup>b</sup>	0.16	I
Fat, kg/d	1.49	1.57	1.53	1.51	0.06	NS
Protein, %	3.28 <sup>a</sup>	3.26 <sup>a</sup>	3.15 <sup>b</sup>	3.26 <sup>a</sup>	0.06	I
Protein, kg/d	1.20 <sup>ab</sup>	1.24 <sup>a</sup>	1.15 <sup>b</sup>	1.28 <sup>a</sup>	0.04	I
MUN, mg/dL	12.8 <sup>a</sup>	12.4 <sup>a</sup>	12.6 <sup>a</sup>	11.2 <sup>b</sup>	0.53	I

<sup>ab</sup>Means with different letters differ (*P* < 0.05).

<sup>1</sup>S = starch effect; I = protein source by starch effect; NS = no significant.

**Key Words:** protein, starch, milk production

**W333 The effect of the supplementation of virginiamycin plus monensin on milk performance under grazing conditions in dairy cattle.** Ramiro Desantadina<sup>1</sup>, Luis Casares<sup>2</sup>, Matias Bailleres<sup>3</sup>, Milton Gorocica<sup>2</sup>, and Alejandro Relling<sup>\*1,4</sup>, <sup>1</sup>Fac Cs Veterinarias, UNLP, Argentina, <sup>2</sup>Phibro Animal Health, Argentina, <sup>3</sup>Ministerio de asuntos Agrarios, Buenos Aires, Argentina, <sup>4</sup>IGEVET, CCT La Plata, CONICET, Argentina.

The aim of the present study was to evaluate the effect of virginiamycin (Vm) plus monensin (Mn) on milk production and composition of grazing dairy cows. Eighty dairy cows, with an average of 2.5 lactations, 129 d in milk and a milk yield (MY) of 27.7 kg per day were randomly distributed into 2 groups; Group 1 (n = 40): 300 mg/cow/d Mn (M), and Group 2 (n = 40): 300 mg/cow/d Vm and 300 mg/cow/d Mn (V). Cows were grazed in a rotational grazing system of 2 grass paddocks per day. The cows were milked twice a day and received 4 kg of a pelleted concentrate and mineral supplement in the parlor at each milking. Both groups were also fed once a d 7 kg corn silage, 6 kg grass haylage, 2 kg corn grain, and 2 kg wheat middlings (all quantities as fed). Milk yield and composition were measured on d 0, 17, 34, 52, 66, 78, 90, 101, 111, 120 and 129, and body condition score (BCS) was measured on d 0, 52 and 129. Four-percent fat corrected milk (FCM) was estimated using the following formula  $MY \times (0.383 \times \% \text{ fat} + 0.242 \times \% \text{ protein} + 0.7832)/3.1138$ . Data were analyzed using a mixed model with repeated measures, using the cow as random effect, and treatment, time and their interaction as fixed effects. The treatment  $\times$  time interaction was not significant in all evaluated variables (Table 1). However, on FCM a trend for time and treatment interaction (*P* = 0.14) was approached: V cows had greater FCM on d 52, 78 (*P* < 0.1) and 101 (*P* < 0.05) than M cows. The V treatment group had numerically greater MY, milk fat and protein content, and BCS at the end of the trial, but differences were not statistically significant (Table 1; all *P* > 0.15). In conclusion, the addition of Vm in diets containing Mn increases FCM during peak- to mid-lactation.

*Contd.*

**Table 1 (Abstr. W333).** Milk yield, composition, and FCM in grazing cows supplemented with Mon (M) and Mon+Vm (V)

Item	Treatment			<i>P</i> -value	
	M	V	Trt	Time	Trt $\times$ Time
MY, kg/d	27.6	28.3	0.46	<0.01	0.64
Milk fat, %	3.34	3.46	0.15	<0.01	0.47
Milk protein, %	3.21	3.24	0.26	<0.01	0.72
FCM, kg/d	25.0	26.1	0.14	<0.01	0.14
d 0 FCM	27.1	26.8	0.80		
d 34 FCM	25.8	26.8	0.33		
d 52 FCM	27.4	25.7	0.06		
d 78 FCM	28.3	26.5	0.07		
d 101 FCM	26.7	24.6	0.04		

**Key Words:** dairy cow, virginiamycin, monensin

**W334 Undegradable aNDFom in non-forage feeds.** Alessandro Maria Zontini\*, Andreas Foskolos, Deborah Ross, and Michael Van Amburgh, Cornell University, Ithaca, NY.

Non-forage fiber feeds are generally highly digestible, however, most calculations of undegradability are made from static calculations based on Chandler et al. (1980) or Weiss et al. (1992). The objective of this study was to analyze the extent of aNDFom degradation in non-forage fiber feeds, to obtain the undegradable aNDFom (uNDF). Samples of 12 feeds (citrus pulp, beet pulp, wheat middlings, soy hulls, corn gluten meal, corn gluten feed, wheat distillers, corn ethanol distillers, flaked corn, rice hulls, soybean meal, and canola meal) were collected, each from 2 providers, and analyzed in duplicate in 3 separate batches for the extent of NDF digestion using the in vitro technique. Samples, 0.5 g, were weighed into Erlenmeyer flasks and 40 ml of Goering and Van Soest (1970) buffer was added to each flask under continuous CO<sub>2</sub>, and incubated in a water bath at 39°C. After 2 h of incubation, 10 mL of mixed rumen fluid from 2 lactating cattle were added to each flask and continuous CO<sub>2</sub> was maintained throughout the fermentation. Fermentations were conducted for 96, 120, and 240 h consistent with previous data from Raffrenato (2011). Residues were filtered on a glass microfiber filter (934-AH, Whataman) with a 1.5 µm pore size to enhance residue recovery. To analyze changes in uNDF the residues were compared with a *t*-test in JMP. For non-forage aNDFom the uNDF was obtained at 120h with the exception of citrus pulp where residues continued to digest out to 240 h compared with 120 h residues (*P*-value = 0.002).

*Contd.*

**Table 1 (Abstr. W334).** The aNDFom (%NDF) residues of feeds after 96, 120, and 240 h of fermentation

Item	Time (h)			SEM	P-value
	96	120	240		
Beet Pulp	22 <sup>a</sup>	19 <sup>b</sup>	17 <sup>b</sup>	0.01	0.004
Canola Meal	40	41	41	0.01	0.79
Citrus Pulp	21 <sup>a</sup>	20 <sup>a</sup>	16 <sup>b</sup>	0.01	0.002
Corn Gluten Feed	16 <sup>a</sup>	14 <sup>ab</sup>	13 <sup>b</sup>	0.01	0.028
Corn Distiller	16	16	14	0.01	0.50
Corn Germ	34	29	27	0.03	0.74
Flaked Corn	14	14	12	0.02	0.73
Rice Hulls	94	93	93	0.01	0.61
Soybean Meal	11	9	9	0.01	0.95
Soy Hulls	10 <sup>a</sup>	9 <sup>ab</sup>	8 <sup>b</sup>	0.01	0.022
Wheat Distiller	28	26	25	0.01	0.20
Wheat Middling	36 <sup>a</sup>	31 <sup>b</sup>	30 <sup>b</sup>	0.01	0.001

<sup>a,b</sup>Values with different letters are statistically different.

**Key Words:** aNDFom

**W335 Effect of two fat supplements differing in saturation on milk production and energy partitioning.** Enhong Liu\*, Courtney L. Preseault, Michael J. VandeHaar, and Adam L. Lock, *Michigan State University, East Lansing, MI.*

Effects of feeding diets containing fat supplements differing in saturation on milk production and energy partitioning were evaluated. Holstein cows ( $n = 32$ ;  $93 \pm 35$  DIM) were randomly assigned to treatment sequence in a crossover design experiment. Treatments were diets containing a saturated fat supplement (2.5% DM palmitic acid-enriched triglyceride [BergaFat T-300], SF) or an unsaturated fat supplement (2.5% DM soybean oil, UF). Diets utilized corn silage and alfalfa silage as forage sources and contained 25% NDF, 18% forage NDF, 32% starch, 18% CP, and 4.6% FA. Treatment periods were 28 d in length with the final 5 d used for sample and data collection. The statistical model included the random effect of cow and fixed effects of treatment and period. Compared with UF, SF increased milk fat concentration (3.07% vs. 2.42%;  $P < 0.01$ ) and yield (1.35 vs. 1.11 kg/d;  $P < 0.01$ ), but reduced milk protein concentration (3.05% vs. 3.12%;  $P < 0.01$ ) and yield (1.40 vs. 1.44 kg/d;  $P < 0.05$ ). Treatment did not alter milk yield (46 kg/d;  $P = 0.6$ ), but SF did increase FCM (41.9 vs. 38.1 kg/d;  $P < 0.01$ ) and ECM (42.6 vs. 39.8 kg/d;  $P < 0.01$ ) compared with UF. DMI and energy intake did not differ between treatments and averaged 25 kg/d and 41.2 Mcal/d, respectively (both  $P > 0.6$ ). However, SF increased the milk to feed ratio (ECM/DMI) compared with UF (1.67 vs. 1.53;  $P < 0.01$ ). Compared with UF, SF reduced BW gain (5.2 vs. 12.8 kg/28 d;  $P < 0.05$ ) but did not alter BCS ( $P = 0.8$ ) or fat thickness over the rump ( $P = 0.7$ ) and rib ( $P = 0.5$ ). SF decreased plasma concentration of insulin (1.18 vs. 1.34  $\mu\text{g/L}$ ,  $P < 0.05$ ), NEFA (122 vs. 137  $\mu\text{Eq/L}$ ,  $P < 0.01$ ), and triglycerides (7.9 vs. 8.5 mg/dL,  $P = 0.05$ ) compared with UF. There was no effect of treatment on plasma concentration of glucose ( $P = 0.3$ ). Compared with UF, SF increased plasma milk energy as a fraction of  $\text{NE}_L$  use (71 vs. 66%,  $P < 0.01$ ) and reduced calculated body energy gain as a fraction of  $\text{NE}_L$  use (3 vs. 8%,  $P < 0.01$ ). In conclusion, the 2 diets resulted in similar  $\text{NE}_L$  intake but the SF diet containing the palmitic acid-enriched triglyceride increased milk fat yield and partitioned more energy toward milk, while the UF diet containing soybean oil reduced milk fat yield and partitioned more energy toward body gain.

**Key Words:** fat supplementation, milk fat, palmitic acid

**W336 Nutrient intake and blood parameters of dairy cows fed sugarcane in different ways of storage.** Viviane B. Ferrari<sup>\*1</sup>, Mauro D. S. Oliveira<sup>2</sup>, and Francisco P. Rennó<sup>1</sup>, <sup>1</sup>University of São Paulo, Pirassununga, São Paulo, Brazil, <sup>2</sup>São Paulo State University, Jaboticabal, São Paulo, Brazil.

This study aimed to evaluate nutrient intake and blood parameters of Holstein cows fed sugarcane based diets. Treatments consisted of fresh chopped sugarcane (FS), hydrolyzed sugarcane (HS) and HS + 6 g live yeast (*Saccharomyces cerevisiae*) (HSY). All treatments contained concentrate with ground corn, soybean meal, cottonseed meal, urea, mineral salt. Roughage:concentrate ratio offered was 60:40. Twelve Holstein cows with body weight of 590 kg were assigned into 4 contemporary  $3 \times 3$  Latin Squares, with 3 periods of 21 d each and 3 treatments. The sugarcane used was IAC 862480 fifth cut and 12 mo of growth, chopped in particle size of 8 mm. The sugarcane was hydrolyzed with 0.5% of hydrated lime (95% of calcium hydroxide) using 0.5 kg of hydrated lime with 2 L of water to each 100 kg of sugarcane. Intake was regulated for animal by daily weighting of diet and orts, allowing for 5 to 10% of orts. Samples of feedstuffs and orts were weekly collected for chemical analyses and determination of dry matter (DMI), neutral detergent fiber (NDFI), crude protein (CPI) and ether extract (EEI) intakes. Blood samples were collected by jugular vein before the first meal for blood parameters determination: glucose (GLU), total protein (TP), total cholesterol (TC), albumin (AL), gamma-glutamyl transferase (GGT), aspartate aminotransferase (AST). Two contrasts were used to test the effect of FS vs. HS and HSY (C1); and HS vs. HSY (C2). Differences between treatments were considered significant at  $P < 0.05$ . FS promoted differences in all nutrient intakes. FS decreased DMI (11.43, 14.78 and 14.38 kg/d;  $P = 0.03$ ); NDFI (5.78, 7.95 and 7.09 kg/d,  $P = 0.02$ ); CPI (1.06, 1.32 and 1.39 kg/d;  $P = 0.03$ ), EEI (0.48, 0.60 and 0.62 kg/d;  $P = 0.03$ ), compared with HS and HSY, respectively. There were no differences for HS vs. HSY on nutrient intakes. There were no differences for none of the studied blood parameters ( $P > 0.05$ ). Fresh chopped sugarcane decreased nutrient intake of Holstein cows compared with hydrolyzed sugarcane.

**Key Words:** crude protein, hydrolysis, neutral detergent fiber

**W337 The effect of increasing concentrations of DL-methionine and HMB on hepatic genes controlling methionine and glucose metabolism.** Dean A. Bowen<sup>\*1</sup>, Nestor D. Luchini<sup>2</sup>, and Heather M. White<sup>1</sup>, <sup>1</sup>University of Wisconsin-Madison, Madison, WI, <sup>2</sup>Adisseo, Alpharetta, GA.

Metabolizable methionine (met) concentrations can be increased by feeding rumen-protected DL-Met (DL) or the isopropyl ester of 2-hydroxy-4-(methylthio)butanoic acid (HMB); however, hepatic response to these compounds have not been comparatively examined. The objective of this experiment was to examine the regulation of key genes in the Met and gluconeogenic pathways in response to doses of DL or HMB that mimic Met deficiency, adequacy, or supplementation in lactating dairy cows. Hepatocytes were isolated from 4 Holstein calves and were maintained as monolayer cultures in FBS for 24h before treatment. Treatments of DL or HMB (0, 10, 20, 40, 60 mM) were added to Met-free media in triplicate and after 24h, cells were collected for RNA isolation and quantification of gene expression by quantitative PCR. Data were analyzed in Proc Mixed of SAS 9.3. Analysis of covariance confirmed equivalent slopes of Met source and final model included source and dose, and random effect of calf within source. There was no main effect of Met source ( $P > 0.1$ ) for any genes examined. Expression of BHMT and MTR, genes that catalyze generation of Met from betaine and homocysteine, decreased ( $P \leq 0.05$ ) with



increasing Met concentration (BHMT: 0.7551, 0.7205, 0.6512, 0.582 ± 0.2563; MTR: 0.8897, 0.8481, 0.7648, 0.6815 ± 0.1763 arbitrary units). When concentrations of Met are high, Met can generate SAM, a methyl donor; however, expression of the gene that catalyzes this reaction, MAT1, was decreased ( $P \leq 0.05$ ) as Met concentration increased (0.7828, 0.7108, 0.5667, 0.4226 ± 0.1554 arbitrary units). Expression of CPT1a, G6Pase, and PEPCKm was not altered ( $P > 0.1$ ) suggesting that increased Met was not used as a gluconeogenic precursor at these concentrations. The decrease in hepatocyte production of Met at higher treatment doses suggest that cellular Met requirements were being met. The lack of influence on gluconeogenic enzymes and the decrease in the enzyme responsible for SAM generation may reflect metabolic priority for Met use at these concentrations, an adequate glucose concentration, or the lack of a Met deficiency before treatment.

**Key Words:** methionine, 2-hydroxy-4-(methylthio)butanoic acid (HMB), hepatocyte

### **W338 aNDFom degradation behavior in nonforage feeds.**

Alessandro Maria Zontini\*, Andreas Foskolos, Deborah A. Ross, and Michael E. Van Amburgh, *Cornell University, Ithaca, NY.*

Previous work conducted in our lab demonstrated that the extent of NDF degradation in nonforage fiber feeds is achieved by 120 h and results in undigested NDF (uNDF). Further, Raffrenato (2011) demonstrated that aNDFom degradation kinetic can be characterized with a dynamic model utilizing 4 data points (0, 30, 120 and 240 h). The objectives of this study were to characterize the degradation behavior of aNDFom, and to determine the time points required to model and calculate the degradation parameters, in nonforage fiber feeds. Samples of 12 feeds (citrus pulp, beet pulp, wheat middlings, soy hulls, corn germ, corn gluten feed, wheat distillers, corn ethanol distillers, flaked corn, rice hulls, soybean meal, and canola meal) were collected, each from 2 providers, and analyzed in duplicates and in 3 separate batches for NDF digestion kinetics using the in vitro technique. Fermentations were conducted for up to 3, 6, 9, 12, 15, 18, 21, 24, 30, 48, 72, 96, 120 h following the methods of Goering and Van Soest (1970). Residues were filtered on a glass microfiber filter (934-AH, Whatman). To study the digestion behavior of non-forage fiber NDF, the degradation curves were plotted on semi-log scale, and analyzed to determine the inflection point of the aNDFom residues. Overall, the natural log-transformation, demonstrated that NDF digestion in nonforage fiber feeds can be described by one degradable pool (no inflection points). To determine the time points, 7 combinations of 4 data points corresponding to: 0 h, 2 intermediate time points, and uNDF were used as inputs. The corresponding slope and intercept of each regression (observed on predicted) were then compared. The accuracy of the model did not allow for detecting statistically significant differences in slope and intercept among time point combinations, however 0, 12, 72, and 120 h were the time points that provided model predictions with an average slope and intercept closest to unity and zero respectively (intercept = 0.04, slope = 0.95). Thus, for routine determination of non-forage fiber digestibility and the estimation of rates of degradation for use in nutrition models, these time points provided adequate information among the feeds analyzed.

**Key Words:** aNDFom

**W339 Can potential digestible fiber affect dietary crude protein level in lactating dairy cows? Milk production and feeding behavior.** H. R. Mirzaei Alamouti\* and B. Mohtashami, *Department of Animal Science, University of Zanjan, Zanjan, Iran.*

Nitrogen (N) excretion from dairy farming systems is a major environmental challenge. An experiment was conducted to determine the effects of dietary potential digestible fiber (pdF) and crude protein (CP) levels in mid lactating Holstein cows on milk production and content, feeding behavior and nitrogen efficiency. Sixteen primiparous ( $n = 8$ ) and multiparous ( $n = 8$ ) cows (BW = kg and DIM = 118 ± 50 d) were allocated to 1 of 4 diets in balanced randomized complete block design in 2 periods (28 d). The diet containing: 1) 19% CP and low pdF, (HPLDF), 2) 17% CP and low pdF, (MPLDF), 3) 15% CP and high pdF, (LPHDF), and 4) 17% CP and high pdF, (MPHDF). The diets were balance for pdF by using different levels of dried alfalfa hay and corn silage in forage proportion of diets. Cows were fed individually and daily dry matter intake was recorded. Daily milk production and bi-weekly milk content were determined. Feeding behavior was recorded in 24 h with exception in milking times. Data were analyzed as repeated measurement with proc MIXED by using SAS software. The diets, periods, times and 2 or 3 ways interactions were constant effects and cows nested in diet and residual errors were random effects. Cows fed the diets with high pdF had greater DMI (17.87, 17.46, 18.44, and 18.60 kg/d, for HPLDF, MPLDF, LPHDF, and MPHDF respectively) and lower (21.8, 17.56, 12.86, and 15.78 for HPLDF, MPLDF, LPHDF, and MPHDF respectively) milk urea nitrogen ( $P < 0.01$ ) than cows fed low pdF diets. Milk Production and content and blood metabolites were not significantly different among the experimental diets. There was no significant difference among diets in parities and periods. There was significant difference ( $P < 0.05$ ) among diets in eating activity and cows fed to high pdF diets had greater eating activity (246.85 vs 261.2 min/d for high pdF vs low pdF). Results from this study indicate that increasing cp content in diets with low pdF and also increasing pdF content of diets with mid or low CP content can improve mid lactating Holstein cows' milk production and efficiency.

**Key Words:** potential digestible fiber, nitrogen efficiency, milk production

**W340 Effects of prill size of a palmitic acid-enriched fat supplement on yield of milk and milk components and nutrient digestibility of dairy cows.** Jonas De Souza\*, Joshua L. Garver, Courtney L. Preseault, and Adam L. Lock, *Michigan State University, East Lansing, MI.*

Effects of prill size of a palmitic acid-enriched fat supplement (PA; 85% C16:0) on feed intake, nutrient digestibility, and production responses of dairy cows were evaluated. Twenty-four Holstein cows (120 ± 44 DIM) were randomly assigned to treatment sequence in a 4 × 4 Latin square design. Treatments were a control diet (CON; no added PA), or 2.0% PA added either as a small prill size (SM; 284 ± 12.4 μm), medium prill size (MD; 325 ± 14.7 μm), or large prill size (LG; 600 ± 17.8 μm) supplement. PA replaced soyhulls in the ration and diets contained 21% forage NDF, 16% CP and 26% starch. Periods were 21 d in length with the final 5 d used for sample and data collection. Indigestible NDF was used as an internal marker to estimate fecal output and nutrient digestibility. The statistical model included the random effect of cow and period and the fixed effect of treatment. Overall, PA increased milk fat content (4.25 vs. 3.99%;  $P < 0.01$ ), milk fat yield (1.48 vs. 1.39 kg/d;  $P < 0.01$ ), tended to increase 3.5% FCM (39.2 vs. 37.7 kg/d;  $P = 0.10$ ), and improved feed efficiency (FCM/DMI; 1.51 vs. 1.42;  $P < 0.01$ ). However, compared with CON, PA treatments did not affect DMI, body condition score, or yields of milk, protein, and lactose. Overall, PA increased NDF digestibility (45 vs. 42%;  $P < 0.01$ ) and reduced the digestibility of 16-carbon FA (72 vs. 79%;  $P < 0.01$ ) and total FA (77 vs. 80%;  $P < 0.01$ ). Compared with CON, PA reduced the yields of de novo-synthesized (<16-carbon) milk FA (251 vs. 297 g/d;  $P < 0.01$ )

and preformed (>16-carbon) milk FA (383 vs. 430 g/d;  $P < 0.01$ ), and increased the yield of 16-carbon milk FA (457 vs. 378 g/d;  $P < 0.01$ ). There was no effect of prill size of PA on DMI, yield of milk and milk components, or feed efficiency. However, LG increased 16-carbon FA digestibility compared with MD (74 vs. 71%;  $P < 0.05$ ) and SM (74 vs. 72%;  $P < 0.05$ ). Additionally, LG increased total FA digestibility compared with MD and SM (78, 76 and 76%, respectively;  $P < 0.05$ ). These results demonstrate that PA increased milk fat concentration and yield and feed efficiency. Although prill size affected FA digestibility, this had no effect on animal performance.

**Key Words:** fatty acid digestibility, feed efficiency, milk fat

**W341 Changes in dairy cattle performance due to addition of a live yeast product.** Heidi A. Rossow<sup>\*1</sup>, Tim Riordan<sup>2</sup>, Andy Riordan<sup>2</sup>, Dennis Ervin<sup>3</sup>, and Dari Brown<sup>3</sup>, <sup>1</sup>University of California, Davis, Davis, CA, <sup>2</sup>Nutri-Systems Inc., Clovis, CA, <sup>3</sup>Phileo Lesaffre Animal Care, Milwaukee, WI.

Live yeast products have been proposed to increase nutrient utilization and milk production. Therefore the purpose of this trial was to determine if a live yeast product (LYP; BioCell 20, Phileo Lesaffre Animal Care, Milwaukee, WI) administered at 3g/cow/d to cows at a commercial dairy could increase milk yield and milk component production. All milking cows were milked 2 times a day and given LYP in the TMR in an OFF, ON, OFF, ON, OFF design where each ON or OFF period lasted 45 d and LYP was added to TMR during the ON periods only. Only data from cows that had at least one Dairy Herd Improvement Association milk test during each period remained in the data analyses (1903). Pens were fed the same respective diet (Fresh cow, milk cow, milk heifer or pregnant cow TMR) twice per day. Statistics were performed using Proc GLM (SAS Institute, 2013). To compensate for differences in days in milk (DIM) and milk production due to a period effect, treatment was nested within period, repeated measures for cow and covariates pen, DIM, diet, relative value, milk yield during period 1 (control period) and fresh date were included in the statistical model. There were no differences in least squares means of parity and small differences in least squares means DIM among periods. Average DIM in periods 2–5 were not different ( $P < 0.05$ ; range 143–298 d). Overall, daily milk yield (31.8 and 32.9.0 kg,  $P < 0.0001$ ) and milk protein (1.02 and 1.08 kg,  $P < 0.0001$ ; 3.25 and 3.31%,  $P < 0.0001$ ) for control and LYP supplemented treatments, respectively, were higher for cows fed LYP. Therefore LYP did increase milk yield and milk protein kg and % but decreased milk fat % (3.71 and 3.60%,  $P < 0.0001$ ), respectively.

**Key Words:** live yeast, dairy cow performance

**W342 Effects of feeding frequency and adding plant oil to diet on performance and feeding behavior of lactating Holstein dairy cows.** H. R. Mirzaei Alamouti<sup>\*</sup> and K. Akbari, *Department of Animal Science, University of Zanjan, Zanjan, Iran.*

The objectives of this experiment were to investigate the effect of feeding frequency and oil supplementation to the diet on performance and feeding behavior of lactating dairy cows. Twenty 4 lactating Holstein cows, 12 primiparous, (BW = 626 ± 58 kg and DIM = 195 ± 44 d) and 12 multiparous, (BW = 617 ± 25 kg and DIM = 207 ± 39 d) were randomly assigned to the 4 diets contained 2.5 percent supplemental oil or no oil and feeding frequency of 3 or 1 time per day in a randomized complete block design with a 2 × 2 factorial arrangement. Frequency of feed delivery and oil supplementation had no effect on dry matter intake. Oil supplementation reduced slug feeding especially with feeding once per

day ( $P < 0.05$ ). Feeding frequency and oil supplementation interaction tended to significant for milk yield ( $P = 0.09$ ), as there was a numerical increase in milk yield in 3 time feeding with oil diet. The cows fed oil diet in 3 time feed delivery had lower milk protein ( $P < 0.05$ ), but milk fat increased with 1 time feeding and oil supplementation ( $P < 0.05$ ). The results showed that oil supplementation to mid lactating cows diet can decrease compromised effects of decreasing feeding frequency.

**Key Words:** feeding frequency, oil supplementation, chewing behavior

**W343 Effect of feeding hay during the nursery phase of calf rearing.** F. Xavier Suarez-Mena<sup>\*</sup>, James D. Quigley, T. Mark Hill, and Rick L. Schlotterbeck, *Nurture Research Center, Provimi North America, Brookville, OH.*

Recommendations on the value of feeding forage to calves before weaning are inconsistent. This study aimed to evaluate the effects of feeding long hay during the nursery phase on calf performance. Treatments were to not feed hay or feed hay to calves throughout the 56-d trial. Forty-eight calves (45.4 ± 1.18 kg BW, 2 to 3 d of age) were fed a milk replacer (MR; 22% CP, 20% fat as-fed basis) powder at 568 g as-fed daily diluted at 12.5% solids. Calves were fed MR daily in 2 equal meals from 0 to 39 d, then at 313 g of MR powder daily (AM meal only) for d 40 to 42. All MR offered was consumed. Starter, hay, and water were fed ad libitum. Hay and starter were fed in separate pails. Calves were housed in individual pens bedded with straw in a nursery with natural ventilation and no added heat through d 56. Feces were scored daily using a 1 to 5 scale with 1 being normal and 5 being watery. Medical treatments were recorded daily. Calves were weighed initially and weekly thereafter. Body condition score (1 being thin and 5 being obese) and hip widths were measured initially and every 2 weeks thereafter. The average temperature in the nursery was 23°C with a range from 12 to 36°C. The average relative humidity was 78% with a range from 40 to 97%. Data were analyzed as a completely randomized design. Repeated measures analysis was performed on pre-weaning (0 to 42 d), post-weaning (42 to 56 d), and overall (0 to 56 d) data. Initial measurements differed for hip widths ( $P < 0.05$ ) among treatments. Pre-weaning hip width change tended ( $P < 0.09$ ) to be less for calves fed hay. Other measurements did not differ ( $P > 0.05$ ). Hay intake was highly variable and averaged 6.1% of total intake overall. Hay intake as a percentage of overall intake ranged from less than 1 to 20% among the 24 calves fed hay. Hay intake was not related to starter intake. Feeding hay did not improve calf performance as measured in this study.

**Key Words:** starter intake, hay, performance

**W344 Effects of non-fiber carbohydrate level in low energy diets on production and health responses in peripartum Holstein cows.** H. R. Mirzaei Alamouti<sup>\*</sup> and P. Panahiha, *Department of Animal Science, University of Zanjan, Zanjan, Iran.*

Effects of rumen fermentability of carbohydrate and nonfiber carbohydrate during late gestation and early lactation on production and metabolic status of periparturient cows were studied. Sixteen multiparous and 40 primiparous cows were used in a randomized complete block design and assigned at random to 1 of 2 treatments in pre- and postpartum. The cows were fed diets as total mixed ration (TMR) with similar crude protein and energy (low energy diets) content, but different source of starch (ground barley vs. corn grain) from 20 ± 2 d relative to expected calving until calving. At parturition, half of the animals from each prepartum treatment were assigned to a lactation diet with 2 level of nonfiber carbohydrate until d 28 and, all cows received the same

high energy lactation diet until d 90. The cows fed prepartum diet had a positive energy balance during the prepartum period. Blood samples were taken at -20, +1 and +21 d relative to calving. Data were separately analyzed for multi- and primiparous cows by proc Mixed of SAS with diet and time as fixed effect and animal nested in diet as random effect. There was no significant difference in milk content except milk lactose in primiparous cows fed prepartum diets. Milk production was higher for primiparous cows fed barley diet in prepartum and high NFC diet in postpartum, and also corn diet in prepartum and low NFC in postpartum. Milk fat content for multiparous cows was different. There was significant difference between  $\beta$ -hydroxybutyrate, albumin and nonesterified fatty acid for primiparous cows and total protein, alanine amino acid transferase and BHBA for multiparous cows in postpartum. There were differences in some reproductive parameters. Generally, the responses of fresh cow to nonfiber carbohydrate content in lactating diet is depends on fermentability of starch in low energy close-up diet.

**Key Words:** transition period, nonfiber carbohydrate

#### W345 Increase in total solids of whole milk and its effects on development of dairy calves.

Rafael Alves de Azevedo\*<sup>1</sup>, Sâmara Raiany de Almeida Rufino<sup>1</sup>, Pâmela Michéli Furini<sup>1</sup>, Fernanda Samarini Machado<sup>2</sup>, Mariana Magalhães Campos<sup>2</sup>, Paulo Campos Martins<sup>1</sup>, Aloma Eitere Leão<sup>1</sup>, Ângela Maria Quintão Lana<sup>1</sup>, and Sandra Gesteira Coelho<sup>1</sup>, <sup>1</sup>Federal University of Minas Gerais, Belo Horizonte, Minas Gerais, Brazil, <sup>2</sup>EMBRAPA Dairy Cattle, Coronel Pacheco, Minas Gerais, Brazil.

Objectives were determine the best total solids (TS) concentration in liquid diet consisting of whole milk (WM) and milk replacer (MR) and its effects on preweaning calves feed intake and performance. Sixty crossbred Holstein-Gyr calves were distributed in 4 treatments, maintaining the balance for initial body weight and total protein concentration in blood serum and genetic composition. The treatments consisted of WM with the increasing addition of MR (Sprayfo Violet SSP), to adjust the TS to 12.50; 15.00; 17.50 and 20.00%, after the measurement the TS in WM using Brix refractometer (DD-3 MISCO Palm Abbe Digital), which had the degree brix value converted to TS (Moore et al., 2009). TS contents verified in the treatments after the addition of MR were 13.50; 16.10; 18.20 and 20.40%. The calves were fed 6 L/d, divided in 2 equal meals (8 and 16h) provided in buckets, from 5 to 55 d of age. From 56 to 59 d, the volume was reduced to 3 L/d (8h). At 60 d the animals were weaned. Starter (Soylac Rumen 20% CP) and water were provided ad libitum throughout the experiment. Feed intake, feed efficiency, average daily gain (ADG), structural body measures and days of diarrhea were measured. Data were analyzed by ANOVA using the PROC GLM procedure of SAS. Increasing TS resulted in a linear increase in water intake, total DMI, ADG, withers height and hearth girth. Starter intake, days of diarrhea, hip width and feed efficiency did not change as the TS increased up to 20.4%.

**Key Words:** milk system, milk replacer

#### W346 Increase in total solids of whole milk and its effects on development in postweaning calves.

Rafael Alves de Azevedo\*<sup>1</sup>, Pâmela Michéli Furini<sup>1</sup>, Sâmara Raiany de Almeida Rufino<sup>1</sup>, Fernanda Samarini Machado<sup>2</sup>, Mariana Magalhães Campos<sup>2</sup>, Aloma Eitere Leão<sup>1</sup>, Paulo Campos Martins<sup>1</sup>, Ângela Maria Quintão Lana<sup>1</sup>, and Sandra Gesteira Coelho<sup>1</sup>, <sup>1</sup>Federal University of Minas Gerais, Belo Horizonte, Minas Gerais, Brazil, <sup>2</sup>EMBRAPA Dairy Cattle, Coronel Pacheco, Minas Gerais, Brazil.

This study aimed to evaluate the effects of intensive whole milk feeding in calves on postweaning feed intake and performance. Sixty crossbred Holstein-Gyr calves were distributed in 4 treatments until the weaning. The treatments consisted of WM with the increasing addition of MR (Sprayfo Violet SSP), to adjust the TS to 12.5; 15.0; 17.5 and 20.0%, after the measurement the TS in WM using Brix refractometer (DD-3 MISCO Palm Abbe Digital), which had the degree brix value converted to TS (Moore et al., 2009). The calves were fed 6 L/d, divided in 2 equal meals (8 and 16h) provided in buckets, from 5 to 55 d of age. From 56 to 59 d, the volume was reduced to 3 L/d (8h). At 60 d the animals were weaned. Starter (Soylac Rumen 20% CP) and water were provided ad libitum throughout the experiment. Corn silage was provided ad libitum to calves from 70 d of age. Feed intake, average daily gain (ADG) and structural body measures were measured in postweaning calves. Data were analyzed by ANOVA using the PROC GLM procedure in SAS. Results are presented in Table 1. Starter, corn silage and water intakes and ADG in postweaning calves did not change as the TS increased up to 20.4%. Increasing TS resulted in a tendency of linear increase in withers height and final body weight.

**Table 1 (Abstr. W346).** Feed intake and performance of postweaning calves fed liquid diet with increasing total solid content during preweaning period

Item	Treatment, % TS				SEM	P-value
	13.5	16.1	18.2	20.4		
Starter intake, g DM/d	2088.91	2073.93	2140.40	2230.93	53.13	0.69
Corn silage intake, g DM/d	201.1	170.4	188.7	201.2	12.33	0.79
Water intake, kg/d	8.4	8.1	9.1	8.9	0.20	0.27
ADG, g/d	999.57	976.77	971.75	1010.23	25.44	0.94
Withers height, cm	91.9	92.5	93.5	94.0	0.40	0.08
Final body weight, kg	101.5	104.9	107.0	110.7	1.60	0.07

**Key Words:** milk system, milk replacer

#### W347 Effect of dietary starch content on the occurrence of subacute ruminal acidosis (SARA) and inflammation in fresh dairy cows.

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Multiparous ruminally cannulated Holstein cows (n = 16) were fed either a lower starch [21% starch, 37% neutral detergent fiber (NDF)] or higher starch (27% starch, 32% NDF) diet from 1 to 21 d in milk (DIM) to assess the effect of dietary starch on rumen fermentation, rumination, energy metabolites, and inflammatory markers. Diets contained 28% corn silage, 22% haycrop silage, 2% straw, and 48% concentrate. Corn meal was replaced partially with soyhulls and wheat middlings in the lower starch diet. Cows were fed a close-up diet (16% starch, 44% NDF) for 21 d. Cows were housed in a bedded pack, fed individually, and milked 3x/d. Rumen pH was measured at 1-min intervals and rumination was measured continuously from -14 to 21 DIM. Rumen and blood samples were collected on -14, 1, 2, 3, 4, 5, 6, 7, 9, 13, 17, and 21 DIM at 0 and 6 h post feeding. Data were analyzed as a completely randomized design by ANOVA with the MIXED procedure of SAS with model effects of treatment, time, treatment x time, and covariate (data from -14 to -8 DIM). Cows averaged 20.4 ± 0.7 kg dry matter intake/d and 42.1 ± 1.9 kg milk/d with 4.6 ± 0.2% fat and 3.5 ± 0.1% true protein. Daily mean rumen pH was higher (6.19 vs. 6.04, SE = 0.05, P = 0.03), time with pH < 5.8 was lower (1.6 vs. 4.5 h/d, SE = 0.7, P < 0.01), and rumen ammonia was higher (10.1 vs. 7.1 mg/dL, SE = 0.6, P < 0.01) for cows fed the lower starch diet. Diet did not affect (P >



0.10) rumination ( $481 \pm 18$  min/d), rumen volatile fatty acids ( $115 \pm 3$  mM), or acetate to propionate ratio ( $3.23 \pm 0.13$ ). Cows fed the lower starch diet had lower ( $P < 0.10$ ) 6-h serum haptoglobin [ $0.10$  (95% CI  $0.06$ – $0.14$ ) vs.  $0.17$  (95% CI  $0.11$ – $0.27$ ) mg/mL] and serum amyloid A [ $22$  (95% CI  $12$ – $38$ ) vs.  $53$  (95% CI  $28$ – $101$ ) ug/mL]. Diet did not affect ( $P > 0.10$ ) 0-h nonesterified fatty acids ( $680 \pm 58$  uEq/L), 0 and 6-h  $\beta$ -hydroxybutyrate ( $1.0 \pm 0.2$  mM), 6-h interleukin (IL)  $-1\beta$  ( $57.3 \pm 5.1$  pg/mL), 6-h IL-6 ( $457 \pm 42$  pg/mL), or 6-h tumor necrosis factor- $\alpha$  ( $40.7 \pm 2.7$  pg/mL). The nutritional strategy that is used during the transition period is critical for minimizing the risk of SARA and controlling inflammation.

**Key Words:** transition cow, starch, acidosis

**W348 Physical and enzymatic hydrolysis characteristics of ruminal protozoal glycogen.** Mary Beth Hall\*, *US Dairy Forage Research Center, USDA-ARS, Madison, WI.*

The characteristics of microbial glycogen have not been well described, but have implications for its analysis and digestion characteristics. A series of analyses, and comparisons carried out as randomized complete block designs were performed on glycogen isolated from protozoa from ruminal inoculum incubated in vitro with glucose. The isolated protozoal glycogen (IPG) was in the form of water and 0.9% NaCl insoluble granules 1.3 to 1.9 mm in length. IPG was not obviously birefringent under polarized light.  $\alpha$ -Glucan was measured by AOAC Official Method 2014.10 as detected glucose  $\times 0.9$ .  $\alpha$ -Glucan contents of beef glycogen, wheat starch (WS), corn starch (CS), and IPG on a dry matter (DM) basis were 98.1, 99.5, 100.0, and 98.3%, respectively. Duplicate samples (each 50 mg in 5 mL water) were incubated at successively increasing temperatures (22, 37, 50, 57, 65, 70, 75, 80, and 85°C) for 30 min with vortexing every 5 min for 5 s. Gelatinization temperature was determined as the point at which transmittance % at 650 nm departed from baseline measurements of unheated samples. WS, CS, and IPG had gelatinization temperatures of 57, 65, and 65°C, respectively. Subsequent enzymatic hydrolysis of 0.2 mL of the sample suspensions was performed with addition of 1 mL 0.1 M Na acetate buffer (pH 5.0), and 4 U amyloglucosidase incubated for 2 h at 39°C. Digested  $\alpha$ -glucan as a percentage of initial CS, WS, and IPG were 8.3, 9.0, and 24.7% for ungelatinized vs. 100.0, 88.9, and 95.6% for gelatinized (85°C) samples, respectively. Effects of sample, gelatinization, and sample  $\times$  gelatinization all were  $P < 0.01$  (standard error of the difference; SED = 0.4). Incubation of ungelatinized suspensions of IPG for 24 h at 57°C with 0, 1.8, or 4 U amyloglucosidase with Na acetate buffer gave  $\alpha$ -glucan values of 0.03, 14.3, and 15.0% of DM, respectively ( $P < 0.01$ , SED = 0.06). Similar to native starch, gelatinization was required to achieve more extensive enzymatic hydrolysis of IPG. This suggests that protozoal glycogen digestion characteristics may be similar to those of native starch, and that IPG requires gelatinization for analysis involving enzymatic hydrolysis.

**Key Words:** rumen microbe, protozoa, glycogen

**W349 The determination of the concentrations of isoforms of vitamin E in tissues, milk and blood via HPLC after short-term feeding in dairy cows.** Y. Qu\*<sup>1</sup>, T. H. Elsasser<sup>2</sup>, J. R. Newbold<sup>3</sup>, E. E. Conner<sup>2</sup>, and K. M. Moyes<sup>1</sup>, <sup>1</sup>University of Maryland, College Park, MD, <sup>2</sup>Agricultural Research Service, US Department of Agriculture, Beltsville, MD, <sup>3</sup>Cargill Innovation Center, Velddriel, the Netherlands.

The objective of this study was to determine the pattern of change in the concentrations of the 4 isoforms of vitamin E ( $\alpha$ -,  $\beta$ -,  $\gamma$ - and

$\delta$ -tocopherol) in bovine tissues (liver, mammary and muscle), blood and milk after short-term feeding of a vegetable-derived oil (Tmix) particularly enriched with  $\gamma$ - and  $\delta$ -isoforms (9%  $\alpha$ -, 1%  $\beta$ -, 24%  $\delta$ - and 62%  $\gamma$ -tocopherol). Healthy multiparous Holstein cows ( $>90$  DIM) were assigned to dietary control (Notoc,  $n = 4$ ) or tocopherol-fed treatment groups ( $n = 5$ : TOC;  $\sim 260$ g Tmix/cow/d, top-dressed) for 9 consecutive days. Milk and blood samples were collected in the mornings on d 0 and d 10 from the 5 TOC cows; tissues were harvested from those same cows on d 10 of treatment and the cohort of physiologically similar Notoc cows. Isoform concentrations were determined in samples by HPLC. Data were analyzed by Student's  $t$ -test and ANOVA. Delta-TOC was not detected in tissues, blood or milk. Concentrations of  $\gamma$ -TOC were increased ( $P < 0.001$ ) in tissues, blood and milk with feeding ( $P < 0.001$ ). In all tissues,  $\alpha$ -TOC concentrations were higher ( $P < 0.001$ ) than  $\gamma$ -TOC before and after feeding. Concentrations of both  $\alpha$ - and  $\gamma$ -TOC were higher in blood and milk after feeding Tmix ( $P < 0.05$ ). The content of  $\alpha$ -TOC was higher than  $\gamma$ -TOC ( $P < 0.001$ ) in milk and blood before and after feeding. In conclusion, short-term feeding of Tmix oil resulted in increases in the  $\gamma$ -isoform, largely no measurable content or significant change in the  $\beta$ - or  $\delta$ -isoforms, and a variable change in the  $\alpha$ -isoform of TOC across various tissues and biological fluids of the cow.

**Key Words:** concentration, cow, vitamin E

**W350 Total replacement with organic minerals regulates endometrial gene expression patterns that improve reproductive performance status in dairy heifers.** Daniel E. Graugnard\*, Allison C. Smith, Sylvie Andrieu, and Kristen M. Brennan, *Alltech Center for Animal Nutrigenomics and Applied Animal Nutrition, Nicholasville, KY.*

Reproductive performance in dairy cows is a major factor determining herd efficiency and profitability. The objective of this study was to evaluate the effect of dietary organic minerals on gene expression profiles of endometrial tissue. A total of 18 Holstein heifers (10–15 mo) were randomly assigned to 2 groups ( $n = 9$ /treatment) and supplemented for 90d with an organic premix (consisting of Bioplex Zn, Bioplex Cu, Bioplex Mn and EconomasE) or a standard inorganic premix (control). Endometrium was biopsied at d90 of supplementation and RNA was hybridized to the Affymetrix Bovine Genome Array. Hierarchical clustering analysis resulted in 2 different groups as a function of the pairwise distances corresponding to the treatments. Relative expression analysis between treatments resulted in a total of 580 differentially expressed genes ( $P < 0.05$ ; 398 upregulated; 182 downregulated), which enriched ( $P < 0.05$ ) different pathways related to reproductive performance: (1) Dendritic cell maturation, which regulates endometrium cell differentiation and the vascular responses associated with implantation; (2) Relaxin hormone signaling, which confers beneficial effects on the endometrium for implantation; (3) G $\alpha$ q signaling function of smooth muscle tissue like endometrium; and (4) Thrombin signaling, which promotes coagulation and the binding and aggregation of platelets and facilitating the rapid adherence of neutrophils, monocytes and lymphocytes to the endothelial cell layer to overcome exogenous challenge or play an essential role in early pregnancy. In conclusion, supplementation with organic minerals potentially benefits reproductive performance, including improved regulation of the estrous cycle, shorter calving intervals, and improved implantation rates.

**Key Words:** organic mineral, endometrium, cattle

**W351 Comparative bioavailability of lysine in three commercial rumen-protected lysine products using the in vivo plasma lysine response method.** Heather A. Tucker<sup>1</sup>, Makoto Miura<sup>2</sup>, Izuru Shinzato<sup>3</sup>, and Catherine S. Ballard<sup>\*1</sup>, <sup>1</sup>William H. Miner Agricultural Research Institute, Chazy, NY, <sup>2</sup>Ajinomoto Co., Inc., Kawasaki-ku, Kawasaki-shi, Japan, <sup>3</sup>Ajinomoto Heartland Inc., Chicago, IL.

Lysine bioavailability of second generation AjiPro-L (A2G; Ajinomoto Heartland, Inc.) has been determined using the plasma free amino acid dose response technique. The objective of this study was to compare bioavailability of MetaboLys (MBL; H. J. Baker & Bro., Inc.) and USA Lysine (USA; Kemin Industries Inc.) relative to A2G following the same technique. Fourteen multiparous lactating Holstein cows (113 ± 28 d in milk) were housed in a tie stall facility and used in a replicated 7 × 7 Latin square design with 7-d periods. Cows were blocked by days in milk and milk production, and assigned randomly to treatment sequence. A common basal diet adequate in Lys was fed proportionately at 3 intervals (33.4% at 0500 h, 33.3% at 1300 h, and 33.3% at 2100 h). Treatments included Lys supplemented at 0, 30, or 60 g/d from A2G, MBL, or USA and were administered 1 h before each feeding on d 2 through 7 of each period in amounts proportional to feed offered to simulate diet inclusion. Blood samples were obtained from each cow on d 6 and 7 of each period from the tail vein at 2-h intervals starting at 0600 h. Plasma, pooled by day, was analyzed for amino acid (AA) concentrations. Data were reduced to a period mean for each cow and analyzed using the MIXED procedure of SAS. REG procedure was used to generate linear regression models for each rumen-protected Lys product using the values of Lys (μmol) and Lys as a percentage of total AA (μmol-basis) to determine the degree of elevation of plasma Lys. Relative to A2G, estimated bioavailability of MBL and USA were determined using the slope ratio assay technique. Dry matter intake and milk yield ( $P > 0.10$ ) did not differ among treatments. The slope for A2G was numerically greater (0.183;  $r^2 = 0.72$ ) when compared with the slope for MBL (0.03;  $r^2 = 0.58$ ) or USA (0.07;  $r^2 = 0.99$ ) when expressing the concentration of plasma Lys as absolute value (μmol). Calculated bioavailability of MBL and USA was 18.3 and 38.2% of the bioavailability of A2G, respectively. Characterizing relative bioavailability of rumen-protected Lys in dairy cattle provides a means for comparing economic value of commercially available products.

**Key Words:** lysine, bioavailability, rumen-protected

**W352 Plane of milk replacer nutrition influences the resistance to an oral *Citrobacter freundii* opportunistic infection in Jersey calves at 10 days of age.** Yu Liang<sup>\*1</sup>, Jeff A. Carroll<sup>2</sup>, and Michael A. Ballou<sup>1</sup>, <sup>1</sup>Texas Tech University, Department of Animal and Food Sciences, Lubbock, TX, <sup>2</sup>USDA-ARS, Lubbock, TX.

This study investigated how early life plane of milk replacer nutrition influences the resistance to an opportunistic enteric challenge with *Citrobacter freundii*. Twenty colostrum-fed Jersey calves were blocked by BW at birth and randomly assigned to either a low (LPN) or high plane of nutrition (HPN) treatment. The LPN calves were fed 400 and 450 g DM/d of a 20% CP and 20% fat milk replacer during the 1st wk and thereafter, respectively. The HPN calves were fed 600 and 700 g DM/d of a 28% CP and 20% fat milk replacer during the 1st wk and thereafter, respectively. All calves were challenged orally with  $1 \times 10^8$  cfu of a stationary phase culture of *Citrobacter freundii* at d 10. Fecal scores were recorded throughout the study and rectal temperatures were recorded daily beginning on d 10. Fecal and plasma samples were collected at d 10, 12, 14, 16, 18, 20, 22, and 24. Fecal samples were analyzed for DM percentage and plasma for urea nitrogen, glucose, and haptoglobin concentrations. All calves were harvested at d 24 and

ileal tissues were analyzed for morphology. All data reported as LPN vs HPN, respectively. Water intakes increased to a greater extent among HPN calves after the challenge ( $P \leq 0.021$ ). There was no difference in DM percentage of feces between treatments (21.9 vs  $19.9 \pm 1.34\%$ ;  $P = 0.291$ ) despite HPN having greater fecal scores ( $2.0$  vs  $2.5 \pm 0.07$ ;  $P = 0.001$ ). Rectal temperatures were greater among the HPN calves ( $38.4$  vs  $38.6 \pm 0.05^\circ\text{C}$ ;  $P = 0.024$ ). Additionally, the HPN calves had numerically greater peak plasma haptoglobin concentrations after the challenge ( $266$  vs  $511 \pm 108.0$  μg/mL;  $P = 0.118$ ) and a tendency to have a greater total mucosal height of the distal ileum ( $752$  vs  $921 \pm 59.1$  μm;  $P = 0.059$ ). Therefore, these data indicate that calves fed a HPN during the neonatal period have a slightly greater acute phase response to an opportunistic bacterial enteric infection. Furthermore, these data reiterate that fecal scores should not be the only measure of enteric health, especially when evaluating the health effects associated with plane of nutrition.

**Key Words:** calf, disease, nutrition

**W353 Pre-weaning calf responses to lysine: I. Development and evaluation of functions explaining nitrogen retention responses to dietary lysine and body weight.** Juan J. Castro Marquez<sup>\*</sup>, Robin R. White, and Mark D. Hanigan, Department of Dairy Science, Virginia Tech, Blacksburg, VA.

The objective of this study was to define the nitrogen retention (NR) response of growing dairy calves as a function of dietary lysine intake (DLI) and body weight (BW). Raw, individual calf data from 3 published papers were collected. Lysine intake and NR were expressed as g per kg BW. Twelve models were derived relating NR, DLI and BW. Three equation forms (logistic, Michaelis-Menten or exponential), 2 effect types (fixed effects or mixed-effects with random study effect) and 2 error assumptions (equal error variance or heterogeneous error variance) were considered. The models were evaluated and compared based on their root mean squared prediction error (RMSPE), concordance correlation coefficient (CCC) and any mean and slope bias revealed by residual analysis. The asymptotic and rate-determining parameters of each function were assumed to vary as a function of BW and the primary explanatory variable within each equation form was DLI. Models varied notably in their predictive capacity and RMSPE ranged from 14.4 to 58.9%. A fixed-effects, logistic model with equal error variance returned the lowest RMSPE (14.4%), highest CCC (0.94) and had no appreciable mean or slope bias. All parameters were statistically significant ( $P < 0.01$ ). This equation,  $\text{NR} = (0.89 - 0.0067 \times \text{BW} + 0.000015 \times \text{BW}^2) \div (1 + e^{-((\text{Lys} - (0.0024 \times \text{BW} + e^{-0.013 \times \text{BW}})) / 0.032))})$ , estimated that a young calf (50 kg BW) could potentially achieve a maximal NR efficiency of 2.72 g/g DLI/kg BW and at 250 kg BW a maximal NR efficiency of 1.25 g/g DLI/kg BW could be reached. Nitrogen retention efficiency reduced substantially as BW increased. Increasing DLI promoted NR at low BW more effectively than at high BW. Although DLI affects short-term protein accretion responses, NR rate was primarily dependent on calf maturity.

**Key Words:** calf, nitrogen retention, amino acid

**W354 Saliva sodium, potassium, and phosphorus concentrations of post-peak lactating Holstein cows are not affected by dietary fiber or protein content.** J. A. D. R. N. Appuhamy<sup>\*1</sup>, M. Niu<sup>1</sup>, T. Teweldebrhan<sup>1</sup>, A. Leytem<sup>2</sup>, R. Dungan<sup>2</sup>, and E. Kebreab<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of California, Davis, CA, <sup>2</sup>USDA-ARS, Northwest Irrigation Research Lab, Kimberly, ID.

Knowledge on saliva sodium (Na), potassium (K), and phosphorus (P) concentrations are important in modeling electrolyte, water, and P partitioning in dairy cows. Saliva mineral concentrations in ruminants are strongly associated with saliva secretion rates, which can be affected by diet composition. The objective of this study was to examine saliva Na, K, and P concentrations in lactating cows fed 2 levels of forage and CP contents. Saliva samples were collected from 12 lactating Holstein cows randomly assigned to a 2 × 2 factorial arrangement of 2 forage (alfalfa hay) levels [38 (LF) vs. 53% (HF)] and 2 CP levels [15.2 (LP) vs. 18.5% (HP)] over 3 periods. Saliva samples were acidified (pH = 5–6) and diluted by a factor of 50 using deionized water (pH = 4.0). The filtered samples were then analyzed for Na, K, P, and calcium (Ca) concentrations by inductively coupled plasma atomic emission spectroscopy. Treatment effects on saliva mineral concentrations were analyzed with mixed-effect models including random cow effect. Mean dietary concentrations and total intake (via feed and water) of Na, K, and P were 0.36 ± 0.01, 1.04 ± 0.10, and 0.40 ± 0.01% of DM and 81 ± 10, 236 ± 40, and 93 ± 9 g/cow/d, respectively. Only total K intake was negatively affected by dietary fiber content ( $P < 0.01$ ). Saliva concentrations of Na and K were on average 70.3 ± 6.7 and 21.9 ± 1.3 mmol/L and were negatively correlated ( $r = -0.40$ ,  $P < 0.01$ ). A unit increase in average daily temperature (T, °C) increased and decreased saliva K ( $P = 0.08$ ) and Na ( $P = 0.05$ ) concentrations by 0.9 ± 0.5 and 4.9 ± 2.4 mmol/L, respectively. Saliva K concentration decreased by 0.010 ± 0.005 mmol/L ( $P = 0.07$ ) for unit increase in BW. Saliva P concentration was on average 1.9 ± 0.4 mmol/L, and increased and decreased by 0.004 ± 0.002 ( $P = 0.10$ ) and 0.61 ± 0.18 mmol/L ( $P < 0.01$ ) for unit increase in BW and T. Saliva P concentration was negatively related to saliva Ca concentration ( $r = -0.37$ ,  $P < 0.01$ ). Dietary fiber or CP contents did not have significant effect on any of the saliva mineral concentrations. Information on BW, T, and relationship with other minerals are important in determining Na, K, and P partitioning in lactating dairy cows.

**W355 Do the viability and dose of *Saccharomyces cerevisiae* affect the digestibility, ruminal fermentation, and performance of lactating dairy cattle?** Y. Jiang<sup>\*1</sup>, R. M. Martins<sup>2</sup>, I. M. Ogunade<sup>1</sup>, M. A. Bamikole<sup>3</sup>, F. Amaro<sup>2</sup>, W. Rutherford<sup>4</sup>, S. Qi<sup>4</sup>, F. Owens<sup>4</sup>, B. Smiley<sup>4</sup>, K. G. Arriola<sup>1</sup>, C. Staples<sup>1</sup>, and A. T. Adesogan<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Florida, Gainesville, <sup>2</sup>Animal Science Department, Federal University of Vicosa, Vicosa, Minas Gerais, Brazil, <sup>3</sup>Department of Animal Science, University of Benin, Benin City, Nigeria, <sup>4</sup>DuPont Pioneer, Johnston, IA.

This study was conducted to examine the effect of the dose and viability of supplemental *Saccharomyces cerevisiae* on ruminal fermentation and performance of lactating dairy cows. Four ruminally cannulated lactating cows (284 ± 18 DIM) were assigned to 4 treatments arranged in a 4 × 4 Latin square design with four 21-d periods. Cows were fed a non-acidotic total mixed ration (46.8% corn silage, 8.5% wet brewers grain and 44.7% concentrate, dry matter basis). The diet was supplemented with no yeast (Control, CON) or with a low dose of live yeast ( $5.7 \times 10^7$  cfu/day; LLY), a high dose of live yeast ( $6.0 \times 10^8$  cfu/day; HLY), or a high dose of killed yeast ( $6.0 \times 10^8$  cfu/day before heating at 80°C; HDY). Ruminal pH and temperature were measured with indwelling probes. Ruminal fluid was collected 0, 2, 4, 6, 8 and 10 h after the morning feeding on d 21 for analysis of volatile fatty acids (VFA) and ammonia-N. All data were analyzed using the GLIMMIX procedure of SAS (SAS Institute, Cary, NC). In vivo digestibility was measured using chromic oxide as a marker. Supplemental LLY increased yields of milk (29.9 vs 31.8 kg/d;  $P = 0.04$ ), milk fat (1.09 vs 1.17 kg/d;  $P = 0.08$ ) and milk protein (0.95 vs 1.02 kg/d;  $P = 0.05$ ), feed efficiency (0.85 vs 0.90;  $P = 0.03$ ), acetate to propionate ratio ( $P = 0.07$ ), and in vivo apparent

digestibility of dry matter and neutral and acid-detergent fiber ( $P = 0.02$ , 0.02 and 0.02, respectively). Feeding HLY instead of LLY decreased milk yield (31.8 vs 30.1 kg/d;  $P = 0.07$ ) and feed efficiency (0.90 vs 0.86 kg/d;  $P = 0.05$ ). Supplementing with killed instead of live yeast decreased concentrations of acetate ( $P = 0.06$ ), propionate ( $P = 0.05$ ) and total VFA ( $P = 0.05$ ). Supplementing with HDY instead of CON increased lactate ( $P = 0.14$ ) concentration and acetate to propionate ratio ( $P = 0.08$ ) and decreased the minimum ruminal pH ( $P = 0.10$ ). The low dose of live yeast increased dry matter and neutral-detergent fiber digestibility, milk production and feed efficiency but the high dose of killed or live yeast did not.

**Key Words:** yeast, dairy cow, milk yield

**W356 Pre-weaning calf responses to lysine: II. Sensitivity and optimization of nitrogen retention responses to dietary lysine and body weight.** Robin R. White\*, Juan J. Castro Marquez, and Mark D. Hanigan, Department of Dairy Science, Virginia Tech, Blacksburg, VA.

The objective of this study was to use a model relating lysine (Lys) intake and weight gain in calves to identify optimal alternative protein sources for use in calf diets to minimize cost of gain (COG). A nonlinear optimization was used to minimize COG by adjusting milk replacer formulation subject to biological and practical constraints. The optimizer balanced milk replacer formulas for calves on a weekly basis from wk 1 through 8 of age. Body weight was diet dependent and updated weekly. Energy and mineral requirements of calves were calculated weekly following NRC (2001). Biological, practical and economic constraints were used to define the feasible set of ingredient combinations. Daily Lys intake was used to estimate nitrogen retention for each age group and change in body protein content was tracked. A literature search of calf slaughter balance studies was used to derive allometric equations relating body protein content to body fat, water and ash content. These functions were differentiated and change in body weight was calculated as the sum of the changes in body protein, fat, water and ash. Diet cost was estimated from current ingredient prices and dietary inclusion rates. Sensitivity of the model outputs to feedstuff Lys, protein content and digestibility was performed by taking 10,000 random draws of these parameters from normal distributions, running the growth model and calculating final weight and COG. Standard deviations in feedstuff Lys, protein content and digestibility were sourced from the National Animal Nutrition Program website. The optimal diet relied on a milk protein base with steadily increased inclusion of wheat gluten as animals aged. Blood meal and meat meal were also identified as key alternative protein sources. Model solutions were very robust. After 8 weeks calves weighed 87.8 ± 0.1 kg and COG was \$0.358/kg ± \$0.001/kg. Protein content and digestibility and Lys significantly ( $P < 0.001$ ) affected COG and BW and protein digestibility was quantitatively the most important parameter to estimate.

**Key Words:** calf, nitrogen retention, optimization

**W357 Effects of corn treated with foliar fungicide at various times of application on in situ ruminal digestibility of corn silage in Holstein cows.** Katie J. Haerr\*<sup>1</sup>, Naina M. Lopes<sup>2</sup>, Japheth D. Weems<sup>1</sup>, Carl A. Bradley<sup>1</sup>, Marcos N. Pereira<sup>2</sup>, Michael R. Murphy<sup>1</sup>, Gary M. Fellows<sup>3</sup>, and Felipe C. Cardoso<sup>1</sup>, <sup>1</sup>University of Illinois, Urbana, IL, <sup>2</sup>Universidade Federal De Lavras, Lavras, MG, Brazil, <sup>3</sup>B.A.S.F. Corporation, Research Triangle Park, NC.



The main objective was to explore the associations of corn plant foliar fungicide application on harvested whole-plant corn silage digestibility. Treatments were control (CON), corn received no foliar fungicide application; treatment 1 (1×), corn received one application of pyraclostrobin foliar fungicide (PYR; Headline; BASF Corp. Florham Park, New Jersey) at corn stage V5; treatment 2 (2×), corn received 2 applications of foliar fungicides, PYR at corn stage V5, and a mixture of pyraclostrobin and metconazole (PYR+MET; Headline AMP; BASF Corp. Florham Park, New Jersey) at corn stage R1; and treatment 3 (3×), corn received 3 applications of foliar fungicides, PYR at corn stage V5, PYR+MET at corn stage R1, and PYR+MET at corn stage R3. Corn was harvested at the same time and ensiled for 7 mo. Three cannulated lactating multiparous Holstein cows (376 ± 28 DIM) were used to measure corn silage in situ digestibility. Dried unground corn silage was put into 288 (3 time points/treatment/cow) 10 × 20 cm bags and incubated for 0, 2, 4, 8, 12, 48, 72 or 96 h. A sample of unground undried corn silage was also placed into a 20 × 40 cm bag and incubated for 48 h. The degradable portion of DM was greater ( $P = 0.01$ ) for corn silages treated with fungicide when compared with CON (0.36, 0.42, 0.40 and 0.47 for CON, 1×, 2×, and 3×, respectively). There was a linear ( $P < 0.001$ ) effect of treatment frequency on the proportion of DM digestibility. The 2 different sizes of bags used (10 × 20 vs. 20 × 40 cm) for the in situ digestibility technique were different ( $P < 0.02$ ) for DM, NDF, ADF, CP, and starch for 48h digestibility. Larger bags had greater ( $P < 0.03$ ) digestibility for DM (33, 35%), and lower digestibility for NDF (42, 35%), and ADF (47, 39%), than smaller bags. In conclusion, corn silage with fungicide application had higher DM digestibility and a trend for a lower rate of digestion as well as linear effects for decreasing DM solubility, increasing DM degradability, and decreasing rate of DM digestion.

**Key Words:** in situ digestibility, corn silage, fungicide

**W358 A multivariate mixed effects model to estimate the energetic efficiencies of synthesizing milk components.** Luis E. Moraes\*<sup>1</sup>, James G. Fadel<sup>1</sup>, David P. Casper<sup>2</sup>, and Ermias Kebreab<sup>1</sup>, <sup>1</sup>University of California-Davis, Davis, CA, <sup>2</sup>South Dakota State University, Brookings, SD.

The current Northern American feeding system for dairy cows suggests that new feeding systems calculate milk energy requirements as individual requirements for milk fat, protein and lactose. Extant models from the literature provide efficiencies that are unrealistically larger than the partial efficiency of utilizing dietary metabolizable energy (ME) for milk production ( $k_L$ ) or require inputs that are often not available to nutritionists when formulating diets. The objective of this study was to develop a model to estimate partial efficiencies of simultaneously synthesizing milk fat, protein and lactose from dietary ME and body stores. The model was developed as a multivariate mixed effects model and it was structured such that the partial efficiencies of synthesizing individual milk components additively determined  $k_L$  and the partial efficiency of utilizing body stores for milk production ( $k_T$ ). The new energy balance function described the trivariate response vector comprising milk fat, protein and lactose energy outputs as functions of the animal's ME intake and tissue energy balance. The model was fitted with 930 energy balance records from 244 Holstein lactating cows. Measurements were conducted through indirect calorimetry at the former USDA Energy Metabolism Unit at Beltsville, Maryland. The partial efficiencies of synthesizing milk fat, protein and lactose additively determined a  $k_L$  of 0.62 (0.009) and a  $k_T$  of 0.80 (0.020) which are in good agreement with the partial efficiencies adopted by the NRC (2001). The net energy requirement for maintenance ( $NE_M$ ) estimated with this

model was 0.34 (0.015) MJ/kg BW<sup>0.75</sup> which is virtually identical to the  $NE_M$  adopted by the NRC (2001). The determination of the ME required to synthesize each milk component may be further evaluated with the estimation of the fraction of the ME intake directed to each milk component and through the estimation of the partial efficiencies of individually synthesizing each milk component. Therefore, the estimated partial efficiencies can be directly implemented in the current Northern American net energy system.

**Key Words:** energy, efficiency, milk component

**W359 Effects of feeding slow-release NPN and microbial fermentation extracts on ruminal pH, ammonia and volatile fatty acids.** Fernando Diaz-Royón\*<sup>1</sup>, Alvaro D. Garcia<sup>1</sup>, Kenneth F. Kalscheur<sup>2</sup>, and Kamal Mjoun<sup>3</sup>, <sup>1</sup>Dairy Science Department, South Dakota State University, Brookings, SD, <sup>2</sup>U.S. Dairy Forage Research Center, USDA-ARS, Madison, WI, <sup>3</sup>Alltech, Brookings, SD.

The objective of this study was to examine the effect of partial substitution of soybean meal with a product containing slow-release NPN and microbial fermentation extracts [(OPT); Optimase Alltech Inc., Nicholasville, KY] on rumen fermentation. This is a companion study to a performance experiment (Díaz-Royón et al., 2014, J. Dairy Sci. 97 (E-Suppl. 1):332). Four cannulated, lactating Holstein cows were randomly assigned to a 4 × 4 Latin square in a 2 × 2 factorial arrangement of treatments. High (HF) and low forage (LF) diets contained, respectively, 61% and 46% forage, with and without OPT. Forages consisted of 75% corn silage and 25% alfalfa hay (DM basis). Experimental diets containing OPT (125 g/cow/day) were designed to partially replace soybean meal 48% CP (0.625 kg) with forage fiber [corn silage (0.225 kg) and alfalfa hay (0.075 kg)] and non-forage fiber [soybean hulls (0.200 kg)]. Increasing forage produced the expected pH increase (6.15 vs. 6.05 for HF and LF, respectively;  $P < 0.01$ ) whereas ruminal pH was not affected by OPT inclusion. Ruminal ammonia concentration tended to be greater in cows fed HF than LF diets (8.07 vs. 7.28 mg/dL;  $P = 0.08$ ), whereas OPT had no effect. Cows fed HF diets had a lower total VFA concentration compared with those fed LF (86.5 vs. 91.6 mM;  $P = 0.02$ ). For cows fed HF diets, molar proportions of acetate (66.0 vs. 64.4%;  $P < 0.01$ ), isobutyrate (0.89 vs. 0.82%;  $P < 0.01$ ), and isovalerate (1.80 vs. 1.58%;  $P < 0.01$ ) were greater, whereas those for propionate were lesser (19.4 vs. 21.1%;  $P < 0.01$ ). Total VFA proportions increased by 6.4% in HF diets supplemented with OPT; however, it decreased by 4.4% on LF diets (forage × OPT,  $P$  value = 0.03). Feeding OPT increased molar proportions of acetate ( $P < 0.01$ ), decreased propionate ( $P < 0.01$ ), isobutyrate ( $P < 0.01$ ), and isovalerate ( $P < 0.01$ ), but did not change molar proportions for butyrate and valerate. Partial replacement of soybean meal with OPT in dairy cow diets altered ruminal fermentation in a manner that suggests improved fiber digestion.

**Key Words:** slow-release NPN, microbial fermentation extract, dairy cows

**W360 Relationship between total-tract starch digestibility and fecal starch content in dairy cows.** Marcos N. Pereira\*<sup>1,2</sup>, Eugenio F. Barbosa<sup>1</sup>, and Renata A. N. Pereira<sup>3,2</sup>, <sup>1</sup>Universidade Federal de Lavras, Lavras, MG, Brazil, <sup>2</sup>Better Nature Research Center, Ijaci, MG, Brazil, <sup>3</sup>Empresa de Pesquisa Agropecuária de Minas Gerais, Lavras, MG, Brazil.

Fecal starch content has utility to estimate starch digestibility in dairy herds. We evaluated the relationship of total-tract starch digestibility and

fecal starch content in individually fed lactating Holsteins. The data set from 5 experiments conducted at the Better Nature Research Center had 190 cow/observations fed at ad libitum intake. Starch content of feces, orts, and feeds were analyzed enzymatically at the Federal University of Lavras laboratory (Hall, 2009). Fecal production was measured by total collection of feces in buckets by trained personal. Feces were collected concurrent to defecation during 3 8 h sampling periods and weighed. The second and third sampling periods begun 8 h later than the previous sampling, to avoid a major disturbance to the animals while still representing a 24-h collection period. Fecal aliquots (equal fresh weight basis) were immediately frozen during the collection period and a composite sample was formed. Diets contained 37 to 42% whole plant corn silage in DM and mature finely ground corn grain and/or high moisture corn at 17 to 26% of DM. Cows BW, DMI, and milk yield (MY) during the period of fecal sampling were obtained. Mean  $\pm$  SD (Min-Max) were: BW 636  $\pm$  73 kg (500–860), DMI 20.74  $\pm$  2.93 kg/d (11.88–27.83), MY 31.16  $\pm$  6.22 kg/d (13.70–47.20), DMI/BW 3.28  $\pm$  0.45% (2.07–4.57), MY/DMI 1.51  $\pm$  0.25 (0.70–2.41), fecal starch content 5.09  $\pm$  2.42% of DM (1.05–13.08), starch intake 6.021  $\pm$  0.985 kg/d (3.374–8.356), consumed diet starch content 29.06  $\pm$  2.84 (22.74–37.72), starch digestibility 94.96  $\pm$  2.60% of intake (86.24–99.18). The correlation coefficient (*r*) of starch digestibility and fecal starch content was  $-0.924$ , higher than with starch intake ( $-0.219$ ), diet starch content ( $0.136$ ), DMI ( $-0.362$ ), DMI/BW ( $-0.376$ ), MY ( $-0.363$ ), and MY/DMI ( $-0.121$ ). Based on stepwise procedure, Starch digestibility (% of intake) =  $100.02 - 0.9945 \times \text{fecal starch content (\% of DM)}$  ( $r^2 = 0.854$ ,  $P < 0.01$ ). Model  $R^2$  was 0.878 when diet starch content was included and 0.882 when MY was further added (0.15 significance required for entry into the model). Fecal starch content was a good predictor of starch digestibility for this data set of cows sampled on similar experimental condition and fed corn based diets.

**Key Words:** starch in feces, starch digestibility, corn digestion

**W361 Relationship between milk urea nitrogen and milk protein ratio with dietary and non-dietary variables in commercial dairy herds.** Liliana Fadul-Pacheco<sup>\*1</sup>, Doris Pellerin<sup>1</sup>, P. Yvan Chouinard<sup>1</sup>, Michel. A. Wattiaux<sup>2</sup>, and Edith Charbonneau<sup>1</sup>, <sup>1</sup>Université Laval, Quebec, Quebec, Canada, <sup>2</sup>University of Wisconsin-Madison, Madison, WI.

Milk urea nitrogen (MUN) can be used as a tool for the nutritional and environmental management of dairy cows as an estimate of the efficiency of protein metabolism. Using MUN to milk protein ratio (MUN/PROT) was suggested to improve the prediction. Thus, the aim of this study was to better understand the relationships between MUN and MUN/PROT with non-nutritional and nutritional variables in commercial dairy herds. Data recorded by Valacta (Dairy Production Center of Expertise Quebec-Atlantic) for the years 2009 to 2011 was used and originally comprised 3,481,705 test-day records (275,758 cows in 3,140 herds). The analysis was restricted to data from Holstein cows, between 1 and 305 DIM, reducing the number of admissible records for the analysis to 1,339,156 (189,913 cows in 2,522 herds). Average ( $\pm$ SD) milk production, fat content and protein content were 30.8  $\pm$  8.5 kg/d, 3.89  $\pm$  0.60% and 3.25  $\pm$  0.33%, respectively. MUN was statistically different between milkings (10.1 and 10.7 mg/dL for AM and PM, respectively;  $P < 0.001$ ). MUN/PROT was also statistically different between milkings (31.1 and 32.1 mg urea-N/kg milk CP for AM and PM, respectively;  $P < 0.001$ ). Average MUN was statistically different between parities (10.2 and 10.5 mg/dL for first and second or more lactation, respectively;  $P < 0.001$ ). MUN/PROT was also affected by parity (30.7 and 32.0 mg urea-N/kg milk CP for first and second or more lactation, respectively;

$P < 0.001$ ). Pearson correlations between MUN or MUN/PROT and diet variables were low, the highest correlation for both parameters being  $r = 0.16$  ( $P < 0.001$ ) with crude protein content. Both parameters were negatively correlated with milk fat percentage ( $r = -0.19$  and  $r = -0.29$  for MUN and MUN/PROT, respectively;  $P < 0.001$ ) and with the estimated breeding value for protein content ( $r = -0.10$  and  $r = -0.22$  for MUN and MUN/PROT, respectively;  $P < 0.001$ ). MUN/PROT was also correlated with daily milk production ( $r = 0.20$ ;  $P < 0.001$ ). These results show that in commercial conditions MUN and MUN/PROT are affected by factors other than diet composition.

**Key Words:** lactating dairy cow, MUN, MUN/protein ratio.

**W362 Effects of corn treated with foliar fungicide at various times of applications on milk production of Holstein cows.** Katie J. Haerr<sup>\*1</sup>, Naina M. Lopes<sup>2</sup>, Marcos N. Pereira<sup>2</sup>, Gary M. Fellows<sup>3</sup>, and Felipe C. Cardoso<sup>1</sup>, <sup>1</sup>University of Illinois, Urbana, IL, <sup>2</sup>Universidade Federal De Lavras, Lavras, MG, Brazil, <sup>3</sup>B.A.S.F Corporation, Research Triangle Park, NC.

The objective of this study was to determine if corn treated with foliar fungicide and ensiled as corn silage would increase milk production and efficiency in dairy cattle. This study utilized 64 Holstein cows with parity 2.53  $\pm$  1.5, BW 653  $\pm$  80 kg, and 161  $\pm$  51 d in milk (DIM). Cows were blocked and randomly assigned to 1 of 4 treatments to be included in the total diet (35% of the DM as corn silage). Treatments were as follows: Control (CON), corn silage with no applications of a foliar fungicide, Treatment 1 (1 $\times$ ), corn silage received application of pyraclostrobin (PYR) foliar fungicide (Headline; BASF Corp.) at corn stage V5; treatment 2 (2 $\times$ ), corn received the same application as 1 $\times$  and another application of a mixture of PYR and metconazole (MET; Headline AMP; BASF Corp.) at corn stage R1; and treatment 3 (3 $\times$ ), corn the same applications as 2 $\times$  as well as third application of PYR and MET at R3. Corn was harvested at the 3/4 milk line stage of growth and ensiled for 200 d. The trial was conducted in 2 consecutive periods each consisting of 1 wk for adaptation (covariate) followed by 5 wk of measurements where cows received assigned treatments. Body weight, BCS, and lame scores were assessed weekly. Milk production, and DMI were measured daily. Milk samples for milk composition analysis were collected during wk 5. Blood samples were taken on d1 (covariate) and d 29 to assess blood metabolites. Data were analyzed using a MIXED procedure in SAS (v9.4). Dry matter intake was 23.78, 22.95, 19.54, and 21.33 kg for CON, 1 $\times$ , 2 $\times$  and 3 $\times$ , respectively. There was a linear ( $P = 0.08$ ) tendency for DMI. Milk yield (34.5, 34.5, 34.2, 34.4 kg/d) and milk components did not differ ( $P > 0.05$ ) among treatments. However, there were trends for increased FCM/DMI (1.65 vs. 1.47.  $P = 0.08$ ) and ECM/DMI (1.60 vs. 1.43.  $P = 0.09$ ) for cows fed corn silage with fungicide compared with CON ( $P < 0.09$ ). Serum glucose was lower for cows receiving treated silage with fungicide when compared with CON (51.1 vs. 63.4 mg/dL,  $P < 0.01$ ). In conclusion, cows receiving corn silage treated with foliar fungicide had increased feed conversion when compared with CON.

**Key Words:** milk yield, corn silage, fungicide

**W363 Increased plasma NEFA lowers the ratio of sphingomyelin to ceramide in Holstein cows.** J. Eduardo Rico<sup>1</sup>, Luciano S. Caixeta<sup>2</sup>, Yves R. Boisclair<sup>2</sup>, and Joseph W. McFadden<sup>\*1</sup>, <sup>1</sup>West Virginia University, Morgantown, WV, <sup>2</sup>Cornell University, Ithaca, NY.

Saturated fatty acids can antagonize insulin sensitivity in rodents by increasing ceramide (Cer) levels in liver and plasma. Cer can be

generated via de novo synthesis, sphingomyelin (SM) hydrolysis, or sphingolipid recycling. In turn, Cer can be glycosylated to form mono-hexosylceramide (GlcCer) and lactosylceramide (LacCer). Analyzing the ratio between sphingolipids, within identical acyl chain length and saturation, can be a means to understand Cer metabolism. Our objective was to evaluate whether an increase in plasma NEFA can modify the sphingolipid ratio in cows. Six nonpregnant, nonlactating Holstein dairy cows (682 kg ± 22), were used in a crossover design with treatments consisting of i.v. infusion (100 ml/h) of either saline (control) or triacylglycerol (TG) emulsion (Intralipid 20%; Frasenius Kabi) for 16 consecutive hours. The feeding level was set at 120% of estimated energy requirement. Blood was collected at routine intervals. LC/MS was used to profile sphingolipids in plasma. Log-transformed data were analyzed using a mixed model with repeated measures. Nonparametric correlations were analyzed. TG infusion increased plasma NEFA by 454% at 3 h relative to control ( $P < 0.01$ ) with no further increase at 16 h. Ratio of C16:0-SM to C16:0-Cer (C16:0 SM: Cer) decreased 9% by 3 h of TG infusion, relative to control ( $P < 0.05$ ). C22:0, C22:1, and C24:0 SM: Cer decreased 27, 52, and 17% by 16 h of TG infusion, relative to control ( $P < 0.01$ ). TG infusion did not modify C16:0-dihydro SM: Cer; however, TG-infusion lowered the ratio of C18:0-SM to C18:0-dihydro-SM ( $P < 0.01$ ). C16:0 and C18:0 Cer:GlcCer decreased 24 and 13% by 16 h of TG infusion, relative to control ( $P < 0.01$ ). In contrast, C24:0 Cer:GlcCer and C26:0 Cer:GlcCer increased with TG infusion, relative to control ( $P < 0.05$ ). C18:0, C22:0, and C24:1 GlcCer: LacCer increased in TG-infused cows, relative to control ( $P < 0.01$ ). C16:0 Cer:GlcCer and C24:0 SM: Cer were negatively correlated with NEFA, and C24:0 Cer:GlcCer was positively correlated with NEFA ( $P < 0.01$ ). Increasing plasma NEFA lowers the ratio of SM to Cer in dairy cows, consistent with a shift in metabolism toward Cer accumulation.

**Key Words:** ceramide, dairy cow, insulin resistance

**W364 Effects of clay (EcoMix) after an aflatoxin challenge on milk production and blood metabolism of Holstein cows.** Saige A. Sulzberger<sup>\*1</sup>, Sergey Melnichenko<sup>2</sup>, and Felipe C. Cardoso<sup>1</sup>, <sup>1</sup>University of Illinois, Urbana, IL, <sup>2</sup>United Minerals Group, Kyiv, Ukraine.

Oral supplementation of clay to dairy cattle has been reported to reduce toxicity of aflatoxin in contaminated feed. The objective of this study was to determine the effects of 3 levels of dietary clay (EcoMix) supplementation after an aflatoxin challenge. Ten multiparous rumen-cannulated Holstein cows (BW = 669 ± 20kg, 146 ± 69 DIM), were assigned to 1 of 5 treatments in a completely randomized replicated 5 × 5 Latin square design. Periods consisted of a 14-d (d 1–14) adaptation period followed by a 3-d aflatoxin challenge (d 14–16; 100 µg of AFB1/kg of diet DM administered intra-ruminally). Cows were sampled daily during the last week of the period (d 14–21). Treatments were: POS, no EcoMix plus aflatoxin challenge; 0.5%, 1%, 2%, percentage of dietary DMI as EcoMix; and CON, no EcoMix and no aflatoxin challenge. Statistical analysis was performed using the MIXED procedure of SAS. Two contrasts CONT1 (POS vs. CON), CONT2 (POS vs. average of 0.5%, 1%, and 2%) were compared along with the linear and quadratic treatment effects. Milk samples were collected daily and tested with AFM<sub>1</sub> snap test (SNP; IDEXX, Inc.). Cows in CON had no positive SNP tests, while cows in POS had 14.2 ± 0.44 ( $P < 0.001$ , CONT2) positive SNP tests. Cows supplemented with EcoMix had lower number of positive SNP tests (0.5% = 13.9, 1% = 13.1, 2% = 12.4, SEM = 0.4,  $P = 0.04$ ) than cows in POS (14.2, CONT2). Cows supplemented with EcoMix through the challenge had lower 3.5% FCM (0.5% = 38.2 kg, 1% = 39.3 kg, 2% = 38.4kg, SEM = 1.8,  $P = 0.06$ ), ECM (0.5% = 37.1 kg, 1% = 37.1

kg, 2% = 37.1 kg, SEM = 1.6,  $P = 0.05$ , and Fat kg/d (0.5% = 1.36 kg, 1% = 1.43 kg, 2% = 1.40 kg, SEM = 0.09,  $P = 0.09$ ) than cows in POS (41.3 kg, 39.6 kg, 1.5 kg, respectively, CONT2). Serum cholesterol and creatine phosphokinase concentrations were higher for cows fed EcoMix (232.6 mg/dL ± 8.3,  $P = 0.08$ , 140.1 U/L ± 9.3,  $P = 0.04$ , respectively) than cows in POS (227.4 mg/dL ± 8.4, 126.3 U/L ± 9.3, respectively). In conclusion, oral supplementation of EcoMix decreased the total number of positive SNP tests after an aflatoxin challenge and seems to work as a flow agent for aflatoxin contamination in dairy cattle.

**Key Words:** clay, aflatoxin, milk yield

**W365 Discrepancies in milk urea nitrogen analysis among milk processing laboratories in Pennsylvania.** Holley L. Weeks<sup>\*</sup> and Alexander N. Hristov, Department of Animal Science, The Pennsylvania State University, University Park, PA.

Milk urea-N is used by dairy nutritionists and producers to monitor dietary protein intake and is indicative of N utilization in lactating dairy cows. Two experiments were conducted to explore discrepancies in MUN results among 3 laboratories and one experiment to evaluate the effect of 2-bromo-2-nitropropane-1,3-diol (bronopol) on MUN. In Experiment 1, 10 replicates of bulk tank milk samples, collected from the Pennsylvania State University's Dairy Center over 5 consecutive days, were sent to 3 milk processing laboratories located in Pennsylvania. Average MUN differed ( $P \leq 0.05$ ) between Laboratory A (Foss 4000; 14.9 ± 0.40 mg/dL), Laboratory B (Foss FT + 600; 6.5 ± 0.17 mg/dL), and Laboratory C (Foss 6000; 7.4 ± 0.36 mg/dL). In Experiment 2, milk samples were spiked with urea at 0, 17.2, 34.2, and 51.5 mg/dL of milk. Two 35-mL samples from each urea level were sent to the 3 laboratories used in Exp. 1. Average analyzed MUN was higher than expected (calculated for each laboratory based on the control; 0 mg added urea): for Laboratory A (23.2 vs. 21.0 mg/dL;  $P = 0.001$ ), Laboratory B (18.0 vs. 13.3 mg/dL;  $P < 0.001$ ), and Laboratory C (20.6 vs. 15.2 mg/dL;  $P < 0.001$ ). In Experiment 3, replicated milk samples were preserved with 0 to 0.30 g bronopol/35 mL milk (at 0.02 g increments) and submitted to one milk processing laboratory that analyzed MUN using 2 methods. Samples with increasing amounts of bronopol ranged in MUN concentration from 7.7 to 11.9 ± 0.27 mg/dL (linear increase due to bronopol;  $P < 0.001$ ) and from 9.0 to 9.3 ± 0.05 mg/dL ( $P = 0.06$ ) when analyzed on Foss 4000 or CL10, respectively. In this experiment, bronopol was calculated to contribute 7.9% of the total N in milk for the average milk vial (0.21 ± 0.003 g bronopol/ 35 mL milk) when milk protein was 3.01% and MUN was 8.5 mg/dL. In summary, MUN concentrations vary due to analytical procedures used by the milk processing laboratories. Amount of bronopol used to preserve the milk sample may also have an effect on MUN. Thus, it is important to maintain consistency in milk sample preservation and analysis to ensure precision and accuracy of MUN results.

**Key Words:** milk analysis, milk urea-N, bronopol

**W366 Plasma long-chain acylcarnitines are elevated in overweight dairy cows experiencing greater lipolysis and insulin resistance during late pregnancy.** J. Eduardo Rico<sup>\*</sup>, Rachel E. Cokeley, and Joseph W. McFadden, West Virginia University, Morgantown, WV.

Excess saturated fatty acids can impair mitochondrial β-oxidation in overweight monogastrics. In turn, long-chain acylcarnitines (LCAC) can accumulate in plasma, biomarkers for insulin resistance in humans. Our objective was to determine whether plasma LCAC are associated



with changes in estimated insulin sensitivity in dairy cows. Our data included multiparous Holstein cows grouped according to BCS at d -30 prepartum: lean (BCS < 3.0; n = 10) or overweight (BCS > 4.0, n = 11; OVER), with blood collected at d -45, -30, -15, -7, and 4, relative to calving. Profiling of LCAC was achieved using LC/MS. Data were analyzed using a mixed model with repeated measures including the random effect of cow and the fixed effects of BCS and time. Nonparametric correlations were analyzed. We previously demonstrated that OVER had greater NEFA mobilization and lower insulin sensitivity at d -30, -15 and -7 prepartum ( $P < 0.05$ ), relative to lean. LC/MS detected C14:0-, C16:0-, C18:0-, and C20:0-LCAC, representing 12, 31, 48, and 9% of total plasma LCAC, respectively. Concomitant with lipolysis, plasma C14:0-, C16:0-, C18:0-, and C20:0-LCAC, and total LCAC increased ( $P < 0.01$ ). Plasma C16:0- and C20:0-LCAC, and total LCAC levels were greater in OVER at d -7 ( $P < 0.05$ ). Plasma C14:0- and C18:0-LCAC levels were higher in OVER ( $P = 0.10$ ). A BCS  $\times$  time interaction was observed, where OVER had greater plasma C18:0- and C20:0-LCAC levels at d -7 ( $P = 0.06$  and  $P < 0.05$ , respectively). Plasma C14:0-, C16:0-, C18:0-, and C20:0-LCAC, and total LCAC were positively correlated with NEFA ( $P < 0.01$ ). Utilizing previously acquired GC/MS data, circulating C16:0- and C18:0-LCAC were positively correlated with mobilized palmitic acid, stearic acid, and lipolysis marker glycerol ( $P < 0.01$ ). Plasma C14:0-, C16:0-, C18:0-, and C20:0-LCAC, and total LCAC were negatively correlated with insulin sensitivity ( $P < 0.01$ ). Our data demonstrate greater plasma LCAC in insulin resistant, overweight periparturient dairy cows; however, the origin and involvement of LCAC in the progression of insulin resistance is unclear.

**Key Words:** acylcarnitine, insulin resistance, periparturient dairy cow

**W367 Effects of the dose and viability of *Saccharomyces cerevisiae* yeast on the diversity of ruminal microbes as analyzed by Illumina MiSeq sequencing and qPCR.** Y. Jiang<sup>\*1</sup>, I. M. Ogunade<sup>1</sup>, S. Qi<sup>2</sup>, F. Owens<sup>2</sup>, B. Smiley<sup>2</sup>, W. Rutherford<sup>2</sup>, C. Staples<sup>1</sup>, and A. T. Adesogan<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Florida, Gainesville, FL, <sup>2</sup>DuPont Pioneer, Johnston, IA.

The objective was to examine the effect of the dose and viability of *Saccharomyces cerevisiae* (SC) on the ruminal microbial population of lactating dairy cows. Four ruminally cannulated lactating cows (284  $\pm$  18 DIM) were used in a study with a 4  $\times$  4 Latin square design with four 21-d periods. Cows were fed a TMR supplemented with no SC (Control) or a low dose of live SC (5.7  $\times$  10<sup>7</sup> cfu/day; LLY), a high dose of live SC (6.0  $\times$  10<sup>8</sup> cfu/day), or a high dose of killed SC (6.0  $\times$  10<sup>8</sup> cfu/day before heating at 80°C). Ruminal fluid collected 0, 2, 4, 6, 8 and 10 h after the morning feeding on d 21 was strained through cheesecloth to obtain liquid (LF) and solid (SF) fractions. Microbial diversity was examined by high throughput Illumina MiSeq sequencing of the V4 region of the 16S rRNA gene. Populations of select ruminal bacteria and protozoa were also quantified by qPCR. Data were analyzed using the GLIMMIX procedure of SAS. In the SF and LF, *Prevotella* was the most abundant genus (23.8 and 49.1%) followed by *Fibrobacter* (10.7 and 1.5%) and *Succinivibrionaceae* (7.1 and 4.6%), respectively. Supplemental LLY increased the prevalence of *Butyrivibrio* ( $P = 0.11$ ) and *Ruminococcus* ( $P = 0.04$ ) in the LF. Supplementing with live instead of dead SC increased the prevalence of *Ruminococcus* in the LF and SF ( $P \leq 0.05$ ). Supplemental killed SC decreased the prevalence of *Lachnospiraceae* ( $P = 0.06$ ) and *Coproccoccus* ( $P = 0.13$ ) in the SF and increased those of *Ruminobacter* and *Porphyromonadaeae* in the SF and LF ( $P \leq 0.12$ , respectively). The qPCR results showed that in the SF, supplementing with LLY or with live instead of killed SC increased the prevalence of *F. succinogenes* ( $P = 0.14$  and 0.04, respectively). Supplemental killed

SC decreased the prevalence of protozoa ( $P = 0.05$ ) and *B. fibrosolvans* ( $P = 0.14$ ). In the LF, supplemental LLY decreased the prevalence of *R. albus* ( $P = 0.12$ ) and increased that of *S. ruminantium* ( $P = 0.14$ ). Unlike the high dose of live or killed SC, feeding the low SC dose or live instead of killed SC increased the prevalence of fiber-degrading bacteria.

**Key Words:** yeast, rumen microbes, MiSeq

**W368 Effects of direct-fed *Bacillus pumilus* 8G-134 during the transition period on health of Holstein cows.** Shaoyu Luan<sup>1</sup>, Elizabeth Galbraith<sup>2</sup>, Megan Duersteler<sup>2</sup>, and Felipe C. Cardoso<sup>\*1</sup>, <sup>1</sup>University of Illinois, Urbana, IL, <sup>2</sup>Dupont Nutrition and Health, Waukesha, WI.

The objective of this study was to evaluate the effects of a direct-fed microbial (DFM; *Bacillus pumilus* 8G-134) fed during the transition period on health of Holstein cows. Forty-three multiparous cows were fed a total mixed ration and assigned to 2 treatments in a randomized completely block design. Cows in the direct-fed microbial treatment (DFMt, n = 21) received 5.0  $\times$  10<sup>9</sup> cfu of *B. pumilus* direct-fed microbial in 28g of maltodextrin carrier, whereas cows in the control treatment (CON, n = 22) received 28g of maltodextrin carrier alone. Treatments were top-dressed daily. Treatments were applied from 21 d before calving to 154 d after calving. Blood samples were collected on d 5 and 14 after calving. Cows that had nonesterified fatty acids (NEFA) serum concentrations higher than 0.7 mEq/L were classified as high (HNEFA). Cows that had blood  $\beta$ -hydroxybutyrate (BHBA) concentrations higher than 1.2 mmol/L were classified as experiencing sub-clinical ketosis (SCK). Cows that had serum haptoglobin concentrations higher than 20  $\mu$ g/mL were classified as positive (POS). The immunoglobulins, IgA, IgG, and IgM were quantified in milk collected during the first week after calving and serum. Statistical analysis was performed using the MIXED, GLIMMIX and FREQ procedures of SAS. Cows on CON tended to have greater odds (OR = 3.55;  $P = 0.09$ ) of being classified as POS than CON cows on d 14. Treatment DFMt had higher (584  $\pm$  34  $\mu$ g/mL;  $P = 0.03$ ) IgA concentrations in milk than CON (478  $\pm$  35  $\mu$ g/mL) cows on the first week after calving. Cows on CON tended to have greater odds of being classified as HNEFA than DFMt cows on d 14 (OR = 3.21;  $P = 0.07$ ). Cows on CON tended to have greater odds of being classified SCK than DFMt cows on d 5 (OR = 3.85;  $P = 0.06$ ). Cows on CON tended to have higher odds for lower (<2) FS than DFMt cows (OR = 1.03;  $P = 0.08$ ). In conclusion, cows receiving DFMt tended to have lower incidence of SCK and to have higher immunity than cows receiving CON. Therefore, supplementation with DFMt seems to contribute for a sound transition period for dairy cows.

**Key Words:** transition period, direct-fed microbial, immunity

**W369 Bacterial communities in rumen fluid from lactating Holstein cows from Washington dairies.** Elizabeth D. Benda<sup>\*1</sup>, Nicola F. Beatty<sup>1</sup>, Janet E. Williams<sup>1</sup>, Matthew L. Settles<sup>1</sup>, John P. McNamara<sup>2</sup>, and Mark A. McGuire<sup>1</sup>, <sup>1</sup>University of Idaho, Moscow, ID, <sup>2</sup>Washington State University, Pullman, WA.

The rumen serves as a fermentation tank for bacteria to break down nutrients from feed for the benefit of the cow. Culture-dependent methods have identified the most prevalent genera as *Ruminococcus*, *Fibrobacter*, *Butyrivibrio*, and *Prevotella*. The objective of the study was to determine if the bacterial community in rumen fluid differed within a herd or across dairies within a given geographic region as assessed by a culture-independent method. In this study, rumen fluid samples from 82 cows across 4 different dairies in Washington were

collected via stomach tube and filtered through cheesecloth. Cows within a herd were in the same pen and fed the same ration. DNA was extracted using the Qiagen QIAamp DNA Stool Mini Kit, amplified via PCR with primers targeting the V1-V3 hypervariable region of the 16S rRNA, and amplicons sequenced using an Illumina Miseq v3 paired-end 300-bp protocol for 600 cycles. Sequence reads were processed using the custom python application dbcAmplicons. Sequencing data at different taxonomic levels were categorized by percent relative abundance for each cow. The least squares means of the top 15 genera were computed and compared among dairies to look at bacterial population differences using Generalized Linearized Mixed Models (SAS v9.3). Significance was declared at  $P \leq 0.05$ . Across all cows, *Prevotella* was the bacterial genus with the greatest relative abundance ( $44.8 \pm 1.06$ ), followed by *Hallella* ( $3.29 \pm 0.11$ ), *Treponema* ( $2.94 \pm 0.15$ ) and *Paraprevotella* ( $1.98 \pm 0.06$ ). Within a herd, the bacterial community of rumen fluid was similar among cows. However, each of the top 15 genera differed in abundance across dairies. These results suggest that cattle within a specific dairy have similar bacterial communities, but bacterial populations can differ across different dairies even within the same geographical region. Further work is necessary to determine the cause of the different levels of abundance of the top genera among dairies.

**Key Words:** rumen, microbial communities, dairy

**W370 Effects cobalt source on rate and extent of DM and NDF degradation *in vitro*.** Claudio F. Vargas-Rodriguez<sup>\*1</sup>, Abigail J. Carpenter<sup>1</sup>, Jeffrey DeFrain<sup>2</sup>, and Barry Bradford<sup>1</sup>, <sup>1</sup>*Kansas State University, Manhattan, KS*, <sup>2</sup>*Zinpro Corp., Eden Prairie, MN*.

Positive effects on fiber degradation have been observed when supplemental cobalt (Co) was added to diets for ruminants, but dose-dependent effects of different Co sources on ruminal fermentation have not been tested. Our objective was to determine the effects of different sources and concentrations of Co on *in vitro* fermentation rate, fermentation end products, and DM and NDF disappearance. Ruminant fluid was collected from heifers fed a wheat straw based diet (49%) with no supplemental Co, and fermentation substrate (46% NFC, 25% NDF, 22% CP) contained no measurable Co. Different inclusion levels (0.0, 0.1, 0.5, 1.0, 2.0, 5.0, 10.0, and 15.0 ppm) of Co glucoheptonate (S1) and Co carbonate (S2) were tested *in vitro* during study 1. Gas production was recorded every 15 min, and after 24 h, pH was measured and contents of each flask were used to determine NDF and DM disappearance (NDFD and DMD). Experiment 2 evaluated the effects of Co (S1 and S2 at 0, 0.33, 1, 3, and 9 ppm) on gas production, ruminal VFA and NH<sub>3</sub> concentration. In both studies, each treatment combination had 4 replicates and samples were incubated for 24 h; asymptotic gas production curves were modeled with NLIN of SAS using the Gauss-Newton fit method. Gas production kinetic values and all other data were modeled to assess the effects of Co concentration, source, and their interaction. Differences were declared at  $P < 0.05$  and tendencies at  $P < 0.10$ . NDFD was increased by S1 at concentrations between 0.1 and 1.0 ppm ( $55.0 \pm 1.8\%$ ), but decreased at 15 ppm (30.9%). S2 had no effect on NDFD across the concentrations tested. Moderate levels of Co tended to increase DMD, and S1 significantly increased this parameter compared with S2 ( $63.5$  vs.  $61.1\% \pm 1.6\%$ ), but interactions were not detected. Effects of Co source on gas production kinetics and pH change were inconsistent between experiments, largely because of dramatic negative effects at 15 ppm. Propionate concentration increased by S1 at 1.0 ppm, but not by S2 (dose by source interaction). Doses of 9 ppm Co decreased branched-chain VFA and NH<sub>3</sub> concentrations after 24-h incubation. In summary, Co does affect ruminal fermentation; however,

concentrations of 9 ppm and greater may be inhibitory, particularly with a highly soluble source of Co.

**Key Words:** mineral, fermentation, ruminant

**W371 Effects of long-term omega 6 fatty acid supplementation on blood metabolites of Holstein cows during transition period and early lactation.** Rodrigo Gardinal<sup>\*1,3</sup>, Gustavo Delfino Calomeni<sup>1</sup>, Filipe Zanferari<sup>1</sup>, Caio Seiti Takiya<sup>1</sup>, Thiago Henrique Aniballi Vendramini<sup>1</sup>, Jose Esler Freitas Junior<sup>2</sup>, Jose Eduardo Portela Santos<sup>3</sup>, and Francisco Palma Renno<sup>1</sup>, <sup>1</sup>*University of Sao Paulo, Sao Paulo, SP, Brazil*, <sup>2</sup>*Federal University of Bahia, Salvador, BA, Brazil*, <sup>3</sup>*University of Florida, Gainesville, FL*.

The objective of this study was determine the effects of feeding omega 6 ( $\omega 6$ ) fatty acids (FA) during an extended period of dry period until early lactation on blood metabolites of dairy cows. Thirty Holstein cows were used in 4 experimental groups in randomized design. The animals were randomly distributed to the following diets: Groups -90, -60, -30 and 0, supplemented with  $\omega 6$  FA source (12% of whole raw soybeans - WRS, on DM basis) 90,60 and 30 d before expected calving date and from the calving date (0) until 90 d in milk. Diets were formulated according to NRC (2001) recommendations. Ether extract (EE) averaged 4.8 and 2.8% for diets containing WRS and without WRS respectively. The blood samples were taken on days -49, -35, -21, -14 and -7 of expected calving date, at calving and on d 7, 14, 21, 35 and 70 of lactation. Blood concentrations of glucose (GLU), total cholesterol (CHOL), very low density lipoprotein (VLDL), low density lipoprotein (LDL), high density lipoprotein (HDL), triglycerides (TRI), total protein (TP), urea, gamma-glutamyl transferase (GGT) and aspartate aminotransferase (AST) were measured. Data were subject to PROC MIXED of SAS 9.3, diet were considered as fixed effect and time, interaction between diet and time, animal and residual error as random effect. The data were analyzed using polynomial regression using PROC REG of SAS 9.3. Linear decrease ( $P < 0.05$ ) in GGT concentration and linear increase of TP and CHOL concentration were observed as the length of  $\omega 6$  FA supplementation was longer. Quadratic effect ( $P < 0.05$ ) was observed in AST blood concentration and the highest value was presented by cows of group -30 (86.4 U/L). Interaction effect ( $P < 0.05$ ) was detected in GLU, TRI and VLDL blood concentration. The higher length of  $\omega 6$  FA supplementation during pre-partum may improve hepatic metabolism of cows during transition period and early lactation.

**Key Words:** dry period, linoleic acid, whole raw soybean

**W372 Organic trace minerals during the transition period: 1. Supplementing Zn, Mn and Cu from AvailaMins and Co from CoPro improves postpartal performance of dairy cows.** J. S. Osorio<sup>\*1</sup>, E. Trevisi<sup>2</sup>, J. K. Drackley<sup>1</sup>, M. T. Socha<sup>3</sup>, and J. J. Loor<sup>1</sup>, <sup>1</sup>*University of Illinois at Urbana-Champaign, Urbana, IL*, <sup>2</sup>*Università Cattolica del Sacro Cuore, Piacenza, Italy*, <sup>3</sup>*Zinpro Corporation, Eden Prairie, MN*.

Beneficial effects of supplementing organic trace minerals (AAC) to lactating dairy cows have been reported. However, there are few studies evaluating AAC supplementation during the transition period. Thirty-seven Holstein dairy cows were enrolled at 60 d prior dry-off (~110 d before calving) and remained on experiment until 30 d in milk (DIM). Cows were offered a common diet supplemented entirely with inorganic trace minerals (INO) from -110 to -30 d before calving. Cows received a common prepartal (1.5 Mcal/kg DM, 15% CP) and postpartal (1.76 Mcal/kg DM, 18% CP) diet. Both diets were partially supplemented with

an INO mix of Zn, Mn, and Cu to supply 35, 45, and 6 ppm, respectively, of the total diet DM. Cows were assigned to treatments in a randomized complete block design, receiving an oral bolus with a mix of INO (n = 21) or AAC (n = 16) containing Zn, Mn, Cu, and Co to achieve 75, 65, 11, and 1 ppm, respectively, in total diet DM. Inorganic trace minerals were provided in sulfate form and AAC were supplied via AvailaZn, AvailaMn, AvailaCu, and CoPro (Zinpro Corp., Eden Prairie, MN). Blood glucose and NEFA were measured at -30, -14, 3, 15, and 30 DIM. Liver biopsies were harvested at -30, -15, 10, and 30 DIM. BHBA was measured via Precision Xtra every other day from 1 to 15 d postpartum. Data were analyzed using the MIXED procedure of SAS. Prepartal DMI was lower ( $P = 0.06$ ) in AAC cows. In contrast, a tendency ( $P = 0.11$ ) was detected for diet by time ( $D \times T$ ) interaction that resulted in ca. 2 kg/d greater postpartal DMI in AAC. Milk and milk protein yield had a  $D \times T$  ( $P < 0.05$ ), because AAC cows produced ca. 3.3 kg/d more milk and 0.14 kg/d more protein during the first 30 DIM. Although blood glucose, NEFA, and liver triacylglycerol were not affected ( $P > 0.56$ ) by diet, the Precision Xtra BHBA was lower ( $P = 0.02$ ) in AAC than INO (1.44 vs 2.18 mmol/L). The positive response in milk yield and milk protein observed in AAC cows might be partly explained by the beneficial effect of AAC on postpartal DMI. Greater BHBA in cows fed INO suggests a mild-to-severe state of ketosis.

**Key Words:** metabolite, trace mineral, transition cow

**W373 Evolution of milk freezing point depression during the year in Holstein and Normande dairy cows.** Catherine Hurtaud<sup>1</sup>, Elise Vanbergue<sup>1,2</sup>, Sophie Lemosquet<sup>1</sup>, Ségolène Colette<sup>3</sup>, Yves Gallard<sup>3</sup>, and Luc Delaby<sup>1</sup>, <sup>1</sup>INRA-Agrocampus Ouest UMR1348 Pegase, Saint-Gilles, France, <sup>2</sup>Institut de l'Élevage, F-35650 Le Rheu, France, <sup>3</sup>INRA, Domaine du Pin-au-Haras, Exmes, France.

Milk freezing point depression (FPD) content is regarded as being relatively stable in cows' milk during lactation. Since the 30s, its measurement is a means to determine if water has been added to the milk. However, results from commercial herds have suggested that changes in milk FPD occurred during the year. The reason of those variations is unclear. However, dairy cow feeding might be an explanation. The objective of this experiment was to compare the characteristics of milk (especially milk FPD measured by MIR spectrometry) based on low input grass-based system compared with corn silage-based system, across 2 breeds of dairy cows (Holstein vs. Normande). Sixty-four dairy cows were observed from calving to drying off. Two feeding systems were compared. The Intensive system was designed to maximize individual performance, with a high energy diet (in winter, corn silage with 30% concentrate; in spring, summer and autumn periods, pasture with 4 kg/d of concentrate supplemented with corn silage from July). The Grass system was designed to decrease inputs (in winter, conserved grass with no concentrate; in spring, summer and autumn, pasture with no concentrate). The experimental design was a continuous design. Data were analyzed by using SAS mixed procedure. There was no significant effect of breed and feeding system on milk FPD. There was a significant effect of month of sampling and month of lactation. Milk FPD did not change from calving to 7th month of lactation. After that date, it largely decreased. These effects could be related to the high temperature in summer, specific feeding (grazing), or restricted access conditions for watering. There was also a significant effect of rank of lactation. FPD was higher for multiparous cows. This difference could be due to the opening of mammary tight junctions and a different repartition of ions and lactose on both sides of the epithelial barrier. Surprisingly FPD did not correlate with lactose content the most important osmotic agent in

mammary epithelial cells. Some investigations have to be done to explain the mechanisms of FPD and its physiological variations.

**Key Words:** milk, dairy cow, freezing point depression

**W374 Influence of calcified seaweed supplementation on rumen pH, digestive efficiency, and health in lactating dairy cows fed an acidosis inducing diet.** B. P. Molloy<sup>\*1</sup>, E. W. Neville<sup>2</sup>, S. J. Taylor<sup>1</sup>, A. W. Fahey<sup>2</sup>, and F. J. Mulligan<sup>2</sup>, <sup>1</sup>Celtic Sea Minerals Ltd., Carrigaline, Cork, Ireland, <sup>2</sup>College of Food Science, Veterinary Medicine and Agriculture, University College Dublin, Dublin, Ireland.

Subacute ruminal acidosis (SARA) is a significant problem on intensively managed dairy farms throughout the world. High producing dairy cows are unable to acquire sufficient nutrients from forage-based diets to meet their needs during early lactation. Therefore, forage based diets are often supplemented with high-energy starch-rich ingredients to meet their caloric demand. High consumption of rapidly fermentable ingredients can cause excessive acidification of the rumen decreasing fiber digestion and milk fat %, inducing rumenitis, laminitis, reduced reproductive performance and liver abscesses. The objective of this experiment was to evaluate the potential for 3 different treatments to prevent SARA and the associated decrease in digestive efficiency. A highly fermentable diet containing 380 g of starch and 300g of total dietary NDF was fed to 4 ruminally fistulated lactating dairy cows. The diet was composed from grass silage, corn silage and concentrates and fed at a 45:55 forage to concentrate ratio. The diets were either not supplemented (control) or supplemented with calcified seaweed (CS), calcified seaweed and marine magnesium oxide (CS + MMgO) or sodium bicarbonate (SB) 4 treatments. A range of parameters investigating the effect of supplementation on rumen physiology were analyzed including pH, volatile fatty acid production, fiber digestion, rate of passage, total-tract digestibility, milk yield and milk quality. Rumen pH was measured every 10 min over 3 d during each experimental period, 25.4% of control, 3.2% of CS, 2.8% of CS + MgO and 13.2% of SB readings were <5.5. All treatments maintained rumen pH above 5.5 for significantly longer ( $P < 0.0001$ , respectively) than the control. The CS and CS + MMgO were significantly more effective ( $P < 0.0001$ ,  $P < 0.000$ , respectively) than the SB in maintaining rumen pH above 5.5 (min). Supplementing with CS or CS + MMgO has the potential to maintain rumen pH above 5.5 for a longer period of the day.

**Key Words:** acidosis, calcified seaweed, rumen

**W375 Evaluation of an on-farm tool to estimate physically effective neutral detergent fiber of forages and total mixed rations.** Sarah E. Schuling<sup>\*1</sup>, Eric J. Staudinger<sup>1</sup>, Jeff A. Rortved<sup>1</sup>, Paul M. Windschitl<sup>1</sup>, Greg L. Golombeski<sup>1</sup>, and Kurt W. Cotanch<sup>2</sup>, <sup>1</sup>Hubbard Feeds Inc., Mankato, MN, <sup>2</sup>William H. Miner Agricultural Research Institute, Chazy, NY.

The objective of this experiment was to evaluate the use of a 3 screen Penn State Particle Separator box (19.0-, 8.0-, and 4.0-mm screens; PSPS4mm) to estimate physically effective factor (pef) and physically effective neutral detergent fiber (peNDF) of forages and total mixed rations (TMR) using the Ro-Tap method as the gold standard. Samples of corn silage (CS; n = 21), alfalfa silage (AS; n = 21), and TMR (n = 20) were collected from 20 Midwest dairy farms in summer/fall 2014. Four gallons of each sample were collected and mixed thoroughly. Samples were split and analyzed for pef using PPS4mm and Ro-Tap with standard shaking methods. Sample nutrient composition was



determined at Dairyland labs (Arcadia, WI). The pef was calculated as percentage of sample retained above 4.0-mm and 1.18-mm screens for PSPS4mm and Ro-Tap, respectively. The peNDF (%DM) of each sample was calculated as pef x NDF. Dry matter intake, milk yield, and milk protein and fat content were recorded for each herd. The REG procedure of SAS was used to determine the relationship between pef and peNDF estimated using PSPS4mm and Ro-Tap. The Stepwise Selection procedure was used to determine variables that affect herd milk components. The PSPS4mm was a good predictor of pef and peNDF in TMR and forages ( $R^2 = 0.93$  and  $0.98$ ; slope =  $0.86$  and  $0.91$ , respectively). For the CS, AS, and TMR samples, average pef estimated with Ro-Tap was (mean  $\pm$  SD)  $87.4 \pm 3.99$ ,  $83.5 \pm 4.34$ , and  $64.3 \pm 6.24$ , respectively, and average pef estimated with PSPS4mm was (mean  $\pm$  SD)  $88.4 \pm 4.58$ ,  $87.6 \pm 2.92$ , and  $64.2 \pm 6.35$ , respectively. Significant variables for predicting herd milk fat content were pef, concentrate intake, and milk yield (model  $R^2 = 0.55$ ). Significant variables for predicting milk protein content were pef and forage intake (model  $R^2 = 0.42$ ). The PSPS4mm is a useful tool to estimate pef and peNDF of forages and TMR. The pef within and across sample type varied, so estimating pef of individual samples on-farm will allow for more precise formulation of ration peNDF, which affects herd milk components.

**Key Words:** physically effective NDF, Penn State Particle Separator, milk component

**W376 The effects of choice feeding during preweaning period on preweaning and postweaning growth performance of dairy calves.** Mohammad Wakil Hassani and Murat Gorgulu\*, *Cukurova University Agriculture Faculty Department of Animal Science, Adana, Turkey.*

The aim of the study was to investigate the effects of choice feeding in preweaning period on growing performance of calf performance pre and postweaning period. Twenty-eight male and 28 female Holstein calves were used to test 2 feeding systems (TMR, total mixed ration, containing 10% alfalfa hay and choice feeding) and 2 sex (male and female) in a factorial arrangement. Before weaning TMR calves were fed with TMR containing 90% calf starter and 10% alfalfa hay and after weaning all calves were fed with the same TMR containing 50% calf grower and 50% alfalfa hay. Choice fed calves were fed with feed ingredients in TMR ad libitum and simultaneously. The choice fed calves before weaning preferred the diet containing lower alfalfa (10% vs. 5.78%,  $P < 0.05$ ) and barley (52.29% vs. 15.87%,  $P < 0.05$ ), and higher wheat bran (17.28% vs. 30.07%,  $P < 0.05$ ) and SBM (17.73 vs. 45.39%,  $P < 0.05$ ). Sex had no significant effects on diet preferences ( $P > 0.05$ ). Choice feeding increased feed and nutrient intake (protein and fiber) and daily gain significantly ( $P < 0.05$ ). After weaning, sex and feeding system during preweaning period had no effects on any parameters investigated ( $P > 0.05$ ). But sex and feeding system interaction had significant effects on daily gain, feed and nutrient intakes ( $P < 0.05$ ). The male calves fed TMR before weaning consumed more feed and nutrients and had higher daily gain than females but, choice fed calves in both sex had similar daily gain, feed and nutrient intake after weaning. When overall performance were evaluated, male calves had higher daily gain than females ( $P < 0.05$ ). Sex  $\times$  feeding system interaction had significant effects on feed and nutrient intake ( $P < 0.05$ ). The male calves fed with TMR consumed more feed and nutrients than the females ones but this differences disappeared in choice feeding group. In conclusion, the results revealed that choice feeding may improve growth performance of calves by increasing protein intake before weaning and this effect may disappear after weaning. The female calf gave better response to

choice feeding in respect to feed intake. This work was supported by Research Fund of the Cukurova University.

**Key Words:** choice feeding, calf, feeding system

**W377 Effects of day of gestation and feeding regimen in Holstein  $\times$  Gyr cows on apparent total-tract digestibility, nitrogen balance, and fat deposition.** Polyana P. Rotta\*<sup>1,2</sup>, Sebastiao C. Valadares Filho<sup>1</sup>, Terry E. Engle<sup>2</sup>, Luiz Fernando Costa e Silva<sup>1,2</sup>, Marcos I. Marcondes<sup>1</sup>, Fernanda S. Machado<sup>3</sup>, Tathyane R. S. Gionbelli<sup>1</sup>, Breno C. Silva<sup>1</sup>, and Marcos V. C. Pacheco<sup>1</sup>, <sup>1</sup>Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil, <sup>2</sup>Colorado State University, Fort Collins, CO, <sup>3</sup>Embrapa Gado de Leite, Juiz de Fora, Minas Gerais, Brazil.

This study investigated how feeding regimen (FR) alters apparent total-tract digestibility, performance, N balance, excretion of purine derivatives, and fat deposition in Holstein  $\times$  Gyr cows at different days of gestation (DG). Forty-four pregnant multiparous Holstein  $\times$  Gyr cows with an average initial body weight of  $480 \pm 10.1$  kg and an initial age of  $5 \pm 0.5$  yr old were allocated to 1 of 2 FR: ad libitum (AL;  $n = 20$ ) and maintenance level (ML;  $n = 24$ ). Maintenance level was considered to be 1.15% of body weight on a dry matter (DM) basis and met 100% of the energy requirements, whereas AL provided 190% of total net energy requirements. Data for hot and cold carcass dressing, fat deposition, average daily gain, empty body gain, and average daily gain without the gravid uterus were analyzed as a  $4 \times 2$  factorial design. Intake, apparent total-tract digestibility, N balance, urinary concentration of urea, and purine derivatives data were analyzed as repeated measurements taken over the 28-d period. Pregnant cows were slaughtered on 4 different DG: 139, 199, 241, and 268 d. Overall, DM intake decreased as DG increased. This decrease observed in DM intake may be associated with the reduction in ruminal volume caused by the rapid increase in fetal size during late gestation. We observed an interaction for DM and organic matter apparent total-tract digestibility between FR and DG; at 150, 178, and 206 d of gestation, ML-fed cows had greater DM and organic matter apparent total-tract digestibility values than AL-fed cows. Rib fat thickness, mesentery, and kidney, pelvic, and heart fat were greater in AL-fed than in ML-fed cows at all DG, with the exception of rib fat thickness on d 139. Ad libitum-fed cows excreted more N in their feces and urine compared with ML-fed cows. Pregnant cows that were fed at maintenance had greater digestibility during some DG, excreted less N in feces and less N and urea in urine, and deposited less fat in the body. We therefore recommend ML (1.15% of body weight with 93% of roughage) as a FR for pregnant dry cows; however, during the last month of gestation, AL seems to be the most appropriate FR to avoid loss of body weight.

**Key Words:** ad libitum, maintenance, performance

**W378 Effects of day of gestation and feeding regimen in Holstein  $\times$  Gyr cows on maternal and fetal visceral organ mass.** Polyana P. Rotta\*<sup>1,2</sup>, Sebastiao C. Valadares Filho<sup>1</sup>, Terry E. Engle<sup>2</sup>, Luiz Fernando Costa e Silva<sup>1,2</sup>, Marcos I. Marcondes<sup>1</sup>, Mariana M. Campos<sup>3</sup>, Tathyane R. S. Gionbelli<sup>1</sup>, Luis H. R. Silva<sup>1</sup>, Edilane C. Martins<sup>1</sup>, Flavia A. S. Silva<sup>1</sup>, and Faider A. C. Villadiego<sup>1</sup>, <sup>1</sup>Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil, <sup>2</sup>Colorado State University, Fort Collins, Colorado, <sup>3</sup>Embrapa Gado de Leite, Juiz de Fora, Minas Gerais, Brazil.

This study investigated the influence of day of gestation (DG) and feeding regimens (FR) on maternal and fetal visceral organ mass in Holstein

× Gyr cows. Forty-four pregnant multiparous Holstein × Gyr cows with an average initial body weight of  $480 \pm 10.1$  kg and an average initial age of  $5 \pm 0.5$  yr were allocated to 1 of 2 FR: ad libitum (AL;  $n = 20$ ) or maintenance level (ML;  $n = 24$ ). Maintenance level was considered to be 1.15% of body weight (dry matter basis) and met 100% of the energy requirements; AL provided 190% of the total net energy requirements. Cows were individually fed a corn silage and concentrate-based diet composed of 93% roughage and 7% concentrate (dry matter basis) as a total mixed ration twice daily. Pregnant cows were slaughtered at 4 DG: 139 ( $n = 11$ ), 199 ( $n = 11$ ), 241 ( $n = 11$ ), and 268 ( $n = 11$ ) d, which was followed by necropsy. Mass of heart, liver, and gastrointestinal tract was greater in AL- than in ML-fed cows. Mammary gland mass was greater in AL- than in ML-fed cows, and the greatest mass was observed at 268 d of gestation. Feeding regimen did not influence fetal body weight in this study. The majority of the visceral organ masses were similar in fetuses from cows fed AL or ML. These data indicate that maternal feed restriction does not affect the development of most fetal organs or fetal development; however, some maternal organs are affected by the FR provided. Moreover, the negative effect on mammary gland mass caused by ML feeding will probably not affect the subsequent lactation because the crude protein concentration in the mammary gland increased with ML feeding. However, we suggest that the AL diet in pregnant dry cows should be provided with caution because the amount of fat in the mammary gland increased at 268 d of gestation.

**Key Words:** fetal development, gastrointestinal tract, mammary gland

**W379 Dietary supplementation of palm- versus high-linoleic safflower oil to mid-lactating Holstein cows: Intake and milk fat yield.** Shahryar Kargar<sup>1</sup>, Clayton M. Stoffel<sup>2</sup>, Lou E. Armentano<sup>3</sup>, and Francisco E. Contreras-Govea<sup>\*3</sup>, <sup>1</sup>Department of Animal Sciences, College of Agriculture, Shiraz University, Shiraz, Iran, <sup>2</sup>Papillon Agricultural Co., Easton, MD, <sup>3</sup>Department of Dairy Science, University of Wisconsin-Madison, Madison, WI.

Cows were fed diets supplemented with either palm oil (PO, rich in C16:0 and C18:1) or high-linoleic safflower oil (SO, rich in C18:2) at 1.5% of dietary DM. Sixty-four primiparous and multiparous, cows with an average of  $100 \pm 21.7$  DIM,  $48.6 \pm 10.3$  kg milk yield per d, and  $657 \pm 70.3$  kg body weight at trial initiation were fed the 2 diets for 56 d, after a 2-week covariance period. Thirty-two primiparous and multiparous cows were assigned to one diet, and the other 32 cows to the other diet. The experimental design was a randomized complete block, blocking by parity. Cows were housed in a free-stall barn equipped with roughage intake control system gates (Insentec BV, Marknesse, the Netherlands), which recorded individual cow feed intake continuously. Milk yield was measured daily, and milk composition and cow's body weight were measured weekly. Data were analyzed as a randomized complete block design with diet, parity, week, and interactions as fixed effects, cow as random effect, and week as repeated measurement (SAS Institute, 2003). There was not difference in DMI (26.6 kg/d) and milk yield (46.5 kg/d) between the 2 diets ( $P > 0.05$ ) but feeding PO instead of SO raised milk fat concentration (3.88 vs. 3.55%) and yield (1.79 vs. 1.65 kg/d) ( $P < 0.05$ ), but milk protein yield tended ( $P = 0.10$ ) to increase for PO (1.39 kg/d) than SO (1.36 kg/d). Feeding SO increased *trans*-C18:1 including *trans*-6/8, *trans*-9, *trans*-10, and *trans*-12. For cows fed PO vs. SO, yields of de novo (<16 carbons; 447 vs. 412 g/d) and preformed (>16 carbons; 701 vs. 662 g/d) fatty acids were no difference between the 2 diets. Yield of mixed origin fatty acids (C16:0 + C16:1) increased for cows fed PO (515 vs. 379 g/d), possibly due to less inhibition of endogenous synthesis of C16 when feeding less dietary C18:2; as well as providing more exogenous dietary C16 from palm

oil. These results confirm the greater milk fat depressing effects of oils containing higher concentration of C18:2, as safflower has, relative to a combination of C18:1 and C16:0 as palm oil.

**Key Words:** palm oil, high-linoleic safflower oil, milk fat

**W380 Short- and medium-term changes in glucose metabolism and insulin sensitivity of dairy calves offered different amounts of milk replacer early in life.** Cristina Yunta<sup>1</sup>, Marta Terré<sup>1</sup>, and Alex Bach<sup>\*2,1</sup>, <sup>1</sup>Department of Ruminant Production, IRTA (Institut de Recerca i Tecnologia Agroalimentàries), Caldes de Montbui, Spain, <sup>2</sup>ICREA (Institució Catalana de Recerca i Estudis Avançats), Barcelona, Spain.

The objective of the present study was to evaluate the consequences of 3 allowances of milk replacer (MR) during the first 2 mo of life on short- and medium-term glucose metabolism and insulin sensitivity (IS) of dairy replacement heifers. Forty-five newborn female Holstein calves ( $40.7 \pm 4.94$  kg) after receiving colostrum were randomly allocated to 4, 6, or 8 L/d of MR until 63 d of life ( $n = 15$ ). A glucose tolerance test (GTT) was performed at 42, 86 and 300 d of life. Area under the curve for plasma insulin (IAUC), clearance rates of glucose (CRG) and insulin (CRI), insulin to glucose rate (ItoG), and IS were calculated. Data were analyzed using a mixed-effects model with repeated measures. There were no differences ( $P = 0.67$ ) in CRG among treatments, although they decreased from  $10.1 \pm 0.55\%/min$  at 42 d of age to  $6.7 \pm 0.56\%/min$  at 300 d of age. Interestingly, CRI was greatest ( $P < 0.05$ ) at 42 d of age, and calves fed 8 L/d had the greatest ( $P < 0.05$ ) ICR throughout the study. Insulin release (measured as IAUC) after a GTT increased ( $P < 0.005$ ) with age, and the increase observed between 42 and 300 d of life was more ( $P < 0.05$ ) marked in calves that received 4 (from  $1,000 \pm 234.0$  to  $3,319 \pm 242.7$   $\mu U/mL \times 60$  min) or 6 L/d (from  $1,538 \pm 226.1$  to  $3,887 \pm 242.6$   $\mu U/mL \times 60$  min) than in those receiving 8 L/d (from  $1,735 \pm 226.1$  to  $2,940 \pm 242.6$   $\mu U/mL \times 60$  min). The amount of MR offered had short- and medium-term effects on ItoG, with calves fed 4 and 6 L/d having lower ( $P < 0.05$ ) values ( $145.2 \pm 16.60$   $\mu U/mg$ ) than calves fed 8 L/d ( $215.3 \pm 16.02$   $\mu U/mg$ ) independently of age. Insulin sensitivity tended ( $P = 0.07$ ) to be lesser in calves fed 8 than in calves fed 4 or 6 L/d ( $1.39 \pm 0.04$  vs  $1.82 \pm 0.05$  mL/min  $\times$   $\mu U/mL$  per kg of BW, respectively). This difference was mainly due to a low IS of calves fed 8 L/d at 42 d, but as age increased, IS tended ( $P = 0.06$ ) to become progressively similar among treatment groups. It is concluded that offering 4 L of MR twice daily elicits a decrease in IS and an increase in ItoG while animals are consuming MR, and the IS returns to normal values over time, but the increase in ItoG is maintained with age.

**Key Words:** calves, enhanced feeding, metabolism

**W381 Energy expenditure in crossbred (Holstein x Gyr) calves differing in phenotypic residual feed intake.** Juliana Mergh Leão<sup>\*1</sup>, Fernanda Samarini Machado<sup>2</sup>, Alexandre Lima Ferreira<sup>2</sup>, Mariana Magalhães Campos<sup>2</sup>, Juliana Campos Carneiro<sup>3</sup>, Paulo Campos Martins<sup>1</sup>, Juliana Aparecida Mello Lima<sup>2</sup>, Thierry Ribeiro Tomich<sup>2</sup>, Luiz Gustavo Ribeiro Pereira<sup>2</sup>, Rayanne Soalheiro de Souza<sup>1</sup>, and Sandra Gesteira Coelho<sup>1</sup>, <sup>1</sup>Universidade Federal de Minas Gerais-UFMG, Belo Horizonte, Minas Gerais, Brazil, <sup>2</sup>Embrapa Dairy Cattle, Juiz de Fora, Minas Gerais, Brazil, <sup>3</sup>Instituto de Ciências Agrárias da UFMG, Montes Claros, Minas Gerais, Brazil.

The aim of this study was to evaluate the energy expenditure in crossbred Holstein-Gyr (F<sub>1</sub>) calves at 50 d of age with different phenotypes for residual feed intake (RFI) by measuring the respiratory gas exchanges

with a face-mask. Eighteen calves were housed in individual sand bed stalls in the experimental farm of Embrapa Dairy Cattle (Coronel Pacheco, Brazil). All animals were fed 6 L/d of whole milk (TS; 11.75%), divided in 2 equal meals (7 and 15h). Solid diet consisting of 95% of pelleted calf starter (88% DM; 20% CP and 3% Fat) and 5% Tifton 85 hay (81% DM; 13.4% CP; 72.8% NDF; 32.3% ADF), and water were provided ad libitum from the first day of life. Phenotypic RFI was calculated for each animal as the difference between actual dry matter intake (DMI) and expected DMI. Expected DMI was computed for each animal by regressing average daily DMI on conceptus-adjusted mean BW<sup>0.75</sup> and conceptus-adjusted ADG over a 60 d period. Twelve animals were ranked by RFI into low (efficient) and high (inefficient) groups. Oxygen consumption, carbon dioxide and methane production data were recorded in calves with 50 d age using Sable System (Sable Systems, Henderson, NV) attached to a facemask over a 2 d period, 3 h after morning milk supply, during 20 min. Heart rate (HR) was recorded during 20 min with Polar equine transmitter and monitor (RS800CX G3, Polar Electro Inc., Finland). Daily energy expenditure was calculated as heat production estimated from Brouwer equation (Brouwer, 1965). Methane production was not detected and urinary nitrogen was neglected in calculations. Data were analyzed as a completely randomized design by ANOVA using the GLM procedure of SAS software (version 9.4; SAS Institute Inc., Cary, NC). High RFI calves had DMI 12.39% higher ( $P < 0.05$ ) than the low RFI group (1.07 and 0.941 Kg/d, respectively), but body weight was higher by 0.6% ( $P < 0.05$ ) for the low RFI group. Heart rate did not differ between the groups. High RFI animals presented higher energy expenditure (177.64 kcal/kg of metabolic weight – MBW) than low RFI animals (144.40 kcal / kg of MBW).

**Key Words:** facemask, heat production, performance

**W382 The effect of different energy and nitrogen sources on in vitro fiber digestion of high and low quality roughages.** Christian W. Cruywagen\* and Mari Strauss, *Stellenbosch University, Stellenbosch, South Africa.*

Three energy sources (starch, sucrose and pectin) were used alone (without any N addition), or in combination with either soybean meal or urea as N source to investigate their effect on the in vitro digestion of NDF from alfalfa hay and wheat straw. The ratio of energy source to forage substrate was 50:50 on a hexose equivalent:NDF basis. Forage control treatments, without any energy or N sources added, were also included. The Goering/Van Soest incubation medium was used with slight modification; ammonium bicarbonate and tryptose were omitted (for the treatments with energy sources alone), or replaced by a similar amount of N from either soybean meal or urea. In the forage control treatments, the medium was used without modification. The trial was repeated in 6 runs. Samples were incubated at 39°C for 6 or 30 h. The substrate control data were analyzed per time according to a main effects ANOVA. For the factorial experiment, the data were also analyzed per time, but according to a 3 way factorial ANOVA with substrate, energy source and nitrogen source as factors. The 3 way interactions were interpreted with LSD multiple comparisons and Bonferroni tests. Significance was declared at  $P < 0.05$ . After 6 h of fermentation, NDF digestion was higher ( $P < 0.01$ ) in alfalfa hay (13.6%) than in wheat straw (8.1%), but energy and nitrogen source had no effect in either forage. After 30 h of fermentation, sucrose with soybean meal (35.8%) did not differ from starch and soybean meal (29.0%), but compared with no supplementation (26.4%) it was the only combination that increased NDF digestion of alfalfa hay significantly ( $P < 0.01$ ). In wheat straw, pectin with soybean meal (16.5%) did not differ from sucrose with soybean meal (12.4%), sucrose with urea (12.6%), pectin alone

(15.0%) or starch alone (13.5%), but compared with no supplementation (11.5%) it was the only combination that increased wheat straw NDF digestion significantly ( $P < 0.02$ ). In the current study, it appeared that sucrose and soybean meal was the best energy-nitrogen combination for alfalfa hay NDF digestion, whereas pectin and soybean meal was the best combination for wheat straw NDF digestion.

**Key Words:** NDF digestion, energy source, nitrogen source

**W383 Milk fatty acid profile of dairy cows fed omega 3 and 6 fatty acid sources during transition period and early lactation.**

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The objective of this study was to evaluate the effects of omega 3(n-3) and 6 (n-6) fatty acids (FA) supplementation on milk FA profile of dairy cows in early lactation. Forty 8 Holstein cows were divided randomly distributed into 4 groups to receive one of the diets: 1) Control (C, n = 12), without FA supplementation; 2) Flaxseed (WF, n = 12), addition of 60 and 80 g/kg diet DM of whole flaxseed (n-3 FA source) during pre and postpartum, respectively; 3) Soybean (WRS, n = 12), addition of 120 and 160 g/kg diet DM of whole raw soybean (n-6 FA source) during pre and postpartum, respectively; 4) Calcium salts of FA (CSFA, n = 12), addition of 24 and 32 g/kg diet DM of Megalac E (n-6 FA source) during pre and postpartum. The diets were offered from 35 d of expected calving date until 84 DIM and were formulated according to NRC (2001). Milk samples were collected weekly, proportionally of each milking (6h00 and 15h00) and proportionally of each cow yield. The extraction of lipids was made according to Feng et al. (2004) and methylated according to Kramer et al. (1997). Fatty acids were quantified by gas chromatography. Data were subjected to PROC MIXED of SAS (2004) using as fixed effect diet and weeks, and using as random effect interaction between week and diet and residual error. Orthogonal contrasts were performed (C vs WRS+CSFA+CSFA; WRS vs CSFA and WF vs WRS+CSFA). Fat supplemented diets increased total saturated FA compared with control ( $P < 0.01$ ) and tended to increase C18 unsaturated FA ( $P = 0.08$ ). Fatty acids with less than 16C were decreased with and FA with higher than 16C were increased FA supplementation. Whole raw soybeans increased C18:3, C18:2 and decreased C18:1 trans-11 FA compared with CSFA. However, SCFA presented higher unsaturated / saturated ratio than cows fed WRS. Increased cis-9, trans-11 FA was observed in cows fed WRS and CSFA when compared with WF. Whole flaxseed did not increase unsaturated FA in milk and SCFA increased the total unsaturated FA acids in milk. Supplementation of n-3 and n-6 FA altered milk fatty acid profile of early lactating cows.

**Key Words:** calcium salts of fatty acid, flaxseed, soybean

**W384 The effect of feeding nucleotides on milk production, reproductive performance and immunity in lactating Holstein dairy cows.**

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Preliminary evidence suggests that nucleotide feeding improves fertility in dairy cows, most likely mediated by an improvement in general and (or) local immunity of the reproductive tract. The objective of this research was to determine the effect of supplementing nucleotides on dairy cow milk production, reproductive performance and immunity. Two hundred and 60 adult ( $\geq 2$  lactations) Holstein dairy cows (87 d in milk) were divided in 2 groups and fed a standard diet (40% forage, 60% concentrate) without (CONTROL) or with nucleotides supplement (NUCLEOFORCE COWS, Biobierica S.A.) included at 4 g/cow/day during a period of 7 mo (May through December, 2014). Milk production (daily) and composition (monthly) were measured throughout the experimental period, and days from calving to first insemination and calving to pregnancy recorded. All cows were re-vaccinated against clostridia after 6 weeks in treatment. Serum IgG ( $n = 160$ ) were determine immediately before and 8 weeks after vaccination, and mucus from the vagina ( $n = 34$ ) was collected on the day of insemination at least 6 weeks after vaccination to measure IgA concentrations. Data were analyzed with PROC GLM of SAS and differences declared at  $P < 0.05$ . There were no differences ( $P > 0.10$ ) in milk production (42.8 kg/d), milk fat content (2.85%) and yield (1.23 kg/d), milk protein content (3.13%) and yield (1.34 kg/d) or somatic cell counts (387,000 counts/mL) between treatments. However, the ratio pre-vaccination vs. post-vaccination serum IgG tended to be higher ( $P < 0.08$ ) in the nucleotides supplementation (1.04) compared with CONTROL (0.93) treatment. Concentrations of IgA in vaginal mucus were higher ( $P < 0.05$ ) in nucleotides supplementation compared with CONTROL (2274 vs. 2078 ng/mL) treatment. Results indicate that the supplementation with nucleotides improved the indicators of general and reproductive tract immunity in dairy.

**Key Words:** immunity, nucleotide, dairy cow

**W385 Evaluation of a proprietary blend of essential oil and cobalt on a commercial dairy.** Olivia Kuester\* and David Casper, *South Dakota State University, Brookings, SD.*

An 18 week field trial was conducted on a commercial dairy in southwest Minnesota equipped with 2 Lely robotic milking units. This study was designed to evaluate the response of feeding a proprietary essential oil and cobalt product (EOC) on the lactational performance and nutrient digestibility of lactating Holstein dairy cows. Cows were divided between 2 pens ( $57 \pm 2$  cows and  $59 \pm 3$  cows for treatment [EOC] and control [C] pens, respectively), based on cow parity ( $2.65 \pm 1.52$  and  $2.33 \pm 1.20$ ), days in milk (DIM) ( $184 \pm 103$  and  $154 \pm 94.2$ ), and milk production ( $35.4 \pm 11.3$  kg/d and  $36.9 \pm 11.3$  kg/d) before study initiation. Cows were fed either an EOC or C total mixed ration (TMR) 2x/d. Cows fed the EOC averaged  $2.72 \pm 0.11$  milkings/d and cows fed C averaged  $2.80 \pm 0.13$  milkings/d. Production data were collected daily from Lely Time for Cows (T4C) robotic milking software and was reduced to weekly observations for each cow. Fecal samples were collected weekly from each pen based on a composite from 15 cows. Weekly TMR samples were collected from EOC and C pens and were composited monthly. Milk samples were collected once every 2 weeks from all cows and were analyzed for milk components and somatic cell counts (SCC) by the Dairy Herd Improvement Association (DHIA). Daily milk production was not different ( $P = 0.26$ ) between EOC and C fed cows, but was numerically greater for cows fed EOC ( $41.9 \pm 1.91$  kg/d) than cows fed C ( $38.8 \pm 2.05$  kg/d). The deviation in milk yield was greater ( $P = 0.01$ ) for cows fed EOC (0.42 kg/d) than cows fed C ( $-0.75$  kg/d). Total-tract NDF digestibility was numerically greater ( $P = 0.36$ ) for cows fed EOC (54.4% NDFD) than cows fed C (50.6% NDFD). Cows fed EOC had lower ( $P = 0.0002$ ) milk urea nitrogen

(MUN) ( $15.2 \pm 0.14$  mg/dL) than cows fed C ( $15.9 \pm 0.14$  mg/dL). Feeding the proprietary EOC product on a commercial dairy operation, which used robotic milking units numerically increased milk production and total-tract NDF digestibility.

**Key Words:** essential oil, cobalt, commercial dairy

**W386 Evaluation of rumen undegradable protein sources on lactational performance of Holstein dairy cows.** Heather A.

Tucker<sup>1</sup>, Shane M. Fredin\*<sup>1</sup>, Heather M. Dann<sup>1</sup>, Kurt W. Cotanch<sup>1</sup>, Catherine S. Ballard<sup>1</sup>, Les W. Berghorn<sup>2</sup>, and Rick J. Grant<sup>1</sup>, <sup>1</sup>William H. Miner Agricultural Research Institute, Chazy, NY, <sup>2</sup>Afgritech LLC, Watertown, NY.

This study compared the effects of feeding a proprietary blend of heat-treated canola and soybean meal (AminoMax; Afgritech LLC; Watertown, NY) or heat-treated soybean meal (AminoPlus; Ag Processing Inc.; Omaha, NE) on lactational performance of dairy cows. Forty-two lactating Holstein dairy cows (21 per treatment) averaging  $866 \pm 98$  kg of BW and  $146 \pm 30$  DIM at study initiation were used in a randomized complete block design with a 2-wk covariate period followed by an 8-wk treatment period. Treatments were diets containing (DM basis) 7.7% AminoMax or AminoPlus. Diets also contained 19.3% conventional corn silage, 19.3% brown midrib corn silage, 14.0% hay crop silage, and 39.7% concentrate mix (excluding AminoMax or AminoPlus). Both diets contained 15.7% CP, 30.9% NDF, and 25.4% starch. Dry matter intake and milk yield were collected daily and milk composition was determined over 3 milkings once per week. Cow served as the experimental unit. Data from the last 7 d of the covariate period were used as a covariate adjustment. Data were analyzed by ANOVA using the MIXED procedure of SAS containing model effects of covariate, treatment, time, and treatment  $\times$  time. Dry matter intake ( $27.4 \pm 0.2$  kg/d), solids-corrected milk (SCM) yield ( $43.8 \pm 0.8$  kg/d), milk fat yield ( $1.76 \pm 0.04$  kg/d), and milk true protein yield ( $1.39 \pm 0.02$  kg/d) were unaffected by diet ( $P > 0.10$ ). Milk lactose yield tended to increase ( $2.14$  versus  $2.06 \pm 0.03$  kg/d;  $P = 0.09$ ) for cows fed AminoPlus. Milk urea N concentration was decreased for cows fed AminoMax ( $11.4$  vs.  $9.6 \pm 0.2$  mg/dL;  $P < 0.01$ ). Efficiency of SCM production (SCM/DMI;  $1.59 \pm 0.02$  kg/kg) and N efficiency ( $0.322 \pm 0.005$  kg/kg) were unaffected ( $P > 0.10$ ) by diet. In conclusion, lactational performance did not differ between the products, but N utilization appeared to be improved for cows fed AminoMax.

**Key Words:** lactation, rumen undegradable protein

**W387 Evaluation of industry growth chart equations from birth until first calving of Holstein heifers fed a high plane of nutrition.** Marie E. Iwaniuk\*<sup>1</sup>, Jill A. Davidson<sup>2</sup>, Catherine M. Bradley<sup>2</sup>, and Tom J. Earleywine<sup>3</sup>, <sup>1</sup>University of Maryland, College Park, MD, <sup>2</sup>Purina Animal Nutrition, Gray Summit, MO, <sup>3</sup>Land O'Lakes

*Animal Milk Products, Shoreview, MN.*

The 2013 Penn State Customized Heifer Growth Chart (PS) was created as a tool to enable dairy producers to create herd-based heifer performance goals and track the performance of individual heifers until first calving. The prediction equations used in the program were derived from body weight (BW) and hip height (HH) benchmarks established by the 2001 *Nutrient Requirements of Dairy Cattle* and as previously reported BW and HH prediction equations (Heinrichs et al., 1992; Kertz et al., 1998). These prediction equations were derived from data sets in which heifers were fed standard diets; however, these prediction equations have yet to be evaluated for heifers fed a higher

plane of nutrition. The objective of this study was to evaluate the PS prediction equations for BW and HH growth using data of heifers fed a high plane of nutrition at the Purina Animal Nutrition Center. PS growth curves for BW and HH were predicted using mature cow herd data as target parameters. To evaluate the PS equations, residuals were regressed on centered predicted values to detect the presence of mean and/or linear biases using methods published by St-Pierre et al. (2006). For BW, the linear bias was not significant ( $P = 0.2131$ ); however, a significant mean bias ( $P < 0.05$ ) indicated that the PS equations overestimate BW by an average of 1.63kg for heifers fed a high plane of nutrition. In regards to HH growth, a significant linear bias (0.53cm;  $P < 0.05$ ) demonstrated that the PS equations overestimate HH from wk 0 to wk 5 and underestimate HH from wk 20 to wk 100 for heifers fed a high plane of nutrition. In addition, a significant mean bias indicated that the PS equations underestimate HH by an average of 5.41cm ( $P < 0.05$ ). Moreover, the presence of mean biases for both BW and HH further indicates that collectively the PS equations do not accurately describe overall growth patterns of heifers fed a high plane of nutrition. Therefore, new BW and HH equations should be developed that adjust for level of nutrition.

**Key Words:** Penn State Customized Heifer Growth Chart, growth curve

**W388 Milk urea nitrogen as a predictor of urinary nitrogen excretion in late lactation dairy cows fed four levels of dietary crude protein.** Tiago Barros\* and Michel A. Wattiaux, *Department of Dairy Science, University of Wisconsin-Madison, Madison, WI.*

Our objective was to assess the relationship between milk urea nitrogen (MUN, mg/dL) and blood urea nitrogen (BUN, mg/dL), and the excretion of urinary nitrogen (UN, g/d), and urinary urea nitrogen (UUN, g/d) in late lactation dairy cows. Holstein cows (means  $\pm$  SD; 736  $\pm$  18 kg BW; 224  $\pm$  54 DIM at the start of the trial) were used in a 16-pen study with 8 cows per pen, and fed one of 4 TMR containing either 11.9, 13.4, 14.4, or 16.3% CP (DM basis) for 12 weeks. Cows ( $n = 4$  per pen) were sampled for BUN on wk 4, 8 and 12. Daily MUN (infrared analysis) was derived from sampling 4 cows per pen in 2 consecutive milkings in wk 3, 7 and 11. In the same 3 weeks, urine volume was estimated using creatinine concentration in spot urine samples collected 6 h before and 6 h after feeding in groups of 4 randomly selected cows in each pen. Linear models were built using PROC REG and differences in slopes were tested using PROC GLM in SAS. Week of sampling did not affect the slope between BUN and MUN ( $P = 0.11$ , Table 1), but affected the slope of the relationship between UN and MUN ( $P = 0.042$ ) and between UUN and MUN ( $P = 0.029$ ). Increased dietary CP resulted in increases in UUN/UN ratio (0.54, 0.68, 0.74, and 0.81). However, there was no difference in slope when UN or UUN were regressed against MUN. Excretion of non-urea-N was constant (40.8 g/d). Four published models showed a slope bias and over-predicted UN when applied to our data set (227, 214, 176 and 125 g/d compared with 116 g/d observed UN). The week effect observed here, and the inadequacy of previous models suggested that best equations to predict UN from MUN should consider stage of lactation.

*Contd.*

**Table 1 (Abstr. W388).**

Y variable	X variable	Week	Intercept	SE	Slope	SE	R <sup>2</sup>	RMSE
BUN	MUN	3	0.19 <sup>1</sup>	0.79	0.96	0.076	0.94	0.94
		7	1.70 <sup>1</sup>	1.23	0.75	0.116	0.75	1.72
		11	-1.99 <sup>1</sup>	2.20	1.18	0.214	0.68	1.81
UN	MUN	3	47.4	7.3	6.95 <sup>b</sup>	0.704	0.87	8.65
		7	29.3	9.1	8.15 <sup>ab</sup>	0.084	0.90	12.50
		11	3.70 <sup>1</sup>	11.4	10.90 <sup>a</sup>	1.106	0.87	9.32
UUN	MUN	3	5.80 <sup>1</sup>	5.98	7.93 <sup>b</sup>	0.576	0.93	7.07
		7	-12.2	9.46	9.62 <sup>ab</sup>	9.46	0.90	13.01
		11	-35.5	10.6	11.54 <sup>a</sup>	1.032	0.90	8.70

<sup>ab</sup>Slopes among weeks for each set of regressions with a different superscript differ at  $P < 0.05$ .

<sup>1</sup>Intercept is not different from zero at  $P < 0.05$ .

**Key Words:** nitrogen excretion, BUN, MUN

**W389 Phosphorous excretion and digestibility in Jersey and Holstein consuming corn milling co-products.** Gabriel Garcia Gomez\*, Alison Foth, and Paulk Kononoff, *University of Nebraska-Lincoln, Lincoln, NE.*

Excess dietary phosphorous (P) in dairy cows diet may result in increased excretion of this mineral. Additionally, P accumulation in the soil may be a result of high concentrations of P when manure is applied to cropland. The objective of this study was to evaluate P intake, digestibility and excretion when dairy cows consumed rations containing reduced fat distillers grains (RFDDGS). Data from this study originated from an energy balance study in which RFDDGS was included at 28.8% of the ration DM. In this study, corn was reduced from 22.9 to 8.95% and soybean meal was reduced from 14.8 to 0% of the ration DM in the control and co-product (Co-P) diet, respectively. The study included 8 Holstein (BW = 693.8  $\pm$  12.9 kg) and 8 Jersey (BW = 429.1  $\pm$  13.0kg) multiparous, lactating cows (93  $\pm$  20 d DIM) in a repeated switchback design. The concentration of P in the test treatments were 0.44% and 0.59%  $\pm$  0.01% DM for the control and Co-P diet, respectively. The intake and excretion of P was estimated through feed sampling and total collection of feces. All feed and fecal samples were analyzed for P. Concentration of P in feces was lower in control diet compared with Co-P (0.97 versus 1.27  $\pm$  0.05%, respectively;  $P < 0.01$ ). Excretion of P was less for cows fed the control diet compared with the Co-P diet (62.34 versus 89.70  $\pm$  3.82 g/d, respectively;  $P < 0.01$ ). The excretion of P per kg of milk yield was higher in cows fed Co-P diet compared with control diet (21.7 and 15.8  $\pm$  1.29 g/kg, respectively;  $P < 0.01$ ). There was no difference between Holstein and Jersey in concentration of P in the feces (1.16 vs 1.08  $\pm$  0.07%, respectively;  $P = 0.36$ ), digestibility (32.3 versus 29.0  $\pm$  2.83%, respectively;  $P = 0.40$ ) and P efficiency (19.4 vs 18.0  $\pm$  1.63g/kg, respectively;  $P = 0.55$ ) across treatments. Results of this study suggest that rations formulated containing RFDDGS should be adjusted for P to reduce P excretion by dairy cows.

**Key Words:** phosphorus, excretion, digestibility.

**W390 Relationship between protein molecular structure and protein metabolic characteristics of co-products from bio-oil processing (rapeseed meal, canola meal and soybean meal) in dairy cattle.** Xinxin Li\*<sup>1,2</sup>, Yonggen Zhang<sup>2</sup>, and Peiqiang Yu<sup>1,2</sup>, <sup>1</sup>*Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, Saskatchewan, Canada,* <sup>2</sup>*College of Animal Science and Technology, Northeast Agricultural University, Harbin, China.*

This study was aimed to detect the relationship between protein inherent molecular structure features and protein rumen degradation kinetics and intestinal digestibility of 3 types of co-products from bio-oil processing including rapeseed meal, canola meal and soybean meal for dairy cattle. Three sources of rapeseed meal, 3 sources of soybean meal and 3 sources of canola meal were collected in 2014. The protein molecular structure in terms of chemical functional groups in these feed samples was analyzed using attenuated total reflectance-Fourier transform infrared spectroscopy (ATR-FT/IR) instrument. Rumen protein degradation kinetics was determined using in situ nylon bag method. Intestinal digestibility of rumen undegraded feed protein were determined according to the 3-step in vitro procedure with 12h pre-rumen incubated feed samples. The PROC MIXED procedure of SAS 9.3 was used to analyze the univariate protein spectral data, protein degradation kinetics and intestinal digestibility data. After normality test of the data, PROC CORR of SAS with a SPEARMAN option was used to investigate the relationship between protein molecular structure and metabolic characteristics in dairy cattle. Significances were declared at  $P < 0.05$ . The results showed that the ratio of protein amide I to amide II spectral peak height had a highly negative correlation with rumen undegraded protein (RUP<sup>NRC</sup>;  $r = -0.67$ ,  $P = 0.05$ ), intestinal absorbable rumen degraded protein (IADP;  $r = -0.67$ ,  $P = 0.05$ ), intestinal digestible protein (IDP;  $r = -0.77$ ,  $P < 0.05$ ) and total digestible protein (TDP;  $r = -0.67$ ,  $P = 0.05$ ). Additionally, the ratio of protein secondary structure  $\alpha$ -helix to  $\beta$ -sheet height was negatively correlated with rumen undegradable protein (RUP<sup>NRC</sup>;  $r = -0.67$ ,  $P < 0.05$ ), rumen degradable protein (EDCP;  $r = -0.83$ ,  $P < 0.05$ ), and total digestible protein (TDP;  $r = -0.93$ ,  $P < 0.05$ ). The results indicated that protein molecular structure was highly related with its nutrient supply to dairy cattle.

**Key Words:** protein molecular structure, protein metabolic characteristics, correlation study

**W391 The effect of decreasing dietary cation-anion difference in the prepartum diet on dry matter intake, milk production and milk composition in multiparous Holstein cows.** B. M. Sweeney<sup>\*1</sup>, C. M. Ryan<sup>1</sup>, K. Zanzalari<sup>2</sup>, D. Kirk<sup>2</sup>, and T. R. Overton<sup>1</sup>, <sup>1</sup>Department of Animal Science, Cornell University, Ithaca, NY, <sup>2</sup>Prince Agri Products Inc., Quincy, IL.

The objective of this study was to determine the effect of decreasing dietary cation-anion difference (DCAD) in the prepartum period on dry matter intake (DMI), milk production and milk composition. Multiparous Holstein cows ( $n = 89$ ) were allocated randomly to one of 3 prepartum diets formulated with decreasing DCAD: CON (+17.5 mEq/100 g DM), MED (+3.6 mEq/100 g DM), or LOW (-10.9 mEq/100 g DM), beginning 24 d before expected parturition. Analyzed DCAD were +18.3, +5.9, and -7.4 mEq/100 g DM. Cows were fed a common postpartum diet from parturition until 63 d in milk. Repeated measures analyses were conducted using the MIXED procedure of SAS with linear and quadratic effects of decreasing prepartum DCAD as contrasts. A quadratic effect of decreasing DCAD on prepartum DMI was observed (CON = 14.6 kg/d, MED = 15.1 kg/d, LOW = 14.1 kg/d;  $P < 0.01$ ). Postpartum DMI as a percent of body weight through wk 3 increased linearly with decreasing DCAD (CON = 2.94%, MED = 3.04%, LOW = 3.15%;  $P = 0.03$ ) and tended to be increased through wk 9 ( $P = 0.07$ ). During wk 1 to 3, milk yield (CON = 40.5 kg/d, MED = 42.1 kg/d, LOW = 43.8 kg/d;  $P = 0.03$ ) and fat yield were increased linearly ( $P = 0.13$ ) by decreasing prepartum DCAD resulting in a trend for greater 3.5% fat-corrected milk ( $P = 0.07$ ). Protein percent decreased linearly ( $P < 0.01$ ) with no effect on yield. Lactose yield ( $P = 0.02$ ), total solids content ( $P = 0.01$ ) and total solids yield ( $P = 0.06$ ) were increased

linearly, with a trend for greater energy-corrected milk for cows fed a lower DCAD ( $P = 0.08$ ). Milk urea nitrogen increased linearly ( $P = 0.04$ ) and there tended to be a quadratic effect on somatic cell score ( $P = 0.06$ ). Effects of prepartum DCAD on milk yield and composition were smaller when assessed from wk 1 to 9. Cows fed lower prepartum DCAD had linearly decreased protein percent ( $P = 0.02$ ) through wk 9 with a trend for a quadratic effect on yield ( $P = 0.11$ ). Ultimately, there was no difference in fat or energy-corrected milk through wk 9. Feeding decreasing prepartum DCAD linearly increased DMI and milk yield during early lactation.

**Key Words:** dietary cation-anion difference, transition cow

**W392 Effect of *Bacillus pumilus* on performance of primiparous dairy cows fed low or high starch diets.** D. N. Lobão da Silva<sup>\*1</sup>, Z. Sawall<sup>1</sup>, J. Guillen<sup>1</sup>, E. Galbraith<sup>2</sup>, T. Parrott<sup>3</sup>, M. Endres<sup>1</sup>, and N. B. Litherland<sup>1</sup>, <sup>1</sup>University of Minnesota, Saint Paul, MN, <sup>2</sup>DuPont Nutrition and Health, Waukesha, WI, <sup>3</sup>DuPont Industrial Biosciences, Waukesha, WI.

*Bacillus* species offer advantages as direct-fed microbials (DFMs) because their spores tolerate the harsh environment of the gastrointestinal tract and some strains can produce enzymes such as xylanase which breaks down xylan and the bonds that link lignin to cellulose, potentially increasing dietary fiber digestibility. This study aimed to investigate the effects of *Bacillus pumilus* 8G-134 (BP) supplementation on performance of primiparous dairy cows fed low (20%, LS) or high (27%, HS) starch diets from calving to 112 DIM. We hypothesized that BP would improve total-tract nutrient digestibility and performance of cows fed LS or HS diets postpartum. Forty-eight ( $n = 12$ /treatment) primiparous cows were assigned to a common prepartum diet 42d before expected calving date. At calving, cows were fed a LS or HS lactation diet and BP carrier or BP was top dressed on the TMR once daily to provide  $5 \times 10^9$  cfu/cow/day until 112 DIM. Factors combined produced 4 treatments: LS+carrier (LSCO); LS+BP (LSBP); HS+carrier (HSCO); HS+BP (HSBP). Fecal samples collected on d 7, 14 were analyzed for NDFd. Data were analyzed using the MIXED procedure of SAS. Apparent total-tract NDFd was greater ( $P = 0.05$ ) for diets supplemented with BP, by 4.4% (LS) and 8.5% (HS) which tended to increase milk yield, 3.5% FCM and 3.5% ECM by 13.9%, 14.9% and 15.2% for HS diets, but increases in LS supplemented diets were less apparent. The 112 d of BP supplementation increased DMI and milk fat yield, and tended to improve yields of 3.5% FCM and milk protein. Results indicated that BP supplementation to primiparous cows can increase fiber digestibility and milk yield resulting in potential increases in profitability.

Contd.



**Table 1 (Abstr. W392).** Intake and lactation performance

Item (kg/d)	Treatments				SEM	<i>P</i> -value		
	LS		HS			S	BP	S × BP
	CO	BP	CO	BP				
<b>28 DIM</b>								
DMI	15.27	16.15	14.23	15.08	0.90	0.16	0.25	0.98
Milk	27.96	28.30	25.92	29.52	1.21	0.73	0.10	0.18
3.5% FCM	28.07	28.57	25.75	29.60	1.24	0.60	0.08	0.17
3.5% ECM	28.33	28.80	26.03	29.99	1.24	0.66	0.08	0.16
<b>112 DIM</b>								
DMI	18.97	20.58	18.85	20.30	0.94	0.73	0.01	0.89
3.5% FCM	31.63	33.00	30.87	33.62	1.71	0.95	0.06	0.51
Milk fat	1.10	1.15	1.06	1.18	0.07	0.87	0.04	0.42
Milk protein	0.97	1.03	1.00	1.07	0.04	0.30	0.06	0.92

**Key Words:** *Bacillus pumilus*, dietary starch, primiparous

**W393 Balancing dairy cattle diets for methionine or all essential amino acids relative to energy at negative and adequate levels of rumen nitrogen.** Ryan J. Higgs<sup>1</sup>, Brian K. Sloan<sup>2</sup>, Larry E. Chase<sup>1</sup>, Charles G. Schwab<sup>3</sup>, and Michael E. Van Amburgh<sup>\*1</sup>,

<sup>1</sup>Cornell University, Ithaca, NY, <sup>2</sup>Adisseo, Alpharetta, GA, <sup>3</sup>Schwab Consulting LLC, Boscobel, WI.

Updates have been made to the Cornell Net Carbohydrate and Protein System which includes estimations of recycled urea nitrogen, rumen protozoa production and yield, endogenous N secretions and a new system for calculating post-ruminal N digestion. The objective was

to evaluate the ability of the updated model to balance diets of high producing dairy cattle below or close to requirements for both rumen N and EAA. Sixty-four high producing dairy cows (100 ± 31 DIM) were randomly assigned to one of 4 treatments: 1) limited in Met, MP and rumen N (Base); 2) adequate in Met but limited MP and rumen N (Base+M); 3) adequate in Met and rumen N, but limited MP (Base+MU); 4) adequate in MP, rumen N and balanced for all EAA (Positive). Dietary CP was 13.5, 13.6, 14.6 and 15.6% DM and model predicted dietary MP balance was -231, -310, -142 and 33 g/d for the Base, Base+M, Base+MU and Positive treatments, respectively. No differences were observed in DMI or milk yield (24.1 to 24.8 and 40.0 to 41.8 kg/d, respectively). Energy corrected milk, fat and true protein yield were greater (3.3, 0.09 and 0.11 kg/d, respectively; *P* < 0.001) in cows fed the Positive compared with the Base treatment. True protein concentration in milk was higher (*P* < 0.001) and milk fat tended to be higher (*P* < 0.10) in cows fed the Positive and Base+MU treatments than cows fed the Base and Base+M treatments. Bacterial growth was predicted to be depressed by 16% and 17% for the Base and Base+M treatments, respectively, due to the predicted rumen N balance and this prediction corresponded with lower (*P* < 0.05) apparent total-tract NDF digestion. The study demonstrated high levels of milk yield can be achieved when diets are formulated on rumen N balance and EAA, even when CP is less than 14% DM provided adequate AA are supplied to the small intestine. The highest milk yield was achieved when considering all EAA in the ration formulation process.

**Key Words:** CNCPS, nitrogen, amino acid

## Ruminant Nutrition: General III

**W394 Effect of heat-treated field beans (*Vicia faba*) on lactation performance of dairy cows.** Mohammad Ramin\*, Annika Höjer, Märten Hetta, and Pekka Huhtanen, *Department of Agricultural Research for Northern Sweden, Swedish University of Agricultural Sciences (SLU), Umeå, Västerbotten, Sweden.*

In Sweden, the availability of organic protein feeds is rather limited (e.g., lack of soybean) and it was therefore the objective of this study to evaluate if the feeding value of heat-treated field beans (FB) could be improved. Other objective was to compare different protein supplements that could be used in organic farming on the performance of lactating dairy cows fed grass silage based diet. Twenty-four lactating Swedish Red cows (95 DIM, milk yield 29.1 kg per day) were used in a cyclic change-over trial with 3 21-d experimental periods. The control diet consisted of grass silage and barley (60:40, DM basis). For experimental diets barley was replaced with rapeseed expeller (RSE; 104 g/kg diet DM), or isonitrogenous supplements of peas (232), untreated FB (UFB; 140), heat-treated FB (TFB; 140) or heat-treated FB that provided the same dietary MP concentrations of UFB (80; TFB-low). Methane (CH<sub>4</sub>) and carbon dioxide emissions were measured with the Green-Feed system. On average, protein supplementation had no significant effect on DMI (18.8 vs. 18.2 kg/d), milk yield (23.8 vs. 23.5 kg/d) or ECM yield (25.6 vs. 24.8 kg/d). This was mainly because peas or FB supplemented diets did not increase milk or protein yield compared with the control diet. The RSE treatment increased milk (24.8 vs. 23.6 kg/d) and protein yield (913 vs. 863 g/d) compared with other protein supplements. Heat-treated FB had no effect on DMI, milk or protein yield compared with UFB. Milk nitrogen efficiency (Milk N / N intake) decreased and MUN increased with protein supplementation compared with the control diet (265 vs. 306 g/kg and 3.92 vs. 3.01 mmol/L, respectively). The RSE supplemented diet tended to decrease ( $P = 0.09$ ) CH<sub>4</sub> production compared with other protein supplements (383 vs. 399 g/d). Overall, this study suggests that in organic farming no improvement on the lactation performance of dairy cows were achieved by the inclusion of heat-treated FB or peas as compared with the control diet without any protein supplement, provided that RDP requirements are met. Only RSE supplementation resulted in significant improvements in animal performance compared with the control diet.

**Key Words:** heat treatment, methane production, organic feed

**W395 Pre- and post-weaning performance of dairy calves fed a milk-wheat-plasma protein milk replacer.** Max Thornsberry<sup>1</sup>, Steve Younker\*<sup>1</sup>, Dave Ziegler<sup>2</sup>, Hugh Chester-Jones<sup>2</sup>, and Jim Linn<sup>1</sup>, <sup>1</sup>*Milk Specialties Global, Eden Prairie, MN*, <sup>2</sup>*University of Minnesota, Southern Research and Outreach Center, Waseca, MN.*

Traditionally, replacing a portion of milk protein in calf milk replacer (CMR) with plant and animal proteins has lowered CMR cost, but reduced calf growth performance. Newer processing methods for wheat protein have improved digestibility and when combined with bovine plasma as a partial replacement for milk protein in a CMR resulted in similar performance of calves fed an all milk protein CMR. The objective of this study was to compare performance of calves fed either a CMR containing a blend (B-CMR) of milk proteins (9% units protein), hydrolyzed wheat gluten protein (5% units protein) and bovine plasma (8% units protein) or all milk protein CMR (AM-CMR). Fifty-two 2- to 5-d-old Holstein heifer calves were randomly assigned to one of 2 CMR treatments. The B-CMR and AM-CMR were similar (DM basis)

in CP (23%) and crude fat (21%) and fed at 0.54 kg DM/d (13% solids) from study d 1 to 35. Both B-CMR and AM-CMR contained Bio-Mos, ClariFly and 1600 g/ton each of Neomycin sulfate and Oxytetracycline the first 14 d of CMR feeding. BovaTec replaced oxy/neo from 15 d until weaning (d 42). From 35 to 42 d of study, CMR was fed at 0.27 kg DM/d. All calves were fed a common 22% CP (DM basis) texturized calf starter from study d 1 to 56. Data were analyzed as a randomized complete block design using PROC MIXED (SAS). Total intake of CMR DM from 1 to 42 d was similar at 20.8 kg DM with no differences in DM intake by weeks. Total calf starter DM intake from study d 1 to 42 was not different ( $P > 0.05$ ) at 27.6 kg for AM-CMR fed calves and 25.4 kg for B-CMR fed calves. Total 56 d calf starter intake was not different between treatments at 64.6 vs. 61.1 kg DM for AM-CMR and B-CMR fed calves. Average daily gain from 1 to 42 d of study tended ( $P = 0.08$ ) to be slightly lower for calves on B-CMR (0.69 kg/d) than calves on AM-CMR (0.75 kg/d). There was no difference ( $P > 0.05$ ) in daily gain from 1 to 56 d between B-CMR treatment calves (0.83 kg/d) and AM-CMR treatment calves (0.87 kg/d). Fecal scores (1 normal, 4 watery) were numerically lower (1.4 vs. 1.6) from d 1 to 14 and scour days were numerically lower (1.27 vs 1.88) from d 1 to 42 for B-CMR calves than AM-CMR calves, respectively.

**Key Words:** calves, milk replacer, protein

**W396 Effects of ruminal dosing of dairy cows with *Megasphaera elsdenii* on milk fat depression and strain persistence.** Paul J. Weimer\*<sup>1</sup>, Luciano da Silva Cabral<sup>2</sup>, and Felipe Cacite<sup>2</sup>, <sup>1</sup>*USDA-ARS, Madison, WI*, <sup>2</sup>*Federal University of Mato Grosso, Cuiabá, MT, Brazil.*

*Megasphaera elsdenii* (*Me*) is a lactate-utilizing bacterium whose ruminal abundance has been shown to be greatly elevated during milk fat depression (MFD). To further examine this association, a total of 23 cannulated multiparous Holstein cows were examined in 3 studies in which strains of *Me* were directly dosed into the rumen ( $\sim 2 \times 10^{12}$  cells/dose); control cows were dosed with sterile lactate-free culture medium. Cows were fed a TMR (30% starch, dry matter (DM) basis) that contained primarily corn silage, alfalfa silage, finely ground high-moisture corn, supplemental protein, and corn oil (3 g/kg DM). Studies differed in stage of lactation of the cows (middle or late), dosing events (single dose, or 4 doses over a 5 d period), timing of dose (pre-feed or 3 h postfeed), and *Me* strain (lab strain Y19 or 3 strains isolated from cows in the same herd). DM intake, milk yield and composition were measured from 5 d to 0 d before dosing and 1 to 7 d after first dosing, plus later time points that varied by experiment. None of these measures were affected by dosing ( $P > 0.05$ ). *Me* was quantified in the liquid phase of ruminal contents by polymerase chain reaction (PCR) with relative quantification (*Me* 16S rRNA gene copy number as a percentage of total bacterial 16S rRNA gene copies). Neither the *Me*-dosed or control cows displayed MFD after dosing, and in almost all cases *Me* populations returned to low baseline levels ( $< 0.02\%$  of 16S rRNA gene copy number) within 24 h of dosing. This rapid decline in *Me* also occurred in several cows that were dosed with a strain that had been isolated from that particular cow during a previous bout of MFD. Ruminal pH, total mM volatile fatty acids (VFA) and mM lactate did not differ between dosed and control cows ( $P > 0.05$ ), although acetate:propionate ratio declined ( $P < 0.01$ ) in both groups, and butyrate increased after dosing with *Me* ( $P < 0.05$ ). The results confirm that establishing exogenously added bacterial strains in the rumen is difficult, even for strains previously isolated from the

recipient cow. The potential role of *Me* as an agent of MFD remains unclear in the absence of successful establishment of the dosed strains.

**Key Words:** dosing, *Megasphaera elsdenii*, milk fat depression

**W397 Effects of Cerrado plant extracts on in vitro rumen microbial fermentation in high inclusion concentrate diets.** Flavia Martins de Souza\*, Barbara Juliana Martins Lemos, Fabiola Alves Lino, Kiria Karolline Gomes Moreira, José Realino de Paula, Victor Rezende Moreira Couto, and Juliano José de Resende Fernandes, *Universidade Federal de Goiás, Goiânia, Goiás, Brazil.*

The objective was to study the effect of the typical plant extracts of the Brazilian Cerrado on in vitro rumen microbial fermentation under a high concentrate diet. Two separate trials were conducted to evaluate 4 doses (0, 30, 300 and 3,000 mg/L) of stem bark extract from *Stryphnodendron adstringens* (Trial 1; 48.1% tannins) and *Lafoesia pacari* (Trial 2; 21.1% tannins). The trials were analyzed as a randomized block design with 5 repetitions, treating incubation as the blocking factor. Treatments were evaluated using a TE-150 incubator ((Tecnal) in a batch culture of rumen fluid with a 10:90 forage:concentrate diet (15% crude protein, 20% neutral detergent fiber) incubated using filter bags of non-woven textile for 2, 4, 8, 12, 24, and 48 h (triplicates each time) to fit the disappearance of dry matter (DM). Volatile fatty acids (VFA), pH, and ammonia nitrogen (NH<sub>3</sub>-N) were evaluated after 48 h of incubation. *S. adstringens* extract at 3,000 mg/L reduced ( $P < 0.05$ ) in 34.5% the potential degradability (a + b) of DM. There was significant reduction in total VFA concentration from 66.3 mM (0 mg/L) to 40.2 and 24.3 mM by adding 300 and 3,000 mg/L of *S. adstringens* extract, respectively. This was also observed for NH<sub>3</sub>-N concentration, representing a decrease of 36.9% (300 mg/L) and 57.8% (3,000 mg/L). The pH significantly increased (5.9 vs 6.1) with the addition of 3,000 mg/L. Regarding *L. pacari* extract, the addition of 3,000 mg/L promoted ( $P < 0.05$ ) reduction of 20.3% in the potential degradability of DM but detrimental effects on the fermentation profile were not observed. The 30 mg/L dose increased ( $P < 0.05$ ) the total VFA concentration (63.4 vs. 85.2 mM), decreased the acetate:propionate ratio (4.3 vs 3.6) and the proportion of isovalerate (1.9 vs 1.5 mM). Changes in the concentration of NH<sub>3</sub>-N were only observed at a dose 3,000 mg/L, with a reduction of 43.0%. The results indicated that Cerrado plant extracts are potential modifiers of rumen fermentation under a high concentrate diet.

**Key Words:** antimicrobial additive, *Lafoesia pacari*, *Stryphnodendron adstringens*

**W398 Effect of fibrolytic and aminolytic enzymes in ruminal metabolism and methane production in cattle.** Ricardo Galbiatti Sandoval Nogueira, Lizbeth Lourdes Collazos Paucar\*, Flavio Perna Jr, Eduardo Cuellar Orlandi Cassiano, Diana Carolina Zapata Vasquez, Lerner Arevalo Pinedo, and Paulo Henrique Mazza Rodrigues, *University of São Paulo, Pirassununga, São Paulo, Brazil.*

Methane (CH<sub>4</sub>), a greenhouse gas, can be held responsible for loss of 2–12% the gross energy of the diet. The objective of this study was to evaluate the effect of enzymes on ruminal metabolism and the production of enteric CH<sub>4</sub> in cattle. Five cows with average weight of 923.04 ± 86.76 kg were assigned to 5 treatments in 5 × 5 Latin square design: (1) diet without enzymes (control); (2) diet with 7.5 g/animal/day of amylase (Amaize, Alltech); (3) diet with 15 g/animal/day of xylanase (Fibrozyme, Alltech); (4) diet with 7.5 g/animal/day of cellulase + protease (Allzyme VegPro PO, Alltech); (5) Diet with 30g/ animal/day

of enzymes mixture (amylase, xylanase, cellulase + protease). Animals were fed twice daily at 0800 and 1600 h. Each experimental period consisted of 21 d, and collections were made in the last 6 d. On d 21, ruminal pH was measured every 10 min using a continuous measurement device and rumen contents were collected at 0, 3, 6, 9 and 12 h after feeding to determine the production of short chain fatty acids (SCFA) and CH<sub>4</sub> by ex situ technique. Data were analyzed using SAS, through the MIXED procedure. The model included the effect of treatment as fixed factors, animal and period effects as random factors. The addition of different enzymes in the cattle diet showed no significant difference ( $P > 0.05$ ) in relation to dry matter intake. There were significant differences ( $P < 0.05$ ) for SCFA production in relation to the control diet. The enzyme mixture improved the production of acetic acid, propionic acid and total SCFA, while protease + cellulase improved the production of propionic acid. The CH<sub>4</sub> production was not significantly difference among treatments ( $P > 0.05$ ). Also no significant differences ( $P > 0.05$ ) were observed for the pH variables, as minimum, average and maximum pH, as well as time and area where pH was below to 5.8, 6.0, 6.2 and 6.5. It is concluded that these additives did not affect the variables DMI, CH<sub>4</sub> production and pH. The association of the enzymes showed improvement in the production of rumen SCFA without increasing the emission of CH<sub>4</sub>.

**Key Words:** enzyme, methane, SCFA

**W399 Effects of diets containing glycerin, derived from biodiesel production, on instrumental meat quality characteristics of lambs.** Rebeca D. X. Ribeiro\*<sup>1</sup>, Ronaldo L. Oliveira<sup>1</sup>, Gleidson G. P. Carvalho<sup>1</sup>, Thadeu M. Silva<sup>1</sup>, André G. Leão<sup>2</sup>, Jonival B. Costa<sup>1</sup>, Sara M. Ribeiro<sup>1</sup>, Elisiane S. Santos<sup>1</sup>, and Abraão S. Nunes<sup>1</sup>, <sup>1</sup>University of Bahia, Salvador City, Bahia State, Brazil, <sup>2</sup>Federal University of Mato Grosso, Rondonópolis City, Mato Grosso State, Brazil.

Glycerin is a low-cost byproduct from biodiesel production, and is also a potential source of carbohydrates. This study was conducted to determine the optimal amount of glycerin in the diets of lambs. Forty-four crossbreed male lambs (23.9 ± 3.4 kg) were distributed in a completely randomized design, and fed with a TMR that contained 50% of *Cynodon sp.* chopped hay and 50% of concentrate composed of corn bran, soybean meal, mineral premix, and the amounts of glycerin: 0; 7; 14 and 21% of total DM. Those amounts were the treatments. The feed trial lasted 78 d. The last day the animals were submitted to feed fasting, and slaughtered. The carcasses were stored in a cold chamber for 24h, and then the *Longissimus lumborum* was collected, stored in a freezer, and all measurements were made on this muscle, after thawing. The pH was measured by a digital potentiometer (skewer type extremity) directly in the muscle. The color measurement was made using a colorimeter to determine the parameters: L\* - luminosity, a\* red content and b\* yellow content. Cooking losses were calculated by the weight difference from the samples before and after cooked, and results expressed in percentages. Tenderness was measured through the shear strength, using a texturometer equipped with a Warner Bratzler blade. The data were subjected to ANOVA and regression testing. Significance was declared as  $P < 0.05$ . There are no effects of glycerin inclusion on instrumental characteristics (Table 1). The similar values of pH, which is correlated with the glycogen source in muscle, can be related to the similarity among the other parameters. Glycerin, derived from biodiesel production, can be added up to 21.0% (DM) in the diets of lambs without compromising meat quality.

*Contd.*



**Table 1 (Abstr. W399).** Cooking losses (CL), shear force (SF), pH, and color of L. lumborum of lambs fed diets containing glycerin from biodiesel production

Item	Glycerin (%DM)				SEM	P-value	
	0.0	7.0	14.0	21.0		Linear	
CL (%)	28.25	25.01	25.74	30.71	1.64	0.27	
SF (Kgf)	3.41	2.41	2.43	3.45	0.30	0.92	
pH	5.60	5.82	5.65	5.67	0.06	0.87	
Color							
L	36.21	35.87	35.82	36.36	0.62	0.89	
a	19.48	19.08	19.36	19.65	0.36	0.63	
b	6.05	5.33	5.74	5.90	0.38	0.98	

**Key Words:** nutrition, ruminant

**W400 Effects of salinomycin and virginiamycin supplementation on ruminal microbial population in Nellore steers fed a high concentrate diet.** Amoracyr J. C. Nuñez<sup>\*1,2</sup>, Vivian V. Almeida<sup>1</sup>, Italo E. Borges<sup>2</sup>, Fabio Pinese<sup>2</sup>, Fernando T. Mercado<sup>2</sup>, Ligia G. Mesquita<sup>2</sup>, Juliane Diniz-Magalhães<sup>2</sup>, Luis Felipe P. Silva<sup>2</sup>, Paulo R. Leme<sup>2</sup>, and José Carlos M. Nogueira Filho<sup>2</sup>, <sup>1</sup>Purdue University, West Lafayette, IN, <sup>2</sup>University of São Paulo, Pirassununga, SP, Brazil.

This experiment was conducted to evaluate the effects of adding salinomycin (SL), virginiamycin (VM), or their combination to finishing diets of Nellore steers on the ruminal population of *Streptococcus bovis*, *Megasphaera elsdenii*, *Selenomonas ruminantium*, and methanogenic archaea. Eight ruminally cannulated Nellore steers (322 ± 26 kg initial BW) were allotted to a 4 × 4 replicated Latin square design with four 16-d periods. Experimental diets had 80% concentrate (DM basis), and treatments were arranged in a 2 × 2 factorial, with 2 SL levels (0 and 13 ppm) and 2 VM levels (0 and 15 ppm) in the diet DM. Animals were housed in individual pens and fed once daily at 0800 h. Ruminal fluid samples were collected at 0 and 4 h post-feeding on d 13 of each period, and DNA was extracted from samples using the Qiagen DNA stool mini kit. Ruminal bacteria and methanogenic archaea populations were assessed by real-time PCR. Statistical analyses were performed using the MIXED procedure of SAS. No interactions between SL or VM levels, nor between time of collection and SL or VM levels were observed. The relative population of *S. bovis* tended to decrease ( $P = 0.09$ ) for animals fed diets containing 15 ppm VM in comparison with those not receiving the additive, but no effects of dietary treatment were observed for any other microbial species (Table 1). There was an effect of time of collection in the relative populations of *M. elsdenii*, *S. ruminantium*, and methanogenic archaea, which were greater ( $P \leq 0.01$ ) in samples collected at 0 h post-feeding. In conclusion, adding SL, VM, or both to the diets of Nellore steers had little effect on the populations of ruminal bacteria and methanogenic archaea.

**Table 1 (Abstr. W400).** Relative populations of ruminal bacteria and methanogenic archaea in Nellore steers fed diets with 2 levels of salinomycin (SL) or virginiamycin (VM)

Item	0 ppm SL		13 ppm SL		SEM	P-value		
	0 ppm	15 ppm	0 ppm	15 ppm		SL	VM	SL × VM
	VZM	VM	VM	VM				
<i>S. bovis</i>	1.00	0.58	0.86	0.49	0.03	0.61	0.09	0.97
<i>M. elsdenii</i>	1.00	1.09	1.09	0.69	0.02	0.43	0.44	0.26
<i>S. ruminantium</i>	1.00	0.79	1.22	0.97	0.04	0.33	0.26	0.99
Methanogenic	1.00	0.64	0.71	1.24	0.08	0.61	0.84	0.11

**Key Words:** antibiotic, bacteria, ionophore

**W401 Productive performance of lambs fed diets containing glycerin from biodiesel production.** Ronaldo L. Oliveira<sup>\*1</sup>, Rebeca D. X. Ribeiro<sup>1</sup>, Gleidson G. P. Carvalho<sup>1</sup>, Thadeu M. Silva<sup>1</sup>, André G. Leão<sup>2</sup>, Jonival B. Costa<sup>1</sup>, Sara M. Ribeiro<sup>1</sup>, Elisiane S. Santos<sup>1</sup>, and Abraão S. Nunes<sup>1</sup>, <sup>1</sup>University of Bahia, Salvador City, Bahia State, Brazil, <sup>2</sup>Federal University of Mato Grosso, Rondonópolis City, Mato Grosso State, Brazil.

Glycerin is a by-product from biodiesel production, an important alternative fuel. It is also a low-cost source of carbohydrates. This study was conducted to determine the optimal amount of glycerin to include in the diets of lambs. Forty-four crossbred male lambs (23.9 ± 3.4Kg) were distributed in a completely randomized design, and fed with a TMR that contained 50% *Cynodon* sp. chopped hay and 50% concentrate composed of corn bran, soybean meal, mineral premix, and the amounts of glycerin: 0; 7; 14 and 21% of total DM. Treatments varied in the amount of glycerin included. During the performance trial the animals were fed ad libitum (diet was offered to allow 10% of orts). The feed and orts were weighed daily. The feedlot trial lasted 78 d, and on the last day the animals were weighed, submitted to feed fasting, and slaughtered. A general linear model was used to perform linear and quadratic regression. Significance was declared as  $P < 0.05$ . The glycerin inclusion promoted increase in final BW, BW gain, and gain:feed ratio up to 5.8, 3.8, and 2.75% of inclusion respectively (Table 1). After that, glycerin inclusion reduced those parameters. The initial increase can be related to the increment on energy intake, because the glycerin used in this study contained 30% of lipids. However, the increase in lipid intake can also reduce fiber digestibility, and then cause decrease in the DM intake, which is related to reduction in productive performance parameters. Glycerin, from biodiesel production, can be added between amounts of 2.75% and 5.8% without compromising the productive performance of lambs.

**Table 1 (Abstr. W401).** Productive performance of lambs fed diets containing glycerin from biodiesel production

Item	Glycerin amount (% of DM)				SEM	P-value	
	0	7	14	21		Lin	Quad
Final BW, kg	37.87	40.45	35.98	32.26	1.21	<0.001	0.0007 <sup>1</sup>
BW Gain, kg	14.91	15.58	12.34	8.00	0.65	<0.001	0.0007 <sup>2</sup>
ADG, g/d	191.24	199.83	158.28	102.62	0.85	<0.001	0.0007 <sup>3</sup>
Gain:Feed	0.155	0.149	0.141	0.102	0.01	<0.0001	0.025 <sup>4</sup>

$$^1Y = -0.0321X^2 + 0.3707X + 38.26.$$

$$^2Y = -0.0256X^2 + 0.1944X + 15.051.$$

$$^3Y = -0.3278X^2 + 2.4924X + 193.04.$$

$$^4Y = -0.0002X^2 + 0.0011X + 0.1541.$$

**Key Words:** by-product, nutrition, ruminant

**W402 Ruminal hydrogen sulfide concentrations on low-quality grass hay by lambs drinking saline water.** Agustín López<sup>\*1,3</sup>, José I. Arroquy<sup>1,2</sup>, Ana V. Juárez Sequeira<sup>2,3</sup>, Nicolás DiLorenzo<sup>4</sup>, Héctor Fissolo<sup>1</sup>, and María C. Barrionuevo<sup>3</sup>, <sup>1</sup>INTA EEA Santiago del Estero, Santiago del Estero, Argentina, <sup>2</sup>CONICET, Santiago del Estero, Argentina, <sup>3</sup>FAyA - Univ. Nac. Santiago del Estero, Santiago del Estero, Argentina, <sup>4</sup>University of Florida, NFREC, Marianna, FL.

The objective of this study was to determine the impact of protein supplementation on dry matter intake and ruminal hydrogen sulfide concentration ([H<sub>2</sub>S]) in lambs consuming low-quality tropical grass hay and drinking low salt water (LS) or high salt water (HS). Twenty

lambs in individual cages were used in a 10 treatments by 2 period (10 × 2) trial. First factor was water quality (WQ): LS (466 mg/kg TDS) vs. HS (8927 mg/kg TDS of which 6680 mg/kg as sulfates). Second factor was 5 levels of soybean meal (SBM; %BW/d). Water quality × SBM interactions were significant for forage (FOMI;  $P < 0.05$ ) and total OM intake (TOMI;  $P < 0.05$ ), but not for water (WI;  $P = 0.60$ ) and sulfur water intake (SWI;  $P = 0.14$ ). At the higher levels of SBM (>0.5%) there was no statistical difference for FOMI and TOMI. Water intake was not affected by WQ ( $P = 0.39$ ) but increased linearly ( $P < 0.01$ ) in response to SBM. Ruminal H<sub>2</sub>S concentration was affected by WQ × SBM interaction ( $P < 0.01$ ). Our results suggest further research is needed on the water quality × protein interaction to understand the mechanisms involved in H<sub>2</sub>S production, to generate efficient supplementation strategies on low-quality forages.

**Table 1 (Abstr. W402).** Effects of SBM supplementation and WQ on sulfur water intake (SWI) and ruminal H<sub>2</sub>S concentration in lambs fed low-quality grass hay

Water quality	SBM	SWI, mg/kgBW <sup>0.75</sup>	H <sub>2</sub> S (ppm) <sup>1</sup>
Low salt	0	5.10	1.23 (16.98)
	0.25	4.90	1.19 (15.49)
	0.50	5.26	1.24 (17.38)
	0.75	5.79	1.20 (15.85)
	1	6.48	1.70 (50.12)
High salt	0	328.67	1.59 (38.90)
	0.25	278.49	2.69 (489.78)
	0.50	359.00	3.32 (2089.30)
	0.75	383.06	2.99 (977.24)
	1	454.38	2.98 (954.99)
SEM		33.74	0.20
Effects			<i>P</i> -value
WQ		<0.01	<0.01
SBM		0.20	<0.01
WQ × SBM		0.14	0.01

<sup>1</sup>Means were back transformed.

**Key Words:** sulfate, hydrogen sulfide, protein supplementation.

#### W403 Effects of Cerrado plant oils on in vitro rumen microbial fermentation in high inclusion concentrate diets.

Flavia Martins de Souza\*, Barbara Juliana Martins Lemos, Fabiola Alves Lino, Kiria Karolline Gomes Moreira, Edemilson Cardoso da Conceição, Victor Rezende Moreira Couto, and Juliano José de Resende Fernandes, *Universidade Federal de Goiás, Goiânia, Goiás, Brazil.*

The objective was to study the effect of the typical plant extracts of the Brazilian Cerrado on in vitro rumen microbial fermentation under a high concentrate diet. Two separate trials were conducted to evaluate 4 doses (0, 30, 300 and 3,000 mg/L) of trunk oil from *Copaifera langsdorffii* (Trial 1; 21.3% β-caryophyllene – essential oil) and fruit oil from *Pterodon emarginatus* (Trial 2; 5.3% β-caryophyllene – essential oil). The trials were analyzed as a randomized block design with 5 repetitions, treating incubation as the blocking factor. Treatments were evaluated using a TE-150 incubator ((Tecnal) in a batch culture of rumen fluid with a 10:90 forage:concentrate diet (15% crude protein, 20% neutral detergent fiber) incubated using filter bags of non-woven textile for 2, 4, 8, 12, 24, and 48 h (triplicates each time) to fit the disappearance of dry matter (DM), volatile fatty acids (VFA), pH, and ammonia nitrogen (NH<sub>3</sub>-N) were evaluated after 48 h of incubation. The inclusion of 3,000 mg/L of oil *C. langsdorffii* reduced (84.7 vs 63.8%) the potential

degradability (a + b) of DM and tended to decrease ( $P < 0.10$ ) the VFA (76.6 vs 60.6 mM), acetate (52.27 vs 42.11 mM) and NH<sub>3</sub>-N (119.12 vs. 101.47 mg/dL) concentration. Acetate:propionate ratio (C<sub>2</sub>:C<sub>3</sub>) was significantly reduced (3.55 vs 2.81) at a dose 300 mg/L of *C. langsdorffii*. The proportion of isovalerate was affected ( $P < 0.05$ ) at a dose 300 (1.93 vs 1.88 mM) and 3,000 mg/L (1.9 vs 1.6mM). No effect ( $P > 0.10$ ) of *P. emarginatus* on the kinetics of disappearance of rumen DM was detected. However, the addition of 30 and 300 mg/L of *P. emarginatus* oil increased ( $P < 0.05$ ) the total VFA concentration by an average of 28.3%. The 3,000 mg/L dose significantly decreased C<sub>2</sub>:C<sub>3</sub> ratio (3.77 vs 2.76 mM) due to a 27.9% reduction in the proportion of acetate. The results indicated that Cerrado plant oil are potential modifiers of rumen fermentation under a high concentrate diet.

**Key Words:** *Copaifera langsdorffii*, essential oils, *Pterodon emarginatus*

#### W404 Effects of the combined use of virginiamycin and salinomycin on ruminal microbial population in Nelore steers fed diets with two concentrate levels.

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Eight ruminally cannulated Nelore steers (434 ± 35 kg initial BW) were allotted to a 4x4 replicated Latin square design (21-d periods) to evaluate the effects of concentrate and virginiamycin (VM) levels in diets containing salinomycin (SL) on the ruminal population of *Streptococcus bovis*, *Megasphaera elsdenii*, *Selenomonas ruminantium*, and methanogenic archaea. Treatments were arranged in a 2 × 2 factorial, with 2 concentrate levels (70 and 90%) and 2 VM levels (0 and 15 ppm) in the diet DM. Animals were fed once daily at 0800 h and SL was included in all diets at 13 ppm (DM basis). Ruminal fluid samples were collected at 0 and 4 h post-feeding on d 18 of each period, and DNA was extracted from samples using the Qiagen DNA stool mini kit. Ruminal bacteria and methanogenic archaea populations were assessed by real-time PCR. Statistical analyses were performed using the MIXED procedure of SAS. No effects of time of collection nor treatment x time of collection interactions were observed for any variable. The relative populations of *S. bovis*, *M. elsdenii*, and *S. ruminantium* were greater ( $P < 0.01$ ) for steers fed the 90% concentrate diet than for animals fed less concentrate, but no effects of VM were observed for those bacteria populations (Table 1). There was a concentrate x VM interaction ( $P = 0.04$ ) for the relative populations of methanogenic archaea, but within each concentrate level, no differences between VM inclusions were observed. In conclusion, the ruminal microbial populations evaluated in this study were more affected by variations in concentrate levels than by VM inclusion in the diets.

**Table 1 (Abstr. W404).** Relative populations of ruminal bacteria and methanogenic archaea in Nelore steers fed diets containing 2 concentrate (C) levels and 2 virginiamycin (VM) levels (0 and 15 ppm)

Item	70% C		90% C		SEM	<i>P</i> -value		
	0 ppm	15 ppm	0 ppm	15 ppm		C	VM	C × VM
<i>S. bovis</i>	1.00	0.92	5.62	8.82	0.27	<0.01	0.61	0.46
<i>M. elsdenii</i>	1.00	0.72	5.70	6.02	0.15	<0.01	0.76	0.67
<i>S. ruminantium</i>	1.00	1.22	5.13	3.07	0.11	<0.01	0.48	0.11
Methanogenic	1.00	2.06	0.44	0.23	0.08	<0.01	0.91	0.04

**Key Words:** antibiotic, bacteria, ionophore

**Table 1 (Abstract W405).**

Item	Inclusion of glycerin, % of DM				SEM	P-value				
	0	7	14	21		Diet	Time	Diet × Time	Lin	Quad
pH	6.31	6.38	6.38	6.32	0.03	0.65	<0.01	<0.01	0.89	0.22
NH <sub>3</sub> , mg/dL	23.74	21.52	18.05	17.54	0.49	<0.01	<0.01	0.19	<0.01	0.39
Total VFA, mM	119.11	111.40	108.16	113.87	2.26	0.41	<0.01	0.85	0.37	0.16
Acetate, mM	74.37	58.82	55.37	53.12	1.42	<0.01	0.04	0.44	<0.01	0.05
Propionate, mM	24.76	25.11	27.06	33.68	0.74	<0.01	<0.01	0.06	<0.01	0.05
Butyrate, mM	14.27	21.16	20.21	19.46	0.44	<0.01	<0.01	<0.01	<0.01	<0.01
C2:C3 <sup>1</sup>	3.05	2.39	2.21	1.73	0.05	<0.01	<0.01	<0.01	<0.01	0.20

<sup>1</sup>Acetate:propionate ratio.

**W405 Ruminal fermentation of dairy cows supplemented with crude glycerin.** Pablo G. Paiva<sup>\*1</sup>, Carlos E. C. Consentini<sup>2</sup>, Fernanda C. R. Santos<sup>2</sup>, Arthur G. B. V. B. Costa<sup>2</sup>, Victor C. Galvao<sup>2</sup>, Guilherme F. Cabral<sup>3</sup>, Elmeson F. Jesus<sup>1</sup>, Tiago A. Del Valle<sup>2</sup>, Thiago H. Silva<sup>2</sup>, Caio S. Takiya<sup>2</sup>, and Francisco P. Renno<sup>2</sup>, <sup>1</sup>Animal Science Department, Universidade Estadual Paulista, UNESP/Jaboticabal-SP, <sup>2</sup>School of Veterinary Medicine and Animal Science, Universidade de São Paulo, USP, <sup>3</sup>School of Animal Science and Food Engineering, Universidade São Paulo, USP/Pirassununga-SP.

The objective of this study was to evaluate the effects of glycerin in replacing corn grain on ruminal fermentation of lactating dairy cows. Eight rumen cannulated Holstein cows (184 ± 50 DIM; 29.38 ± 0.89 kg/d milk yield; 594 ± 39 kg BW) were assigned in a replicated 4 × 4 Latin square design, with 21 d of experimental periods. Cows were assigned within each square to receive one following diets: Control or 0% glycerin; 7% of glycerin; 14% of glycerin; and 21% glycerin added to diets in replacing corn ground. On 20 d of experimental period, ruminal fluid samples were collected before (0), 2, 4, 6, 8, 10 e 12 h after morning feeding for analysis of the ruminal pH, NH<sub>3</sub> and volatile fatty acids (VFA). Data were analyzed as repeated measures in PROC MIXED (SAS for Windows 9.0), in the statistical model the effects of animal, period, square, treatment, beyond the effects of time with their interactions with treatment were considered. There was no effect of glycerin on total VFA ( $P > 0.05$ ); but glycerin inclusion in the diet increased propionate and butyrate, and decreased acetate concentrations, resulting in lower acetate: propionate ratio ( $P < 0.05$ ). Cows fed glycerin had lower ruminal pH in the first time of collect ( $P < 0.05$ ). Ruminal NH<sub>3</sub> was linearly decreased as glycerin levels increased ( $P < 0.05$ ). Glycerin inclusion in the diets of lactating dairy cows changed ruminal fermentation, with increase in propionate concentration, that indicate better efficiency the use of the energy diet.

**Key Words:** by-product, glycerol, performance.

**W406 Dry matter intake and nutrient digestibility of dairy cows supplemented with crude glycerin.** Pablo G. Paiva<sup>\*1</sup>, Fernanda C. R. Santos<sup>2</sup>, Arthur G. B. V. B. Costa<sup>2</sup>, Victor C. Galvao<sup>2</sup>, Guilherme F. Cabral<sup>3</sup>, Carlos E. C. Consentini<sup>2</sup>, Elmeson F. Jesus<sup>1</sup>, Tiago A. Del Valle<sup>2</sup>, Thiago H. Silva<sup>2</sup>, Caio S. Takiya<sup>2</sup>, and Francisco P. Renno<sup>2</sup>, <sup>1</sup>Animal Science Department, Universidade Estadual Paulista, UNESP/Jaboticabal-SP, <sup>2</sup>School of Veterinary Medicine and Animal Science, Universidade de São Paulo, USP, <sup>3</sup>School of Animal Science and Food Engineering, Universidade São Paulo, USP/Pirassununga-SP.

The aim of this study was to evaluate the effects of glycerin in replacing corn grain on dry matter intake (DMI) and nutrient digestibility of lactating dairy cows. Twenty-four Holstein cows (184 ± 50 DIM;

29.38 ± 0.89 kg/d milk yield; 594 ± 39 kg BW) were assigned in 4 × 4 Latin square design, with 21 d of experimental periods. Cows were assigned within each square to receive one following diets: Control or 0% glycerin; 7% of glycerin; 14% of glycerin; and 21% glycerin added to diets on replacing corn ground. On d 16 to 18 of each period, fecal samples were taken from each cow, twice daily after milking, comprising a composite sample. The indigestible acid detergent fiber was used as internal marker to estimate fecal excretion and total-tract apparent digestibility of the nutrients. Data were analyzed with PROC MIXED (Statistical Analysis System for Windows 9.0), diet, period, and square were considered as fixed effect, animal within square and residual error as random effect. The inclusion of glycerin in the diets decreased DMI ( $P < 0.05$ ). However, total-tract digestibility of dry matter, crude protein, and ether extract was linearly increased, and neutral detergent fiber (NDF) linearly decreased with glycerin inclusion in the diet ( $P < 0.05$ ). High glycerin levels in corn silage based diets affected NDF digestibility and decreased DMI of lactating dairy cows.

**Table 1 (Abstr. W406).**

Item	Inclusion of glycerin, % of DM				SEM	P-value	
	0	7	14	21		Lin	Quad
DMI, kg/d	21.97	21.97	21.42	21.06	0.32	0.04	0.73
DMI, % of BW	3.54	3.63	3.49	3.37	0.05	0.01	0.07
Digestibility, %							
DM	69.63	70.39	71.72	72.40	0.30	<0.01	0.85
CP	73.38	73.60	75.57	77.24	0.37	<0.01	0.16
EE	72.93	76.15	84.17	86.93	0.82	<0.01	0.82
NDF	59.99	58.00	57.13	54.30	0.50	<0.01	0.71

**Key Words:** biodiesel, by-product, energy

**W407 Nitrogen utilization of Nelore bulls supplemented with energy sources associated crude glycerin on finishing phase.** Antônio José Neto<sup>\*1</sup>, Joanis Tilemahos Zervoudakis<sup>2</sup>, Pedro Veiga Rodrigues Paulino<sup>2</sup>, Luciana Keiko Hatamoto-Zervoudakis<sup>2</sup>, Luciano da Silva Cabral<sup>2</sup>, Juliane Quenoizoré Soares<sup>2</sup>, Rosemary Lais Galati<sup>2</sup>, Renata Pereira da Silva-Marques<sup>2</sup>, and Lilian Chambó Rondena Pesqueira Silva<sup>2</sup>, <sup>1</sup>Department of Animal Science; São Paulo State University-UNESP, Jaboticabal, SP, Brazil, <sup>2</sup>Department of Basic Sciences and Animal Production; Federal University of Mato Grosso-UFMT, Cuiabá, MT, Brazil.

Glycerin has the potential to partially replace starch-based ingredients in the diet, such as corn, because glycerol is converted to propionate in the rumen and acts as a precursor for hepatic glucose synthesis. The objective of this study was to evaluate the effects of crude glycerin (Gly) combined with protein sources on balance of nitrogen compounds of



Nellore bulls fed tropical pasture during the rainy season. Five Nellore bulls (448.2 ± 14 kg; 25 ± 3 mo) were evaluated in a 5 × 5 Latin square design with 5 treatments and 5 periods. The animals were allocated into 5 paddocks of 0.25 ha, consisting of *Brachiaria brizantha* 'Marandu' in the rainy season. Crude glycerin (83.9% of glycerol) was used in all supplements to replace (15% of DM) corn. The supplements were: T1 - Gly with urea, T2 - Gly with soybean meal, T3 - Gly with cottonseed meal, and T4 - Gly with gluten, and the control supplement received mineral mixture *ad libitum*. Animals were individually supplemented at the rate of 300 g · 100 kg<sup>-1</sup> of BW, daily, at 1000 h. The experimental period was the 17 d, divided in adaptation (14 d) and 3 d for collections the feces in different times. Urine samples were collected 4 h after the supplementation to determine the concentration of nitrogen, on the last day of period. Data were analyzed using the PROC MIXED of the SAS with  $\alpha = 0.05$  and F-test. Nitrogen intake (g · d<sup>-1</sup>) was greater ( $P < 0.01$ ) for Gly-urea compared with control and protein sources. There were differences in total grams of N excreted across treatments ( $P < 0.01$ ). The increase in urinary and fecal N output by animals fed gly-urea contributed to greater total N output compared with other supplements. Nitrogen retained (g · d<sup>-1</sup> or % of N intake) was not affected by protein sources, but there were greater than control supplement ( $P < 0.01$ ). Independently of protein source utilized, there was an increase of 59.5% ( $P < 0.01$ ) in the N utilization (N retained, g · d<sup>-1</sup>) for animals supplemented when compared with control group. Crude glycerin associated with protein sources can be efficient for improve the N utilization.

**Key Words:** beef cattle, glycerol, nitrogen

**W408 In vitro production of gas in diets containing different levels of ethanolic extract of propolis.** Sandra M. Yamamoto\*, Cintia M. Araujo, Fernanda G. da Silva, Marcela A. Formiga, Layse A. Gordiano, and Daniel R. Menezes, *Federal University of Vale do São Francisco, Petrolina, Pernambuco, Brazil.*

In this study, we evaluated propolis as a safe alternative for ruminants feeding free of antibiotic additives and growth promoters. Phenolic compounds with antibiotic activity mainly against gram-positive bacteria, in the same manner as ionophores, are present in propolis. We evaluated the effects of different levels of ethanolic extract of propolis (EEP) at 30% on the in vitro production of gases by bacteria. Feed was formulated to contain 50% ground elephant grass (*Pennisetum purpureum*) and 50% concentrate, consisting of corn, soybean meal and core minerals and vitamins. EEP was added to the concentrate at 0, 6, 12, 24, and 36 mL/kg. A sample of 1000mg was weighed in nylon bags, with a weight of 20 mg/cm<sup>2</sup>, sealed and added to the fermentation vials (160mL) previously injected with CO<sub>2</sub>, with 4 replicates per treatment. To each vial was added 90mL of buffer solution and 10 mL of ruminal fluid spray kept under CO<sub>2</sub>. The vials were sealed, placed in a room at 39°C and stirred every 2 h. Gas pressure readings were made at 2, 4, 6, 8, 9, 11, 12, 14, 17, 20, 24, 28, 34, 48, 72, 96, and 120 h after incubation. Statistical analyzes were performed using SAS 9.1 (SAS Institute Inc., Cary, NC). Data were previously checked for normality of residuals by Shapiro-Wilk test (PROC UNIVARIATE) and the variances compared by orthogonal contrasts (PROC GLM) with significance level of 5%. When significant, the regression equations were determined (PROC MIXED). As EEP levels were not equidistant, it was generated vectors of each contrast using PROC IML. There were no effects ( $P > 0.05$ ) of EEP levels to the fibrous (0.0715 ± 0.02 mL/hour), non-fibrous (0.013 ± 0.01 mL/hour), and total (0.083 ± 0.02 mL/hour) carbohydrate fermentation rates. For the potential production of total carbohydrate gases (Vt) there was a quadratic effect ( $P < 0.05$ ). The lowest value of gas output (77.69 mL/g of DM) was obtained for the inclusion of 23

mL of EEP/kg of concentrate. For the gas production from non-fibrous carbohydrates, the lowest value (63.12 mL/g of DM) was obtained for the inclusion of 24.78 mL of EEP/kg of concentrate. The results suggests that EEP was efficient at inhibiting total gas production by ruminal microorganism in vitro.

**Key Words:** fermentation, growth promoters, propolis

**W409 Influences of SmartCare in milk replacer and XPC in calf starter on the performance and health of pre-weaning Holstein calves challenged orally with an opportunistic infection with *Citrobacter freundii*.** Tyler L. Harris\*<sup>1</sup>, Yu Liang<sup>1</sup>, Matt D. Sellers<sup>1</sup>, Cameron R. Nightingale<sup>1</sup>, Kate P. Sharon<sup>1,2</sup>, Jeff A. Carroll<sup>2</sup>, Ilkyu Yoon<sup>3</sup>, Mark F. Scott<sup>3</sup>, and Michael A. Ballou<sup>1</sup>, <sup>1</sup>Department of Animal and Food Sciences, Texas Tech University, Lubbock, TX, <sup>2</sup>USDA-ARS, Lubbock, TX, <sup>3</sup>Diamond V, Cedar Rapids, IA.

This study investigated how supplementing milk replacer with SmartCare (SC) and calf starter with original XPC (XPC) would affect performance and health following an oral challenge with *Citrobacter freundii*. The study was performed over a 54 d period with 30 Holstein bull calves. Calves were randomly assigned to treatments that included: CON: milk replacer and calf starter with no added products, SC: milk replacer with 1 g SC/calf/d and base calf starter, and SC+XPC: milk replacer with 1 g SC/calf/d and calf starter with 0.5% of DM as XPC. Calves were fed 350 g of milk replacer at 0700 and 1700 from d 0 to 42. Calf starter and water were offered *ad libitum* and recorded daily. Calves were challenged with 10<sup>8</sup> cfu of *Citrobacter freundii* orally on d 16 of the study. Blood samples were collected on d 0, 7, 14, 16, 18, 21, 24, 28, and 35 and analyzed for hematology. All data were reported as CON, SC, and SC+XPC, respectively. LSMeans with various superscripts differ ( $P \leq 0.05$ ). Calf starter intake and ADG did not differ overall ( $P \geq 0.127$ ); however, the SC+XPC calves consumed more calf starter from d 36 to 54 (0.865<sup>a</sup>, 0.840<sup>a</sup>, 0.981<sup>b</sup> ± 0.053 kg/d;  $P = 0.054$ ) and had increased ADG from d 15 to 21 (0.677<sup>a</sup>, 0.696<sup>a</sup>, 0.840<sup>b</sup> ± 0.046 kg/d;  $P = 0.006$ ). There was treatment x time interaction in neutrophil to lymphocyte ratio (N:L;  $P = 0.043$ ), whereas CON calves had greater ( $P \leq 0.019$ ) N:L than SC and SC+XPC on d 7 (1.21<sup>a</sup>, 0.83<sup>b</sup>, 0.72<sup>b</sup> ± 0.15) and d 16 (1.43<sup>a</sup>, 0.93<sup>b</sup>, 0.99<sup>b</sup> ± 0.15). Additionally, the SC+XPC calves had lower hemoglobin throughout the study (10.9<sup>a</sup>, 11.2<sup>a</sup>, 10.3<sup>b</sup> ± 0.19 mg/dL;  $P = 0.006$ ). Average fecal scores by period were greatest among CON calves after the challenge on d 17 to 21 (2.7<sup>a</sup>, 2.1<sup>b</sup>, 1.9<sup>b</sup> ± 0.003;  $P \leq 0.010$ ) and CON was also greater than SC+XPC calves on d 22 to 28 (2.8<sup>a</sup>, 2.4<sup>ab</sup>, 2.1<sup>b</sup> ± 0.15;  $P \leq 0.002$ ). The *Citrobacter* challenge increased scours, but did not result in systemic disease. Feeding SC in milk replacer and XPC in calf starter improved calf performance, health, and reduced the risk of scours during neonatal life.

**Key Words:** calf, health, yeast culture

**W410 Effect of adding *Enterococcus faecalis* on in vitro ruminal fermentation profiles.** Lovelia L. Mamud, Ashraf A. Biswas, and Sang Suk Lee\*, *Sunchon National University, Suncheon, Jeonnam, South Korea.*

*Enterococcus faecalis* is one of the beneficial microorganisms which produces enterolysin and converts fumarate to succinate. Hence, this study was conducted to determine the effect of adding *E. faecalis* on in vitro ruminal fermentation, methane concentration, microbial diversity and population. Ruminal samples were collected from ruminally cannulated Holstein Friesian cattle, and a substrate consisting of rice straw and concentrate mixture at 40:60 DM was offered at 1g DM/100mL

buffered ruminal fluid. Fresh culture of *E. faecalis* ( $7.5 \times 10^8$  cfu/mL) at different inclusion rates were investigated using in vitro ruminal fermentation. The treatments tested were: non addition (control), 0.1% (T1), 0.5% (T2), and 1.0% (T3) of *E. faecalis*. All treatments were conducted in triplicates and analyzed by ANOVA for randomized complete block design. Duncan's Multiple Range Test (DMRT) and Orthogonal Polynomial Contrast were used to identify differences among treatments and control. All analyses were carried out using SAS version 9.1 (SAS, 2002). Total gas production and ammonia nitrogen concentration of cultures were directly proportional to incubation times, while culture pH was inversely proportional to incubation times. Addition of *E. faecalis* lowered ( $P < 0.05$ ) the total gas production after 24 h but the opposite was observed after 48 h of incubation having the highest ( $P < 0.05$ ) in T3 followed by T4, T2 and control with 93.80, 91.00, 88.33 and 82.20 ml, respectively. Higher concentrations of acetate were detected in *E. faecalis* treatments than control at 12 and 48 h but the opposite was observed at 24 h, while propionate was highest ( $P < 0.05$ ) in T3 after 48 h with 12.59 mM. Butyrate concentration was higher ( $P < 0.05$ ) in *E. faecalis* treatments than control after 12 and 24 h of incubations and the highest ( $P < 0.05$ ) concentrations were detected in T2 at 24 h and T3 at 48 h with 14.61 and 14.88 mM, respectively. Higher ( $P < 0.05$ ) concentrations in *E. faecalis* treatments than control were also observed in total volatile fatty acid (VFA) concentration after 12 and 48 h. Addition of *E. faecalis* enhanced in vitro ruminal fermentation by increasing the butyrate and total VFA concentrations in dose dependent manner.

**Key Words:** *Enterococcus faecalis*, in vitro, pyrosequencing

**W411 Acetohydroxamic acid did not influenced ruminal microbiota but altered urea metabolism.** P. P. Wang<sup>1</sup>, J. Q. Wang<sup>1</sup>, D. P. Bu<sup>\*1,2</sup>, D. Jin<sup>1</sup>, J. Zhang<sup>1</sup>, and X. M. Nan<sup>1,3</sup>, <sup>1</sup>State Key Laboratory of Animal Nutrition, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>2</sup>World Agroforestry Centre, East and Central Asia, Beijing, China, <sup>3</sup>Synergetic Innovation Center Of Food Safety and Nutrition, Harbin, China.

The objective of this experiment was to investigate the effects of urea and acetohydroxamic acid (AHA) on in vitro ruminal urea-nitrogen (urea-N) metabolism and microbial community using total mixed ration as a substrate. Treatments were arranged in a  $2 \times 2$  factorial design with urea supplemented at 0 or 2% dry matter (DM), and AHA equivalent to 0 or 450 mg/kg DM. Ruminal fluids were collected from 3 Chinese Holstein dairy cows through permanent ruminal fistula, diluted with artificial saliva (1:2, v/v), and incubated anaerobically at 39°C for 0, 1, 2, 4, 6, and 12 h. Each treatment was performed in 3 serum bottles and experiment was run 3 times. Supplementation of urea increased ( $P < 0.01$ ) ruminal pH, ammonia-nitrogen (NH<sub>3</sub>-N) concentration and urease activity, while addition of AHA inhibited ( $P < 0.01$ ) their increments. Acetohydroxamic acid was still stable within 6 h of incubation. When AHA was added, urea-N concentration of fermentation fluid in the treatment with urea supplied was gradual decline ( $P < 0.05$ ). The peak of NH<sub>3</sub>-N concentration was not delayed by AHA addition, comparing with treatment with urea supplementation only. The bacterial PCR-DGGE profiles of 4 treatments were similar to each other before and after incubation. Urea stimulated ( $P < 0.01$ ) the decrements of *Ruminococcus albus*, *R. flavefaciens*, *Fibrobacter succinogene*, and *Butyrivibrio fibrisolvens* populations, but had no effect on *Prevotella* population ( $P = 0.18$ ). However, all those functional bacterial populations were not influenced by AHA addition. It was concluded that AHA could slow down the degradation of urea by the inhibition of urease activity, and the dose of 450 mg/kg DM could not alter ammonia formation pattern in

this fermentation condition. In addition, AHA had no effect on ruminal microbiota, which could be altered by urea supplementation.

**Key Words:** acetohydroxamic acid, in vitro fermentation, urea metabolism

**W412 Effect of interaction of cereal and forage on rumen fermentation characteristics in the diet of cattle.** S. Y. Cao<sup>1</sup>, C. W. Yang<sup>2</sup>, Z. B. Yang<sup>\*1</sup>, and Y. Wang<sup>3</sup>, <sup>1</sup>College of Animal Science, Shandong Agricultural University, Tai-an, Shandong, China, <sup>2</sup>College of Life Science, Shandong Agricultural University, Tai-an, Shandong, China, <sup>3</sup>Agriculture and Agri-Food Canada, Lethbridge Research Centre, Lethbridge, AB, Canada.

Four ruminally cannulated Limousin  $\times$  Luxi beef cattle were used in a  $4 \times 4$  Latin square design to monitor the changes in rumen fermentation during the feeding cycle and compare the differences caused by the type of cereal(C) and forage(F) in the diet. The 4 experimental diets contained 60% of either *Leymus chinensis* (LC) or sweet potato vine (SP) as forage and 40% of concentrate with either wheat (W) or maize (M). Cattle were fed twice daily and samples from rumen fluid were taken before morning feeding on the first 10 d after changing the diet and then at 0, 3, 6, 9 and 12 h after the morning feeding for 3 d. Significant variability in ruminal pH with the WSP diet was more pronounced on which pH dropped from 6.81 on d 1 (before the dietary changes) to 6.57 by d 10 ( $P = 0.03$ ). The NH<sub>3</sub>-N and total VFA concentrations of the cattle fed the *Leymus chinensis* were decreased ( $P < 0.05$ ) after diet changes and then increased to values similar to that before changing. Whereas the total VFA concentrations of the cattle fed the sweet potato vine have the opposite trend. Mean rumen pH, acetate and propionate proportion and the ratio of acetate to propionate were not affected by the treatments ( $P > 0.05$ ). The cattle fed WSP diet had greater ( $P < 0.05$ ) NH<sub>3</sub>-N and total VFA concentrations and greater ( $P < 0.05$ ) proportion of butyrate. The average pH values and the ratio of acetate to propionate were affected by cereal ( $P = 0.004$ ,  $P = 0.046$  respectively) and the concentrations of total VFA, the proportion of propionate and butyrate were affected by forage ( $P = 0.006$ ,  $P = 0.045$ ,  $P < 0.001$ , respectively). No cereal  $\times$  forage interactions were detected among all rumen fermentation characteristics ( $P > 0.05$ ). The rumen pH values and concentrations of NH<sub>3</sub>-N and total VFA were fluctuated at certain sampling days after the abrupt change of diets, however, at the end of the adaptation period the rumen fermentation characteristics were similar to the values before diets changes. The result also indicated that the type of cereal and forage had apparent effects on rumen fermentation characteristics, but no interactions were observed.

**Key Words:** cereal, forage, rumen fermentation

**W413 Effect of dietary energy source and level on nutrient digestibility, rumen microbial protein synthesis, and milk performance in lactating dairy cows.** X. Q. Zhou<sup>1,2</sup>, J. Q. Wang<sup>1</sup>, D. P. Bu<sup>\*1,3</sup>, Y. D. Zhang<sup>1</sup>, M. Zhao<sup>1</sup>, P. Sun<sup>1,4</sup>, and T. Zhang<sup>1</sup>, <sup>1</sup>State Key Laboratory of Animal Nutrition, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>2</sup>College of Animal Science and Technology, Northeast Agricultural University, Harbin, China, <sup>3</sup>Synergetic Innovation Center of Food Safety and Nutrition, Harbin, China, <sup>4</sup>CAAS-ICRAF Joint Lab on Agroforestry and Sustainable Animal Husbandry, Beijing, China.

This study examined the effects of dietary energy source and level on feed intake, digestion, rumen microbial protein synthesis and milk performance of lactating dairy cows fed corn stover as the main forage

source. Eight primiparous Chinese Holstein cows (138 ± 19.4 DIM, 29 ± 0.8 kg/d milk, and 589 ± 57.6 kg of BW), 4 of which were fitted with ruminal cannula, were used in a replicated 4 × 4 Latin square design with a 2 × 2 factorial treatment arrangement (energy level: NE<sub>L</sub> 1.52–1.53 Mcal for LE and 1.71–1.72 Mcal for HE; energy source: SFC and GC). The NE<sub>L</sub> was calculated based on Chinese recommendation (2004). Changes to dietary energy level were induced by feeding the cows diets of either 35.6% corn stover plus 1.24% EB100 or 13% corn stover, 22% corn silage plus 1.84% EB100 (a saturated free fatty supplement). Milk yield [27.8 vs. 24.6 kg/d (0.45 SEM), *P* = 0.007], milk protein content and yield [0.86 vs. 0.76 kg/d (0.01 SEM), *P* = 0.006], and milk lactose yield [1.34 vs. 1.20 kg/d, (0.02 SEM) *P* = 0.008] increased in response to increased levels of dietary energy, while contents of milk fat [4.22 vs. 4.24% (0.06 SEM), *P* = 0.433] and milk lactose [4.94 vs. 4.90% (0.08 SEM), *P* = 0.323] were not affected. Cows fed HE diets had a higher microbial protein yield [1.57 vs. 1.34 kg/d (0.54 SEM), *P* = 0.003] than those fed LE diets. Apparent digestibilities of nutrients were higher in HE than in LE diets. Milk yield [25.8 vs. 23.3 kg/d (0.434 SEM), *P* = 0.047], milk protein yield [0.79 vs. 0.72 kg/d (0.02 SEM), *P* = 0.043] and microbial protein yield [1.42 vs. 1.25 kg/d (0.33 SEM), *P* = 0.038] were also higher when SFC was used to replace GC as the main energy source for lactating cows fed with LE diets. This study suggest that increased dietary energy level and ruminal degradation rate are beneficial to milk protein production when cows are fed corn stover as a forage source, and this can be attributed to the resulting increased supply of microbial protein.

**Key Words:** energy, corn stover, microbial protein

**W414 Physico-molecular structures and metabolic characteristics of protein in brown and yellow flaxseed with altered nutrient traits for dairy cattle.** Nazir A. Khan<sup>1</sup>, Helen Booker<sup>2</sup>, Yajing Ban<sup>1</sup>, and Peiqiang Yu<sup>\*1</sup>, <sup>1</sup>Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, SK, Canada, <sup>2</sup>Department of Plant Sciences, University of Saskatchewan, Saskatoon, SK, Canada.

Flaxseed (*Linum usitatissimum* L.), also known as linseed, is one of the most important commercial oilseed crop of Western Canada, providing valuable inputs for food, feed, manufacturing and pharmaceutical industries. The objectives of this study were to investigate chemical profiles; protein subfractions; ruminal CP degradation and intestinal digestibility of RUP of flaxseed in dairy cattle; and protein molecular structures of newly developed yellow-seeded flax. Four flaxseed genotypes with 2 yellow breeding lines and 2 brown varieties obtained from Crop Development Center were evaluated. Each genotype were sampled from 2 replicate plots at 2 different locations. The flaxseed samples were analyzed for protein molecular profiles in terms of amide I and II,  $\alpha$ -helix and  $\beta$  sheet structures using a JASCO FT/IR-ATR. The rumen and intestinal digestion of flaxseed were determined using in situ and 3-step in vitro methods with 4 lactating Holstein dairy cows (body weight, 680 ± 10 kg). The cows were fed TMR with forage to concentrate ratio 50 to 50. The data were analyzed as a RCBD with in situ cows as a random effect using the PROC MIXED procedure of SAS. The results showed that the yellow-seeded lines had higher (*P* < 0.001) contents of oil (44.5 vs. 41.4% DM, SEM = 0.35) and CP (24.9 vs. 20.9% DM, SEM = 0.96) compared with the brown-seeded varieties. The CP in yellow-seeds contained lower (*P* < 0.01) contents of true protein subfraction (81.3 vs. 92.7% CP, SEM = 1.46), and more (*P* < 0.001) extensively degraded (70.8 vs. 64.9% CP, SEM = 0.46) in rumen resulting in lower (*P* < 0.001) content of RUP (29.2 vs. 35.1% CP, SEM = 0.71) than the brown-seeded varieties. However, the total supply of

digestible RUP was not significantly different between the 2 seed types. Regression equations based on protein molecular structural features gave relatively good estimation for the contents of CP (*R*<sup>2</sup> = 0.87), soluble CP (*R*<sup>2</sup> = 0.92), RUP (*R*<sup>2</sup> = 0.97) and intestinal digestibility of RUP in dairy cattle (*R*<sup>2</sup> = 0.71). In conclusion, molecular spectroscopy can be used to rapidly characterize flaxseed protein molecular structures and predict their nutritive value in dairy cattle.

**Key Words:** protein molecular structure, protein subfraction, flaxseed

**W415 Effect of rubber seed oil and flaxseed oil on milk performance, fatty acid composition and oxidative stability of milk fat.** Y. Pi<sup>1</sup>, J. Q. Wang<sup>1</sup>, D. P. Bu<sup>\*1,3</sup>, L. Ma<sup>1,2</sup>, Y. X. Zhu<sup>4</sup>, and J. C. Xu<sup>4</sup>, <sup>1</sup>Institute of Animal Science, State Key Laboratory of Animal Nutrition, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>2</sup>CAAS-ICRAF Joint Lab on Agroforestry and Sustainable Animal Husbandry, World Agroforestry Centre, East and Central Asia, Beijing, China, <sup>3</sup>Synergetic Innovation Center of Food Safety and Nutrition, Harbin, China, <sup>4</sup>Kunming Institute of Botany, Chinese Academy of Sciences, Kunming, Kunming, China.

The experiment was conducted to investigate the effect of rubber seed oil compared with flaxseed oil when fed alone or in combination on the milk yield, milk composition, fatty acid composition and oxidative stability of milk fat from dairy cows. Forty-eight mid-lactation Holstein dairy cows were randomly assigned to one of 4 treatments according to a completely randomized block design. Cows were fed a basal diet (Control) or the basal diet supplemented with 4% rubber seed oil (RO), 4% flaxseed oil (FO), or 2% rubber seed oil plus 2% flaxseed oil (RFO) on a dry matter (DM) basis for 9 weeks. Feed intake (DMI), milk protein percentage, and milk fat levels did not differ among the treatments. Cows fed the RO, FO or RFO treatments had a higher (*P* < 0.001) milk yield than the control group (up to 6.5% more), while milk fat percentages were decreased. Compared with the Control, milk concentration of  $\alpha$ -linolenic acid (ALA) content was substantially higher in cows receiving RO or RFO, and was double in cows receiving FO (*P* < 0.001). Both C<sub>18:1</sub> *trans*-11 (VA) and C<sub>18:2</sub> *cis*-9, *trans*-11 (CLA) levels were higher (*P* < 0.001) in cows fed added flaxseed or rubber seed oil. The increasing in VA, ALA, and CLA was greater in cows fed RFO than in cows fed RO alone. Compared with the Control, the milk fat from cows fed any of the dietary supplements had a higher concentration of unsaturated fatty acids (UFA), monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA), while the average saturated fatty acids (SFA) levels in milk fat were 30% lower. Compared with the Control, the superoxide dismutase (SOD) and glutathione peroxidase (GSH-Px) activities were all decreased (*P* > 0.05) in RO, FO and RFO treatments, while the catalase (CAT) activity was decreased (*P* < 0.05) in FO and RFO groups. The concentration of malondialdehyde (MDA) tended to increase (*P* < 0.10) in RO, FO and RFO treatments. These results indicated that the supplement of rubber seed oil and flaxseed oil in dairy diet could increase milk yield and the functional fatty acids content (ALA, VA, and CLA) in milk fat, while decrease the content of saturated fatty acids and oxidative stability of milk fat.

**Key Words:** milk fat, oxidative stability, rubber seed oil

**W416 Varying the degrees of synchrony of energy and nitrogen release in rumen affect the synthesis of microbial protein in continuous culture system.** J. Zhang<sup>1</sup>, J. Q. Wang<sup>1</sup>, D. P. Bu<sup>\*1,2</sup>, S. G. Zhao<sup>1</sup>, P. P. Wang<sup>1</sup>, and X. M. Nan<sup>1,3</sup>, <sup>1</sup>State Key Laboratory of Animal Nutrition, Institute of Animal Sciences, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>2</sup>World Agroforestry Centre,



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A continuous rumen simulation system apparatus with 16 1000 mL fermentation vessels was used to investigate the effects of different degrees of synchrony between ruminal available energy and nitrogen on rumen fermentation and microbial protein synthesis (MPS). The basal substrate was diet of (g DM/day) 18.50 corn straw, 10.00 corn, 0.26 premix, and 11.24 soybean meal containing 67.5 g N/kg DM, and diets were divided into 2 equal feedings at 09:00 and 21:00 h. The treatments were: the basal diet with (1) 10.0 g maltodextrin which was in the McDougall's buffer infused continuously into the fermenter (CONT), (2) 10.0 g maltodextrin as 2 6-h infusions starting at 09:00 and 21:00h (FAST) and (3) 10.0 g maltodextrin given as 2 6-h infusions starting at 15:00 and 03:00h (SLOW). Rumen inoculum was obtained from 4 ruminal fistulated cows in early lactation that were fed TMR with 45:55 forage to concentrate ratio. A 8-d incubation period was used, with the first 5 d serving as an adaptation period followed by 3 d of sampling with solid and liquid dilution rates in the fermenters set at approximately 10.0 and 8.0%/h, respectively. Data were analyzed using the MIXED procedure of SAS. All 3 infusion treatments had no significant effect on the apparent digestibility of OM (65.0% for CONT, 62.2% for FAST, and 61.2% for SLOW), DM (66.1%, 63.4% and 63.6%), and CP (71.5%, 68.2% and 68.6%). The pH values among 3 treatments were similar (6.70, 6.68 and 6.71). The concentrations of total volatile fatty acid for CONT and FAST treatments have higher tendency ( $P = 0.05$ ) than SLOW (79.51 and 77.58 vs. 72.31, mmol/L). However, the CONT and FAST treatments increased ( $P < 0.05$ ) MPS compared with SLOW (4.0 and 3.8 vs. 2.2, g/day). Compared with SLOW, the lower ( $P < 0.01$ ) concentration of ammonia in CONT and FAST treatments (7.90 and 8.44 vs. 16.29, mg/dL) may indicate better utilization of ammonia for the growth of ruminal microbe. It is concluded that altering the degree of synchrony in the rates of ruminal release of energy and nitrogen had a marked effect on MPS.

**Key Words:** synchrony, energy, nitrogen

**W417 Effects of wilting and additives on fermentation quality of alfalfa (*Medicago sativa* L.) silage.** H. Liu<sup>1,2</sup>, J. Q. Wang<sup>2</sup>, D. P. Bu<sup>\*2,4</sup>, Z. W. Lv<sup>2</sup>, and P. Sun<sup>2,3</sup>, <sup>1</sup>College of Animal Science and Veterinary Medicine, Heilongjiang Bayi Agricultural University, Daqing, Heilongjiang, China, <sup>2</sup>State Key Laboratory of Animal Nutrition, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>3</sup>Synergetic Innovation Center Of Food Safety and Nutrition, Harbin, China, <sup>4</sup>CAAS-ICRAF Joint Lab on Agroforestry and Sustainable Animal Husbandry, Beijing, China.

The effect of wilting and different additives on alfalfa silage fermentation and nutritive value were studied under laboratory conditions. Lucerne (at the early bloom stage of maturity), wilted for 3 and 12 h respectively, was ensiled at 27.4% and 39.2% dry matter (DM) contents. Chopped grasses at each moisture content were either untreated (control) or treated with (1)inoculant (LAB) containing *Lactobacillus plantarum* LP70, *Lactobacillus casei* LC05, and *Enterococcus faecium* EF08 (a final application rate of  $2 \times 10^5$  cfu/g of fresh forage); (2)sugar beet pulp (SB) at 50g kg<sup>-1</sup> of fresh forage weight; (3)sodium formate (SF) at 6g kg<sup>-1</sup> of fresh forage weight; (4)a combination of inoculant and sugar beet pulp (LAB+SB), and (5)sodium formate and sugar beet pulp combined (SF+SB). Forages were packed into triplicate 1-L laboratory silos per treatment at a density of 650 g/L. Silos were opened and analyzed after 65 d of ensiling. The results showed that all treatments had lower pH and lower concentrations of acetic and ammonia-N (of % TN), but higher concentrations of lactic acid than did untreated silages ( $P < 0.001$ ).

These effects were stronger in the LAB+M-treated silage. Addition of LAB+M resulted in silage with the highest concentrations of lactic acid ( $5.12 \pm 0.36$ ;  $P = 0.011$ ) and the lowest ammonia-N ( $1.87 \pm 0.07$ ;  $P < 0.001$ ) over treatments. Treated silages had similar concentrations of neutral detergent fiber (NDF), acid detergent fiber (ADF) and dry matter recovery compared with untreated silage. The addition of sugar beet pulp (SB) greatly reduced crude protein (CP) concentrations ( $20.53 \pm 0.12$  vs.  $21.57 \pm 0.12$ ;  $P < 0.001$ ) compared with control. Water-soluble carbohydrate concentrations were found increased in silage from alfalfa herbage treated with LAB+SB ( $2.09 \pm 0.04$  vs.  $1.80 \pm 0.04$ ;  $P < 0.001$ ) and SF-treated silage ( $2.04 \pm 0.04$  vs.  $1.80 \pm 0.04$ ;  $P < 0.001$ ) compared with untreated silage. Wilting reduced the rate of decline of silage pH ( $P < 0.001$ ) and produced silages with lower acetic acid ( $P < 0.001$ ) and ammonia-N contents ( $P < 0.001$ ). As silage wilting was increased, silage quality was improved. Inoculant and sugar beet pulp combined (LAB+SB) was found to be the most effective additive.

**Key Words:** wilting, additive, alfalfa silage

**W418 Effect of choline inclusion on lamb performance and meat characteristics.** Julio Godinez-Cruz<sup>1</sup>, Oswaldo Cifuentes-Lopez<sup>1</sup>, Jorge Cayetano<sup>1</sup>, Hector Lee-Rangel<sup>\*1</sup>, German Mendoza<sup>2</sup>, Anayeli Vazquez<sup>1</sup>, and Alejandro Roque<sup>1</sup>, <sup>1</sup>Universidad Autónoma de San Luis Potosí, Facultad de Agronomía y Veterinaria, San Luis Potosí, San Luis Potosí, Mexico, <sup>2</sup>Universidad Autónoma Metropolitana, Departamento de Producción Animal, Distrito Federal, Mexico.

The objective of this experiment was to determine the effect of rumen-protected choline (RPC Reashure; Excential RumenPass CH) and a vegetal Bio choline (Indian Herbs) on lamb performance in finishing rations. The experimental units were 24 Rambouillet lambs ( $23.4 \pm 1.1$  kg initial BW) housed in individual metabolic cages. The experiment lasted 42 d and treatments were: no choline (T1, control); 4 g/d biocholine (T2); 4 g/d RPC (T3). Lambs were assigned to one of the 3 experimental treatments according to a completely randomized design and data were analyzed using PROC MIXED with initial BW as a covariable. There were no differences ( $P = 0.05$ ) on average daily gain and feed conversion; however, final BW (T1 = 32.7a, T2 = 33.9a, and T3 = 35.6b kg) and daily feed intake (T1 = 1.06a, T2 = 1.07a, and T3 = 1.22b kg) were different ( $P < 0.05$ ). In carcass characteristics (cold carcass weight, hot carcass weight and rib area), only for meat color the L value were different (41.18a, 42.68b, and 42.58b to T1, T2, and T3, respectively). Therefore, addition of a choline did not change performance of finishing lambs, but could change some meat characteristics and final weight.

**Key Words:** lamb, choline, carcass

**W419 Effects of *Yucca schidigera* extract on Rumen Fermentation Parameters in vitro.** J. L. Niu<sup>1</sup>, L. Ma<sup>1,2</sup>, D. P. Bu<sup>\*1,3</sup>, J. N. Li<sup>1</sup>, L. Pan<sup>1</sup>, and J. C. Xu<sup>4</sup>, <sup>1</sup>State Key Laboratory of Animal Nutrition, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>2</sup>CAAS-ICRAF Joint Laboratory of Agroforestry and Sustainable Animal Husbandry, World Agroforestry Centre, East and Central Asia, Beijing, China, <sup>3</sup>Synergetic Innovation Center of Food Safety and Nutrition, Harbin, China, <sup>4</sup>Kunming Institute of Botany, Chinese Academy of Sciences, Kunming, Kunming, Yunnan, China.

To investigate the effects of *Yucca schidigera* extract on fermentation characteristics in vitro, 5 treatments consisted of supplemental *Y. schidigera* extract at 0, 0.5, 2.5, 12.5 and 62.5 mg, which were assigned ran-

domly to 6 of 30 incubation bottles together with 0.5 g TMR (therefore, the adding levels of *Y. schidigera* were 0, 1, 5, 25, 125 g/kg TMR), 50 mL basal media and 25 mL rumen fluid obtained from rumen-cannulated lactation Holstein dairy cows. Cumulative gas production was continuously measured in an automated trace gas recording system (AGRS-III, Beijing) lasted for 30 h at a 39°C incubator. All fermentation vessels were cultured in shaking incubator at 39°C for 30 h. All the data were analyzed by SAS9.2. Results showed that pH and microbial crude protein (MCP) were not affected by the treatments, while the average pH in the treatments were  $6.6 \pm 0.05$ ,  $6.6 \pm 0.05$ ,  $6.6 \pm 0.06$ ,  $6.6 \pm 0.03$  and  $6.6 \pm 0.04$ , the average MCP were  $1.58 \pm 0.025$ ,  $1.61 \pm 0.034$ ,  $1.58 \pm 0.049$ ,  $1.61 \pm 0.018$  and  $1.67 \pm 0.055$  mg/mL, while the total gas production, in vitro dry matter degradability (IVDMD), neutral detergent fiber degradation rate (DNDF) and acid detergent fiber degradation rate (DADF) were improved ( $P = 0.028$ ,  $P = 0.042$ ) at the level of 25 and 125 g/kg TMR. The averages of the total gas production were  $121.85 \pm 0.034$ ,  $119.68 \pm 0.037$ ,  $120.22 \pm 0.037$ ,  $135.29 \pm 0.041$  and  $130.35 \pm 0.042$  mL/g, respectively. The average percent of IVDMD were  $60.48 \pm 0.888$ ,  $62.45 \pm 1.045$ ,  $62.22 \pm 0.686$ ,  $64.55 \pm 0.796$  and  $64.55 \pm 0.765$ , while the DNDF were  $43.24 \pm 0.004$ ,  $43.28 \pm 0.003$ ,  $43.69 \pm 0.001$ ,  $44.72 \pm 0.004$  and  $44.32 \pm 0.003$ , and the DADF were  $31.66 \pm 0.003$ ,  $31.87 \pm 0.007$ ,  $32.10 \pm 0.001$ ,  $32.95 \pm 0.001$  and  $33.01 \pm 0.003$ , respectively. With the increasing levels of *Y. schidigera* extract, the concentration of  $\text{NH}_3\text{-N}$  was decreased ( $P = 0.042$ ), while the acetate, propionate and total volatile fatty acids (TVFA) were increased ( $P = 0.030$ ,  $P = 0.009$ ,  $P = 0.048$ ) after 30 h incubation. The mean values of  $\text{NH}_3\text{-N}$  were  $39.60 \pm 0.839$ ,  $36.18 \pm 1.063$ ,  $35.07 \pm 1.576$ ,  $34.28 \pm 1.698$  and  $34.12 \pm 1.073$  mg/mL, while the TVFA were  $65.72 \pm 1.388$ ,  $67.04 \pm 1.473$ ,  $73.52 \pm 1.584$ ,  $67.82 \pm 1.824$  and  $72.57 \pm 1.080$  mmol/L, respectively. The results indicated that *Y. schidigera* extract was beneficial to manipulate rumen fermentation in vitro.

**Key Words:** *Yucca schidigera* extract, in vitro fermentation, gas production

**W420 Influence of Indian odd fruit oil and combination with yucca saponin or rubber seed oil on in vitro rumen fermentation parameters.** J. L. Niu<sup>1</sup>, L. Ma<sup>1,2</sup>, D. P. Bu<sup>\*1,3</sup>, L. Pan<sup>1</sup>, and J. C. Xu<sup>4</sup>, <sup>1</sup>State Key Laboratory of Animal Nutrition, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>2</sup>CAAS-ICRAF Joint Laboratory of Agroforestry and Sustainable Animal Husbandry, World Agroforestry Centre, East and Central Asia, Beijing, China, <sup>3</sup>Synergetic Innovation Center of Food Safety and Nutrition, Harbin, China, <sup>4</sup>Kunming Institute of Botany, Chinese Academy of Sciences, Kunming, Yunnan, China.

The objective of this experiment was to investigate the effects of Indian odd fruit oil (IO) and combination with *Yucca schidigera* (YS) or rubber seed oil (RO) on in vitro rumen fermentation parameters. The treatments were as follows: 5 doses (0, 1, 2, 3 and 4% DM) of IO, 5 doses (0, 0.5, 1, 1.5 and 2% DM) of IO mixed with YS (12.5 mg) (IO+YS), respectively, and 5 doses (0, 0.5, 1, 1.5 and 2% DM) of both IO and RO (IO+RO) (4 replicates per treatment). All treatments were evaluated in vitro 24 h batch culture of 25 mL rumen fluid with 0.5 g TMR (48% forage and 52% concentrate) and 50 mL basal medium. The cumulative gas production (GP) was continuously measured in an automated trace gas recording system (AGRS-III, Beijing). After 24 h, pH was determined, and samples were collected to analyze ammonia N ( $\text{NH}_3\text{-N}$ ), in vitro dry matter degradability (IVDMD), neutral detergent fiber degradation rate (DNDF), acid detergent fiber degradation rate (DADF) and volatile fatty acids (VFA). Results showed that 3% and 4% DM of IO reduced the DNDF and DADF ( $P < 0.05$ ), without effecting pH,  $\text{NH}_3\text{-N}$ ,

IVDMD, GP, total and individual VFAs, 2% DM of IO increased the concentration of acetate and propionate ( $P < 0.05$ ). 4% DM of IO+12.5 mg YS supplement reduced the DNDF and DADF ( $P < 0.05$ ), and did not affect pH, IVDMD, GP, the concentration of  $\text{NH}_3\text{-N}$  or total and individual VFAs. Supplementation of IO+RO increased pH ( $P < 0.05$ ), while did not affect IVDMD, GP, the concentration of  $\text{NH}_3\text{-N}$  or total and individual VFAs. However, 0.5% of IO+RO increased DNDF and DADF ( $P < 0.05$ ), 1% of IO+RO decreased DADF ( $P < 0.05$ ). It was concluded that supplements of IO, IO+YS and IO+RO nearly had no effect on rumen fermentation.

**Key Words:** Indian odd fruit oil, *Yucca schidigera*, rubber seed oil

**W421 Influence of rubber seed oil on in vitro rumen fermentation parameters, fatty acid composition and methane production.** J. L. Niu<sup>1</sup>, L. Ma<sup>1,2</sup>, D. P. Bu<sup>\*1,3</sup>, L. Pan<sup>1</sup>, and J. C. Xu<sup>4</sup>, <sup>1</sup>State Key Laboratory of Animal Nutrition, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>2</sup>CAAS-ICRAF Joint Laboratory of Agroforestry and Sustainable Animal Husbandry, World Agroforestry Centre, East and Central Asia, Beijing, China, <sup>3</sup>Synergetic Innovation Center of Food Safety and Nutrition, Harbin, China, <sup>4</sup>Kunming Institute of Botany, Chinese Academy of Sciences, Kunming, Yunnan, China.

A completely randomized block experiment was designed to investigate the effect of rubber seed oil on in vitro rumen fermentation, concentration of unsaturated fatty acids and gas production. Rubber seed oil was added to total mixed ration (TMR) at the rate of 0, 1, 2, 3 and 4% dry matter of the diet (4 replicates per treatment). The culture medium consisted of 25 mL rumen fluid collected from 3 fistulated Holstein cows, 50 mL basal medium and 0.5 g TMR (48% forage and 52% concentrate), and each treatment was incubated for 24 h at 39°C. Gas production, fermentation kinetics, in vitro dry matter digestibility (IVDMD), neutral detergent fiber degradation (DNDF), acid detergent fiber degradation (DADF) and volatile fatty acids (VFA) were determined. The results showed as follows, gas production, pH,  $\text{NH}_3\text{-N}$ , IVDMD, DNDF, DADF, total and individual VFAs were not affected by rubber seed oil ( $P > 0.05$ ). However, the DNDF tended to decrease linearly with increasing levels of rubber seed oil ( $P < 0.1$ ). After 24 h incubation, 3% and 4% DM of rubber seed oil increased the concentrations of t11C18:1, c9c12C18:2, c9t11CLA, C22:2 and total fatty acids (TFA) ( $P < 0.05$ ), 4% DM of rubber seed oil increased the concentrations of t9C18:1, C18:3, t10c12CLA ( $P < 0.05$ ). Rubber seed oil seemed to enhance the accumulation of unsaturated fatty acids in rumen but did not affect rumen fermentation.

**Key Words:** rubber seed oil, rumen fermentation parameter, unsaturated fatty acid

**W422 Changes of the rumen microbial profiles as affected by urea and acetohydroxamic acid addition in vitro.** D. Jin<sup>1,4</sup>, J. Q. Wang<sup>1</sup>, D. P. Bu<sup>\*1,2</sup>, P. P. Wang<sup>1</sup>, S. G. Zhao<sup>1</sup>, and X. M. Nan<sup>1,3</sup>, <sup>1</sup>State Key Laboratory of Animal Nutrition, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>2</sup>CAAS-ICRAF Joint Laboratory of Agroforestry and Sustainable Animal Husbandry, World Agroforestry Centre, East and Central Asia, Beijing, China, <sup>3</sup>Synergetic Innovation Center of Food Safety and Nutrition, Harbin, China, <sup>4</sup>Gembloux Agro-Bio Tech, University of Liège, Gembloux, Belgium.

In ruminants, urea was broken down rapidly to ammonia by rumen bacteria and urease inhibitors were used for increasing the efficiency of

urea utilization by inhibiting ruminal urease. However, the effect of urea and urease inhibitors on the rumen microbes was not clear. This study investigated the effect of urea and AHA (acetohydroxamic acid) addition in the diets on rumen microbial diversity using dual-flow continuous culture systems. Eight fermenters were used in a period of 10 d (7 d for adaptation and 3 d for sampling) experiment and TMR (containing alfalfa hay 17.72%, corn silage 17.5%, steam corn 7.39%, soybean meal 2.64%) were placed into each fermenter twice a day. Based on this diet, the fermenters were assigned to a 2 × 2 factorial arrangement of treatments with urea supplemented at 0 or 0.5% dry matter intake (DMI), and AHA equivalent to 0 or 450 mg/kg DMI. While the urea and AHA were dissolved in the artificial saliva and infused into the vessels twice daily. On each sampling day, fermentation fluids were collected at 0 h, 2 h, 6 h and 10 h from each fermenter. Total DNA of rumen microbes were extracted and subjected for DGGE and 16S rRNA gene sequencing. Distinct bacterial profiles were observed with urea addition and little differences were found with AHA addition. UPGMA analysis showed that samples with urea and AHA addition were clustered together. Group with urea addition showed a higher Shannon diversity compared with other groups ( $P < 0.01$ ). 16S rRNA gene sequencing analysis revealed that the dominant ruminal bacteria shared by all 4 groups belonged to phyla *Firmicutes*, *Bacteroidetes* and *Proteobacteria*. However, in urea adding groups, the bacteria *Lachnospiraceae*, *Clostridiaceae*, and *Succinivibrionaceae* were found in highest abundance compared with the other 2 groups ( $P < 0.01$ ). In contrast, the *Paraprevotellaceae* and *Veillonellaceae* bacteria were abundant in treatments without urea ( $P < 0.01$ ). Little difference of the bacteria abundance was found with AHA addition. In conclusion, adding urea to the diet could change the ruminal bacteria diversity while AHA addition had little effect on the rumen microbiota.

**Key Words:** rumen bacteria diversity, urea, acetohydroxamic acid (AHA)

**W423 Milk production and composition responds to dietary neutral detergent fiber and starch ratio in dairy cows.** M. Zhao<sup>1</sup>, J. Q. Wang<sup>1</sup>, D. P. Bu<sup>\*1,3</sup>, X. Q. Zhou<sup>1</sup>, D. Zhu<sup>1</sup>, and P. Sun<sup>1,2</sup>, <sup>1</sup>State Key Laboratory of Animal Nutrition, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, <sup>2</sup>CAAS-ICRAF Joint Laboratory of Agroforestry and Sustainable Animal Husbandry, World Agroforestry Centre, East and Central Asia, Beijing, China, <sup>3</sup>Synergetic Innovation Center of Food Safety and Nutrition, Harbin, China.

The objective of this study was to investigate effects of dietary carbohydrate composition with different dietary neutral detergent fiber (NDF):starch ratio by altering the forage to concentrate ratio on milk production and milk composition synthesis in dairy cows. Eight primiparous (146 ± 21.6 d in milk) including 4 rumen cannulated dairy cows were assigned to 4 total mixed rations (TMRs) in a replicated 4 × 4 Latin square design. Each experiment period was 21 d containing 14 d of adaptation and 7 d of measurement. Four dietary treatments were designed in which corn grain was gradually replaced with corn silage and oat hay. The NDF:starch ratios of TMRs were 0.86, 1.18, 1.63 and 2.34 from group T1 to T4, respectively. Dry matter intake (DMI) and milk production was averaged 20.8 kg/d and 31.4 kg/d, and decreased by 21.1% and 14.8% from group T1 to T4 ( $P < 0.01$ ). Digestibility of dry matter (DM), organic matter (OM), NDF, crude protein (CP) were linearly decreased from group T1 to T4 ( $P < 0.01$ ), and averaged 69.9%, 72.1%, 50.5% and 72.8%, respectively. However, digestibility of starch showed no difference among groups and averaged 93.7% ( $P > 0.05$ ). As NDF:starch ratio increased, milk protein content and production, and

milk lactose content and production were linearly reduced ( $P < 0.01$ ), and averaged 3.1%, 0.97 kg/d, 4.95% and 1.56 kg/d, respectively. However, milk fat content was linearly increased from 3.72% to 4.25% from group T1 to T4 ( $P < 0.01$ ). Quadratic effect was observed on milk fat production ( $P < 0.01$ ), which increased from 1.23 kg/d to 1.31 kg/d from group T1 to T3, and then decreased to 1.20 kg/d in group T4. Therefore, it implied that great attention should be directed at this dietary factor (NDF:starch ratio) in practical diet formulation.

**Key Words:** carbohydrate composition, NDF:starch ratio, milk production

**W424 Evaluation of enhanced mineral block on in vitro rumen microbial fermentation.** Chang Dae Jeong, Lovelia L. Mamuad, Catherine G. Avedoza, Sang Suk Lee\*, Bang Geul Kim, and Maro Lee, *Sunchon National University, Suncheon, Jeonnam, South Korea.*

Mineral block is a strategic feed supplement for ruminants that provides a constant source of minerals to promote growth of rumen microbes. Hence, this study was conducted to investigate the effect of enhanced mineral blocks on in vitro rumen fermentation. Ruminal samples were collected from 3 ruminally cannulated Holstein Friesian cattle, and a commercial concentrate substrate was offered at 2g DM/100mL buffered ruminal fluid. Five enhanced mineral blocks were investigated and the treatments were: non addition (control), commercial mineral block (T1), T1+0.6ppm selenium (T2), T1+100ppm ammonium chloride (T3), T1+300ppm sodium bicarbonate (T4) and T1+0.03% molasses (T5). Each serum bottle was filled anaerobically and sealed before incubation at 0, 3, 6, 12 and 24 h. All treatments were conducted in triplicates and analyzed by ANOVA for randomized complete block design. Duncan's multiple range test (DMRT) was used to identify differences among treatments and control. All analyses were carried out using SAS version 9.1 (SAS, 2002). All treatments resulted in decreased pH and increased gas production as incubation time increases. Total gas production was higher ( $P < 0.05$ ) in T1 to T4 than that of T5 and control at 12 h of incubation. Ammonia-nitrogen (NH<sub>3</sub>-N) concentration was lowest ( $P < 0.05$ ) in T4 (11.54 mM) after 24 h incubation followed by T5 and then T3. Also, propionate concentrations were higher ( $P < 0.05$ ) in T2 to T5 at 12 h and T1 to T5 at 24 h incubation with T4 and T3 being the highest after 12 and 24 h of incubation with 22.56 mM and 27.55 mM, respectively. Acetate to propionate ratio was inversely proportional as the time increases in all treatment groups wherein lowest concentration was observed in T3 (1.47 mM) after 24 h incubation. Total volatile fatty acid (TVFA) was relatively higher in all treatments than the control group wherein T4 have the highest result after 12 and 24 h incubation with 72.13 and 86.56 mM, respectively. In conclusion, enhanced mineral block such as sodium bicarbonate (T4) can improve gas production, propionate and total volatile fatty acid for better rumen microbial fermentation.

**Key Words:** enhanced mineral block, in vitro, total volatile fatty acid

**W425 Effects of cassava peel as a replacement for corn on nutrient digestibility and lactating performance of dairy cows.** Viviany Lúcia Fernandes dos Santos<sup>1</sup>, Marcelo de Andrade Ferreira<sup>2</sup>, Geraldo Tadeu dos Santos<sup>3</sup>, Raphael Eduardo Moretti<sup>3</sup>, Tobias Tobit de Barros Melo<sup>2</sup>, Leônia Régia Costa da Silva<sup>1</sup>, Thaynah Vasconcelos Gracindo<sup>1</sup>, Michelle de Oliveira Maia Parente<sup>4</sup>, and Valdi de Lima Júnior<sup>\*1</sup>, <sup>1</sup>Universidade Federal do Rio Grande do Norte, Natal, Rio Grande do Norte, Brazil, <sup>2</sup>Universidade Federal Rural de Pernambuco, Recife, Pernambuco, Brazil, <sup>3</sup>Universidade Estadual



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Fresh cassava peel (*Manihot esculenta*) has a potential used as a ruminant feed due to advantages such as a high amount of soluble carbohydrates with high digestibility. This agroindustrial residue could provide the energy necessary for animal production in situations where the availability of feed is limited, because it is cheaper than commodities such as corn. Eight Holstein cows ( $480 \pm 24$  kg BW and  $88 \pm 17$  d in milk) were allotted in a double Latin-Square design ( $4 \times 4$ ) to evaluate the effect of the replacement of corn by cassava peel in the diets on performance of dairy cows. Each experimental period lasted 21 d, 14 d for adaptation and 7 d for sampling. Diets were isonitrogenous ( $12.8\% \pm 1.0$  CP, DM basis) and composed of 37.5% concentrate and 62.5% corn silage and Tifton hay. Treatments were defined by 4 replacement levels of corn by cassava peel: 0, 33, 66, and 100% (DM basis). The data were analyzed by using the MIXED procedure (SAS Inst. Inc.). Orthogonal polynomials for diet responses were determined by linear and quadratic effects. Effects were declared significant at  $P \leq 0.05$ . The replacement of corn by cassava peel linearly decreased ( $P \leq 0.05$ ) dry matter (DM) intake (17.0, 17.0, 16.0, and 16.07 kg.d<sup>-1</sup> for 0, 33, 66, and 100% of replacement) and organic matter (OM) intake. Crude protein (CP) intake and neutral detergent fiber (NDF) intake did not differ ( $P \geq 0.05$ ) among treatments. DM digestibility (65.2, 54.4, 46.7 and 44.2% for 0, 33, 66, and 100% of replacement corn by cassava peel), CP digestibility, OM digestibility and NDF digestibility also linearly decreased ( $P \leq 0.05$ ) with the replacement of corn by cassava peel. In addition, there was a linear decrease ( $P \leq 0.05$ ) in milk production (20.0, 20.0, 19.0 and 18.0 kg.d<sup>-1</sup> of replacement corn by cassava peel). Although cassava peel decreased parameters evaluated, the replacement of corn with cassava peel may be interesting in locations near to starch industry, as well as cows with low nutritional requirements.

**Key Words:** digestibility, industry residue, milk production

**W426 Creatinine excretion in Nelore heifers grazing during dry season.** Jarbas Miguel da Silva Junior\*, Luciana Navajas Rennó, Sebastião de Campos Valadares Filho, Edenio Detmann, Mario Fonseca Paulino, Rilene Ferreira Diniz Valadares, Taiane da Silva Martins, Lyvian Cardoso Alves, and Ricardo Marostegan de Paula, Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.

The creatinine excretion through the sampling spot of urine has been studied as an indicator of urinary volume. However, no studies in the literature have reported its use in animal grazing. The aim of this study was to evaluate the excretion of creatinine in beef cattle. Five heifers with  $300 \pm 15$  kg average body weight were distributed in three  $5 \times 5$  Latin squares and kept in individual paddocks equipped with feeding and drinking troughs. The experimental treatments were defined to represent those commonly used in the dry season, as follows: control (mineral salt), concentrated with 20.31% crude protein (CP) on dry matter (DM) being offered (OF) level of 0.5 to 1% of body weight fasted (BWF) OF5 and OF10, respectively; and 2 concentrated self-regulating (SR) consumption, containing 69.38% CP on a DM basis (20% urea and 20% salt) offered ad libitum (SR70) and other concentrate containing 39.73% CP based on MS being offered ad libitum (SR40). The experiment included four 17-d periods; the first 12 d were for adaptation, and the remaining 5 d were for data collection. The total collection of urine was carried out between d 13 and 17 of each experimental period, for which a Foley catheter number 26 2-way was used, with 50-mL flask placed on the d 13 of each experimental period. At the free end, the probe was coupled by conductive polyethylene hose to 2 L collection bag (polyethylene bag) with secured with ropes in a sturdy cotton fabric bag to the animal's

neck in the ventral region. The animals were led to a weigh crate every 2 h from 0800 to 2000 h, and every 4 h from 2000 to 0800 h to facilitate the emptying of the bag. A urine sampling was performed every 4 h, obtaining a sample of 10 mL diluted in 0.036 N H<sub>2</sub>SO<sub>4</sub>. The creatinine content was assessed through an kinetic colorimetric test. Data were analyzed using the PROC MIXED procedure of SAS 9.1. The creatinine excretion was not affected by treatments or day and time of sampling ( $P > 0.05$ ) with an average rate of  $23.03 \pm 0.30$  (22.73 to 23.33) mg/kgBW. The spot sampling technique may be used to estimate the daily excretion of creatinine in animals kept in a pasture system.

**Key Words:** creatinine, bovine, urine

**W427 Influence of varying level of fibrolytic enzyme on nutritive value of oat grass silage in cannulated buffalo bulls.** Mahr un Nisa\*<sup>1</sup>, Osman Ahmad Khan<sup>2</sup>, Sarwar Muhammad<sup>3</sup>, and Muhammad Sharif<sup>3</sup>, <sup>1</sup>Institute of Home and Food Science, Govt. College University, Faisalabad, Punjab, Pakistan, <sup>2</sup>Livestock and Dairy Development, Lahore, Punjab, Pakistan, <sup>3</sup>Institute of Animal Nutrition and Feed Technology, University of Agriculture, Faisalabad, Punjab, Pakistan.

This experiment was conducted to examine the influence of varying levels of fibrolytic enzyme on oat grass silage nutrient digestibility and ruminal characteristic in ruminally cannulated buffalo bulls using  $4 \times 4$  Latin square design. The enzyme used was blend of xylanase (350000 BXU/g, where 1 BXU is the amount of enzyme that will release 0.06 micromole of reducing sugar (xylose equivalent) from birch xylan per minute at pH 5.3 and 50°C) and cellulase (10000 ECU/g, where 1 ECU is the amount of enzyme that will release 0.06 micromole of reducing sugars as glucose from hydroxethyl cellulose per minute at pH 4.8 and 50°C) and was derived from *Trichoderma reesei*. The control diet (C) was without enzyme, whereas diets containing 1, 2 and 3 g enzyme per Kg of silage's dietary neutral detergent fiber (NDF) were denoted as low (LE), medium (ME) and high enzyme (HE) diets, respectively. Animals were fed only silage twice daily at 1% of their body weight. The apparent silage DM digestibility by animals fed ME diet ( $63.4 \pm 3.70$ ) was significantly higher ( $P < 0.05$ ) and was lowest in animals fed C diet ( $60.7 \pm 3.11$ ). Similar trend was noticed in CP, NDF and ADF digestibility. The ruminal pH was higher ( $P < 0.05$ ) in animals fed LE ( $6.73 \pm 0.05$ ), ME ( $6.64 \pm 0.04$ ) and HE ( $6.65 \pm 0.06$ ) diets than those fed C diet ( $6.52 \pm 0.04$ ). Similarly, ruminal NH<sub>3</sub>-N concentration (mg/dL) was higher ( $P < 0.05$ ) in animals fed LE ( $20.4 \pm 2.05$ ), ME ( $20.6 \pm 2.21$ ), HE ( $21.1 \pm 1.96$ ) diets than those fed C diet ( $18.7 \pm 1.91$ ). The blood glucose was higher ( $P < 0.05$ ) in animals fed LE, ME and HE diets than those fed C diet at 3 and 6 h postprandial. However, blood urea nitrogen remained unaltered across all diets. In conclusion, enzyme supplementation increased nutrient digestibility, ruminal NH<sub>3</sub> and blood glucose.

**Key Words:** fibrolytic enzyme, cannulated buffalo bull, silage

**W428 Adaptation of dairy cows to grazing after TMR feeding during early lactation: I. Effects on endocrine profile and hepatic expression of IGF system genes.** Ana L. Astessiano\*<sup>1</sup>, Pablo Chilibroste<sup>2</sup>, Diego A. Mattiauda<sup>2</sup>, Ana Meikle<sup>3</sup>, and Mariana Carriquiry<sup>1</sup>, <sup>1</sup>School of Agronomy, UDELAR, Montevideo, Uruguay, <sup>2</sup>School of Veterinary Medicine, UDELAR, Paysandú (EEMAC), Uruguay, <sup>3</sup>School of Veterinary Medicine, UDELAR, Montevideo, Uruguay.

Multiparous cows ( $n = 18$ ) were used in a randomized complete block design to study endocrine profile and hepatic gene expression related to

IGF system on the adaptation to grazing after TMR feeding during early lactation. During the first 60 d postpartum (DPP), cows were assigned to 1) TMR (30 kgDM/d offered; 45% forage, 55% concentrate) and 2) 50% pasture in one (am) grazing session (6 h; pasture allowance 4 cm above ground level = 15 kgDM/d) + 50% TMR (15 kgDM/d offered) (G1). At 61 DPP and during 21 d, all cows were managed as G1 group determining 2 treatments (TREAT): PostTMR and G1. Plasma and liver biopsies were collected pre (+55 DPP) and post (+75 DPP) dietary change. Gene expression was quantified by SYBR-Green real time RT-PCR. Means from a repeated analysis using a mixed model were considered to differ when  $P < 0.05$ . Average milk energy output and BCS from 61 to 82 DPP did not differ between TREAT (27.5 and  $25.7 \pm 1.4$  Mcal/d and 2.8 and  $2.7 \pm 0.1$  units for PostTMR and G1, respectively). Plasma IGF1 concentrations at +75 DPP were greater in PostTMR than G1 cows ( $161$  vs.  $127 \pm 26$  ng/mL) as decreased from +55 at +75 DPP only in the latter cows. Plasma insulin and leptin as well as hepatic IGF1, IGF2 IGFBP2, and IGFBP4 mRNA were not affected by TREAT, DPP or its interaction. Before the diet change (+55 DPP), PostTMR cows had greater hepatic IGFBP1 mRNA but lower IGFBP3 mRNA than G1 cows. However, IGFBP1 mRNA had a 2.1-fold decrease while IGFBP3, IGFBP5 and IGFBP6 mRNA had 0.4 to 0.9-fold increase in PostTMR cows with the dietary change (+55 to +75 DPP). In addition, hepatic IGFBP3 mRNA decreased by 65% during this period in G1 cows. These changes determined that at +75 DPP, hepatic IGFBP3, IGFBP5 and IGFBP6 mRNA were greater while IGFBP1 mRNA was less in PostTMR than G1 cows. Decreased plasma IGF1 and hepatic IGFBP3 mRNA in G1 cows were associated with the increased milk yield from +55 to +75 DPP in these cows. In addition, while milk yield, BCS, and plasma IGF1 were maintained after the diet change in PostTMR cows, expression of IGFBP mRNA in the liver was modified in these cows

**Key Words:** nutrition, gene expression, grazing

**W429 Effects of feeding 3-nitrooxypropanol (NOP) on rumen microbial profiles in lactating dairy cows and beef cattle.** Mi Zhou\*<sup>1</sup>, Yanhong Chen<sup>1</sup>, Jennifer Haisan<sup>1</sup>, Atmir Romero-Perez<sup>1,2</sup>, Karen A. Beauchemin<sup>2</sup>, Masahito Oba<sup>1</sup>, Maik Kindermann<sup>3</sup>, Stephanie Duval<sup>4</sup>, and Le Luo Guan<sup>1</sup>, <sup>1</sup>Department of Agricultural, Food, and Nutritional Science, University of Alberta, Edmonton, Alberta, Canada, <sup>2</sup>Agriculture and Agri-Food Canada, Lethbridge, Alberta, Canada, <sup>3</sup>DSM Nutritional Products, Animal Nutrition & Health, Basel, Switzerland, <sup>4</sup>DSM Nutritional Products France, Research Centre for Animal Nutrition and Health, Saint Louis Cedex, France.

3-Nitrooxypropanol (NOP) can reduce CH<sub>4</sub> production from the rumen of dairy cows and beef cattle, but the effect of NOP on the rumen microbiome is unknown. The current project aimed to evaluate the microbial profile changes in 4 studies with NOP fed to beef and dairy cows. Rumen contents were collected from 2 dairy (Study 1: Haisan et al., 2014; Study 2: unpublished data) and 2 beef (Study 3: Romero-Perez et al., 2014; Study 4: Romero-Perez et al., 2015) studies, and subjected to genomic DNA extraction. Bacterial, archaeal and protozoal rRNA fragments were amplified and subjected to pyrosequencing analysis using 454 Titanium FLX (Roche). The main bacteria phyla in the rumen were found to be affected by NOP treatments: in dairy, Firmicutes was greater in control than in NOP dairy cows for Study 1 (58 vs 55%,  $P = 0.03$ ); while in beef, Bacteroidetes proportions varied among different NOP levels for Study 3 (from 31% for Control to 36% for NOP,  $P < 0.01$ ) and among NOP treatment periods for Study 4 ( $P = 0.04$ ). NOP also influenced other minor bacterial phyla, with the magnitude of impact differing among studies. The major methanogen genera were similar for all studies, and similar trends were found for both beef and dairy

studies: *Methanobrevibacter ruminantium* was greater ( $P < 0.05$ ) in all 4 studies (57 vs 46%; 66 vs 51%; 52 vs 37%; 30 vs 20%) while *Mbb. gottschalkii* was less ( $P < 0.01$ ) in cattle receiving NOP (Studies 2 to 4: 36 vs 20%; 59 vs 42%; 68 vs 54%). *Mbb. smithii* and *Methanosphaera stadtmanae* responded to NOP feeding in different ways depending upon the study. Entodiniomorphida predominated the protozoal community (>90%). The protozoa composition was not affected by NOP, and no clusters formed according to NOP dosage. In conclusion, NOP may reduce enteric CH<sub>4</sub> though changing rumen microbial community, but its effect of each phylotype was different, requiring further experimentation to understand its mode of action in altering the rumen microbiome.

**Key Words:** 3-nitrooxypropanol, rumen microbiome

**W430 Effect of protein supplementation to low-quality forage diets on enteric methane production and ruminal microbial community structure of beef steers.** Adam L. Shreck\*<sup>1</sup>, Nirosh D. Aluthge<sup>3</sup>, Jenny S. Jennings<sup>2</sup>, Samodha C. Fernando<sup>3</sup>, and N. Andy Cole<sup>1</sup>, <sup>1</sup>USDA-ARS, Bushland, TX, <sup>2</sup>Texas Agrilife Research, Amarillo, TX, <sup>3</sup>University of Nebraska-Lincoln, Lincoln, NE.

British-cross steers (n = 23; initial BW:  $344 \pm 33.9$  kg) were used in a 3-period crossover design to evaluate the effect of protein supplementation to low-quality forage on ruminal methane (CH<sub>4</sub>) and metabolic carbon dioxide (CO<sub>2</sub>) emissions, forage intake, and microbial composition. Steers were individually given ad libitum access to low-quality bluestem hay (3.9% crude protein) and provided one of 3 supplements: (dry matter basis): no protein (CON), cottonseed meal (CSM; 0.29% of body weight (BW) daily) or dried distillers grains with solubles (DDGS; 0.41% of BW daily). Ruminal CH<sub>4</sub> and metabolic CO<sub>2</sub> fluxes were obtained  $6.3 \pm 1.6$  times/steer daily using a GreenFeed unit (C-Lock Inc., Rapid City, SD). Microbial community structure of ruminal fluid collected using stomach tubes was determined using 16S rRNA based sequencing. Forage intake increased ( $P < 0.01$ ) by 35.0% with protein supplementation; however, no difference ( $P = 0.14$ ; SEM: 0.36) was observed between CSM (5.82 kg/d) and DDGS (5.50 kg/d). Flux of CO<sub>2</sub> (g/d) was greater ( $P < 0.01$ ; SEM: 172.0) for steers fed CSM (5,520) and DDGS (5,453) than for steers fed CON (4,895). Steers supplemented with CSM (204.9) had greater ( $P < 0.01$ ; SEM: 5.8) CH<sub>4</sub> emissions (g/d) than DDGS (189.2), both of which were greater ( $P < 0.01$ ) than CON (174.1). Methane emissions, as a proportion of GE intake (Y<sub>m</sub>), were lower ( $P < 0.01$ ; SEM: 0.25) for DDGS (7.27%) and CSM steers (7.80%) than CON (10.10%). Principal coordinate analysis revealed shifts in microbial community structure with CON having greater abundance of specific operational taxonomic units (OTU) classified as Firmicutes and Streptococcus while CSM contained greater Bacteroidetes and Prevotella and DDGS steers had greater abundance of specific OTUs belonging to Clostridiales and Anaerobiviro. Results of this study suggests that the common practice of supplementing protein to cattle consuming low-quality forage increases overall gas emissions but yields beneficial decreases in greenhouse gas emissions per unit of intake and alters microbial community structure.

**Key Words:** enteric methane, protein, 16S rRNA

**W431 Effect of a Saccharomyces cerevisiae preparation on in vitro ruminal fermentation of four fibrous substrates.** Karla Y. Valdés<sup>1</sup>, Iván Mateos<sup>1</sup>, Cristina Saro<sup>1</sup>, Alexey Díaz<sup>1,4</sup>, María Dolores Carro<sup>2</sup>, Abdelfattah Z. M. Salem<sup>3</sup>, and María José Ranilla\*<sup>1,4</sup>, <sup>1</sup>Animal Production Department, University of León, León, Spain, <sup>2</sup>Agriculture Production Department, Technical University of Madrid, Madrid, Spain, <sup>3</sup>Faculty of Veterinary Medicine and Animal Science,



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Yeast cultures, especially *Saccharomyces cerevisiae*, are beneficial in the rumen, and have been progressively introduced into the feed industry. They can affect microbial activities, thus improving fiber digestion, modifying VFA production and increasing animal performance. Most of studies have focused on yeast effects when good quality and high concentrate diets are used, but much less attention has been paid to the possible effects with fibrous feeds. In this study, the effects of a preparation of *S. cerevisiae* (strain SC-47) on in vitro ruminal fermentation of 4 fibrous feeds differing in quality and chemical composition were analyzed. Two low-quality forages (barley straw and *Pennisetum purpureum* clon Cuba CT-115) and 2 medium-quality forages (alfalfa hay and grass hay) were used. Samples of the forages (500 mg) were accurately weighed into 120-mL serum bottles, and incubated with buffered rumen fluid (50 mL) at 39°C for 24 h. Based on previous experiments with the same product, 4 doses of the *S. cerevisiae* preparation were tested as feed additive in batch cultures at 0, 25, 50 and 100 µL/bottle. VFA and gas production, pH, ammonia-N concentrations and true in vitro digestibility (TIVD) were measured. There were no additive x substrate interactions ( $P > 0.05$ ) for any variable. TIVD, ammonia-N concentration and acetate:propionate ratio were not affected ( $P > 0.05$ ) by SC-47 addition to the batch cultures. Gas production and total VFA production were linearly increased ( $P < 0.001$ ) by increasing amounts of SC-47, whereas pH was linearly decreased ( $P = 0.010$ ). Also, the production of all individual VFA linearly increased with the addition of the different doses of SC-47 (acetate,  $P = 0.001$ ; propionate,  $P < 0.001$ ; butyrate,  $P < 0.001$ ; and the sum of isobutyrate, isovalerate and valerate,  $P < 0.001$ ). The results show that *S. cerevisiae* stimulate ruminal fermentation in vitro of these fibrous forages of different quality, and so, studies analyzing their possible influence on fermentation of low-quality substrates are recommended.

**Key Words:** *Saccharomyces cerevisiae*, batch culture, forage

**W432 Nutritional evaluation of corn wet feed in cannulated Nili-Ravi buffalo bulls.** M. Nisa<sup>\*1</sup>, M. Osman<sup>3</sup>, S. Najeeb<sup>4</sup>, and M. Sarwar<sup>2</sup>, <sup>1</sup>Department of Food Science, Nutrition and Home Economics, Govt. College University, Faisalabad, Pakistan, <sup>2</sup>Institute of Animal Sciences, University of Agriculture, Faisalabad, Pakistan, <sup>3</sup>Livestock and Dairy Development Board, Lahore, Pakistan, <sup>4</sup>Star Farm Pvt Ltd., Sargodha, Pakistan.

The study was aimed to examine the influence of replacing corn wet feed (CWF) with corn grains on feed intake, nutrient digestibility, nitrogen balance and ruminal characteristics in ruminal cannulated buffalo bulls. The animals were fed 1% of their body weight. All diets were isocaloric (2.30 Mcal/kg) and isonitrogenous (12.55%CP). The control diet CWF0 did not contain CWF, whereas CWF10, CWF20 and CWF30 diets had 10, 20, and 30% CWF, respectively. Animals were housed on concrete floor in separate pens and water was made available round the clock. The experiment lasted for 80 d. The 4 × 4 Latin square design was used in this experiment and means were compared by Duncan's new multiple range test. Dry matter (DM) and crude protein (CP) intakes were not differ ( $P > 0.05$ ) in animals fed diets containing with and without CWF. Neutral detergent fiber (NDF) intake was higher ( $P < 0.05$ ) in buffalo bulls fed diets contained different levels of CWF. Dry matter, CP and NDF digestibilities were highest ( $P < 0.05$ ) in animals fed CWF10 diet than those fed CWF0, CWF20 and CWF30 diets while DM, CP and NDF digestibilities remained same ( $P > 0.05$ ) among animals groups fed CWF0, CWF20 and CWF30 diets. All animal were in positive nitrogen balance. However, there was high nitrogen retention observed in animal

fed CWF10 and CWF20 diets. Ruminal pH in buffalo bulls at 0, 3, 6 and 9 h remained same across all diets. Ruminal NH<sub>3</sub>-N at 0 h was also same while significant ( $P < 0.05$ ) difference was observed in buffalo bulls at 3, 6 and 9 h across all diets. In this study 10% replacement of corn grain with CWF showed better nutrient digestibility, nitrogen balance and ruminal characteristics of Nili-Ravi buffalo bulls.

**Key Words:** corn wet feed, corn replacement, buffalo bulls

**W433 Effect of direct-fed microbial supplementation and different diets on performance and carcass characteristics in beef feedlot heifers.** Laura F. Prados<sup>\*2,1</sup>, Galen E. Erickson<sup>1</sup>, Nirosh Aluthge<sup>1</sup>, Samodha C. Fernando<sup>1</sup>, Curtis J. Bittner<sup>1</sup>, and Fred H. Hilscher<sup>1</sup>, <sup>1</sup>University of Nebraska-Lincoln, Lincoln, NE, <sup>2</sup>Universidade Federal de Viçosa-MG, Viçosa, Minas Gerais, Brazil.

The objective of this study was to measure feedlot performance and carcass characteristics of heifers fed a newly developed direct-fed microbial (DFM) or not within 2 different basal diets. Three hundred thirty-six heifers (348 ± 33 kg) were used in a randomized block designed study with 24 pens (6 replications/treatment). The treatment design was a 2 × 2 factorial arrangement consisting of 2 basal diets based on corn (CON) or 40% modified distillers grains plus solubles replacing corn (MDGS) and presence (1 × 10<sup>9</sup> cfu/mL of each bacteria species/animal per day) or absence of DFM. The DFM was isolated from prior experiments, grown in the lab, and consisted of members of *Bacteroides* and *Lachnospiraceae* that were top-dressed on feed immediately after feeding to ensure accurate supply daily to pens. One block (4 pens) of heifers was harvested after 133 d on feed and 3 blocks (20 pens) were harvested after 147 d on feed. Hot carcass weight was obtained the day of harvest, and LM area and fat thickness collected after a 48-h chill. Carcass adjusted final BW was calculated using HCW divided by a common dressing percentage (63%), and used for ADG and G:F. All data were analyzed using the MIXED procedure of SAS with an  $\alpha = 0.05$  and pen as the experimental unit. There were no interactions ( $P > 0.25$ ) between diet and DFM supplementation for DMI, ADG, G:F, and HCW; thus, main effects of basal diet and DFM are presented. There were no differences in DMI, ADG, and carcass characteristics ( $P > 0.23$ ) due to supplementation of DFM. Heifers fed DFM tended ( $P = 0.10$ ) to be less efficient. Feeding MDGS increased ( $P < 0.01$ ) DMI, ADG, and HCW of heifers. Feed efficiency was improved ( $P < 0.01$ ) by 7% for heifers fed MDGS diets compared with CON. Heifers fed MDGS were fatter ( $P < 0.01$ ) compared with CON likely due to greater ADG. The DFM developed for this study did not enhance performance, while feeding modified distillers grains compared with corn improved performance.

**Key Words:** carcass characteristics, direct-fed microbial, modified distillers grains plus solubles

**W434 Effect of Econase on rumen fermentation patterns, diet digestibility and performance pre-and postpartum of primiparous dairy cows.** D. N. Lobão da Silva<sup>\*1</sup>, R. Riewer<sup>2</sup>, A. Gander<sup>1</sup>, N. Walker<sup>2</sup>, B. Ellison<sup>2</sup>, M. Endres<sup>1</sup>, and N. B. Litherland<sup>1</sup>, <sup>1</sup>University of Minnesota, Saint Paul, MN, USA, <sup>2</sup>AB Vista, Marlborough, UK.

The objective of this study was to examine the effect of feeding a fibrolytic enzyme preparation (Econase) via a molasses-based liquid feed (MLF) on digestibility, ruminal patterns and milk production of primiparous dairy cows. Thirty-six (n = 18) dairy heifers were blocked by BW, BCS, mature 305 ME and assigned to 2 treatments from d 45 pre- to 56 postpartum: 1) untreated MLF diet (CON); 2) enzyme treated MLF diet (ECO). Treated MLF preparation conferred activity of 3,500



BXU/g of xylanase. An amount of 0.5mL of treated or untreated MLF was mixed to each kg of DM of pre- and postpartum TMR before feeding. Data were analyzed using the MIXED procedure of SAS. Prepartum DMI was similar (14.2 vs. 15.0 kg/d;  $P = 0.17$ ) between CON and ECO treatments and no differences were observed on ruminal pH (7.1 vs. 7.0;  $P = 0.64$ ), %NDFd (60.0 vs. 57.9;  $P = 0.48$ ) or %DMd (64.9 vs. 61.3;  $P = 0.15$ ) for CON compared with ECO treatment, respectively. However, enzyme supplementation tended ( $P = 0.09$ ) to increase colostrum yield by 32% (3.9 vs. 5.2 kg) although no changes were observed on total colostrum solids concentration. Postpartum DMI (20.6 vs. 20.9 kg/d;  $P = 0.78$ ), 3.5% FCM yield (28.5 vs. 29.9 kg/d;  $P = 0.27$ ) and milk fat (3.6 vs. 3.5%;  $P = 0.59$ ) were similar for CON and ECO treatments, respectively. Postpartum NEFA and BHBA were also similar (181.0 vs. 161.1  $\mu$ Eq/L;  $P = 0.41$ ) and (5.8 vs. 5.6 mg/dL;  $P = 0.66$ ), respectively for CON and ECO. Postpartum ruminal fluid pH tended ( $P = 0.08$ ) to be greater for ECO (6.53 vs. 6.71) compared with CON, but % NDFd was similar (53.7 vs. 52.1;  $P = 0.58$ ). Although numerical increases in milk yield (28.3 vs. 30.1 kg/d;  $P = 0.15$ ) for ECO treatment, the design did not have enough statistical power to report a significant effect. Future research is needed to investigate whether different doses and feeding period length of this fibrolytic enzyme may affect performance and growth of first lactation animals.

**Key Words:** fibrolytic enzyme, primiparous, xylanase

**W435 Effect of supplementation of rumen-protected vitamins (choline, riboflavin, and folic acid) in heat-stressed dairy cows in northern Mexico (La Laguna Region).** Pedro A. Robles-Trillo<sup>\*1</sup>, Janet Garcia<sup>1</sup>, Francisco G. Veliz-Deras<sup>1</sup>, Carlos Arechiga-Flores<sup>2</sup>, Rafael Rodriguez-Martinez<sup>1</sup>, and Karla Fernandez<sup>1</sup>, <sup>1</sup>Universidad Autonoma Agraria Antonio Narro, Torreon, Coahuila, Mexico, <sup>2</sup>Universidad Autonoma de Zacatecas, Calera, Zacatecas, Mexico.

Purpose of present work was to evaluate the effect of supplementation of rumen-protected vitamins (choline, riboflavin and folic acid) on incidence and appearance metabolic diseases. Dairy cows ( $n = 105$ ), on the transition period (fresh and challenged cows). Cows were randomly allotted from May to August into 2 groups: 1) Control cows (cows receiving the regular total-mixed ration without vitamin supplementation; TMR), and 2) RPV-cows (cows receiving a supplement containing rumen-protected vitamins; i.e., 50 g/cow/day). Five blood samplings were performed to determine incidence of metabolic disorders and/or postpartum diseases (d 0, 19, 30, 50, and 80 after R-PV supplementation). Three periparturient diseases were monitored: retained fetal membranes, metritis, and ketosis, defined as follows: (1) retained-fetal membranes (RFM), was considered when fetal membranes were not expelled 12 h after parturition; (2) metritis (M) was diagnosed when cows presented a blood-purulent vaginal discharge at 7 d after parturition; (3) ketosis: mount of  $\beta$ -hydroxybutyrate was determined through evaluation of a blood sample using reactive bands for ketone bodies (Abbott, Precision Xtra), and using a Ketometer (Ketometer Precision Xtra). Data were analyzed using a 2-independent samples comparison analysis, using a 2  $\times$  2 contingency table (considering year and treatment). Supplementation of rumen-protected vitamins (RPV: choline, riboflavin and folic acid), during the transition period of dairy cows, reduced incidence of retained fetal membranes from 52.24 to 12.30%; incidence of ketosis from 29.52 to 6.67% and incidence of metritis from 79.05 to 15.24% ( $P < 0.05$ ). In conclusion, under present study conditions, supplementation 50 g/cow/d of rumen-protected vitamins (choline, riboflavin and folic acid), during the transition period, significantly reduced incidence of

retained fetal membranes, metritis and ketosis, in heat-stressed dairy cows at northern Mexico (La Laguna dairy region).

**Key Words:** dairy cow, metabolic disorder, vitamins

**W436 Effect of reducing dietary calcium, phosphorus and trace minerals on intake and performance of finishing Nellore bulls.** Laura F. Prados<sup>\*2,1</sup>, Breno Silva<sup>2</sup>, Herlon M. Alhadas<sup>2</sup>, Marcos P. Rodrigues<sup>2</sup>, Ana C. O. Lopes<sup>2</sup>, Danilo F. T. Sathler<sup>2</sup>, Paloma M. Amaral<sup>2</sup>, Lays D. S. Mariz<sup>2</sup>, Sebastião C. Valadares Filho<sup>2</sup>, and Galen E. Erickson<sup>1</sup>, <sup>1</sup>University of Nebraska-Lincoln, Lincoln, Nebraska, <sup>2</sup>Universidade Federal de Viçosa-MG, Viçosa, Minas Gerais, Brazil.

Mineral nutritional studies are important to establish dietary needs and avoid excesses for improved excretion and economics. The aim of this experiment was to evaluate the effect of decreasing calcium, phosphorus and trace mineral concentration in finishing performance and carcass characteristics using weaned Nellore bulls ( $n = 36$ ;  $273 \pm 37$  kg) in a RCBD-designed study. Treatments A and B contained 96.3 and 97.9% of the Ca and P requirement by supplementation of limestone and dicalcium phosphate whereas treatments C and D contained 47.8 and 66.8% of the BR-CORTE published requirement for Ca and P, respectively. Treatments A and B contained 56.8 ppm of Zn, 49.2 ppm of Mn, and 15.4 ppm of Cu and C and D contained 31.2 ppm of Zn, 42.3 ppm of Mn, and 5.8 ppm of Cu. The diets were isonitrogenous (13.3% CP). Intake was monitored every day. The cattle were slaughtered (84 and 147 d) and LM area and 12th rib fat thickness measured. Feed samples were analyzed for DM, ash, CP, Ca, P, and trace minerals (Zn, Mn and Cu). Treatment differences were declared significant by Tukey's test at  $P \leq 0.05$ . Calcium, P and trace mineral concentration did not affect DM and OM, CP, NDF, EE, and NFC intake ( $P > 0.42$ ). Calcium and P intake were different among the treatments ( $P < 0.01$ ) with supplemented bulls consuming more Ca and P in treatments A and B compared with treatments C and D. Final BW, empty BW, and ADG were similar ( $P > 0.16$ ) among treatments. Carcass characteristics were not influenced ( $P > 0.70$ ) by dietary treatment. These data suggest that dietary mineral concentration did not influence cattle performance. These results indicate that councils overestimate the requirements of Ca, P, and trace minerals, so supplementation is not necessary in conventional feedlot diets. Dietary reductions in these minerals would represent a significant decrease in the costs of feedlot and reduction in mineral excretion.

**Key Words:** mineral requirement, phosphorus, trace mineral

**W437 Overfeeding energy alters visceral fat miRNA profiles in Holstein cow.** Afshin Hosseini<sup>1</sup>, Hua Bao<sup>2</sup>, Khuram Shahzad<sup>\*1</sup>, James K. Drackley<sup>1</sup>, Leluo Guan<sup>2</sup>, and Juan J. Loo<sup>1</sup>, <sup>1</sup>University of Illinois, Urbana, IL, <sup>2</sup>University of Alberta, Edmonton, AB, Canada.

MicroRNAs (miRNAs) are small regulatory molecules (~22 nt) which cause post-transcriptional gene silencing. Our objective was to examine the effect of overfeeding a moderate-energy diet on miRNA expression in mesenteric adipose tissues of dry non-pregnant cows. Fourteen Holstein cows were assigned to treatments in a randomized complete block design. All cows were fed individually a control diet (CON; NEL = 1.3 Mcal/kg) to meet 100% of NRC requirements for 3 wk, after which half of the cows were assigned to a moderate-energy diet (OVE; NEL = 1.6 Mcal/kg) and half of the cows continued on CON for 6 wk. The OVE diet was fed ad libitum and resulted in cows consuming energy at ~180% of NRC. CON cows were fed to consume only to 100% of NRC. The BW was measured from wk -3 to 6 wk. Cows were slaughtered after 6 wk, and samples of mesenteric fat collected for total RNA plus

miRNA extraction, and miRNA sequencing. The differential expression of miRNAs in OVE vs. CON was assessed using *EdgeR* with an FDR adjusted *P*-value ( $P < 0.05$ ). The analysis of predicted miRNA target genes was performed by estimating a Total Context+ Value ( $TC+V \leq -0.7$ ) using the TargetScan database. OVE cows had greater ( $P < 0.001$ ) BW at slaughter (758 vs. 693 kg). Fourteen miRNAs were differentially expressed ( $P < 0.05$ ); the OVE cows had a greater expression of miRNAs bta-miR-378, 22-3p, 107, 103, 149-5p, 378c, 30c, 1307 and 6529a, whereas, the bta-miR-199a-5p, 205, 339b, 339a and bta-miR-27a-3p were upregulated in CON cows. The TargetScan analysis predicted the transcriptional change of 47 genes related to cellular ion transport and binding (e.g.,  $Na^+/K^+$  and calcium), fat metabolism (*PPARGC1B*) and feeding behavior (*GPR178*). Overfeeding energy did not alter insulin sensitivity but seemed to affect adipogenesis and fatty acid metabolism in mesenteric adipose tissue. In OVE cows, *PPARG* was the main target gene triggered by bta-miR-30c upregulation. The results suggest that miRNAs can respond to dietary energy level and likely play a role in regulating adipose tissue metabolism and energy expenditure.

**Key Words:** dairy cow, plane of energy, visceral fat

**W438 Microbial protein yield of Nelore bulls supplemented with protein sources associated with crude glycerin on finishing phase.** Antonio Jose Neto\*<sup>1</sup>, Joanis Tilemahos Zervoudakis<sup>2</sup>, Luciana Keiko Hatamoto-Zervoudakis<sup>2</sup>, Luciano da Silva Cabral<sup>2</sup>, Juliane Quenoizoré Soares<sup>2</sup>, Rosemary Lais Galati<sup>2</sup>, Renata Pereira da Silva-Marques<sup>2</sup>, Everton Riva Donida<sup>2</sup>, and Lilian Chambó Rondena Pesqueira Silva<sup>2</sup>, <sup>1</sup>Department of Animal Science; São Paulo State University-UNESP, Jaboticabal, SP, Brazil, <sup>2</sup>Department of Basic Sciences and Animal Production; Federal University of Mato Grosso-UFMT, Cuiabá, MT, Brazil.

Crude glycerin (Gly) has the potential to partially replace starch-based ingredients in the diet, such as corn, because glycerol is converted to propionate in the rumen and acts as a precursor for hepatic glucose synthesis. The objective of this study was to evaluate the effects of Gly combined with protein sources on microbial protein yield of Nelore bulls fed tropical pasture during the rainy season. Five Nelore bulls (448.2 ± 14 kg; 25 ± 3 mo) were evaluated in a 5 × 5 Latin square design with 5 treatments and 5 periods. The animals were allocated into 5 paddocks of 0.25 ha, consisting of *Brachiaria brizantha* 'Marandu' in the rainy season. Crude glycerin (83.9% of glycerol) was used in all supplements to replace (15% of DM) corn. The supplements were: Gly with urea (GU), Gly with soybean meal (GS), Gly with cottonseed meal (GC), and Gly with corn gluten (GG), and the control received mineral mixture ad libitum. Animals were individually supplemented at the rate of 300 g · 100 kg<sup>-1</sup> of BW, daily, at 1000 h. Urine samples were collected 4 h after the supplementation to determine the concentration of urea, creatinine, nitrogen and purine derivatives, on the last day of period. Data were analyzed using the PROC MIXED of the SAS. Purines derivatives absorbed, microbial N, microbial protein and CP intake per digestible organic matter intake was lower ( $P < 0.05$ ) for control compared with protein sources (Table 1). Crude glycerin associated with protein sources can be efficient to improve microbial protein yield.

*Contd.*

**Table 1 (Abstr. W438).** Effect of supplemental nitrogen sources associated with crude glycerin on microbial protein yield of Nelore bull in tropical pasture

Item	Supplements					SEM	<i>P</i> -value
	Control	GU	GS	GC	GG		
Purines derivatives, mmol/d							
Allantoin	108.74 <sup>b</sup>	168.39 <sup>a</sup>	150.01 <sup>a</sup>	162.57 <sup>a</sup>	156.61 <sup>a</sup>	14.03	0.04
Uric acid	25.40 <sup>b</sup>	44.14 <sup>a</sup>	35.34 <sup>ab</sup>	27.15 <sup>b</sup>	24.14 <sup>b</sup>	5.94	0.01
Absorbed	110.13 <sup>b</sup>	201.99 <sup>a</sup>	170.54 <sup>a</sup>	175.93 <sup>a</sup>	164.69 <sup>a</sup>	18.02	0.02
Microbial protein yield, g/d	500.44 <sup>b</sup>	917.83 <sup>a</sup>	774.94 <sup>a</sup>	799.45 <sup>a</sup>	748.36 <sup>a</sup>	81.88	0.02
CP intake per DOMI <sup>1</sup> , g/kg	184.36 <sup>b</sup>	220.39 <sup>a</sup>	238.93 <sup>a</sup>	240.75 <sup>a</sup>	239.05 <sup>a</sup>	10.31	<0.01

<sup>1</sup>DOMI = digestible organic matter intake.

**Key Words:** *Brachiaria brizantha*, glycerin, supplement

**W439 Effects of supplemental protein sources associated with crude glycerin in Nelore bulls finished on pasture on apparent digestibility.** Antonio Jose Neto\*<sup>1</sup>, Joanis Tilemahos Zervoudakis<sup>2</sup>, Luciana Keiko Hatamoto-Zervoudakis<sup>2</sup>, Luciano da Silva Cabral<sup>2</sup>, Renata Pereira da Silva-Marques<sup>2</sup>, Rosemary Lais Galati<sup>2</sup>, Juliane Quenoizoré Soares<sup>2</sup>, Everton Riva Donida<sup>2</sup>, and Felipe Cacite<sup>2</sup>, <sup>1</sup>Department of Animal Science; São Paulo State University-UNESP, Jaboticabal, SP, Brazil, <sup>2</sup>Department of Basic Sciences and Animal Production; Federal University of Mato Grosso-UFMT, Cuiabá, MT, Brazil.

Previous studies demonstrated that crude glycerin (Gly) was an excellent energy source and can partially replace corn grain in ruminant diets. The aim of this study was to evaluate the effect of supplemental protein sources associated with Gly on digestibility of OM, NDF and CP of Nelore bulls in tropical pasture during the rainy season. Five Nelore bulls (448.2 ± 14.8 kg) were used in a 5 × 5 Latin square design (5 periods and 5 treatments). The pastures were divided into 5 paddocks of 0.25 ha each, consisting of *Brachiaria brizantha* 'Marandu'. Crude glycerin was used in all supplements to replace (15% of DM) corn. The supplements were: T1 - Gly with urea, T2 - Gly with soybean meal, T3 - Gly with cottonseed meal, and T4 - Gly with gluten, and the control received mineral mixture ad libitum. Animals were individually supplemented at the rate of 300 g · 100 kg<sup>-1</sup> of BW, daily, at 1000 h. To estimate feed intake, chromium oxide was used as external marker, and indigestible neutral detergent fiber (iNDF) was used as internal marker (forage intake). Fecal grab samples were collected on d 15–17 in different times. Fecal samples were composited on a wet-basis by period for each bull. Fecal and ingredients samples were analyzed for OM, NDF and CP contents. Data were analyzed using the PROC MIXED of the SAS. There was no effect of Gly associated protein sources on digestibility of OM and NDF (Table 1). However, there was effect on digestibility of CP ( $P = 0.04$ ). Crude glycerin showed be an good energy source and can partially replace corn grain (15% of DM) in ruminant diets. Independently of protein source utilized, there was an increase of 21.3% ( $P = 0.04$ ) on digestibility of CP for animals supplemented when compared with control group. Urea associated with Gly can be effective for improving CP digestibility of Nelore bulls in tropical pasture.

*Contd.*

**Table 1 (Abstr. W439).** Effect of supplemental nitrogen sources associated with glycerin on nutrients digestibility of Nellore bulls

Item	Dietary treatments					SEM	P-value
	Control	T1	T2	T3	T4		
OM, %	54.87	62.50	60.41	59.62	59.99	2.72	0.39
NDF, %	60.58	64.56	62.05	60.07	62.05	2.11	0.45
CP, %	55.38 <sup>b</sup>	70.55 <sup>a</sup>	73.00 <sup>a</sup>	69.78 <sup>a</sup>	68.01 <sup>a</sup>	3.76	0.04

**Key Words:** *Brachiaria brizantha*, digestibility, glycerin

**W440 Nutrient intake of Nellore bulls in tropical pasture supplemented with crude glycerin and different sources of protein.**

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This study was aimed to evaluate the effect of supplemental protein sources associated with glycerin on DM and nutrient intake of Nellore bulls in tropical pasture during the rainy season. Five Nellore bulls (448.2 ± 14.8 kg) were used in a 5 × 5 Latin square design (5 periods composed of 17 d and 5 treatments). The pastures were divided into 5 paddocks of 0.25 ha each, consisting of *Brachiaria brizantha* 'Marandu'. All diets consisted of 15% of crude glycerin (DM) associated with protein sources: urea (GU), soybean meal (GS), cottonseed meal (GC), gluten (GG), and the control. Animals had a free access to mineral supplement. Animals were individually supplemented at the rate of 300 g:100 kg<sup>-1</sup> of BW, daily, at 1000 h. To estimate feed intake, chromium oxide was used as an external marker, and indigestible neutral detergent fiber (iNDF) was used as an internal marker to estimate forage intake. All data were analyzed using the MIXED procedure of SAS. Dry matter intake (% of BW) was higher ( $P < 0.01$ ) when animals were supplemented with GU (Table 1). Forage intake was increased ( $P = 0.02$ ) fed animals with GU supplements compared with other treatments. Supplement intake was similar ( $P = 0.89$ ) among treatments. Feeding GU increased ( $P < 0.01$ ) OM, CP, NDF and TDN intake of bulls. Crude protein intake were greater in bulls consuming supplements ( $P < 0.01$ ) compared with control, which is logical due the lower content of CP in forage. Providing supplement with association of crude glycerin and urea for grazing cattle in the rainy season improved dry matter and nutrient intake. Crude glycerin can partially replace (15% of DM) energy source in ruminant diets.

**Table 1 (Abstr. W440).** Effect of supplemental nitrogen sources associated with glycerin on nutrients intake of Nellore bull in pasture

Item	Dietary treatments					SEM	P-value
	Control	GU	GS	GC	GG		
DM, intake							
% of BW	1.68 <sup>c</sup>	2.24 <sup>a</sup>	1.98 <sup>b</sup>	2.01 <sup>b</sup>	1.94 <sup>b</sup>	0.08	<0.01
Forage, g/d	8,363 <sup>b</sup>	9,416 <sup>a</sup>	7,975 <sup>b</sup>	8,034 <sup>b</sup>	7,865 <sup>b</sup>	426.72	0.02
Supplement, g/d	0.00	1,832	1,862	1,858	1,854	15.73	0.89
OM, g/d	7,656 <sup>c</sup>	10,309 <sup>a</sup>	9,002 <sup>b</sup>	9,067 <sup>b</sup>	8,877 <sup>b</sup>	390.65	<0.01
NDF, g/d	5,565 <sup>b</sup>	6,870 <sup>a</sup>	5,872 <sup>b</sup>	5,861 <sup>b</sup>	5,830 <sup>b</sup>	283.91	<0.01
CP, g/d	754.4 <sup>c</sup>	1,402 <sup>a</sup>	1,292 <sup>b</sup>	1,293 <sup>b</sup>	1,268 <sup>b</sup>	38.51	<0.01
TDN, g/d	4,686 <sup>c</sup>	6,560 <sup>a</sup>	5,793 <sup>b</sup>	5,844 <sup>b</sup>	5,712 <sup>b</sup>	239.09	<0.01

**Key Words:** *Brachiaria brizantha*, glycerin, nutrient intake

**W441 Protein molecular structure and nutritive value of yellow and black canola seed.** Katerina Theodoridou<sup>1,2</sup>, Ban Yajing\*<sup>2</sup>, and Peiqiang Yu<sup>2</sup>, <sup>1</sup>University of Saskatchewan, Saskatoon, SK, Canada, <sup>2</sup>Queen's University, Belfast, UK.

Oilseeds and their products are the most valuable agricultural crops in world trade. Canola includes the yellow and the black-seeded varieties. Apart from the chemical composition of oilseed crops, their protein secondary structure profiles may also influence protein quality, nutrient utilization and availability. Although data are rare, is vital to study protein secondary structure to understand crop's digestive behavior and nutritional value. A new approach is the use of Fourier-transformed infrared-vibration spectroscopy (FT/IR), a technique for studying the secondary structural composition, stability and conformational changes. The objective was to evaluate the nutritive value of canola seed, for ruminants, in terms of: 1) chemical and nutrient profiles 2) rumen degradation kinetics, 3) in vitro intestinal protein digestibility and 4) protein molecular structures. Yellow (CS-Y) and black (CS-B) canola seeds (n = 4) were collected from 2 harvest years (2010, 2011) and used as feed sources. Three dry Holstein cows fitted with rumen cannula were used in an in situ trial; then a 3-step in vitro procedure was conducted to determine protein intestinal digestibility. CS-Y was lower in nitrogen detergent fiber and acid detergent fiber ( $P < 0.05$ ). Crude protein content was not different between the 2 varieties but the non-protein nitrogen was lower ( $P < 0.05$ ) for CS-Y compared with CS-B. The net energy for lactation, the digestible and metabolisable energy were higher ( $P < 0.05$ ) for CS-Y compared with CS-B. The C20:1 eicosaenoic acid content (n-9) was lower ( $P < 0.05$ ) while the total polyphenols were tended to be lower ( $P \leq 0.10$ ) for the CS-Y than for the CS-B. No significant differences were observed for the effective degradability of protein and the protein's inherent molecular structural make up, between the CS-Y and the CS-B. The in vitro protein intestinal digestibility of CS-Y was higher ( $P < 0.05$ ) compared with that of CS-B. In conclusion, the breeding of CS-Y has the potential to be a promising route to reducing fiber and hull content, while at the same time increasing the level of oil.

**Key Words:** canola, protein molecular structure, ruminal digestion

**W442 Effect of supplementation of two sources and two levels of copper on performance, copper status and ruminal fermentation in Nellore bulls.** Lisia Bertonha Correa, Marcus Antônio Zanetti, Janaina Silveira Silva, Brenda Barcelos\*, and Arlindo Saran Netto, University of Animal Science and Food Engineering, Pirassununga, São Paulo, Brazil.

Thirty-five Nellore bulls were used to determine the effect of supplementation of 2 levels and 2 sources of copper (organic and inorganic) on performance, copper status in the body and ruminal parameters. The 5 treatments used were: 1) Control (C) - without copper supplementation; 2) 10 mg of Cu / kg of DM (as Cu sulfate; CuI10), 3) 40 mg of Cu / kg of DM (as Cu sulfate; CuI40); 4) 10 mg of Cu / kg of DM (as Cu proteinate; CuO10); 5) 40 mg of Cu / kg of DM (as Cu proteinate; CuO40), for a period of 84 d. Body weight and blood samples were collected every 28 d for copper and ceruloplasmin analysis. At slaughter samples collected consisted of rumen fluid for short chain volatile fatty acids (VFA) determinations and ammonia nitrogen, also liver and muscle samples were collected for copper analysis. The experimental design was a completely randomized design with 7 replicates. Data were analyzed using PROC GLM of SAS. Means were compared by contrasts at a 5% significance level. There were no significant effects of copper level in the muscle (4.45, 4.24, 4.82, 4.21 and 5.79 with 1.05 of SEM for C, I10, I40, O10 and O40, respectively) and copper concentrations in the serum (0.873, 0.885, 0.967, 0.896 and 0.901 with



0.03 of SEM for C, I10, I40, O10 and O40, respectively). However, there was a quadratic effect for supplementation with different sources and Cu levels, with respect to time. The Cu concentration in the liver was higher ( $P < 0.05$ ) in animals supplemented with CuO40 with the values 237.3, 435.7, 607.5, 466.6 and 765.4 with 48.71 of SEM for C, I10, I40, O10 and O40, respectively. The CuI40 treatment showed the highest serum activity of ceruloplasmin ( $P < 0.05$ ). There was no significant effect for feed efficiency between treatments. The ruminal pH, VFA and ammonia nitrogen were not affected by treatments. In general, copper supplementation altered the Cu "status" in the animal with a greater accumulation of Cu in the liver of animals receiving the CuO40. These results show that feeding higher amounts of organic Cu make the mineral more bioavailable.

**Key Words:** cattle, mineral, performance

**W443 Effects of dietary tannin extracts levels during a thirteen-week period on lactating cow performance and N use efficiency.** Matias J. Aguerre\*<sup>1</sup>, Benjamin D. Duval<sup>2</sup>, Mark Powell<sup>1</sup>, Peter Vadas<sup>1</sup>, and Michel A. Wattiaux<sup>1</sup>, <sup>1</sup>Department of Dairy Science, University of Wisconsin-Madison, Madison, WI, <sup>2</sup>Department of Biological Systems Engineering, University of Wisconsin-Madison, Madison, WI.

Our objective was to determine the long-term effect of feeding tannin extracts to lactating cows on performance and nitrogen (N) utilization efficiency. Thirty-four multiparous lactating cows (711 ± 59 kg BW; 113 ± 19 DIM) were randomly assigned to 3 dietary treatments in a randomized complete block design. Following a 2-wk covariate adjustment period, cows were fed their assigned treatment diets for 13 weeks. Rice hull was removed from a 54:46 forage to concentrate ratio (%DM) TMR as a tannin extract mixture from Quebracho and Chestnut trees (2:1 ratio) was included at 0 (control), 0.45, and 1.8% of dietary DM. Urinary excretion (g/d) was calculated as  $0.0283 \times \text{BW (kg)} \times \text{MUN (mg/dL)}$ . There was no interaction between dietary treatments and experimental week. Overall, milk yield (48.7 kg/d), fat-and-protein corrected milk (46.7 kg/d), milk fat content (3.89%) and yield (1.86 kg/d) and true protein yield (1.45 kg/d) were not affected by dietary treatments. There was a linear increase in DMI (29.2 to 30.9 kg/d), a linear decrease in milk/DMI (1.67 to 1.57 kg/kg) and a linear decrease in MUN (12.2 to 10.8 mg/dL) with incremental levels of tannin extracts in the diet (all  $P < 0.01$ ). There was a quadratic effect ( $P < 0.01$ ) for milk true protein content, (2.96, 3.13, and 3.00% for control, 0.45 and 1.8% tannin extract, respectively) and a tendency for linear ( $P = 0.07$ ) and quadratic ( $P = 0.06$ ) response for BW gain (0.31, 0.16 and 0.44 kg/d for control, 0.45 and 1.8% tannin, respectively). Intake of N increased linearly (782, 795, and 820 g/d) and N utilization efficiency (milk N/intake N) decreased linearly (0.30, 0.30 and 0.28) for control, 0.45 and 1.8% tannin, respectively. An 11% reduction ( $P < 0.01$ ) in estimated urinary N excretion was observed between control and 1.8% tannin extract. In this study, adding tannin to the diet reduced feed efficiency, but had a positive effect on milk protein content at 0.45% tannin extract level. Feeding tannins at 1.8% in the diet may reduce dietary protein degradation and urinary N excretion without affecting milk protein yield.

**Key Words:** tannin, nitrogen efficiency, dairy

**W444 Evaluation of inoculation method on rumen in vitro gas production kinetics.** F. O. Scarpino-van Cleef\*<sup>1</sup> and J. P. Keim<sup>2</sup>, <sup>1</sup>São Paulo State University, Jaboticabal, São Paulo, Brazil, <sup>2</sup>Universidad Austral de Chile, Valdivia, Región Los Rios, Chile.

This study evaluated the effect of pooling rumen fluid (RF) or using fluid of one cow across incubations on in vitro gas production (GP) on parameters of feedstuffs. A complete randomized block design was used and arranged in a  $4 \times 2$  factorial scheme. Concentrate (corn grain, barley, soybean meal, mineral and vitamin premix) grass silage, grass hay and grass pasture were dried, ground and placed into 160-mL glass bottles. Duplicates of each substrate (1g) plus 2 blanks were incubated for three 48-h runs (considered as technical replicates). Bottles with 85 mL of Goering-Van Soest buffer solution were purged with CO<sub>2</sub> and 10 mL of RF was added to them. One treatment was incubated with RF from one cow (different for each run; not-pooled), whereas the other, a pool of RF from the same 3 cows was used for all runs. The volume of gas was extracted with a syringe, until the pressure in the digital display of the transducer reached 0, at times 2, 3, 4, 5, 6, 8, 10, 12, 18, 24 and 48h after incubation. A generalized Michaelis-Menten (MM) model was used to estimate fermentation kinetics, considering A: asymptote of GP (mL/g OM); N: shape of the curve; C: half-life rate degradation (%/h); K: time to ferment 50% of the substrate (h); MDR: maximum degradation rate (%/h); t25, t75: time to ferment 25 and 75% of asymptotic GP, respectively (h); 48GP: GP at 48 h. MIXED procedure of SAS was used, with the incubation method and substrate as fixed, and the run as random effect. The variance components (substrate, incubation, and error) were determined for each method with the varcomp procedure of SAS. There was no interaction between substrates and inoculation method; therefore, the variables were analyzed independently. The use of pooled RF promoted greater values of A (218) and t75 (23.3) compared with not-pooled RF (A = 212, t75 = 21.4), with  $P$  values and SED of 0.02 and 2.3, and 0.03 and 0.8 for A and t75, respectively. All other parameters did not differ between inoculation methods. Substrate effect was significant ( $P < 0.001$ ) for all MM parameters. For both inoculation methods, more than 94% of the variance was due to the substrates for all MM parameters. The variance accounted for incubation-runs was similar among inoculation method. Gas production parameters of different substrates follow a same trend regardless of the inoculation method.

**Key Words:** batch culture, gas production

**W445 In vitro evaluation of a treatment to flaxseed for reducing bio-hydrogenation of the n-3 fatty acid  $\alpha$ -linolenic.** Scott L. Kronberg\*<sup>1</sup> and Eric J. Scholljegerdes<sup>2</sup>, <sup>1</sup>USDA-ARS, Northern Great Plains Research Laboratory, Mandan, North Dakota, <sup>2</sup>Department of Animal and Range Sciences, New Mexico State University, Las Cruces, New Mexico.

Given the human health benefits of increasing n-3 fatty acid intake, inadequate intake of these fatty acids by many people, and problems associated with supplying these fatty acids from marine sources, there is a need to increase amounts of n-3 fatty acids in foods that are frequently eaten and affordable. Therefore, this study evaluated the amount of the  $\alpha$ -linolenic acid (ALA) that was bio-hydrogenated by bovine ruminal microbes to evaluate the potential effectiveness of a consumer-acceptable protective treatment to flaxseed. The proprietary treatment to flaxseed was evaluated with an in vitro fermentation trial that was conducted for 12 or 24 h. A small amount of quebracho tannin was included in the in vitro medium of some fermentation tubes to determine if this material could help reduce bio-hydrogenation of ALA. Residue from each tube was lyophilized then analyzed for fatty acids using direct trans-esterification with methanolic-HCl. The trial had a completely randomized design with a factorial arrangement of treatments. The treatment applied to flaxseed reduced ( $P < 0.01$ ) bio-hydrogenation of ALA with 65% of ALA remaining after 24 h of fermentation versus 23% remaining for the non-treated flaxseed. Inclusion of tannin in fermentations

did not help reduce bio-hydrogenation ( $P = 0.15$ ), and the interaction of flaxseed treatment and tannin was not significant ( $P = 0.56$ ). These results indicate that the protective treatment applied to flaxseed may reduce bio-hydrogenation of ALA in cattle and this may lead to greater amounts of n-3 fatty acids in their muscles if they consume the treated flaxseed daily for several months before harvest.

**Key Words:** n-3 fatty acids, cattle, ruminants

**W446 Evaluation of botanical extracts supplemented or not with live yeast compared with monensin supplementation on rumen fermentation in lactating Jersey cows.** Bertrand Medina<sup>\*1</sup>, Phillip Meiring<sup>2</sup>, Bruno Ghilardi<sup>1</sup>, and Lourens Erasmus<sup>2</sup>, <sup>1</sup>Laboratoires Phodé, France, <sup>2</sup>Dept of Animal & Wildlife Sciences, University of Pretoria, South Africa.

More and more countries have banned the use of antibiotic growth promoters in livestock nutrition stimulating the neo-investigation for natural alternative additives. The objectives of this study were (i) to compare the effects of monensin sodium (MO), botanical extracts (BE) and its combination with a yeast product (BE+LY) on dairy cow performance by determining their effect on rumen pH and general rumen fermentation and (ii) to determine whether natural alternatives have the potential to replace ionophores as the primary fermentation altering feed additive for lactating Jersey cows. Four rumen cannulated lactating Jersey cows were used in a 4 × 4 Latin square design experiment. The 4 experimental treatments were: 1) Control (C), a lucerne hay/maize based TMR (10.7 MJ ME/kg, 17% CP, 32.1% NDF), 2) MO (15 mg/kg of Rumensin, Elanco Animal Health), 3) BE (1g/d/cow, Oleobiotec, Laboratoire PHODE, France) and 4) BE + LY (group 3 combined with 1x10<sup>10</sup> cfu yeast (*Saccharomyces cerevisiae* CNCM I-1077)/d/cow. The experimental periods were 25 d with the last 4 d for milk performance, rumen sampling and the *in sacco* nutrient disappearance. Results (Table 1) suggest that the tested BE has potential to be a natural alternative to ionophore antibiotics in dairy diets.

**Table 1 (Abstr. W446).** Effect of feed additives

Parameter	C	MO	BE	BE + LY	SEM
DMI (kg/d)	22.0 <sup>cd</sup>	21.3 <sup>d</sup>	22.7 <sup>c</sup>	22.3 <sup>cd</sup>	0.5
Milk production (kg/d)	22.9 <sup>cd</sup>	22.4 <sup>d</sup>	23.2 <sup>c</sup>	23.4 <sup>c</sup>	0.37
Fat %	4.32 <sup>b</sup>	4.45 <sup>ab</sup>	4.52 <sup>a</sup>	4.46 <sup>ab</sup>	0.05
Protein %	3.93	3.93	3.91	3.91	0.25
Rumen fermentation parameters measured at 1400 h (7 h post-feeding)					
Ruminal pH	6.02 <sup>d</sup>	6.28 <sup>c</sup>	6.30 <sup>c</sup>	6.10 <sup>c</sup>	0.09
Total VFA (mmol/L)	120.6 <sup>cd</sup>	106.4 <sup>d</sup>	128.1 <sup>c</sup>	121.7 <sup>cd</sup>	6.52
<i>In sacco</i> disappearance (%)					
NDF 24 h (Lucerne)	50.6 <sup>d</sup>	51.8 <sup>cd</sup>	54.7 <sup>c</sup>	50.6 <sup>d</sup>	1.35
Starch 24 h (TMR)	90.5 <sup>b</sup>	93.1 <sup>ab</sup>	95.7 <sup>a</sup>	90.2 <sup>b</sup>	1.55
N-nitrogen 24 h (TMR)	84.5 <sup>ab</sup>	85.1 <sup>a</sup>	84.8 <sup>a</sup>	82.8 <sup>b</sup>	0.50

Means in the same row without a common superscript differs (<sup>abP</sup> < 0.05; <sup>cdP</sup> < 0.1).

**Key Words:** botanical extract, monensin, lactating cow

**W447 Fertilization and dried distillers grains supplementation effects on performance and nitrogen recovery by stocker cattle grazing old world bluestem.** Phillip A. Gunter<sup>\*</sup>, Brody D. Wallis,

Phillip A. Lancaster, and Gerald W. Horn, Oklahoma Agricultural Experiment Station, Stillwater, OK.

A 2-yr study evaluated the efficacy of using dried distillers grains plus solubles (DDGS) as a substitute for nitrogen (N) or N and phosphorus (P) fertilizer in stocker cattle grazing Plains Old World bluestem. Cattle were allotted to 1 of 4 treatments: 1) Old World bluestem pastures with no N or P fertilizer and low stocking rate of 325 kg BW/ha (CONT), 2) Old World bluestem pastures fertilized with 90 kg/ha of N and no P with high stocking rate of 650 kg/ha (NFERT), 3) Old World bluestem pastures fertilized with 90 kg/ha of N and 40 kg/ha of P with high stocking rate of 650 kg BW/ha (NPFERT), and 4) unfertilized Old World bluestem pastures with the same stocking rate as NFERT and NPFERT with cattle receiving 0.75% BW of corn DDGS per day for a 5 d/week feeding schedule (SUPP). Average forage mass in yr 1 and 2 was 3,170 and 6,051 kg/ha, respectively. In yr 1 final BW ( $P < 0.05$ ), total BW gain ( $P < 0.05$ ), overall ADG ( $P < 0.05$ ), and gain/ha ( $P < 0.05$ ) were greater for SUPP compared with CONT, NFERT, and NPFERT. Nitrogen recovery as cattle weight gain was greatest ( $P < 0.05$ ) for CONT (28.32%), and SUPP (14.22%) was greater than NFERT (4.42%) and NPFERT (4.77%). In yr 2 there were no differences ( $P > 0.05$ ) between final BW, total BW gain, or overall ADG between treatments. This may have been due to greater forage mass in yr 2. Gain per ha was greater ( $P < 0.05$ ) for SUPP, NFERT, and NPFERT compared with CONT and may be due to increased stocking rates for those treatments. Nitrogen recovery was greater ( $P < 0.05$ ) for CONT (45.45%), intermediate for SUPP (18.20%), and lowest for NFERT (6.91%) and NPFERT (6.81%). Dried distillers grains can be used as a substitute for forage and N fertilizer by improving performance and N recovery by stocker cattle grazing Old World bluestem.

**Key Words:** stocker cattle, Old World bluestem, nitrogen recovery

**W448 Effect of altered nitrogen excretion by condensed tannin supplementation on steer energy losses.** Adam L. Shreck<sup>\*1</sup>, Pake J. Ebert<sup>2</sup>, Eric A. Bailey<sup>3</sup>, Jenny S. Jennings<sup>2</sup>, and N. Andy Cole<sup>1</sup>, <sup>1</sup>USDA-ARS, Bushland, TX, <sup>2</sup>Texas Agrilife Research, Amarillo, TX, <sup>3</sup>West Texas A&M University, Canyon, TX.

Uncertainty remains regarding the effect of excess dietary protein on the energetic cost of urea excretion in beef cattle. We individually fed British-cross steers (n = 27; initial BW = 350 ± 32 kg) a finishing diet based on steam-flaked corn (14.4% CP) top dressed with commercially-available condensed tannin extract (CT) at 3 levels (0, 0.5, and 1.0% of diet, DM basis). Ruminal methane (CH<sub>4</sub>) and metabolic CO<sub>2</sub> fluxes were measured using a GreenFeed unit (C-Lock Inc., Rapid City, SD) for two 20-d sampling periods, that coincided with fecal and urine sampling. Diet digestibility and N balance were estimated approximately 30 d after the experiment began (EARLY) and 30 d before the animals were harvested (LATE), using TiO<sub>2</sub> as a marker of fecal output and urinary creatinine:BW ratio as a marker for urine output, respectively. From this, we previously determined that fecal N excretion as a percentage of total N excretion increased approximately 10% as CT supplementation increased to 1.0%. Urine energy loss was estimated from urine N excretion, assuming all excreted N was urea. Heat production was estimated from the Brouwer (1965) equation. Oxygen consumption was estimated from CO<sub>2</sub> production assuming a respiratory quotient of 1.05. Flux of CO<sub>2</sub> (10,279, 10,537, and 10,478, g/d; SEM: 542.5) and CH<sub>4</sub> (144, 154, and 158 g/d; SEM:13.2) were similar ( $P \geq 0.23$ ) among treatments during both sampling periods for 0, 0.5, and 1.0% CT, respectively. Percentage of GE intake lost as CH<sub>4</sub> was not different for 0 (3.27%), 0.5 (3.32%) or 1.0% (3.71%) CT. Proportion of GE intake lost in urine averaged 1.03, 1.01, and 0.97% for CT levels of 0, 0.5, and

1.0%, respectively but was not different among treatments ( $P \geq 0.41$ ; SEM: 0.08). Heat production was similar across treatments (27.3, 28.0, 27.8 Mcal/d;  $P \geq 0.52$ ; SEM: 1.37) and no difference was observed for heat production lost as a percent of GE intake (47.0%, 45.7%, 50.1%) for 0, 0.5, and 1.0% CT, respectively. The results of this study suggest that while N excretion was altered by 10% from urine to feces by tannin supplementation, no difference in energy losses was observed.

**Key Words:** condensed tannin, net energy, nitrogen

**W449 Organ mass in pure and crossbred mature beef cows grazing different herbage allowances of grasslands.** Alberto Casal\*<sup>1</sup>, Ana L. Astesiano<sup>1</sup>, Ana I. Trujillo<sup>1</sup>, Ana C. Espasandin<sup>2</sup>, Pablo Soca<sup>2</sup>, and Mariana Carriquiry<sup>1</sup>, <sup>1</sup>Facultad de Agronomía, Universidad de la República, Montevideo, Uruguay, <sup>2</sup>Facultad de Agronomía EEMAC, Universidad de la República, Paysandu, Uruguay.

The aim of this study was to evaluate the effect of controlling the intensity of grassland grazing (Campos biome), through control of herbage allowance (HA), on mass of internal organs of beef cows of different genotype. Mature beef cows (n = 32) were used in a randomized complete block design with a factorial arrangement of herbage allowance (2.5 vs. 4 kg DM/d; LO vs. HI) of native pastures (52% DM, 8.4% crude protein, 39.7% acid detergent fiber,) and cow genotype (CG; pure breed: Angus-Hereford vs. F1 reciprocal crosses; PU vs. CR). The experiment was conducted for 3 years and at the end of the third year at  $192 \pm 10$  d postpartum cows were slaughtered and weight of all tissues and organs were collected. Means from a mixed model with repeated measure analysis were considered to differ when  $P \leq 0.05$ . The gastrointestinal tract (GIT) mass (kg) was greater ( $P = 0.02$ ) and the diaphragm and lungs mass tended ( $P \leq 0.09$ ) to be greater in HI than LO cows, while liver and heart mass were greater ( $P \leq 0.01$ ) in CR than PU cows. Absolute kidney mass was not affected by HA or CG. However, when expressed as a proportion of the carcass, GIT and kidney weights tended to be lower ( $P < 0.08$ ) and diaphragm weight was lower ( $P = 0.04$ ) in CR than PU cows ( $151.6$  vs.  $161.1 \pm 4.0$ ;  $8.2$  vs.  $8.9 \pm 0.3$  and  $12.4$  vs.  $13.3 \pm 0.3$  g/kg carcass for GIT, kidney and diaphragm, respectively). In addition, relative weights of heart and kidney were lower ( $P \leq 0.05$ ) in HI than LO cows ( $8.2$  vs.  $8.9 \pm 0.2$  and  $8.1$  vs.  $9.0 \pm 0.3$  g/kg carcass for heart and kidney, respectively). Neither absolute nor relative head weights differed between cow groups. Absolute weight of the skin tended ( $P = 0.08$ ) to be greater and relative weight was greater ( $P = 0.01$ ) in LO than HI cows ( $30.3$  vs.  $32.3 \pm 1.0$  g/kg carcass). Although absolute weight of legs was not affected by HA or CG, its relative weight was 16.7% less in CR than PU and 14.8% less in HI than LO cows. Results suggest that LO and PU cows could have increased maintenance energy requirements through increased relative organ weights, which could affect cow-calf system productivity.

**Key Words:** cattle, organ mass, rangeland

**W450 Effect of cow parameters on enteric methane production in dairy cows: an individual variation approach.** Edward H. Cabezas-García\*<sup>1</sup>, Sophie J. Krizsan<sup>1</sup>, Kevin J. Shingfield<sup>2</sup>, and Pekka Huhtanen<sup>1</sup>, <sup>1</sup>Swedish University of Agricultural Sciences, Department of Agricultural Research for Northern Sweden, Umeå, Västerbotten, Sweden, <sup>2</sup>Aberystwyth University, Institute of Biological, Environmental and Rural Sciences, Aberystwyth, Ceredigion, UK.

The current knowledge about the biological basis of between-animal variation in methane (CH<sub>4</sub>) production is still limited. The aim of this

study was to evaluate between-animal variability and repeatability animal factors expected to affect CH<sub>4</sub> emissions using a meta-analytical approach. The data set were collected from 35 studies conducted in Nordic countries involving 126 individual cows and 566 cow/period observations. All studies were conducted either using Latin square or change over designs. Diets comprised principally grass silage and cereal grain concentrates with the mean forage: concentrate ratio of 60:40 on DM basis. CH<sub>4</sub> emissions per mol VFA were calculated based on VFA stoichiometry (Wolin, 1960). The variance components analysis was conducted with MIXED procedure of SAS. Repeatability (R) was calculated as  $R = \delta^2_{\text{cow}} / (\delta^2_{\text{cow}} + \delta^2_{\text{Resid}})$ . Of animal variables affecting CH<sub>4</sub> emissions DMI showed the greatest variation and repeatability (Table 1), whereas both OMD and CH<sub>4</sub>VFA displayed small variability. Efficiency of microbial N synthesis (ENS) and passage rate of iNDF were more variable and repeatable than CH<sub>4</sub>VFA. Increased feed intake was negatively ( $P < 0.001$ ) associated with reduced OMD and CH<sub>4</sub>VFA and positively ( $P < 0.01$ ) with EMPS (H<sub>2</sub> sink). Reduced CH<sub>4</sub>/kg DM with increased DMI can be attributed to these factors. Efficiency of MPS decreased ( $P < 0.001$ ) with improved OMD that can result from increased digesta retention time in the rumen. It is concluded that between-animal CV in rumen fermentation pattern cannot account observed variation in CH<sub>4</sub>. Between-animal differences in passage rate can contribute to variation in CH<sub>4</sub> as it influences digestibility and partitioning of fermented substrate between VFA and microbial cells.

**Table 1 (Abstr. W450).** Between cow variability of variables possibly related to CH<sub>4</sub> production

Item	DMI <sup>1</sup>	OMD	ENS	iNDF-kp	Acet	Prop	CH <sub>4</sub> VFA
Mean	19.4	738	23.8	0.023	648	182	356
CV							
Total	14.0	4.6	18.1	21.9	8.1	13.1	5.0
Cow	8.7	1.50	6.5	8.4	1.25	2.52	0.96
Cow <sup>1</sup>		1.39	6.2	8.6	1.25	2.35	0.97
Repeatability	0.71	0.37	0.40	0.39	0.42	0.11	0.10

<sup>1</sup>DMI as covariate.

**Key Words:** digestibility, passage rate, rumen fermentation

**W451 Rumen fermentation responses to plant extracts rich in tannins.** Barbara J. M. Lemos\*, Flavia M. Souza, Fabiola A. Lino, Victor R. M. Couto, and Juliano J. R. Fernandes, Universidade Federal de Goiás, Goiania, Goiás, Brazil.

Effects of extracts derived from the husk of *Lafloensia pacari* (27.5% tannins, mainly hydrolysable) and *Stryphnodendron adstringens* (52.2% tannins, mainly condensed) on rumen fermentation were evaluated in 2 independent in vitro experiments. The doses were CTL, LOW, MEDIUM and HIGH (0, 30, 300, and 3000 mg/L of buffered rumen fluid, respectively). The diet (50:50 concentrate:roughage, 91% DM, 20% CP, 1.6% EE, 39% NDF) was ground (1 mm) and incubated using filter bags for 3, 6, 12, 24, 48, 72 and 96 h. The rumen fluid donor was fed with a 50:50 diet. The incubation was repeated 4 times in each experiment. The effective degradability (ED, passage rates of 5 and 8%/h) and potential degradability (PD) of DM were calculated based on DM disappearance. Culture fluid was sampled after 96 h of incubation. The responses of CTL, LOW, MEDIUM and HIGH doses of *L. pacari* were, respectively: ED5%/h = 51, 50, 50 and 28% (SEM = 0.75,  $P < 0.01$ ); ED8%/h = 44, 43, 42 and 26% (SEM = 0.69,  $P < 0.01$ ); DP = 81, 82, 81 and 48% (SEM = 2.57,  $P < 0.01$ ); N-NH<sub>3</sub> = 12.3, 11.7, 9.8 and 6.5 mg/L (SEM = 0.88,  $P < 0.01$ ); Total VFA = 62.9, 57.6, 50.9 and 42.3 mM (SEM = 4.28,  $P = 0.03$ ); Acetate = 40.6, 37.1, 33.5 and 27.3 mM (SEM = 2.02,



$P = 0.01$ ); Propionate = 11.9, 11.2, 9.7 and 8.4 mM (SEM = 0.81,  $P = 0.05$ ). The HIGH dose reduced ED5%/h (46%), ED8%/h (41%) and PD (40%), compared with CTL. The N-NH<sub>3</sub>, total VFA, acetate and propionate decreased with HIGH and also with MEDIUM and LOW doses, which were similar. In the *S. adstringens* study, responses of CTL, LOW, MEDIUM and HIGH doses were, respectively: ED5%/h = 51, 52, 52 and 25% (SEM = 1.59,  $P < 0.01$ ); ED8%/h = 44, 45, 44 and 23% (SEM = 1.58,  $P < 0.01$ ); DP = 81, 82, 83 and 37% (SEM = 1.74,  $P < 0.01$ ); N-NH<sub>3</sub> = 10.2, 10.6, 9.8 and 6.4 mg/L (SEM = 0.83,  $P = 0.01$ ); Total VFA = 55.7, 58.6, 52.8 and 32.7 mM (SEM = 4.40,  $P < 0.01$ ); Acetate = 36.8, 37.1, 33.5 and 20.9 mM (SEM = 2.63,  $P < 0.01$ ). The HIGH dose reduced the ED5%/h (52%), ED8%/h (48%), PD (54%), total VFA (41%) and acetate (43%), compared with CTL. CTL and LOW doses showed similar concentration of N-NH<sub>3</sub>, which decreased with MEDIUM and HIGH doses. There was no effect on propionate ( $P = 0.13$ ). In conclusion, these plant extracts can be potentially useful to modify rumen fermentation patterns.

**Key Words:** degradability, in vitro

**W452 Carcass characteristics of Nellore beef cattle fed with whole corn diet.** Andrea M. Mobiglia<sup>1</sup>, Fernando R. Camilo<sup>1</sup>, Victor R. M. Couto<sup>1</sup>, Wallace V. S. Santos<sup>1</sup>, Lucas D. Silva<sup>1</sup>, Bruno P. C. Mendonça<sup>2</sup>, Flávio G. F. Castro<sup>2</sup>, and Juliano J. R. Fernandes<sup>1</sup>, <sup>1</sup>Escola de Veterinária e Zootecnia da Universidade Federal de Goiás, Goiânia, Goiás, Brazil, <sup>2</sup>Agrocricia Nutrição Animal e Sementes, Goiânia, Goiás, Brazil.

The objective was to evaluate effects of virginiamycin (VM) and monensin sodium (MON) in whole corn diet, with or without the use of roughage during adaptation on carcass characteristics of feedlot cattle. The animals were kept in the feedlot in group pens for 106-d with a 21-d adaptation period. One hundred Nellore bulls were used in a randomized complete block design with a 2 × 2 factorial arrangement of 4 treatments, being 2 kinds of additive (VM and MON) and 2 adaptation strategies. Each treatment consisted of 5 pens with 5 bulls per pen. Treatments were defined by VM (25 mg/kg of DM) and MON (30 mg/kg of DM), with or without 10% (DM basis) sugarcane bagasse (SB) during adaptation period as follows: VM 10% SB; VM 0% SB; MON 10% SB and MON 0% SB. After the adaptation period, animals were fed ad libitum once daily with a diet containing 85% whole corn, 15% pelleted feed (soybean, minerals and the additive). Probability of 10% was considered as statistical difference and 15% of probability was discussed as tendency. There were no differences ( $P > 0.10$ ) between VM and MON treatments for any variables studied (Table 1). However, differences were observed ( $P < 0.10$ ) for average daily gain (ADG) when the animals were fed with SB in the adaptation period. There was no interaction between the additives and SB for any variables studied; in contrast, animals fed with SB at adaptation period tended ( $P < 0.15$ )

to have higher carcass gain (CG) and carcass daily gain (CDG) than treatments without SB, independent of the additive used. Therefore, VM and MON had a similar effect on carcass characteristics, but the treatments containing SB showed great carcass gain when the tendency was discussed. The use of roughage source (SB) may increase the performance and carcass gain due to a greater adaptation of the Nellore bulls to whole corn diet.

**Table 1 (Abstr. W452).** Carcass data of bulls feedlot fed with whole corn diet

Variable (kg)	MON		VM		SEM	P-value		
	10%SB	0% SB	10%SB	0% SB		Additive	Roughage	A × R
Initial carcass weight	197.24	197.37	196.99	196.99	1.375	0.827	0.9631	0.963
Final carcass weight	290.98	297.54	291.48	289.30	5.865	0.521	0.715	0.470
Average daily gain	1.455	1.384	1.577	1.354	0.075	0.551	0.074	0.331
Carcass daily gain	0.895	0.877	0.977	0.842	0.048	0.631	0.138	0.249

**Key Words:** adaptation, additive, feedlot

**W453 Effect of the use of virginiamycin and monensin sodium in high concentrate diet on carcass characteristics of beef cattle feedlot.** Fernando R. Camilo<sup>1</sup>, Andrea M. Mobiglia<sup>1</sup>, Gustavo R. Siqueira<sup>2</sup>, Flávio D. Resende<sup>2</sup>, Regina K. Grizotto<sup>2</sup>, Victor R. M. Couto<sup>1</sup>, and Juliano J. R. Fernandes<sup>1</sup>, <sup>1</sup>Escola de Veterinária e Zootecnia da UFG, Goiânia, Goiás, Brazil, <sup>2</sup>APTA - Agência Paulista de Tecnologia dos Agronegócios, Colina, São Paulo, Brazil.

Beef cattle feedlot is a strategy to increase the productivity, and to offer a great feedstock for the consumers. Ionophore and non-ionophore antibiotics have been used as feed additives for enable to supply of the high concentrate diets. The objective of this study was to evaluate the isolated and combined effects of the virginiamycin (VM) and monensin sodium (MON) in diets of crossbreed bulls feedlot. The animals were kept in feedlot in group pens for 105-d with a 28-d adaptation period. Three hundred thirty-nine 1/2 Nellore × 1/2 Guzera beef steers (402.7 ± 1.3 kg of initial BW) were used in a randomized complete block design with 5 treatments and 7 replicates (pens). The blocks were defined by initial BW. The treatments are defined by levels of the VM and MON (mg/kg of dry matter) as follows: 30MON; 15VM+30MON; 25VM+30MON; 34VM+30MON and 34VM. Animals were fed ad libitum twice daily with isonitrogenous and isoenergetic diets, with 88:12 concentrate:roughage (sugarcane bagasse) ratio. Bulls were weighted at the beginning, after the adaptation period and at the end of the trial. Nine animals

**Table 1 (Abstr. W453).** Carcass data of the feedlot bulls fed with virginiamycin (VM) and monensin sodium (MON) in high concentrate diets

Variables	Additive (mg/kg DM)				SEM	Contrast (P-value)				
	MON 30					VM	VM vs. MON	L	Q	VM vs. VM/MON
	0	15	25	34						
ICW (kg)	215	215	215	215	216	0.653	0.3142	0.2334	0.2706	0.2620
FCW(kg)	322	317	318	320	320	8.785	0.6798	0.7171	0.1076	0.9849
Dressing (%)	56.63	56.74	56.51	56.76	56.92	0.267	0.4125	0.8772	0.7957	0.6560
CG (kg)	106.33	102.43	102.18	105.07	104.07	3.701	0.5088	0.7079	0.1670	0.7695
BWG (kg)	166.56	159.02	160.36	162.88	159.53	5.500	0.1446	0.5162	0.1403	0.4795
GY (%)	64.04	65.63	64.19	64.53	65.38	1.203	0.4364	0.9963	0.6080	0.6206
ADCG (kg/d)	1.02	0.98	0.98	1.00	0.99	0.038	0.5029	0.7084	0.1706	0.7643

were slaughtered at the beginning of the trial to determine initial hot carcass weight (HCW). At the end of the experiment, 330 bulls were slaughtered and determined the final HCW (Table 1). There was not statistical difference ( $P > 0.05$ ) between the treatments for all variables studied. Therefore, the use of additives, MON and VM, isolated and combined, do not influenced the carcass characteristics.

**Key Words:** additive, feedlot, Nellore

#### W454 Effect of supplementation with different levels and rates of protein degradation on performance of Nellore calves.

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The objective of the present study was to evaluate the effect of different levels and rates of protein degradation on the performance of Nellore animals in their post-weaning phase on *Brachiaria brizantha* 'Marandu' pastures. The following treatments were used: protein-energy supplement containing 25% CP (C-25); protein-energy supplement containing 40% CP, with 1/3 of the CP from urea, and 2/3 plant-derived (40-1/3NPN); protein-energy supplement containing 40% CP, with 1/2 CP from urea, and 1/2 plant-derived (40-1/2NPN); and protein-energy supplement containing 40% CP, with 1/3 of the CP plant-derived, and 2/3 from urea (40-2/3NPN). The experimental period was from June to September 2013, divided into a period for acclimation, and three 28-d periods. The experimental areas consisted of 12 paddocks averaging 2.3 ha, totaling 28 ha. Eighty-four (84) uncastrated Nellore cattle at 15 mo of age, with an initial weight of  $332 \pm 6.19$  kg, were used. Animals were weighed every 28 d to determine the daily weight gain (DWG). The experimental design was completely randomized, wherein the initial animals' BW was used as a co-variable, and paddocks were considered the experimental unit. Data were analyzed as repeated measures over time in mixed models, with means compared by the *t*-test at 10% significance. No effect of supplements was found on DWG (0.673 kg;  $P = 0.96$ ). There was also no interaction between supplements and experimental periods ( $P = 0.76$ ); however, the periods altered the animal performance ( $P = 0.02$ ). The initial period provided a 30% higher DWG than the others (0.842, 0.573 and 0.603 kg/d in the first, second, and third periods, respectively). Treatment did not alter BW significantly ( $380 \text{ kg} \pm 5.24$ ;  $P = 0.90$ ). In conclusion, different levels and rates of protein degradation do not modify the animal performance.

**Key Words:** protein degradation rate, protein levels, supplement

#### W455 Ruminal parameters of Nellore steers fed with high grain diet containing virginiamycin or monensin sodium.

Andrea M. Mobiglia\*<sup>1</sup>, Fernando R. Camilo<sup>1</sup>, Alexandre A. Miszura<sup>1</sup>, Ricardo A. Souza<sup>1</sup>, Victor R. M. Couto<sup>1</sup>, Flávio G. F. Castro<sup>2</sup>, Bruno P. C. Mendonça<sup>2</sup>, and Juliano J. R. Fernandes<sup>1</sup>, <sup>1</sup>Escola de Veterinária e Zootecnia da Universidade Federal de Goiás, Goiânia, Goiás, Brazil, <sup>2</sup>Agrocria Nutrição Animal e Sementes, Goiânia, Goiás, Brazil.

The objective of this study was to evaluate the effects on ruminal parameters of Nellore steers fed with high-grain diet using virginiamycin

(VM) and monensin sodium (MON). Twelve Nellore steers ( $268 \pm 38$  kg BW) with ruminal cannula were used in completely randomized design with 2 treatments and 6 replicates. Treatments were defined by kind of additive used in diet, VM (25 ppm) and MON (30 ppm). The animals were kept in feedlot in individual pens for 21 d. Ruminal ammonia nitrogen (NH<sub>3</sub>-N), short-chain fatty acids concentration (SCFA) and ruminal fluid pH were measured before feeding (T0), 6 and 12 h after feeding (T6 and T12, respectively). Animals were fed ad libitum twice daily with a diet containing 85% whole corn and 15% pelleted feed (soybean, minerals and the additive). Data are shown in Table 1. There were no effects ( $P > 0.05$ ) on NH<sub>3</sub>-N and SCFA concentration and pH values when MON or VM were added in diet. Acetate:propionate ratio (A:P) did not show differences ( $P > 0.05$ ) between treatments. However, the pH value decreased after feeding for both treatments. In conclusion, VM and MON did not affect pH, NH<sub>3</sub>-N, SCFA and A:P ratio, however, the pH values were next to 6 and there was not wide variation between the measurement times.

**Table 1 (Abstr. W455).** Ruminal parameters of Nellore steers fed with high-grain diet containing virginiamycin or monensin sodium

Ruminal pH	VM	MON	SEM	<i>P</i> -value
T 0	6.11	6.08	0.307	0.952
T 6	6.07	6.15	0.322	0.868
T 12	5.98	6.02	0.256	0.921
SCFA (mM)				
T 0	120.34	114.99	5.073	0.476
T 6	103.74	100.00	3.969	0.534
T 12	108.95	105.23	4.025	0.536
A:P ratio				
T 0	2.25	2.44	0.228	0.556
T 12	2.07	2.12	0.166	0.812
NH <sub>3</sub> -N (mg/dL)				
T 0	9.28	8.76	1.491	0.813
T 6	12.31	12.98	3.28	0.889
T 12	10.63	13.88	1.545	0.187

**Key Words:** adaptation, additive, feedlot

#### W456 Characterization of the variation in linoleic acid (18:2) in corn silage and grain hybrids in test plots.

Michel Baldin\*, Yun Ying, Gregory Roth, and Kevin J. Harvatine, Penn State University, University Park, PA.

Dietary unsaturated fatty acids (FA) provide many challenges to ruminants as they are toxic to rumen microbes and result in formation of bioactive *trans* FA in the rumen that reduce milk fat yield. Corn silage and high moisture corn are commonly recognized as a risk factor for milk fat depression (MFD). The mechanism of increased risk of MFD is not clear, but may be due to the high concentration of linoleic acid (18:2) in corn. Corn silage and corn grain have a low concentration of FA, but due to their high inclusion rate in diets they contribute substantially to the polyunsaturated FA intake. Limited literature has reported the variation in FA profile of commercial hybrids as well as the contribution of individual plant parts to total FA in corn. To determine the location of FA in the corn plant, 4 stalks from 4 different commercial hybrids were separated into: stalk, husk + shank, leaves, cob, and kernels. On a DM basis, 80.3% of total FA were in kernels, 11.9% in leaves, 5.1% in the stalk, 1.7% in the cob, and 1.0% in husk + shank. More than 96% of the 18:1 and 92.4% of the 18:2 was in kernels, whereas 71.3% of the 18:3 was in the leaves. Second, we screened 67 corn silage and 36 corn grain hybrids from commercial test plots. Corn silage total FA (% of

DM) averaged  $2.54 \pm 0.41$  (mean  $\pm$  SD, percentiles: 10th = 2.0 and 90th = 3.1). Corn silage 18:2 (% of DM) averaged  $1.24 \pm 0.24$  (percentiles: 10th = 0.94 and 90th = 1.60) and 18:2 (% of FA) averaged  $48.66 \pm 3.0$  (percentiles: 10th = 45.2 and 90th = 52.2). In the corn grains, total FA (% of DM) averaged  $3.56 \pm 0.27$  (percentiles: 10th = 3.0 and 90th = 4.2), 18:2 (% of DM) averaged  $2.05 \pm 0.15$  (percentiles: 10th = 1.8 and 90th = 2.4), and 18:2 (% of FA) averaged  $57.5 \pm 1.6$  (percentiles: 10th = 53.4 and 90th = 60.8). In conclusion, moderate variation in 18:2 concentration (both % of DM and % of FA) was observed in commercially available hybrids. Corn FA profile may provide an additional tool to improve nutritional value of corn silage and other corn products for ruminants.

**Key Words:** corn silage, fatty acid, milk fat

#### W457 Performance of beef cattle fed with different levels of roughage in high whole grain diet during the adaptation period.

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The objective of this study was evaluated the performance of beef cattle fed with whole grain diet and different levels of roughage during the adaptation period. The animals were kept in the feedlot in group pens (6 pens/treatment) for 120-d, with 21-d adaptation period and 99-d finishing period. Seventy 2 Nellore bulls were used in a randomized complete block design with 3 treatments and 6 replicates. All animals were fed with 85% of whole grain and 15% of pelleted feed (soybean, minerals and the additive, virginiamycin)(WG), and the treatments were defined by level of roughage (R) includes in diet, as follow: 0%R (animals fed with 100%WG during adaptation and finishing period); 15%R (animals fed with 85%WG:15%R during adaptation and 100% WG during finishing period) and 5% R (animals fed with 85%WG:15% R during adaptation and 95% WG:5% R during finishing period). In this study was evaluated the dry matter intake (DMI), average daily gain (ADG) and gain:feed (G:F). There were no differences ( $P > 0.05$ ) for those variables between treatments. All animals showed the same performance and G:F, independent of roughage level in the diet. The roughage may be a problem in finishing cattle diet due to its management and storage. Therefore, animals fed without a roughage source may show the same performance when the roughage is added in the high grain diet.

**Table 1 (Abstr. W457).** Performance of beef cattle fed with different levels of roughage in high whole grain diet

Variable	Treatment			SEM	P-value
	0% R	15% R	5% R		
DMI (kg/d)	7.658	7.525	7.116	0.219	0.238
IBW (kg)	316	315	316	0.765	0.658
FBW (kg)	485	479	477	7.964	0.766
ADG (kg/d)	1.412	1.365	1.348	0.064	0.773
G:F	0.187	0.182	0.190	0.007	0.748

**Key Words:** adaptation, feedlot, whole corn

#### W458 Microalgae meal affects heifer performance and plasma fatty acids.

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Microalgae meal (MA; *Schizochytrium limacinum* CCAP 4087/2) was fed to finishing heifers (n = 285;  $453 \pm 24$  kg initial BW) to assess impact on growth performance, carcass traits, and fatty acid composition in plasma. Heifers were stratified by weight, randomly assigned to pens of 8 animals each, and fed diets based on steam-flaked corn and corn gluten feed for 89 d. Treatments consisted of 0, 50, 100, or 150 g/animal daily of MA (Alltech, Inc., Nicholasville, KY) that was included in the feed additive premix and subsequently mixed in the total ration. Approximately 6 h before feeding, blood was collected via jugular venipuncture from 3 animals/pen on d 21 and 82 for analysis of plasma concentrations of eicosapentanoic, docosapentaenoic, and docosahexaenoic acids (EPA, DPA, and DHA, respectively), which are long chain omega-3 fatty acids that are most active within the body. Feeding MA did not affect ADG ( $P = 0.58$ ), but decreased DMI ( $P = 0.16$ ) and increased G:F, HCW, yield grade, and percent of USDA Choice carcasses in a dose dependent manner (linear,  $P < 0.08$ ). Plasma concentrations of EPA (3.5, 23.6, 86.3,  $153.5 \pm 4.64$ , respectively), DPA (13.4, 15.4, 22.4,  $30.8 \pm 1.04$ , respectively), and DHA increased in proportion to the amount of MA fed (linear,  $P < 0.01$ ). Data were analyzed as a randomized complete block design using the PROC MIXED procedure of SAS 9.4 (SAS Inst. Inc., Cary, NC) with pen as the experimental unit and animal as the observational unit. Microalgae meal has potential for improving growth performance and assimilation of omega-3 fatty acids into beef tissues.

**Table 1 (Abstr. W458).** Performance, hot carcass weight, and plasma DHA concentrations

Item	0	50	100	150	SEM
DMI, kg/d	9.81	9.62	9.45	9.54	0.163
Gain:Feed <sup>1</sup>	0.0844	0.0889	0.0873	0.0929	0.0031
HCW, kg <sup>1</sup>	702.0	687.6	715.0	723.1	12.2
DHA, $\mu$ g/g plasma <sup>2</sup>	5.3	26.5	38.8	46.8	1.42

<sup>1</sup>Linear effect ( $P < 0.07$ ).

<sup>2</sup>Linear effect ( $P < 0.01$ ).

**Key Words:** omega-3, algae, beef

#### W459 Effect of concentrate supplementation on milk yield and fatty acid profile of crossbred dairy cows on tropical pasture.

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The aim of this study was to evaluate the supplementation rate on yield, composition and fatty acid (FA) profile of milk from 12 crossbred (1/2 *Bos taurus* – 1/2 *Bos indicus*) dairy cows at 60 d in milk and assigned to 4 treatments: 0, 150, 300 y 450 g concentrate (as-fed basis)/kg daily milk production, in a crossover design, with 3 periods of 15d. The cows were fed grazed tropical pasture (28% native grasses, mainly *Paspalum* spp. and *Axonopus* spp. and 72% *Brachiaria* spp. and *Cynodon neumftuensis*). The trial was conducted at the research station in Martínez de la Torre, Veracruz, Mexico. The daily milk yield was recorded through the experiment and weekly milk sampling was evaluated for composition, percentages of milk fat, protein, lactose, solid not fat (SNF), density (kg/m<sup>3</sup>) and fatty acids profile (g/100 g of fat). The data were analyzed with the SAS MIXED procedure and tested by means of orthogonal polynomials. Increasing concentrate at 0, 150, 300 and 450g/kg milk, increase (linear effect,  $P < 0.05$ ) milk yield (7.35, 8.62, 8.79,  $8.75 \pm$



0.049 kg) and quadratically affected ( $P < 0.05$ ) for SNF (6.54, 6.53, 6.64, 6.85  $\pm$  0.047) and density (1022.5, 1022.4, 1022.8, 1023.46  $\pm$  0.63). The C15:0 and C:17:0 decreased (linear effect,  $P < 0.05$ ) with increasing concentrate. The fatty acid profile particularly beneficial for human health as oleic (18:1N9C), Trans-vaccenic (18:1N11T), linoleic (C18:2N6C), linolenic (C18:3N3) and conjugated linoleic acid (C18: cis-9, trans-11 CLA and C18: trans-10, cis-12 CLA) were not modified ( $P > 0.05$ ) by increasing concentrate. Overall, this study indicates that the concentrate supplementation of grazing crossbred dairy cows increase milk production without compromising the FA profile or milk composition.

**Key Words:** milk, fatty acid profile, crossbred cow

**W460 Composition and nutritive value of palm kernel meal for ruminants.** Jennifer K. Cuenca<sup>1</sup>, Diego A. Vela<sup>1</sup>, Darren D. Henry<sup>2</sup>, Nicolas DiLorenzo<sup>2</sup>, and Christian H. Ponce\*<sup>1</sup>, <sup>1</sup>*Departamento de Ciencias de la Vida y Agricultura, Universidad de las Fuerzas Armadas ESPE, Sangolquí, Ecuador*, <sup>2</sup>*North Florida Research and Education Center, Department of Animal Sciences, University of Florida, Marianna, FL*.

Palm kernel meal originated from oil processing is readily available in Ecuador. However, information related to the nutritive value for ruminants is limited. Therefore, 2 experiments were conducted to evaluate the nutritive value of palm kernel meal (PKM) from oil palm extract plants for ruminants. For Exp. 1, 3 regional plants were selected, and PKM were collected weekly during 12 wk. Proximate analysis was performed on individual samples (mean values  $\pm$  standard errors across plants for DM, OM, CP, fat and crude fiber were 94.5  $\pm$  0.33, 95.8  $\pm$  0.23, 14.8  $\pm$  0.36, 8.9  $\pm$  0.47, and 68.68  $\pm$  2.42%, respectively). Significant variations were detected for all nutrients analyzed between plants ( $P \leq 0.016$ ). Additionally, particle size was different between plants (403  $\pm$  2  $\mu$ m;  $P < 0.001$ ). For Exp. 2, 2 ruminally cannulated cows (BW = 445  $\pm$  49.5) were used to determine ruminal in situ degradability of DM, OM, NDF, ADF and CP of PKM, and in vitro ruminal degradability parameters. Cows had ad libitum access to forage, composed of Pennisetum clandestinum. Composite samples of PKM within each plant, from the 12-wk period of Exp. 1 were used as substrate for determination of in situ degradation kinetics. Substrates were incubated in the ventral sac of the rumen for 0, 3, 6, 9, 12, 18, 24, 48, and 72 h, and in situ degradability was replicated in 2 different weeks. In situ digestibility fractions (i.e., immediate soluble, potentially degradable, undegradable, and effective degradability) were not affected by plant for all nutrients analyzed ( $P \geq 0.231$ ), with exception of rate of digestion for OM and ADF (0.036  $\pm$  0.001;  $P \leq 0.042$ ). Calculated effective degradability for DM and OM were 42.3  $\pm$  4.7, and 16.6  $\pm$  2.1%, respectively. Crude protein effective degradability was 16.1  $\pm$  3.9%. For the fiber contents, NDF and ADF effective degradability were 39.8  $\pm$  5.0%; and 23.1  $\pm$  4.2%, respectively. Additionally, 24 h IVDMD (44.7  $\pm$  2.8%) and total gas production (154.1  $\pm$  59.1 mL/g of substrate) were not different among plants ( $P = 0.897$ ). Results from these experiments provide relevant nutritive information of PKM as feedstuff for ruminants, and reveal that significant variation between processing plants can be expected in nutrient profile.

**Key Words:** nutritive value, palm kernel meal, ruminants

**W461 Effects of  $\beta$ -acid extract of hops on heifer performance and ruminal fermentation.** Justin E. Axman\*, Cadra L. Van Bibber-Krueger, Christian A. Alvarado-Gilis, Jake D. Thieszen, and James S. Drouillard, *Kansas State University, Manhattan, KS*.

Beta-acid extract of hops was fed to finishing heifers (n = 80; 389  $\pm$  23.6 kg initial BW) to assess effect on growth performance and ruminal fermentation. Heifers were randomly assigned to individual pens and fed diets based on steam-flaked corn and corn gluten feed for 147 d. Treatments consisted of a control (no additive); monensin (Elanco Animal Health, Greenfield, IN) fed at 33 mg/kg diet DM; and  $\beta$ -acid extract of hops (DSM Nutritional Products, France) fed at 10, 25, or 50 mg/kg diet DM. Ruminal fluid was collected on d 44 and 86 by rumenocentesis for analysis of VFA and NH<sub>3</sub> concentrations. Feeding  $\beta$ -acid or monensin did not alter ruminal fermentation, growth performance, DMI, or gain efficiency ( $P > 0.05$ ). Performance data were analyzed using a mixed model with diet as a fixed effect and weight block as a random effect. Pair-wise comparisons were performed using the PDIF option. Categorical data were analyzed using the Glimmix procedure with diet as a fixed effect, weight block as the random effect, and feedlot pen as the experimental unit. In conclusion, under conditions of this experiment  $\beta$ -acid extract did not alter performance or ruminal fermentation.

**Table 1 (Abstr. W461).**

Item	Control	Beta acid extract, mg/kg DM			Monensin	SEM
		10	25	50		
DMI, kg/d	9.90	9.91	9.89	9.98	9.94	0.38
G:F	0.1394	0.1405	0.1465	0.1394	0.1457	0.0049
NH <sub>3</sub> , mM	0.40	0.40	0.47	0.37	0.19	0.105

**Key Words:**  $\beta$ -acid, hops, rumen

**W462 Effect of a ruminal acidosis challenge on methane emission rate in lactating cows.** Matias J. Aguerre\*, Fei Sun, Shayna Welch, and Michel A. Wattiaux, *Department of Dairy Science, University of Wisconsin-Madison, Madison, WI*.

The objective of this study was to determine the effect of a subacute ruminal acidosis (SARA) challenge on CH<sub>4</sub> emission rate from lactating cows. We hypothesized that low ruminal pH resulting from SARA would reduce CH<sub>4</sub> emission substantially especially in cows fed a low forage diet. Eleven ruminally cannulated Holstein cows (711  $\pm$  59 kg BW; 103  $\pm$  59 DIM) were randomly assigned to either a 45 or 67% forage diet (DM basis) in a complete randomized design. After 2-wks of adaptation to a GreenFeed CH<sub>4</sub> measuring unit, 4 cows on each treatment were selected to undergo the SARA induction protocol, which included 4 periods: baseline (4 d), feed restriction (50%; 1 d), challenge (TMR + wheat-barley pellet at 20% of baseline DMI; 2 d), and recovery (normal dietary treatments feeding; 2 d). Rumen pH was recorded every 5 min for 7 of the 9 d of protocol using wireless pH meters. Methane (g/h) was measured at 12 p.m., 5 p.m., 9 p.m., and 6 a.m. during the induction protocol. Overall, milk yield (40.4  $\pm$  3.6 kg/d,  $P = 0.16$ ), DMI (23.1  $\pm$  1.8 kg/d,  $P = 0.26$ ), mean ruminal pH (6.08  $\pm$  0.15,  $P = 0.84$ ), nadir ruminal pH (5.49  $\pm$  0.15,  $P = 0.31$ ), hours below pH 5.6 (3.2  $\pm$  1.78 h/d,  $P = 0.31$ ) and CH<sub>4</sub> emission rate (12.2  $\pm$  1.0 g/h,  $P = 0.29$ ) were not affected by dietary treatments. There was no treatment by period interaction but a significant period effect for reported variables. Compared with baseline (27.3 kg/d) DMI was not different ( $P = 0.82$ ) during the challenge (26.1 kg/d) but was lower ( $P = 0.05$ ) during recovery (25.2 kg/d). Milk yield dropped ( $P < 0.01$ ) from 45.5 kg/d during baseline to 32.5 kg/d during challenge and returned almost to baseline during recovery (41.2 kg/d,  $P = 0.10$ ). Methane emission (g/h) rate were 14 and 26% lower during challenge (11.7,  $P = 0.03$ ) and recovery (10.1,  $P < 0.01$ ) compared with baseline (13.6). During recovery, CH<sub>4</sub>/DMI (g/kg) was lower (9.7,  $P = 0.05$ ) compared with baseline (12.2). Hours below pH 5.6 increased from 1.5 to 6.4/d from baseline to challenge

( $P < 0.01$ ). In this study, SARA reduced  $\text{CH}_4$  emission but there was a complex temporal relationship between rumen pH and  $\text{CH}_4$  emission during and after an acidosis episode.

**Key Words:** ruminal acidosis, methane, dairy

**W463 Co-products (Micelio and Raffinate) from the corn processing industry for finishing cattle.** Murillo Alves Porto Meschatti<sup>\*1</sup>, Maria Erika Picharillo<sup>1</sup>, Camila Delveaux Batalha<sup>1</sup>, Julia Silveira Pereira<sup>2</sup>, and Flavio Augusto Portela Santos<sup>1</sup>, <sup>1</sup>University of São Paulo, São Paulo, Brazil, <sup>2</sup>Cargill.

The objective of this experiment was to evaluate the inclusion (7 and 15%, DM basis) of 2 corn co-products derived from the citric acid industry: Micelio (77% NDF, 0.5% lignin, DM basis) and Raffinate (25.5% soluble sugars, 32% soluble fiber, DM basis) in diets with high levels of flint corn fed to 200 finishing Nelore bulls (initial BW =  $396.4 \pm 37.7$  kg). Animals were blocked based on initial BW and randomly allocated to 25 pens. The control diet contained 11.73% sugarcane bagasse, 67.2% corn, 18% corn gluten feed, 2.07% mineral supplement and 1% urea. The other 4 treatments were Micelio 7, Micelio 15, Raffinate 7 and Raffinate 15. The co-products partially replaced corn in the diets. The parameters evaluated were dry matter intake (DMI), average daily gain (ADG), feed efficiency (ADG/DMI) and carcass traits. The data were analyzed using PROC MIXED of SAS and means were compared by Tukey test considering the block as random effect and treatment as fixed effect. There was no effect of treatments ( $P > 0.05$ ) on cattle performance and carcass traits. Average values for dry matter intake, average daily gain and feed efficiency were 10.5 kg/d, 1.6 kg/d, and 0.156 respectively. Average values for dressing (%), back fat thickness (mm) and rib eye area ( $\text{cm}^2$ ) were 53.79, 2.84, and 64.08, respectively. The inclusion of 7 or 15% of Micelio or Raffinate in partial replacement of ground flint corn in finishing diets does not affect cattle performance

**Key Words:** co-product, feedlot, performance

**W464 The effect of heating temperature on ruminal protein digestion kinetics and estimated intestinal digestibility of canola meal.** Katarzyna Burakowska<sup>\*1</sup>, Pawel Górka<sup>2</sup>, and Gregory B. Penner<sup>1</sup>, <sup>1</sup>University of Saskatchewan, Saskatoon, Canada, <sup>2</sup>University of Agriculture, Kraków, Poland.

The objective of this study was to determine the optimal heat treatment temperature of canola meal to increase the rumen undegradable protein concentration without negatively affecting intestinal digestibility. Twelve samples of canola meal (25 kg/sample) were subjected to 1 of 4 heat treatments: an unheated control (H0), or heat-treated in a tumble dryer (POS, Saskatoon) to 100°C (H100), 110°C (H110) or 120°C (H120) for 10 min after reaching the specified temperature. Canola meal was placed in nylon bags (50- $\mu\text{m}$  pore size) and incubated in the rumen of 4 heifers for 0, 2, 4, 8, 12, 16, 24 and 48 h to evaluate rumen degradation kinetics. Estimated intestinal digestibility was determined using samples incubated in the rumen for 12 h, followed by an *in vitro* gastric and enzymatic digestion. Samples were analyzed for DM and CP content. Data were analyzed using SAS 9.3 as completely randomized design with polynomial contrasts to determine whether provision of heat affected the response linearly or quadratically. The rate of DM

disappearance in the rumen decreased (quadratic,  $P = 0.02$ ,  $\text{SE} = 0.214$ ) from 4.5%/h for H0 to 3.06 for H120. The rate of CP degradation ( $P < 0.01$ ,  $\text{SE} = 0.590$ ) and the degradable fractions of DM ( $P < 0.01$ ,  $\text{SE} = 1.906$ ) and CP responded quadratically ( $P < 0.01$ ,  $\text{SE} = 3.531$ ) with values of 4.89, 60.0 and 79.0 for H0, 4.59, 57.1 and 70.0 for H100, 3.54, 52.7 and 59.0 for H110, and 7.77, 34.1 and 22.3 for H120, respectively. Assuming a constant passage rate (5%/h), heat treatment decreased the effectively degradable DM (quadratic,  $P < 0.01$ ,  $\text{SE} = 0.776$ ) from 54.2 to 31.7% for H0 to H120 and decreased the effectively degradable CP (quadratic,  $P < 0.01$ ,  $\text{SE} = 2.652$ ) from 55.5 to 25.4%. The estimated intestinal CP digestibility (quadratic,  $P = 0.03$ ,  $\text{SE} = 2.550$ ) increased from H0 (45.9%) to H110 (51.0%) and decreased for H120 (37.2%). The results suggest that the temperature of 110°C decreases DM and CP degradation in the rumen without negative effects on intestinal digestibility.

**Key Words:** canola meal, heat treatment, digestibility

**W465 Effects of postruminal AA supply and roughage removal on growth performance during growing phase of beef calves.**

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Previous research demonstrated benefit in beef calves growth performance fed no roughage diets balanced to meet AA requirement. The study objective was to separate the effects of balancing diets for AA:effective energy (AA:EE) and roughage removal on steers growth performance (initial BW  $338 \pm 23$  kg) during growing phase (GP; 84 d). We hypothesized balancing diets for postruminal AA:EE supply as well as roughage removal would improve feed efficiency and growth performance. Crossbred steers ( $n = 117$ ) were randomly assigned to 3 corn-based diets. Control, common feedlot diet (CON; 13.5% CP, 20% NDF, 10% roughage, 60% corn, 27% dried distillers grains with solubles); roughage and AA:EE balanced (RBAL; 16% CP, 16% NDF, 10% roughage, 62% corn, 12% Aminoplus, 10% dried distillers grains with solubles, 2% soybean meal); or no roughage and AA:EE balanced (NRBAL; 16% CP, 10% NDF, 72% corn, 14% Aminoplus, 9% dried distillers grains with solubles, 2% soybean meal). Steers had *ad libitum* access to feed and water. Individual feed intake was measured using GrowSafe feed intake system and BW was recorded every 28 d. Data were analyzed as repeated measure. Dry matter intake (kg/d) was greater ( $P < 0.01$ , SEM 0.28) for CON (10.5) and RBAL (10.4) compared with NRBAL (9.0). Average daily gain (kg/d; 28 d) was greater ( $P < 0.03$ , SEM 0.09) for NRBAL (2.02) and RBAL (2.06) compared with CON (1.77) however throughout GP ADG did not differ among treatments ( $P = 0.3$ ). Gain:Feed (28 d) was greater ( $P < 0.01$ , SEM 0.009) for NRBAL (0.22) compared with CON (0.18) however RBAL (0.20) did not differ. During total period (84 d) NRBAL (0.19) had greater ( $P < 0.03$ , SEM 0.005) G:F compared with RBAL (0.17) and CON (0.16). Balancing diets for postruminal AA:EE supply improved G:F by 11% and roughage removal improved G:F an additional 11%. In conclusion balancing diets for postruminal AA:EE supply can improve growth performance and feed efficiency when lean tissue deposition potential results in postruminal AA demand. Balancing diets to allow roughage removal also benefits growth performance throughout the feeding period.

**Key Words:** postruminal AA, feed efficiency

## Ruminant Nutrition: Other ruminants

**W466 Dynamic changes of main rumen microflora and ruminal fermentation in sheep supplemented with molasses-urea.** Alatengdalai, Shuyuan Xue, Ming Hu, and Changqing Li\*, *Inner Mongolia Academy of Agricultural and Animal Husbandry Sciences, Huhhot, Inner Mongolia, China.*

The digestion of diet and utilization of nutrients by sheep depends on rumen microbial fermentative digestion. Reproduction and growth rate of microbes is mainly determined by the availability of nutrient and energy levels in rumen. Therefore, rumen environment affects the extent of microbial protein (MCP) synthesis. The aim of this study is to evaluate the effects of licking the molasses-urea feed block on the rumen of sheep, including microbial biomass and fermentative efficiency. Eight sheep were selected and divided into 2 groups (control group and treatment group). The treatment group animals were supplied with molasses-urea feed block for ad libitum consumption. Rumen fluid was collected every 2 h and rumen fermentation parameters were measured. In addition, the population of majority bacteria were investigated by real-time PCR. The results showed that the population of majority bacteria is increased in rumen of treatment group animals ( $P < 0.05$ ). The trend of daily change of rumen bacteria was same in all animals. Each bacterium quantity decreased gradually after feeding, and reached the lowest level after 2 h intake. It then increased slowly and reached the highest level after intake for 8 h. Finally, each bacterium quantity was back to same level as before intake. In contrast, the protozoa number raised to the highest in 4 h feeding and declined gradually, the concentration of protozoa in treatment group sheep was significantly higher than that of control group ( $P < 0.05$ ). The pH of rumen liquids were in normal range and were not different between both groups. However, pH decreased from the highest level before feeding to the lowest level within 4 h. It was increased after intake for 8 h. The concentration of  $\text{NH}_3\text{-N}$  and MCP synthesis in the rumen liquids were both significantly higher than that of control group ( $P < 0.05$ ), the highest concentration of  $\text{NH}_3\text{-N}$  and MCP were reached after feeding by 2 h and 4 h respectively. Block containing urea and molasses can be well utilized by sheep, it is conducive for rumen fermentation by the microbes in ruminant.

**Key Words:** molasses-urea, sheep, rumen

**W467 Effects of corn levels on TMR on growth performance and economic efficiency in growing Korean black goats.** S. U. Kim\*, M. J. Ku, Y. S. Choi, S. K. Lee, D. H. Park, and S. G. Park, *Jeollanam-do Livestock Research Institute, Gangjin-gun, Jeollanam-do, Republic of Korea.*

This study was conducted to investigate the growth performance and economic efficiency in dietary corn mixing level on total mixed ration (TMR) in growing goats. A total of 32 castration goats with similar age (average 4 mo) and body weight (average 16 kg) were equally allocated to dietary 4 treatments in a randomized complete block design. Dietary treatments included a controls (conventional) group and 3 corn mixing treatments groups (fed diets containing 15, 25, 35% corn mixing level of TMR). Average daily gain (ADG) was significantly ( $P < 0.05$ ) higher in control group (103.5 g). Within treatments, ranked the highest in 25% (91.2 g), 15% (84.8 g) 35% (72.6 g) corn mixing treatments ( $P < 0.05$ ). Daily feed intakes was significantly ( $P < 0.05$ ) higher in control group (1,076 g). Within treatments, ranked the highest in 15% (1,050 g), 25% (1,041 g), 35% (1,015 g) corn mixing treatments ( $P < 0.05$ ). The amounts of feed required for body weight gain ranked the lowest in control group (10.4),

followed by 15% (11.4), 25% (12.4) and 35% (14.0) corn mixing treatments ( $P < 0.05$ ). However, comparing the feed cost contrast with market price for economic analysis by comparing the income results, the income ranked the highest 396 Won (36 cents) in 25% corn mixing treatments, followed by 15% corn mixing treatments 344 Won (31.2 cents), control group 322 Won (29.2 cents) and 35% corn mixing treatments 254 Won (23 cents) ( $P < 0.05$ ). In conclusion, it was estimated that the optimal level of corn mix on TMR might be 25% feed intake, when diets based TMR were fed to growing Korean black goats for their improvements of economic efficiency.

**Key Words:** Korean black goats, corn mixing levels, total mixed ration (TMR)

**W468 Effects of water restriction following feeding on microbial N supply and excretion of urinary purine derivatives in Corriedale ewes under heat stress condition.** J. Ghassemi Nejad, B. W. Kim\*, B. H. Lee, J. L. Peng, D. H. Ji, and K. I. Sung, *College of Animal Life Sciences, Kangwon National University, Chuncheon, Kangwon, South Korea.*

Urinary excretion of purine derivatives (PDs) is used to estimate the microbial N supply to sheep. The objective of this study was to determine the effect of water restriction following feeding on microbial N supply, and urinary excretion of PDs in Corriedale ewes under heat stress condition. Nine Corriedale ewes (average BW =  $45.0 \pm 4.5$  kg) were fed diets based on maintenance requirements individually in metabolic crates. Sheep were assigned to 3 treatment groups according to a  $3 \times 3$  Latin square design for 3 periods of 21 d duration each (9 sheep per treatment). Treatments were free access to water (FAW), 2h water restriction (2hWR), and 3h water restriction (3hWR) following feeding. No differences were found in body weight gain among all treatment groups ( $P > 0.05$ ). Daily water intake decreased linearly as water restriction time following feeding increased ( $P < 0.05$ ). Urine weight and volume were higher ( $P < 0.05$ ) in FAW than restricted groups ( $P > 0.05$ ). Water restriction following feeding decreased the urinary N (g/d) and urinary N as a percentage of N intakes ( $P < 0.05$ ); however, fecal N (g/d) and fecal N as a percentage of N intake were not affected by water restriction following feeding ( $P > 0.05$ ). Allantoin concentration tended to be higher ( $P = 0.07$ ) in FAW group than 2hWR and 3hWR groups while no differences ( $P > 0.05$ ) were observed in uric acid, xanthine + hypoxanthine and creatinine concentrations. Sum of PDs tended to be higher in FAW ( $P = 0.08$ ) but water restriction following feeding reduced total PDs excretion compared with the FAW group ( $P < 0.01$ ). It is concluded that water restriction following feeding for less than 3 h improved microbial N supply in ewes under heat stress condition.

**Key Words:** ewe, heat stress, microbial nitrogen

**W469 Comparative study between two traditional camel rearing systems effects on milk fat and its fatty acids profile.** Amir Ahmadpour<sup>1</sup>, Abdol Hamid Karimi<sup>2</sup>, Rupert Bruckmaier<sup>3</sup>, and Mousa Zarrin\*<sup>4</sup>, *<sup>1</sup>Department of Animal Science, Bu-Ali Sina University, Hamadan, Iran, <sup>2</sup>Department of Animal Science, Fars Agricultural and Natural Resources Research Center, Shiraz, Fars, Iran, <sup>3</sup>Vetsuisse Faculty, University of Bern, Bern, Switzerland, <sup>4</sup>Department of Animal Science, Yasouj University, Yasouj, Iran.*



Raising awareness about harmfulness of high saturated fatty acid content of milk fat caused to increase the study to improve milk fat quality. The dromedary (*Camelus dromedarius*) is an important domestic animal in eastern Iran. In spite of importance of this specie, few studies have been reported on camel milk. The objective of the present study is comparison of the effects of different rearing systems on fat content and fatty acids profile of milk fat in camel. Eighteen lactating camels were assigned in 2 treatment groups include very extensive (VE, n = 9) and extensive (E, n = 9) rearing system. In the EV group, camels depended merely on pasture, but in the E group camels received 750g/animal barley after daily grazing. Animals watered twice daily and received 250 g/animal salt weekly. Animals were milked twice a day. Milk samples (250 mL from each camel) collected in the morning and analyzed for milk constituents. Thin-layer chromatography used for separation of triacylglycerol and fatty acids profile measured by gas chromatography method. Data are presented as means  $\pm$  SEM. The fat content was  $33.2 \pm 9.2$  g/L and  $32.5 \pm 11.6$  g/L for VE and E group respectively. The amounts of short-chain (C4 – C8) and medium-chain (C10 – C13) fatty acids were higher in E group compared with VE group ( $P < 0.05$ ), whereas the C14 like C16:0 and C16:1n-7 were higher in VE group ( $P < 0.01$ ). Although C18:0 had no significant difference between groups, C18:1n-9 and C18:2n-6 in E and C18:3n-3 in VE were higher ( $P < 0.05$ ). Moreover 20:0, 20:5n-3, 22:3n-3, and 22:6n-3 of VE was higher ( $P < 0.05$ ). Milk fat of VE was less saturated ( $P < 0.05$ ). Except free cholesterol and phospholipids that were lower and higher respectively in VE ( $P < 0.05$ ), no significant difference observed between groups for cholesteryl esters, free fatty acids, triacylglycerols, diacylglycerols, and monoacylglycerols. The results demonstrated that the VE rearing system increases total polyunsaturated fatty acids and decreases free cholesterol compared with E group rearing system. In conclusion, the VE rearing system could produce healthier milk fat in camels.

**Key Words:** camel, milk, fat

**W470 Influence of zilpaterol hydrochloride and enriched-Cr yeast supplementation on feedlot performance lambs.** Miguel A. Rodríguez<sup>1,2</sup>, Ignacio A. Domínguez-Vara<sup>1</sup>, Ernesto Morales-Almaráz<sup>1</sup>, and Rubén Barajas\*<sup>2</sup>, <sup>1</sup>Universidad Autónoma del Estado de México, FMVZ, Toluca, Estado de México, México, <sup>2</sup>FMVZ-Universidad Autónoma de Sinaloa, Culiacán, Sinaloa, México.

Twenty-eight Rambouillet cross lambs ( $27.85 \pm 2.91$  kg) were used in a 74-d feedlot experiment to evaluate the influence of zilpaterol hydrochloride (Zilmax, Merck Animal Health) and enriched-Cr yeast (BioChrome, Alltech) supplementation on feedlot performance of lambs. Sheep were weighed and individually allotted in  $1.2 \times 2.5$  m concrete floor pens. They were randomly assigned to 4 treatments as follows: 1) A 85% concentrate diet, contained 15.5% CP and 1.87 Mcal NEm/kg, and formulated from corn grain and soybean meal (Control); 2) Control added with 0.2 mg of zilpaterol hydrochloride/ kg BW during last 28 d in the feedlot (ZH); 3) Control plus 0.3 mg of Cr/kg DM (YCr); and 4) Control plus ZH and YCr (ZHCr). Results were analyzed as a completely randomized design with a  $2 \times 2$  factorial arrangement. Pen was the experimental unit; data were analyzed with initial weight as a covariate in the model. Final body weight tended ( $P = 0.10$ ) to be increased by ZH supplementation ( $45.4$  vs.  $44.1$  kg). Average daily gain tended ( $P = 0.07$ ) to be higher for lambs in ZH treatment ( $0.306$  vs.  $0.280$  kg/d). Dry matter intake was not affected by treatment ( $P > 0.20$ ). Gain/feed ratio was increased ( $P = 0.02$ ) by 11% ( $0.236$  vs.  $0.213$  kg/kg) with ZH supplementation. Yeast-Cr supplementation did not modify any variable analyzed ( $P > 0.20$ ). No interaction of ZH x Yeast-Cr was observed ( $P > 0.20$ ). It is concluded, that zilpaterol hydrochloride supplementation

promoted feedlot performance of lambs, whereas chromium supplementation from enriched-Cr yeast did not shown benefits to feedlot lambs both alone or in combination with zilpaterol hydrochloride.

**Key Words:** chromium, lamb, zilpaterol

**W471 Influence of zilpaterol hydrochloride and enriched-Cr yeast supplementation on carcass characteristics and meat composition of feedlot lambs.** Miguel A. Rodríguez<sup>1,2</sup>, Ignacio A. Domínguez-Vara<sup>1</sup>, María A. Mariezcurrena-Berasain<sup>1</sup>, Juan E. Sánchez-Torres<sup>1</sup>, and Rubén Barajas\*<sup>2</sup>, <sup>1</sup>Universidad Autónoma del Estado de México, FMVZ, Toluca, Estado de México, México, <sup>2</sup>FMVZ-Universidad Autónoma de Sinaloa, Culiacán, Sinaloa, México.

Twenty-eight Rambouillet cross lambs ( $27.85 \pm 2.91$  kg) were used in a 74-d feedlot experiment to evaluate the influence of zilpaterol hydrochloride (Zilmax, Merck Animal Health) and enriched-Cr yeast (BioChrome, Alltech) supplementation on carcass characteristics and meat composition of feedlot lambs. Lambs were weighed and individually allotted. They were randomly assigned to 4 treatments as follows: 1) A 85% concentrate corn-SBM diet, contained 15.5% CP and 1.87 Mcal NEm/kg, (Control); 2) Control with 0.2 mg of zilpaterol hydrochloride/ kg BW during last 28 d in the feedlot (ZH); 3) Control with 0.3 mg of Cr/kg DM (YCr); and 4) Control with ZH and YCr (ZHCr). Lambs were harvested, hot carcass weight measured and carcass dressing obtained. After 24 h chilling, *longissimus dorsi* muscle was cross sectioned at the 12th rib, and then back fat thickness and rib eye area were recorded. *Longissimus dorsi* samples were taken and meat chemical composition determined. Results were analyzed as a completely randomized design in a  $2 \times 2$  factorial arrangement. Hot carcass weight tended to be greater ( $P = 0.10$ ) in ZH ( $20.9$  vs.  $20.3$  kg). Hot carcass dressing ( $46 \pm 0.35\%$ ) was not different across treatments ( $P > 0.20$ ). Back fat thickness was not affected by ZH ( $P > 0.20$ ), but Cr tended ( $P = 0.10$ ) to increase back fat thickness ( $2.02$  vs.  $1.67$  mm). Rib eye area ( $25 \pm 0.68$ ) was similar across treatments ( $P > 0.20$ ). Meat dry matter, ash, and CP content were not altered by treatments ( $P > 0.10$ ). ZH decreased ( $P = 0.03$ ) fat content ( $28.3$  vs.  $39.14$  g/kg). Shear force of the meat from lambs fed ZH treatments was greater ( $P < 0.01$ ) than in meat lambs not fed ZH ( $5.8$  vs.  $2.8$  kg/cm<sup>2</sup>). Cooking losses were similar across treatments ( $P > 0.10$ ). In conclusion, zilpaterol hydrochloride supplementation contributed to greater carcass weight and reduced fat content of meat, but shear force was greater. Yeast-Cr tended to increase back fat thickness without benefit in other carcass or meat characteristics.

**Key Words:** chromium, lamb, zilpaterol

**W472 The nutritional value of confectionary sunflower hulls as forage source for finishing lambs.** Rae-Leigh A. Pederzolli\* and Gregory B. Penner, *University of Saskatchewan, Saskatoon, Saskatchewan, Canada.*

The objective of this study was to determine whether sunflower hulls (SFH) could be used as a partial replacement for barley silage in diets for finishing lambs. Eighteen Canadian Arcott wether lambs were randomly assigned to 1 of 3 treatments: 0SFH (60% barley grain, 30% barley silage and 10% lamb mineral pellet), 7.5SFH (60% barley grain, 22.5% barley silage and 10% lamb mineral pellet) and 15SFH (60% barley grain, 15% barley silage and 10% lamb mineral pellet). The lambs were weighed on 2 consecutive days at the start and end of the 25-d feeding period. Total fecal collection was performed from d 21 to 24, and behavior was monitored on d 25. Samples of feed and refusals were analyzed to evalu-

ate the sorting index. Data were analyzed as a completely randomized design using mixed model of SAS. Dry matter intake was not affected ( $P = 0.32$ ) averaging 1.4 kg/d but ADG increased (quadratic,  $P = 0.03$ ) from 160 g/d for 0SFH to 215 g/d for 7.5SFH followed by a decrease for 15SFH (143 g/d). Sheep fed 15SFH sorted against particles retained on the 8 mm sieve ( $P < 0.01$ ), tended to sort for particles retained on the 1.18 mm sieve ( $P = 0.09$ ) and sorted for particles retained on the pan ( $P < 0.05$ ) showing there was preference for the 19mm, 1.8mm and  $< 1.8$ mm particles in the diet. There was no difference in time spent

ruminating or eating between the treatments ( $P > 0.1$ ) but 15SFH spent less time drinking than lambs fed 0SFH ( $P = 0.03$ ). Apparent total-tract digestibility was not affected by treatments ( $P > 0.1$ ) with DM, CP, NDF, and ADF digestibility averaging 72.7, 71.6, 45.8, and 39.7%, respectively. The data in this study indicate that sunflower hulls can be used as a partial replacement for barley silage without negatively affecting DMI, G:F, feeding behavior and digestibility.

**Key Words:** ADG, lamb, sunflower hulls

## Small Ruminant III

**W473 Effects of the level of fish oil in the diet on lamb performance, ruminal fermentation and leptin gene expression.** Pedro Hernandez<sup>1</sup>, German Mendoza\*<sup>2</sup>, Nallely Sanchez<sup>2</sup>, Jose Martinez<sup>2</sup>, and Fernando Plata<sup>2</sup>, <sup>1</sup>Universidad Autonoma del Estado de Mexico, Amecameca, Mexico, Mexico, <sup>2</sup>Universidad Autonoma Metropolitana Unidad Xochimilco, Mexico, D.F., Mexico.

The objective of this study was to evaluate the effects of fish oil on lamb performance, carcass yield, ruminal fermentation, and leptin gene expression. Thirty-two lambs (24.10 kg initial BW, Katahdin × Pelibuey) were used in a completely randomized design. Dietary treatments were: 1) 0 fish oil and 30% corn (DM), 2) 1% fish oil and 25% corn, fish oil substituted for corn (DM), 3) 2% fish oil and 20.5% corn (DM), and 4) 3% fish oil and 17% corn (DM). The lambs were weighed on consecutive days at the beginning (d 0 and 1) and at the end (d 55 and 56) of the trial. Ruminal fluid samples were collected on d 56 to evaluate the ruminal fermentation pattern. The lambs were slaughtered on d 56; the perirenal adipose tissue samples were collected, and the carcass yields were recorded. The VFA, ammonia N and leptin mRNA expression were not affected ( $P > 0.05$ ) by the dietary treatments. However, the DMI, ADG, final BW and the hot carcass yield (HCY) showed linear or quadratic responses ( $P < 0.05$ ) as fish oil was increased in the ration; the estimated optimal responses were obtained with fish oil levels of 1.16% for the final BW, 1.03% for the HCY, 1.12% for the ADG, and 0.78% for the DMI. Additionally, the feed efficiency and the back fat showed a quadratic response ( $P < 0.05$ ) as fish oil was increased in the diet; the estimated optimal responses were obtained with fish oil levels of 1.4% for feed efficiency and 1.6% for back fat. Increasing fish oil in the diet did not have a negative impact on ruminal fermentation or leptin mRNA expression; however, levels of 1% fish oil improved lamb performance and allowed for a reduction in the amount of grains in the rations.

**Key Words:** finishing lamb, fish oil, leptin mRNA

**W474 Growth rates of females from a Romane sheep breed flock reared in rangelands and effects of an early or late first mating regime on their offspring.** Eliel González-García\*<sup>1</sup>, Didier Foulquié<sup>2</sup>, François Bocquier<sup>1</sup>, Dominique François<sup>3</sup>, and Dominique Hazard<sup>3</sup>, <sup>1</sup>INRA UMR Systèmes d'élevage Méditerranéens et Tropicaux (SELMET), Montpellier Cedex 1, France, <sup>2</sup>INRA UE0321, Domaine de La Fage, Roquefort-sur-Soulzon, France, <sup>3</sup>INRA UMR1388 Génétique, Physiologie et Systèmes d'Élevage (GenPhySE), Castanet-Tolosan Cedex, France.

Benefits in lifetime production of the ewe is often determined by the particular characteristics of the growth during the first stages of their life, including the related decision of the age at first mating. The objective of this work was to characterize the growth rates of the young female (from birth to first mating) from the Romane sheep breed, in a flock reared under the Mediterranean rangelands conditions of France. The effects of 2 contrasted first mating regimens (Early; 7 mo vs. Late; 19 mo old) on the individual growth rate of the ewe lamb and their first offspring were also evaluated. A database was built based on historical data that were extracted from the INRA database (GEEDOC). Data from 1359 females born during the period 2002–2012 were collected, processed and interpreted using SAS (9.3). From the total, 762 and 597 females were submitted to the Early or Late regimen, respectively. The effects of the litter size at their birth (singletons; SING vs. multiple; TWIN) and the age of their dams at birth (primiparous, PRIM vs. multiparous,

MULT) were considered. The growth curves were built for both population (Early;  $39.7 \pm 0.07$  kg and Late;  $50.6 \pm 0.04$  kg BW at first mating) of ewes lambs. Significant interactions of the litter size at birth and the dam's parity were found for BW at birth and BW at weaning. Overall, the ewes lambs born from MULT × SING and PRIM × TWIN dams showed the highest ( $4.7 \pm 0.03$  and  $26.2 \pm 0.15$  kg) and lowest ( $3.3 \pm 0.01$  and  $22.5 \pm 0.09$  kg) BW at birth and weaning, respectively. Even if submitting the ewes lambs from the actual flock to the Late regimen constituted a change on the reproductive management policy of the farm (from 2010), data showed that females submitted to Late corresponded to those having a weaker growth from birth to weaning. When analyzing the growth traits of their first offspring, the first litter size ( $1.8 \pm 0.01$  vs.  $2.1 \pm 0.01$  lambs/lambing) and its total weight ( $6.2 \pm 0.03$  vs.  $7.4 \pm 0.02$  kg) were different ( $P < 0.0001$ ) for ewes submitted to the early or late regimen, respectively. Other related carry over effects during the whole productive life of this females' population and their offspring are currently being analyzed.

**Key Words:** growth rate, Romane female lamb, mating regimen

**W475 Effects of maternal lines and mating systems on susceptibility to parasitism in a pasture-lambing, low-input production system.** Shuna A. Jones\*<sup>1</sup>, Chadwick C. Chase<sup>1</sup>, Michael Heaton<sup>1</sup>, Karen K. Shuck<sup>2</sup>, Kreg Leymaster<sup>1</sup>, and John Keele<sup>1</sup>, <sup>1</sup>US Meat Animal Research Center, Clay Center, NE, <sup>2</sup>Great Plains Veterinary Educational Center, Clay Center, NE.

Internal parasites are a concern for industry as they negatively affect growth, survival, and reproduction. The objective was to evaluate the susceptibility of maternal lines to parasite infection, with the goal of developing genetic tests for this trait in US sheep. Purebred and Texel-sired lambs were born to Polypay, Katahdin, and Easycare (1/2 Romanov, 1/4 Katahdin, 1/4 White Dorper) ewes. Lambs were born on one of four 10-acre pastures, in a low-input system, 391 lambs were weaned at 70 d of age. Variation was accounted for by having all combinations of maternal lines and mating systems in each pasture. At weaning, a fecal sample was collected from individual lambs and the McMaster fecal egg count test (FEC) was performed on each sample. Data from 282 lambs were analyzed. For genetic evaluation, parentage was determined by allelic exclusion with genotype data from an international panel of 109 parentage SNPs. Data,  $\log(\text{FEC} + 1)$ , were analyzed using a mixed animal model with maternal line, mating system, maternal line × mating system, sex, and pasture as fixed effects. Variance components for additive genetic and environmental effects were estimated by REML using the iterative MIVQUE algorithm. Variance component estimates converged at positive values ensuring that the estimates were REML. Heritability was calculated as the additive genetic variance divided by the phenotypic variance after adjustment for fixed effects. The pasture effect for  $\log(\text{FEC} + 1)$  was highly significant ( $P = 9.38 \times 10^{-16}$ ). Mean  $\log(\text{FEC} + 1)$  of the 4 pastures were 2.66, 3.29, 3.90, and  $2.93 \pm 0.12$ , and the corresponding median FEC (back-transformed from log-normal distribution) were 454, 1944, 7927, and 845 eggs/g. However, breed cross and sex were not significant, with P values of 0.99 and 0.49, respectively. The heritability estimate was  $0.28 \pm 0.17$  which is moderate and significantly different from 0 ( $P = 0.0025$ ) based on the restricted likelihood ratio test. These results indicate that the fecal



egg count phenotype for sheep raised in central Nebraska is affected by parasite exposure and host genetics.

**Key Words:** sheep, internal parasite, heritability

**W476 GnRH at moment or 24 hours after hCG treatment upon reproductive outcomes in anestrus goats.** VH Gonzalez-Alvarez<sup>1</sup>, CA Meza-Herrera<sup>2</sup>, AS Alvarado-Espino<sup>1</sup>, JM Gillen-Muñoz<sup>1</sup>, R. Rodriguez-Martinez<sup>1</sup>, G. Arellano-Rodriguez<sup>1</sup>, PA Robles-Trillo<sup>1</sup>, and FG Veliz\*<sup>1</sup>, <sup>1</sup>Antonio Narro Autonomous Agricultural University, Torreon, Coahuila, Mexico, <sup>2</sup>Chapingo Autonomous University, Regional University Unit on Arid Lands, Bermejillo, Durango, Mexico.

The aim of this study was to evaluate if GnRH incorporation to the hCG treatment improves the reproductive parameters of seasonal anestrus goats in northern Mexico (26°N). Adult anovulatory adult (n = 36) were divided into 4 groups (n = 9), according to live weight and body condition score. On May 2, all goats were treated with progesterone (20 mg) intramuscular (im). Twenty-four hours before (d 0), Gc was treated with physiological saline (0.5 mL), Gh with hCG im (100 IU), G3 with hCG (100 IU) + GnRH (8.4 µg), and G4 with hCG (100 UI) + GnRH (8.4 µg) 24 h before hCG administration. The same day, all goats received PGF2α (7.5 mg) intravenously. Estrus was determined twice a day, from d-0 to d-5, using multiracial bucks provided with an apron; females were considered to be in estrus when allowed to be mounted. To determine ovulation occurrence, a transrectal ultrasonographic scanning (US) (Aloka SSD 500, Richmond, BC, Canada) was performed on d-10 in all goat; ovulation considered the presence of well defined corpus luteum at either ovary. Chi-squared test was performed to determine the differences among all groups concerning the reproductive traits measured (SYSTAT, Version 10, 2006), with a 95% interval confidence. The Gc group had no response in estrus, ovulation and gestation (0%, 0/9). The Gh-group had the highest percentages ( $P < 0.05$ ) of both estrus (88.8%, 8/9) and pregnancy (71.4%, 5/9) and, along with G4, the highest ovulating rate (55.5%, 5/9). Results suggest that GnRH inclusion at moment or 24-h before hCG treatment negatively affects the reproductive outcomes of seasonally anestrus goats.

**Key Words:** goat, hCG, GnRH

**W477 Application of 40 mg progesterone prior to the ram effect induces estrus response and ovulatory activity in Dorper sheep.** LM Tejada\*<sup>1</sup>, CA Meza<sup>2</sup>, O. Angel<sup>1</sup>, R. Rodríguez-Martínez<sup>1</sup>, MV Perales<sup>1</sup>, E. Carrillo<sup>3</sup>, G. Arellano<sup>1</sup>, and FG Véliz<sup>1</sup>, <sup>1</sup>Universidad Autónoma Agraria Antonio Narro, Torreon, Coahuila, México, <sup>2</sup>Universidad Autónoma Chapingo Unidad Regional Universitaria de Zonas Áridas, Bermejillo, Durango, México, <sup>3</sup>Instituto Tecnológico de Torreón, Torreón, Coahuila, México.

The aim of this study was to determine whether the application of intramuscular progesterone induces estrus activity in anovulatory Dorper sheep exposed to the male effect. A total of 20 ewes diagnosed as anovulatory, were divided into 3 homogeneous groups in terms of age and body condition. Ewes had no contact with males at least 3 weeks before exposure to the male. While a group of ewes (G1; n = 6) received a single dose of 20 mg i.m. progesterone (day -2 of breeding), a second group (G2; n = 7) received a single dose of 40 mg i.m. of progesterone (day -2 of breeding), and the third group (GE n = 7) were inserted with a intravaginal sponge with 20 mg of progesterone (day -6 breeding) which was removed one day before introduction of the male. This experimental breeding was performed with males previously

treated daily with testosterone (25 mg i.m.) during 15-d before mating to induce an intense sexual activity. Three males (one per group) were rotated every 12 h among experimental groups. Estrus activity was evaluated twice daily 1 h during 15-d. On d-12 after male introduction, an ultrasonographic scanning was performed to determine the percentage of ovulated females. Percentage of ewes depicting estrus and ovarian activity were compared using chi<sup>2</sup>, using the program SYSTAT 12. While no females from the G1 group expressed estrus activity (0/6,  $P < 0.05$ ), all ewes of both the G2 and GE showed estrus activity ( $P > 0.05$ ). In addition, most of the females from all the experimental groups depicted ovulatory activity (G1 5/6; G2 5/7 and GE 7/7  $P > 0.05$ ). The application of 40 mg i.m. of progesterone before the male effect was demonstrated to be as effective as the traditional use of intravaginal sponges to induce estrus response and ovulation in previously anovulatory females exposed to the male effect. Yet, the use of i.m. progesterone is significantly cheaper and easier to apply.

**Key Words:** sheep, progesterone, ram effect

**W478 Evaluation of the use of hCG to promote the induction of reproductive activity in anovulatory Alpine goats.** Alan S. Alvarado-Espino<sup>1</sup>, Cesar A. Meza-Herrera<sup>2</sup>, Vicente H. Gonzalez-Alvarez<sup>1</sup>, Juan M. Guillen-Muñoz<sup>1</sup>, Martha V. Perales-García<sup>1</sup>, Raymundo Rivas-Muñoz<sup>3</sup>, Evaristo Carrillo<sup>3</sup>, and Francisco G. Veliz\*<sup>1</sup>, <sup>1</sup>Universidad Autónoma Agraria Antonio Narro, Torreón, Coahuila, Mexico, <sup>2</sup>Universidad Autónoma Chapingo-Unidad Regional Universitaria en Zonas Áridas, Bermejillo, Durango, Mexico, <sup>3</sup>Instituto Tecnológico de Torreón, Torreón, Coahuila, Mexico.

The aim of this study was to evaluate the efficacy of the use of human chorionic gonadotrophin hormone (hCG) to induce reproductive activity in Alpine goats during seasonal anestrus in northern Mexico (26°N). Adult anovulatory goat (n = 30) were randomly divided into 3 groups according to body condition score ( $1.96 \pm 0.32$ ) and weight ( $37.36 \pm 8.48$ ). On July 23 (day -1), all goats received intra-vulvo-submucosal (ivs) administration of 20 mg of progesterone. On day 0 (D0), goats in the groups G300, (n = 10) received an ivs administration of 300 UI of hCG, the group G100, (n = 10) received 100 UI of hCG while the control group GC, (n = 10) received 0.5 mL of saline solution plus intramuscular administration (im) of 7.5 mg of synthetic prostaglandin. After D0, the onset of estrus behavior was evaluated twice a day for 5 min during the first 5-d after hCG administration by a sexually active buck. Goats were scanned by transrectal ultrasound (7.5 MHz probe) on d-45 after breeding for detection of pregnancy. Goats depicting estrus behavior were similar between groups (G100: 100% and G300: 90%,  $P > 0.05$ ), with not estrus behavior in the GC-goats. The onset of estrus occurred earlier in G100 than G300 ( $54 \pm 6.32$  h vs  $76 \pm 30$  h, respectively;  $P < 0.05$ ). No differences were found ( $P > 0.05$ ) in pregnancy rate between G100 (100%) and G300 (70%) groups. The kidding rate and prolificacy were similar ( $P > 0.05$ ) in the G100 and G300 ( $80$  vs  $70\%$  and  $1.5 \pm 0.56$  vs  $1.57 \pm 0.53$ , respectively). These results indicate that administration of 100 UI of hCG could be used without a reduction in both estrus behavior and reproductive outcomes in anestrus Alpine goats at 26° N.

**Key Words:** hCG, induction, estrus

**W479 Effects of mixing two different tannin-containing diets to evaluate rumen fermentation and microbial population changes in goats.** D. Perkins\*<sup>1</sup>, B. R. Min<sup>1</sup>, C. Wright<sup>1</sup>, N. Gurung<sup>1</sup>, T. Terrill<sup>2</sup>, J.-S. Eun<sup>3</sup>, W. H. McElhenney<sup>1</sup>, R. Shange<sup>1</sup>, and Y. Uyeno<sup>4</sup>, <sup>1</sup>Tuskegee University, Tuskegee, AL, <sup>2</sup>Fort-Valley

State University, Fort Valley, GA, <sup>3</sup>Utah State University, Logan, UT, <sup>4</sup>Shinshu University, Minamiminowa, Japan.

Study was performed to investigate the influence of different sources of condensed tannins (CT) supplementations on ruminal fermentation and rumen microbial diversity changes of goats. The objective of this study was to determine whether the tannin-containing sericea lespedeza leaf pellet (SL, *Lespedeza cuneata*; 13.6% CT), ground pine bark (PB; *Pinus*; 16.3% CT), or its combination (SL+PB) would have effects on rumen fermentation and microbial diversity in meat goats. Twenty-four Kiko-crossbreed intact male goats (*Capra hircus*; BW = 38.6 ± 2.7 kg) were randomly assigned to 4 treatments (n = 6): 1) 30% bermudagrass hay and 70% grain mix, 2) 30% PB and 70% grain mix, 3) 30% SLP and 70% grain mix, and 4) 15% PB, 15% SL pellet, and 70% grain mix. Each treatment diet (30% DM) was mixed with remainder of each diet (70% DM) contained 70% commercial sweet feed and 30% alfalfa pellets. Feed intake and BW were monitored every 2 wks for 42 d. Rumen fluid samples were taken using a stomach tube at the d 0, 20, and 42 for microbial DNA analysis. Overall, there were no differences in DMI and BW gain among treatments. However, goats supplemented with PB and SLP+PB diets decreased ( $P < 0.05$ ) concentrations of acetate, isobutyrate, and isovalerate compared with those in the control and SLP diets. There were no differences in rumen microbial kingdom (fungi, bacteria, and archaea) and phylum diversity. However, green algae population in rumen fluid was greater ( $P < 0.01$ ) for control diet than for other treatments. In this study, *Bacteroides* (30 to 55%) and *Firmicutes* (30 to 47%) were the major bacterial phylum, while *Prevotella* spp. was the most predominant rumen bacterial species in the percentage of 22.1, 42.2, 28.9, and 23.9 for control, PB, SLP, and mixed diets, respectively. The population of rumen bacterial species in PB-supplemented group was greater for *Bacteroides* spp. ( $P < 0.02$ ), *Marinifilum* spp. ( $P < 0.04$ ), and *Oribacterium* spp. ( $P < 0.03$ ) compared with other treatment groups. However, population of *Fibrobacter succinogenes* was greater ( $P < 0.05$ ) for SLP than for other treatments. Supplementing tannins in goat diets such as PB and SLP diets has a potential to modify rumen bacterial population.

**Key Words:** goat, rumen microbial diversity, tannins

**W480 Reproductive efficiency of anovulatory goats through bucks treated with testosterone and (or) estrogenized females in northern México.** Juan M. Guillén-Muñoz<sup>1</sup>, César A. Meza-Herrera<sup>2</sup>, Rafael Rodríguez-Martínez<sup>1</sup>, Pedro A. Robles-Trillo<sup>1</sup>, Carlos Leyva-Orasma<sup>1</sup>, Evaristo Carrillo<sup>3</sup>, Francisco Gerardo Veliz<sup>1</sup>, and Gerardo Arellano-Rodríguez\*<sup>1</sup>, <sup>1</sup>Universidad Autónoma Agraria Antonio Narro, Torreón, Coahuila, México, <sup>2</sup>Universidad Autónoma de Chapingo, Bermejillo, Durango, México, <sup>3</sup>Instituto Tecnológico de Torreón, Torreón, Coahuila, México.

The aim of this study was to evaluate the effect of treated males with testosterone and (or) estrogenized female upon reproductive efficiency in anovulatory goats in northern México (26°N). Multiparous mix-dairy breed anestrus goats (n = 80) were randomly assigned to one 4 treatment groups: (GT; n = 20) exposed to testosterone-treated bucks (25 mg, 3 d x 3 weeks); (GTH; n = 20) joined to testosterone-treated bucks and expose to estrogenized females x 3-d (pen of bucks were separated of females by a metal mesh); (GH; n = 20) exposed to estrogen-treated females (kept in a pen aside to another pen with 2 estrogenized females x 2-wk) and (GC; n = 20) exposed to untreated bucks. All goats in this study received an I.M. injection of 25 mg progesterone, 24-h before exposure to males. Response variables considered estrus response during 14-d, ovulatory response and pregnancy rate of goats. Percentages were analyzed through a chi-squared test (SYSTAT 12). Results from the

treatment groups (GC; GT; GTH; GH) give important evidence that both treatments: testosterone-treated bucks and (or) exposure of males to estrogenized females were equally effective to promote not only estrus induction (75–90%) but also both an increased ovulatory response (70–90%) as well as pregnancy rates (70–90%;  $P < 0.05$ ) respectively, in dairy mix-breed anovulatory goats under subtropical conditions.

**Key Words:** buck, testosterone, reproductive efficiency

**W481 Impact of adequate or reduced mineral supplementation on rumen function and growth in lambs.** Abigail M. Temple\*<sup>1</sup>, Gbenga A. Ayangbile<sup>1</sup>, Dustin R. Vandermyde<sup>1</sup>, Calvin R. Vandermyde<sup>2</sup>, and Dan J. Schauff<sup>1</sup>, <sup>1</sup>Agri-King Inc., Fulton, IL, <sup>2</sup>Morrison Veterinary Clinic, Morrison, IL.

Mineral supplementation is vital for optimum production and efficiency in livestock, but it is an added feed cost. We hypothesized a reduction in supplemental minerals in a ration may have minimal effect on rumen function and performance of lambs fed Ru-Mend, a product designed to enhance nutrient absorption. Sixty Katahdin wethers (average BW 13 ± 3 kg) 55 d of age were fed an ad lib corn silage plus alfalfa haylage-based diet with Ru-Mend, and either adequate (FULL) or reduced (RED) mineral supplements for 52 d. The total mixed rations were analyzed and RED diet found to be 30% lower in Co, Cu, Mn and Zn, 15% in S, 10% in Ca and Mg, and 5% in P as compared with the FULL diet. Diets were balanced for K, Na and Cl, but Na was 10% lower in the RED diet. Lambs were penned in groups of 15 with 2 pens per treatment. ADG was calculated for each animal from their initial and 44 d weight. DMI was calculated from pen daily intake. Lambs were euthanized on d 52, and tissues collected for analysis. Data were analyzed by ANOVA completely randomized design. Pearson correlations were used for comparison. Final rumen content pH was between 5.2 and 6.0 for all lambs. Lambs fed the FULL diet had higher DMI (640.71 vs. 513.13 g/d;  $P < 0.0001$ ) and ADG (102.15 vs. 72.17 g/d;  $P = 0.003$ ) than RED lambs. FULL lambs' ADG correlated with lower rumen ammonia ( $P = 0.02$ ). The rumen fluid contents of FULL-fed lambs had higher total amino acids ( $P = 0.004$ ) and essential amino acids [arginine, isoleucine, lysine, phenylalanine, threonine and valine] ( $P \leq 0.03$ ) than RED-fed lambs. The lower ammonia and higher amino acids may indicate positive effect of minerals due to increased DMI and reduced protein degradation. Rumen osmolality and acetate were higher ( $P = 0.05$ ) for FULL lambs. RED lambs had lower rumen and ileum contents Co, Cu and Mn ( $P \leq 0.02$ ) and rumen Fe, Mg, P and S ( $P \leq 0.03$ ). Lower ADG of RED lambs correlated with reduced ileum K, P and S ( $P \leq 0.05$ ) and numerically less rumen Mg, Ca and Co ( $P \leq 0.10$ ). This may indicate lambs fed the RED diet did not benefit from Ru-Mend, thus minerals needed for energy metabolism, growth and weight gain were unavailable.

**Key Words:** mineral, amino acid, rumen

**W482 Tissue mineral deposition in growing lambs fed adequate or reduced levels of mineral supplements.** Abigail M. Temple<sup>1</sup>, Gbenga A. Ayangbile\*<sup>1</sup>, Dustin R. Vandermyde<sup>1</sup>, Calvin R. Vandermyde<sup>2</sup>, Dan J. Schauff<sup>1</sup>, and Jeff G. Horst<sup>1</sup>, <sup>1</sup>Agri-King Inc., Fulton, IL, <sup>2</sup>Morrison Veterinary Clinic, Morrison, IL.

Ru-Mend is a supplemental product designed to enhance nutrient absorption in the ruminant. Previous research shows animals fed Ru-Mend had increased mineral uptake and blood glucose versus the control. The objective of this study was to determine whether Ru-Mend supplementation would be a cost-effective way to improve mineral uptake and prevent performance issues in ruminants fed inadequate minerals by examining

blood metabolites and tissue mineral deposition. Sixty Katahdin wethers (average BW  $13 \pm 3$  kg) approximately 55 d of age were split into groups and fed an ad lib corn silage plus alfalfa haylage-based diet with Ru-Mend, and either adequate (FULL) or reduced (RED) minerals for 52 d. The total mixed rations (TMR) were analyzed and RED diet found to be 30% lower in Co, Cu, Mn and Zn, 15% in S, 10% in Ca and Mg, and 5% in P as compared with the FULL diet. Diets were balanced for K, Na and Cl, but there was a 10% reduction in Na for the RED TMR. Lambs were penned in groups of 15 with 2 pens per treatment. Jugular blood samples were analyzed for each animal during wk 0, 2, 5, and 7. Animals were euthanized on d 52, tissues collected and prepared for analysis. Data were analyzed by ANOVA completely randomized design. Lambs on FULL supplement had higher blood hemoglobin and hematocrit ( $P = 0.02$ ) and a tendency for higher glucose ( $P = 0.09$ ). However, lambs on RED supplement had higher blood Cu ( $P = 0.0001$ ) and a tendency for higher Fe ( $P = 0.06$ ). Lambs fed RED treatment had significantly lower liver Ca, Cu, Mn and S ( $P \leq 0.03$ ) and a tendency for lower Zn ( $P = 0.06$ ). In addition, RED lambs had lower duodenum tissue Cu and Mn ( $P < 0.03$ ), but higher spleen Ca, Cu, K, Mg, P and Zn ( $P < 0.04$ ). This may suggest the spleen acted as an organ of sequestration. FULL TMR-fed lambs had higher hoof covering minerals of Co, Mn and Na ( $P \leq 0.01$ ), whereas hoof bones were higher in Ca, Mn and P ( $P \leq 0.04$ ). These results may indicate lambs receiving RED supplementation did not benefit from Ru-Mend as observed by lower blood hemoglobin and hematocrit as well as lower liver, duodenum, and hoof mineral levels compared with FULL-fed lambs.

**Key Words:** mineral, tissue, glucose

**W483 Nutrient digestibility in growing lambs fed adequate or reduced levels of mineral supplements.** Abrigail M. Temple, Gbenga A. Ayangbile, Dustin R. Vandermyde, Dan J. Schauff\*, and David A. Spangler, *Agri-King Inc., Fulton, IL.*

Minerals can be a costly addition to a livestock ration, and farmers may choose to reduce mineral supplementation to reduce feed costs. Ru-Mend is a product designed to enhance nutrient absorption in the ruminant. Previous research demonstrated animals fed Ru-Mend had improved rumen osmolality, mineral digestibility and blood glucose versus the control. We hypothesized Ru-Mend supplementation could be a cost-effective way to improve mineral uptake and prevent performance issues in ruminants fed inadequate mineral levels. The objective of this study was to observe the effect of Ru-Mend on nutrient digestibility in ruminants fed reduced levels of supplemental minerals. Sixty Katahdin wethers (average BW  $13 \pm 3$  kg) approximately 55 d of age were split into groups and fed an ad libitum corn silage plus alfalfa haylage-based diet with Ru-Mend, and either adequate (FULL) or reduced (RED) minerals for 52 d. The total mixed rations (TMR) were analyzed and RED diet observed to be lower by 30% in Co, Cu, Mn and Zn, 15% in S, 10% in Ca and Mg, and 5% in P compared with the FULL diet. Diets were balanced for K, Na and Cl, but there was a 10% reduction in Na for the RED TMR. Lambs were penned in groups of 15 with 2 pens per TMR treatment. For determination of DMI, digestibility, and nutrient utilization, 9 animals from each treatment were placed in digestibility crates for 5 d sample collection of orts, fecal and urine. ADG was calculated for each animal from their initial and 44 d body weight (BW). Data were analyzed by ANOVA completely randomized design. Lambs fed FULL treatment had significantly higher DMI (427.49 vs. 332.68 g/d;  $P < 0.0001$ ) and ADG (102.15 vs. 72.17 g/d;  $P = 0.003$ ) compared with RED. Also, FULL TMR-fed lambs had significantly higher digestibility of ADF, CP and NDF ( $P \leq 0.03$ ) and numerically higher DMD ( $P = 0.14$ ). In addition, FULL lambs had significantly improved digestibility

of Ca, Cu, Fe, K and S ( $P < 0.04$ ) compared with RED. The data suggests lambs did not adequately benefit from Ru-Mend in the analyzed nutrient digestibility parameters or BW gain when the diet's mineral level was insufficient, as in the RED diet.

**Key Words:** mineral, digestibility

**W484 Adipocyte differentiation-related protein promotes lipid accumulation in goat mammary epithelial cells.** Hengbo Shi, Kang Yu, and Jun Luo\*, *College of Animal Science and Technology, Northwest A&F University, Yangling, Shaanxi, China.*

Milk fat originates from the secretion of lipid droplets (LDs) synthesized within mammary epithelial cells. Adipocyte differentiation-related protein (ADRP; also known as PLIN2) is an LD binding protein that is crucial for synthesis of mature LD. The hypothesis was ADRP regulate LD production and metabolism in goat mammary epithelial cells (GMEC), thus, play a role in determining milk fat content. The objective of the present study was to investigate the function of ADRP in milk fat metabolism by ADRP overexpression and knockdown in GMEC using an adenovirus system. The expression of ADRP mRNA was determined by RT-PCR, and concentration of triacylglycerol (TG) extracted from GMEC was quantified on a micro-titer plate reader. Immunocytochemical staining revealed that ADRP localized to the surface of LDs. Supplementation with oleic acid (OA) enhanced its localization on the LDs surface and lipid accumulation. Overexpression of ADRP increased lipid accumulation and the concentration of triacylglycerol in GMEC. In contrast, morphological examination revealed that knockdown of ADRP decreased lipid accumulation even when OA was supplemented. This response was confirmed by the reduction in mass of cellular TG when ADRP was knockdown. The fact that knockdown of ADRP did not completely eliminate lipid accumulation at a morphological level in GMEC without OA suggested that some other compensatory factors may also aid in the process of LD formation. ADRP reversed the decrease of LD accumulation induced by Adipose triglyceride lipase (ATGL). This indicated that ADRP promote TG stability within LD by preventing access to ATGL. Collectively, these data provide direct in vitro evidence that ADRP play a key role in LD formation and stability in GMEC. Further experiments need to explore the mechanisms for enlargement of LD via ADRP activity in GMEC.

**Key Words:** milk lipid droplet, Adipocyte differentiation-related protein (ADRP), goat mammary epithelial cells

**W485 Activation of liver X receptor  $\alpha$  and SREBP-1 promotes fatty acid synthesis in goat mammary epithelial cells.** Huifeng Xu and Jun Luo\*, *College of Animal Science and Technology, Northwest A&F University, Yangling, Shaanxi, China.*

Liver X receptor  $\alpha$  (LXR $\alpha$ ) and sterol regulatory element binding protein-1 (SREBP-1) are key transcription factors regulating lipid synthesis in mammals, but the joint function of LXR $\alpha$  activation and SREBP-1 maturation in milk fatty acid synthesis of lactating goat remains unclear. The objective of the present study was to determine the regulating function of LXR $\alpha$  and SREBP-1 in fatty acid metabolism of goat mammary gland using RT-qPCR. Goat mammary epithelial cells (GMEC) were treated by T0901317, a synthetic agonist of LXR $\alpha$ , to explore the regulatory function of LXR $\alpha$ -SREBP-1 pathway in goat mammary gland. The results showed that there was no significant change in the mRNA level of LXR $\alpha$  following treatment with different concentrations of T0901317, but a dose-dependent increase were observed for levels of mRNA, protein and relative luciferase activity of SREBP-1.



Immunofluorescence results showed a clear accumulation of mature *SREBP-1* in cell nucleus by the addition of T0901317. Activation of *LXRα-SREBP-1* pathway increased significantly the mRNA expression of genes related to de novo fatty acid synthesis, fatty acid desaturation, elongation, transportation, and TG synthesis including *FASN*, *ACCA*, *SCD1*, *IHD1*, *ACSS2*, *Elovl6*, *FABP3*, *DGAT1*, *DGAT2*, *AGPAT6*, *LPIN1* and *PLIN2* ( $P < 0.01$ ). One micro Mole T0901317 treatment caused a 20% increase of total content of cellular TG compared with the control ( $P < 0.01$ ). Furthermore, addition of T0901317 remarkably increased the proportion of C16:1 and C18:1, while decreased that of C16:0 and C18:0. In conclusion, the *LXRα* regulates the expression and proteolytic maturation of *SREBP-1* in GMEC, which implied that the crosstalk between *LXRα* and *SREBP-1* may play an important role in the transcriptional regulation of de novo fatty acid synthesis and TG synthesis in goat mammary gland.

**Key Words:** *LXRα*, fatty acid synthesis, goat mammary epithelial cell

**W486 Evaluation of different doses of intramuscular progesterone to induce reproductive activity in anestrus goats.**

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The use of different doses of intramuscular progesterone for sexual activity induction in anestrus goats (26°N) was evaluated. During late May, mix-breed anovulatory adult goats (n = 15) were divided in 2 homogeneous groups regarding body condition score and body weight; 4 mix-breed bucks treated with testosterone were also used. Does grazed from 1000h to 1600h. Group 1 (10 mg; n = 7) received 10 mg i.m. progesterone + 200 IU i.m. eCG, while group 2 (20 mg; n = 8) received 20 mg i.m. progesterone + 200 IU i.m. eCG. Does were penned and kept there during 5-d and were fed with alfalfa hay ad libitum, 200 g commercial mix (14% CP, per animal/d) and trace mineral salt blocks and water ad libitum. Estrus activity was evaluated by introducing a male in each experimental group 15 min (morning) 15 min (noon) and 15 min (afternoon). Females depicting signs of heat were moved to a different pen with 2 males. A transectal ultrasonographic scanning (TUS) was performed to detect ovulation (10 d) and pregnancy (45 d) after estrus detection. The percentage of females depicting estrus, ovulation and pregnancy were compared with a chi2. Reproductive response from both experimental groups is presented on Table 1. Results obtained allow us to conclude that administration of intramuscular progesterone, at 10 or 20 mg, induces sexual activity of goats during the normal anestrus season.

**Table 1 (Abstr. W486).** Reproductive response of mixed-breed anestrus goats receiving either 10 or 20 mg i.m. progesterone

Sexual response	Group	
	10 mg	20 mg
Estrus (no.)	6/7 <sup>a</sup>	7/8 <sup>a</sup>
Ovulation (no.)	7/7 <sup>a</sup>	8/8 <sup>a</sup>
Gestation (no.)	5/6 <sup>a</sup>	5/8 <sup>a</sup>

<sup>a,b</sup>Different superscript within variables denote differences  $P < 0.05$ .

**Key Words:** progesterone, anestrus, goat

**W487 Induction of sexual behavior in Dorper rams treated with glutamate and/or testosterone during the natural sexual resting season at 26°N.**

Ma. Guadalupe Calderón-Leyva<sup>\*1</sup>, Cesar A. Meza-Herrera<sup>2</sup>, Oscar Ángel-García<sup>1</sup>, Juan Ramón Luna-Orozco<sup>3</sup>, Raymundo Rivas-Muñoz<sup>4</sup>, Gerardo Arellano-Rodríguez<sup>1</sup>, F. Gerardo Véliz-Deras<sup>1</sup>, and Rafael Rodríguez-Martínez<sup>1</sup>, <sup>1</sup>Universidad Autónoma Agraria Antonio Narro, Torreón, Coahuila, México, <sup>2</sup>Universidad Autónoma Chapingo, Unidad Regional Universitaria de Zonas Áridas, Bermejillo, Durango, México, <sup>3</sup>Centro de Bachillerato Tecnológico Agropecuario No. 1, Torreón, Coahuila, México, <sup>4</sup>Instituto Tecnológico de Torreón, Torreón, Coahuila, México.

The study was performed in northern Mexico (25°64' N, 103°26' LW) to evaluate the effectiveness of different treatments to stimulate sexual behavior of rams during spring. Dorper rams (n = 20; 81 ± 0.32 kg BW; 3.8 ± 0.16 BCS) were divided in 4 homogeneous groups in terms of body weight (BW), body condition score (BCS) and odor, and were randomly assigned to 4 experimental groups: Glutamate Group (GG, n = 5; treated 7 mg kg<sup>-1</sup> BW of L-Glutamate i.m.), Testosterone Group (TG, n = 5; treated with 25 mg of testosterone i.m.), Glutamate + Testosterone Group, (GTG, n = 5; receiving 7 mg kg<sup>-1</sup> BW of L-Glutamate + 25 mg of testosterone i.m.) and Control Group (CONT, n = 5; which received 0.5 mL of saline i.m.). Odor score of rams was evaluated every 2 weeks by smelling the base of the horns at a distance of 15 cm and using a 0–3 scale. After 30 d of treatments, 2 males were randomly selected from each treated group and placed in one of 4 groups of anovulatory ewes (n = 14 each; 42 ± 2.36 kg BW; 3.04 ± 0.22 BCS) to evaluate the male sexual behavior, considering the appetitive sexual behavior (ASB), the consummatory sexual behavior (CSB) and isolation behavior (ISL), during 2-h daily x 2 d. While odor data were evaluated by ANOVA, sexual behavior considered X<sup>2</sup> (SYSTAT 12.0). No differences ( $P > 0.05$ ) regarding odor score was observed in the first sample among experimental groups. Yet, at the end of the experimental period, the GTG showed the highest values (1.5 ± 0.29,  $P < 0.05$ ) compared with groups GG and TG (0.75 ± 0.34 and 0.65 ± 0.75, respectively;  $P > 0.05$ ). In the ASB, the GTG depicted the highest percentage of performed behaviors regarding GG, TG and CONT (43 vs 29, 11 and 18% respectively); only statistical differences ( $P < 0.05$ ) occurred with respect to TG and CONT. In the CSB-phase, GG and GTG (37%) and the CONT (23%) did not differ ( $P > 0.05$ ), yet, the TG depicted the lowest CSB performance ( $P < 0.05$ ). In the case of ISL, the CONT accumulated the highest percentage (62%;  $P < 0.05$ ) regarding GTG, GG and TG (25, 13 and 0%, respectively). Results demonstrate that Dorper rams treated with glutamate + testosterone promoted not only an increased sexual behavior but also an augmented odor score during spring.

**Key Words:** rams, glutamate, testosterone

**W488 Reproductive outcomes in nulliparous ewes exposed to dorper rams treated with glutamate and/or testosterone during increased photoperiods.**

Ma. Guadalupe Calderon-Leyva<sup>1</sup>, Cesar A. Meza Herrera<sup>2</sup>, Oscar Angel-Garcia<sup>1</sup>, Juan Ramon Luna-Orozco<sup>3</sup>, Raymundo Rivas-Muñoz<sup>4</sup>, Gerardo Arellano-Rodríguez<sup>1</sup>, F. Gerardo Veliz-Deras<sup>1</sup>, and Rafael Rodriguez-Martinez<sup>\*1</sup>, <sup>1</sup>Universidad Autonoma Agraria Antonio Narr, Unidad Laguna, Torreon, Coahuila, Mexico, <sup>2</sup>Universidad Autonoma Chapingo, Unidad Regional de Zonas Aridas, Bermejillo, Durango, Mexico, <sup>3</sup>Centro de Bachillerato Tecnológico Agropecuario No 1, Torreon, Coahuila, Mexico., <sup>4</sup>Instituto Tecnológico de Torreon, Torreon, Coahuila, Mexico.

The aim of this study was to evaluate the ability to induce sexual activity in nulliparous Droper ewes during the anestrus season throughout the male effect in northern Mexico (26° N). Dorper rams (n = 8; 83 ± 3.6

kg BW) and 55 nulliparous anovulatory ewes ( $42 \pm 2.36$  kg BW) were used. Before to be in contact with males, ewes received 60 mg i.m. of progesterone to avoid short cycles and expect ovulation once estrus activity occurred, then ewes were divided in 4 homogeneous groups in terms of BW and BCS. Prior exposure to females, males were randomly divided in 4 experimental groups and treated during 30-d with: Glutamate (GG,  $n = 14$  ewes; 2 males treated with  $7 \text{ mg kg}^{-1}$  BW of Glutamate, i.m.), Testosterone (TG,  $n = 14$  ewes; 2 males treated with 25 mg i.m. of testosterone), Glutamate + Testosterone (GTG,  $n = 14$  ewes; 2 males received  $7 \text{ mg kg}^{-1}$  BW of L-Glutamate and 25 mg of testosterone) and Control (CONT,  $n = 13$ ; 2 males received 0.5 mL of saline). Since the first day of contact with males, estrus response and interval to estrus onset (h) were registered. Thereafter, embryo implantation rate was determined on d-45 from male exposure throughout transrectal ultrasound scanning. Data regarding the onset of estrus were analyzed using *t*-test, while the estrus response and implantation rate considered  $\chi^2$  (SYSTAT 12.0). Ewes exposed to rams treated with glutamate or glutamate + testosterone showed the best reproductive outcomes ( $P < 0.5$ ), depicting, in average, the shortest interval to estrus (37.7 h) while the largest implantation (100%) and estrous response (100%) rates. Therefore, glutamate administration in rams emerges as an interesting option to improve the male effect. Nonetheless, it is further required to elucidate the role played by testosterone in the GTG group, because ewes exposed to rams treated with testosterone alone (TG group) had the lowest reproductive outcomes.

**Key Words:** nulliparous ewe, male effect, glutamate

**W489 Quality parameters of goat meat as influenced by dietary condensed tannins from pine bark.** Jolethia O. Jones<sup>\*1</sup>, Jung Hoon Lee<sup>1</sup>, Byeng R. Min<sup>2</sup>, Govind Kannan<sup>1</sup>, and Brou Kouakou<sup>1</sup>, <sup>1</sup>Fort Valley State University, Fort Valley, GA, <sup>2</sup>Tuskegee University, Tuskegee, AL.

This study was conducted to evaluate the quality characteristics of goat meat (chevon) from meat goats fed ground pine bark (PB, *Pinus* spp.) containing up to 13% condensed tannins (CT) on a DM basis. Twenty-four intact male Kiko goats (8 mo of age;  $\text{BW} = 39.7 \pm 2.55$  kg) were randomly assigned to pastures for a grazing trial. Goats were grazed in a winter rye grass-dominant pasture, and supplemented with either bermudagrass hay (BG) or PB pellet ( $n = 12$  goats/supplementation). Each supplement consisted of alfalfa pellet, molasses, and mineral mixtures with either BG or PB powder, provided at 1.5% of BW at individual feeding stations. The dietary supplements were isocaloric and isonitrogenous. After 50 d grazing, goats were slaughtered and kept at 2°C for 24 h. *Longissimus muscle* (LM) pH was measured from individual carcasses and each carcass was fabricated to obtained 2.5-cm thick loin chops for meat quality analysis. All data were analyzed as a completely randomized design. The LM pH of goats was not significantly different ( $P > 0.05$ ) between BG-hay and PB-pellet supplementations (5.65 and  $5.70 \pm 0.048$ , respectively). No significant differences were found in the CIE L\* (lightness) and b\* (yellowness) values of loin chops from goats supplemented either BG-hay or PB-pellet. However, the CIE a\* (redness) values of chops from goats fed BG-hay were higher ( $P < 0.05$ ) than those from goats fed PB-pellet. No differences ( $P > 0.05$ ) were found in the proximate composition of LM from the loin chops of goats fed the 2 different supplements. Furthermore, the thiobarbituric acid reactive substances (TBARS) values of LM from the loin chops were not significantly different between the 2 different supplements. No significant differences were found in the Warner-Bratzler shear values ( $3.90$  vs  $3.86 \pm 0.13$  kg) and cooking losses ( $20.1$  vs  $22.4 \pm 1.12\%$ ) of loin chops from goats supplemented either BG-hay or PB-pellet.

The results indicate that supplementing with pine bark pellet did not change the quality of chevon but it might influence the fresh red meat color of chevon.

**Key Words:** quality, goat meat, pine bark

**W490 Fatty acid composition of different fat depots from hair and wool x hair sheep supplemented with soy hull on pasture.**

Beruk B. Lemma<sup>1</sup>, Jung Hoon Lee<sup>\*1</sup>, Stephan A. Wildeus<sup>2</sup>, Brou Kouakou<sup>1</sup>, and Govind Kannan<sup>1</sup>, <sup>1</sup>Fort Valley State University, Fort Valley, GA, <sup>2</sup>Virginia State University, Petersburg, VA.

Terminal sire mating and supplementation were used to improve growth rate and muscling in hair sheep breeds. However, their effect on the fatty acid profile of different fat depots in lambs has not been reported. Forty-seven lambs (5-mo old), 23 purebred hair sheep (Barbados Blackbelly or BB,  $\text{BW} = 16.2 \pm 1.9$  kg; St.Croix or SC,  $\text{BW} = 19.5 \pm 2.0$  kg) and 24 crossbred wool (Dorset, D) x hair (BB;  $\text{BW} = 21.7 \pm 2.3$  kg or SC;  $\text{BW} = 21.7 \pm 2.9$  kg) of both sexes were rotationally grazed on predominately tall fescue with or without soy hull supplementation during summer. Soy hull was provided at 2.0% of BW at individual feeding stations. After 90 d of grazing, lambs were harvested, and intramuscular, subcutaneous and kidney fats were obtained from each carcass. Total lipids from each fat depot sample were extracted by the chloroform-methanol method. Extracted lipids were prepared for the fatty acid methyl esters (FAME) and then analyzed by a gas chromatography. All data were analyzed as a completely randomized design with breed type (pure- or cross-bred), supplement (with or without soy hull), and sex (male or female) as main effects. The fatty acid profiles of different fat depots from experimental lambs were significantly influenced by supplementation and sex. Compared with lambs fed supplement, pasture-only fed lambs had higher ( $P < 0.01$ ) concentrations of linoleic (C18:2n6, 6.83 vs 4.96%), arachidonic (C20:4n6, 4.08 vs 2.27%), docosapentaenoic (C22:5n3, 1.41 vs 0.73%) and docosahexaenoic (C22:6n3, 0.45 vs 0.25%) acids in intramuscular fat; a higher ( $P < 0.01$ ) concentration of stearic (C18:0, 26.0 vs 22.4%) acid, but lower ( $P < 0.01$ ) concentrations of C16:0 (19.4 vs 22.2%) and C18:1n9 (31.4 vs 37.2%) in subcutaneous fat; and a higher ( $P < 0.01$ ) concentration of C18:1n9 (38.7 vs 33.1%), but lower ( $P < 0.01$ ) concentrations of C18:0 (23.7 vs 29.4%) and C18:2n6 (1.13 vs 1.76%) in kidney fat. The results indicate that fresh lamb from pasture only fed-lambs might have healthier fatty acid profiles compared with that from lambs supplemented with soy hull, regardless of breed types.

**Key Words:** wool and hair sheep, fat depot, soy hull

**W491 Relationship among somatic measurements, body condition score, live weight and internal fat depots in Sarda ewes and Saanen goats in early lactation.** Leonardo S. Knupp<sup>1</sup>, Sheila N. R. Knupp<sup>1</sup>, Mauro Ledda<sup>2</sup>, Davide Rondina<sup>1</sup>, Marco Acciaro<sup>3</sup>, Carla Manca<sup>3</sup>, Gesumino Spanu<sup>1</sup>, Mondina F. Lunesu<sup>1</sup>, and Antonello Cannas<sup>\*1</sup>, <sup>1</sup>Dipartimento di Agraria, University of Sassari, Sardinia, Italy, <sup>2</sup>Dipartimento di Medicina Veterinaria, University of Sassari, Sardinia, Italy, <sup>3</sup>Agris Sardegna, Dipartimento di Ricerca nelle Produzioni Animali, Sardinia, Italy.

Very little information is available on the relationship between BCS and LW in many dairy sheep and goat breeds. For these reasons, this work aimed to study in Sarda dairy ewes and in Saanen dairy goats: i) the relationship between BCS and LW; ii) the relationship between BCS, LW and the amount of internal body fat and its distribution in the abdominal cavity. Eight Sarda ewes (from 38.0 to 53.5 kg of LW and from 2.5 to 3.0 of BCS) and 8 Saanen goats (from 44.0 to 67.5 kg

of LW and from 2.25 to 3.0 of BCS) in the second month of lactation were used in this study. Nine different somatic measurements were taken on each animal. At slaughter, omental, mesenteric, kidney and other internal fat were separated and weighed. LW linearly increased as BCS increased ( $R^2 = 0.85$ ;  $P < 0.001$ ) in Sarda ewes. BCS was a better predictor of LW ( $R^2 = 0.92$ ;  $P < 0.001$ ) than somatic body measurements. For each unitary change of BCS, the LW increased 23.45 kg. In contrast, in Saanen goats the relationship between LW and BCS was low ( $R^2 = 0.25$ ) and not significant. In this species, chest of girth was instead a very good predictor of LW ( $R^2 = 0.91$ ). Kidney, omental, mesenteric and others fat weighted, respectively, 0.2 kg, 0.9 kg, 0.4 kg, and 0.1 kg and represented 12.4%, 53.5%, 24.3%, and 9.7% of the total visceral fat depots in Sarda ewes. In Saanen goat they weighted, respectively, 0.3 kg, 1.0 kg, 0.7 kg, and 0.2 kg and represented 13.7%, 41.9%, 36.2%, and 8.2% of the total visceral fat depots. The weight of the total internal fat in relation of the EBW was very similar in ewes and in goats (0.4% and 0.5%, respectively). The kidney fat was highly correlated with the total visceral fat in sheep and goats ( $R^2 = 0.92$  and 0.87, respectively). BCS and LW predicted equally well total internal fat (in kg) ( $R^2 = 0.65$ ) in Sarda ewes, while in Saanen goats the BCS was a slightly better predictor ( $R^2 = 0.63$ ) than LW ( $R^2 = 0.59$ ). In conclusion, BCS was the best predictor of total internal fat in both species.

**Key Words:** body condition score, somatic measurement, visceral fat

**W492 Assessment of RNA stability within six ovine tissues postmortem.** Fiona M. McGovern<sup>\*1</sup>, Tommy M. Boland<sup>1</sup>, Francis P. Champion<sup>1</sup>, Marion T. Ryan<sup>1</sup>, and Torres Sweeney<sup>2</sup>, <sup>1</sup>*School of Agriculture and Food Science, University College Dublin, Dublin, Ireland*, <sup>2</sup>*School of Veterinary Medicine, University College Dublin, Dublin, Ireland*.

Transcriptome analysis is commonly employed to evaluate biological processes in both human and livestock species. One of the prerequisites for this type of analysis is the possession of high purity, intact RNA. Postmortem tissue collection has inherent time delays and hence it is important to understand the temporal variation, both in the stability of total RNA and individual gene transcripts, with respect to particular tissues. The objective of this experiment was to both qualitatively and quantitatively assess the integrity of both total and mRNA species derived from ovine liver, spleen, thyroid, skeletal muscle, ileum and perirenal adipose tissue, which has been stored at ambient temperature and sampled at time points 0, 3, 6 and 9 h post-mortem. One hour after parturition, 6 lambs ( $5.12 \pm 0.27$  kg) were euthanized. Samples were collected from the liver, spleen, thyroid, skeletal muscle, ileum and perirenal adipose tissue and stored on a sterile Petri dish at ambient temperature. Approximately 1–2 g of tissue was then harvested at each time point (0, 3, 6, 9h), held for 24h in RNAlater and then stored at  $-80^\circ\text{C}$ . The quality and quantity of total RNA was assessed on the NanoDrop spectrophotometer and Agilent 2100 Bioanalyzer, respectively. While postmortem sampling time had no effect on RNA quantity ( $P \geq 0.05$ ) in 5 of the 6 tissue types analyzed, the RNA integrity number decreased over time and was significantly lower at 6 and/or 9 h postmortem in the spleen, thyroid, skeletal muscle, ileum and perirenal adipose tissues relative to the 0-h time point ( $P < 0.05$ ). A reduction in the normalization factor in the liver, spleen, ileum and perirenal adipose tissues was observed over the time period ( $P < 0.05$ ). In summary, the stability of total RNA remained intact within the first 3 h postmortem, regardless of tissue type, however tissue specific variation was evident in the RNA integrity across the 4 postmortem sampling times.

**Key Words:** ovine, RNA stability, postmortem

**W493 A meta-analysis of net protein and energy requirements for growth of dairy goats.** Anaiane Souza<sup>\*1</sup>, Normand St-Pierre<sup>2</sup>, Marcia Fernandes<sup>1</sup>, Amélia Almeida<sup>1</sup>, Julián Vargas<sup>1</sup>, and Isabelle Teixeira<sup>1</sup>, <sup>1</sup>*Universidade Estadual Paulista, Jaboticabal, Sao Paulo, Brazil*, <sup>2</sup>*The Ohio State University, Columbus, OH*.

Several factors including age, genotype, and sex affect the rate of body protein and fat deposition and, thus, the nutritional requirements for growth. Thus, the objective of this meta-analysis was to develop equations for estimating net protein ( $\text{NP}_g$ ) and net energy ( $\text{NE}_g$ ) requirements for growth of different sexes in dairy goats. A database of 244 individual records of Saanen goats weighing between 4.61 and 51.0 kg of body weight, from 7 comparative slaughter studies was used. Allometric equations were developed for body protein and energy contents in the empty body weight (EBW) as dependent variables and EBW as the allometric predictor. Parameter estimates were obtained using a linearized (log-transformation) expression of the allometric equations using the MIXED procedure of SAS 9.4. The model used included the random effect of study, and the fixed effects of sex (intact male, castrated male, and female;  $n = 93, 79, \text{ and } 72$ , respectively). Net requirements for growth were estimated as the first derivative of the allometric equations. The  $\text{NP}_g$  equations differed across sexes ( $P = 0.083$ ). The allometric equation for intact and castrated males was  $\log_{10} \text{EBW protein (g)} = 2.221 \pm 0.0224 + 1.015 \pm 0.0165 \times \log_{10} \text{EBW (kg)}$ ; and for females:  $\log_{10} \text{EBW protein (g)} = 2.277 \pm 0.0288 + 0.958 \pm 0.0218 \times \log_{10} \text{EBW (kg)}$  ( $\sigma_e = 0.0374$ ;  $\sigma_s = 0.0196$ ). The  $\text{NP}_g$  requirements for males were nearly 10% greater than those of females. The  $\text{NE}_g$  equations also differed between sexes ( $P < 0.001$ ). The allometric equation for intact males was:  $\log_{10} \text{EBW energy (kcal)} = 2.988 \pm 0.0323 + 1.240 \pm 0.0238 \times \log_{10} \text{EBW (kg)}$ ; for castrated males:  $\log_{10} \text{EBW energy (kcal)} = 2.873 \pm 0.0377 + 1.359 \pm 0.0283 \times \log_{10} \text{EBW (kg)}$ ; and for females:  $\log_{10} \text{EBW energy (kcal)} = 2.820 \pm 0.0377 + 1.442 \pm 0.0281 \times \log_{10} \text{EBW (kg)}$  ( $\sigma_e = 0.0472$ ;  $\sigma_s = 0.0309$ ). The  $\text{NE}_g$  requirements of castrated males were nearly 14% greater than those of intact males, and nearly 15% less than those of females. These results indicate that  $\text{NP}_g$  and  $\text{NE}_g$  requirements differ among sexes in growing dairy goats. This distinction is not done in the prevalent nutritional requirement systems (FAPESP grant # 2014/14734–9).

**Key Words:** allometry, Saanen, sex

**W494 Effect of selenium supplementation on reproductive activity of Saanen bucks.** Khoboso C. Lehloeny<sup>\*</sup>, Mamokou M. Mojapelo, and Jannes B. van Ryssen, *University of Pretoria, Pretoria, Gauteng, South Africa*.

Some parts of South Africa have soils deficient in selenium leading to selenium deficiency in animals grazing pastures or fed forages produced from these areas. Selenium deficiency is not only associated with poor fertility but also occurrences of several ruminant diseases. This study evaluated the effect of selenium supplementation on reproductive activity of Saanen bucks. Forty Saanen kids aged  $\pm 3$  mo were allocated into 2 groups comprising of selenium supplemented and the control. The study was conducted over a period of 6 mo. The Kids were fed lucerne ad libitum and had free access to water throughout the experiment. The supplemented group received sodium selenite orally at 3-mo intervals. Body weight and testicular measurements were recorded every 2 weeks while sexual behaviors were evaluated at weekly intervals. Semen was collected bi-weekly using an electro ejaculator from 5 mo of age. The supplemented group were significantly ( $P < 0.01$ ) heavier ( $21.09 \pm 4.1$  kg) than the control group ( $19.03 \pm 1.5$  kg) and also they had larger ( $P < 0.05$ ) scrotum circumference ( $22.77 \pm 3.2$  cm vs  $20.94 \pm 3.7$  cm) and testicular length ( $P < 0.01$ ) ( $8.92 \pm 1.61$  cm vs  $8.20 \pm 1.71$  cm).



Body weight had a positive correlation with the scrotal circumference; testicular length and width while it had a negative correlation with the expression of the mounting sexual behavior. Semen volume, mass motility and pH were not affected by selenium supplementation. However, the supplemented group had higher sperm concentration ( $2063.07 \times 10^6 \text{ mL}^{-1}$  vs  $1748.91 \times 10^6 \text{ mL}^{-1}$ ), live sperm ( $77.50 \pm 13.0\%$  vs  $76.64 \pm 10.5\%$ ) than the control group. The percentage of dead sperm ( $22.50 \pm 13.0$  vs  $23.36 \pm 10.5$ ) and secondary abnormalities ( $8.07 \pm 7.6$  vs  $8.48 \pm 9.7$ ) was lower in the supplemented group than the control group. The higher morphologically abnormal sperm cells observed from the supplemented group was mostly due to higher percentage of primary abnormalities, which cannot be contributed by selenium supplementation. It is therefore concluded that selenium supplementation increased body weight however heavier bucks expressed mounting behavior at later age than lighter bucks. Selenium supplementation also improves the sperm quality and viability.

**Key Words:** goat, testicular measurement, semen characteristic

**W495 Fatty acid profile of the meat of goats fed diets with soybean meals substituted with peanut cake.** Thadeu M. Silva<sup>\*1</sup>, Ariosvaldo N. Medeiros<sup>2</sup>, Ronaldo L. Oliveira<sup>1</sup>, Severino Gonzaga Neto<sup>2</sup>, Rita C. R. E. Queiroga<sup>3</sup>, and Rebeca D. X. Ribeiro<sup>1</sup>, <sup>1</sup>Federal University of Bahia, Salvador, Bahia, Brazil, <sup>2</sup>Federal University of Paraíba, Areia, Paraíba, Brazil, <sup>3</sup>Federal University of Paraíba, João Pessoa, Paraíba, Brazil.

Peanut cake is a by-product of biodiesel and contains relatively elevated lipid content. The inclusion of vegetable lipid sources in the diets can cause changes to the animal's fatty acid profile. This study was conducted to determine the effect of substituting soybean meal with peanut cake in the feeding of goats on meat fatty acid profile. Forty goats were assigned at a completely random design, to 1 of 4 diets. The concentrate was composed of corn bran, soybean meal, and mineral premix; soybean meal was substituted with peanut cake at substitution rates of 0.0, 33.33, 66.67, and 100%. Tifton-85 (*Cynodon* sp.) hay, was used at 50% of TMR. After 62 d of feeding, the animals were slaughtered. The left loin of each animal was collected and subjected to lipid extraction, methylation, and the quantification of the fatty acid esters were obtained through gas chromatography analysis. A general linear model was used to perform linear and quadratic regression with 5% significance. The sum of fatty acid and atherogenicity index was not influenced by the substitution of soybean meal with peanut cake (Table 1). Peanuts possess an elevated content of oleic acid (C18:1) compared with soy, which is rich in linoleic acid (C18:2). However, it is important to consider that these ingredients compose, at most, 21% of the total diet, so their lipid characteristics were diluted by the other ingredients. Another important factor is the ruminal biohydrogenation, which tends to convert oleic, linoleic and linolenic acids into stearic acid (C18:0). The peanut cake can replace completely the soybean meal in goat diet with no effect on fatty acid profile of the meat.

**Table 1 (Abstr. W495).** Sum of fatty acids as the percentage of total fatty acids and atherogenicity index in the loin of goats fed diets with soybean meal substituted with peanut cake

Item	Amount of substitution (%DM)				SEM	P-value	
	0.0	33.3	66.6	100.0		Lin	Quad
S Saturated	53.41	58.28	55.4	56.55	1.25	0.29	0.17
S Unsaturated	46.59	41.72	44.6	43.45	1.25	0.29	0.17
S Polyunsaturated	3.36	3.75	3.09	3.53	0.45	0.96	0.97
Unsaturated:							
Saturated	0.87	0.72	0.81	0.77	0.04	0.22	0.16
n-6:n-3	15.89	14.81	15.47	19.52	2.86	0.13	0.18
Atherogenicity index	0.89	1.00	0.91	0.97	0.06	0.59	0.73

**Key Words:** biodiesel, omega fatty acid, ruminant

**W496 Digestibility of nutrients in rams diets feed with fruit residue.** Elenice Conceição dos Santos<sup>1</sup>, Darcilene Maria Figueiredo<sup>\*1</sup>, Dorismar David Alves<sup>2</sup>, Aldrin Vieira Pires<sup>1</sup>, Paulo Gustavo Macedo de Almeida Martins<sup>1</sup>, Janaína Leite Barbosa<sup>1</sup>, Mônica Lopes Paixão<sup>3</sup>, Cassiane Gomes dos Santos<sup>1</sup>, and Mari-anne Schorer<sup>1</sup>, <sup>1</sup>Universidade Federal dos Vales do Jequitinhonha e Mucuri, Diamantina, MG, Brazil, <sup>2</sup>UNIMONTES, Montes Claros, MG, Brazil, <sup>3</sup>Universidade Federal de Viçosa, Viçosa, MG, Brazil.

The objective of this study was to evaluate the digestibility of nutrients after the inclusion of pineapple (*Ananas comosus* L.), banana (*Musa* sp.), mango (*Mangifera indica*) and passion fruit (*Passiflora* sp.) residue in diet for confined rams replacing 75% corn silage. Twenty 5 Santa Ines crossbred rams, rumen fistulated, castrated were used with approximate initial age of 24 mo, and initial body weight of  $45.4 \pm 6.1$  kg. Experiment was conducted in Latin square design ( $5 \times 5$ ), with 5 treatments, 5 animals per treatment, and 5 experimental periods of 13 d each. The forage: concentrate diet was 40:60 (% dry matter), and the forage in control diet was composed of 100% corn silage. To estimate the fecal nitrogen, animal feces were collected during 96 h using bag collectors. Samples of feces, bulky concentrates, and scraps of food provided were recorded. In all the samples we analyzed the chemical composition to estimate the digestibility of dry matter (DM), organic matter (OM), crude protein (CP), ether extract (EE), neutral detergent fiber (NDF), neutral detergent fiber corrected for ash and protein (NDFap), total carbohydrates (TC), and non-fibrous carbohydrates (NFC). The results were analyzed by ANOVA and Dunnett test at 5% probability, using the Statistical Analysis System. The inclusion of pineapple residue promoted increase ( $P < 0.05$ ) in digestibility of OM, and TC, respectively, 2.5 and 4.5%, compared with control diet, which may be related to fermentation processes caused by this food, as there was a greater ( $P < 0.05$ ) digestion of NDF, and NDFap with their inclusion in the diet. The diet containing banana residue decreased ( $P < 0.05$ ) the EE digestibility by 23.6%, however provided an increase ( $P < 0.05$ ) of the digestibility of NDFap in 7.78%. With the addition of mango residue, there was a reduction ( $P < 0.05$ ) in DM by 1.5%. We concluded that the use of fruit residue instead of 75% corn silage (DM) do not cause reduction in the total apparent digestibility of nutrients in rams.

**Key Words:** alternative foods, confinement, sheep

**W497 Nitrogen balance in sheep fed with fruit residue.** Elenice Conceição dos Santos<sup>1</sup>, Darcilene Maria Figueiredo<sup>\*1</sup>, Paulo Gustavo Macedo de Almeida Martins<sup>1</sup>, Aldrin Vieira Pires<sup>1</sup>, Dorismar David Alves<sup>2</sup>, Mônica Lopes Paixão<sup>3</sup>, Janaína Leite Barbosa<sup>1</sup>, and Marianne

Schorer<sup>1</sup>, <sup>1</sup>Universidade Federal dos Vales do Jequitinhonha e Mucuri, Diamantina, MG, Brazil, <sup>2</sup>UNIMONTES, Montes Claros, MG, Brazil, <sup>3</sup>Universidade Federal de Viçosa, Viçosa, MG, Brazil.

The objective of this study was to evaluate the effect of inclusion of pineapple (*Ananas comosus* L.), banana (*Musa* sp.), mango (*Mangifera indica*), and passion fruit (*Passiflora* sp.) residue in replacement of 75% corn silage in diets for confined sheep on nitrogen balance. Twenty 5 Santa Ines crossbred sheep male, rumen fistulated, castrated were used with approximate initial age of 24 mo, and initial body weight of 45.4 ± 6.1 kg. Experiment was conducted in Latin square design (5 × 5), with 5 treatments, 5 animals, and 5 experimental periods of 13 d each. The diets were formulated to contain approximately 72% total digestive nitrogen, and 14.7% crude protein. The forage: concentrate diet was 40:60 (% dry matter), and the forage in the control diet was composed of 100% corn silage. We carried out daily records of food provided and the remains of these, to ensure 10% of leftovers. To estimate the fecal nitrogen, total feces were collected during 96 h, using bag collectors. The urinary nitrogen were estimated collecting urine during 72 h. Nitrogen balance was measured by subtracting the total nitrogen intake and total nitrogen excreted in feces, and urine. The results were analyzed by ANOVA, and Dunnett test at 5% probability, using the Statistical Analysis System. There was no difference ( $P > 0.05$ ) among fruit residue, and corn silage for nitrogen values (intake, fecal, urinary, absorbed, and retained). Nitrogen balance was positive, and presented a mean value of 15.73 g day of nitrogen retained in animal body, representing 51.27% ingested nitrogen, indicating better utilization of nitrogen by sheep. The amount of nitrogen intake averaged 30.69 g day<sup>-1</sup>, and fecal and urinary losses represented, respectively, 28.87 and 19.87% ingested nitrogen. We conclude that the use fruit residue to replace 75% corn silage DM is viable and do not interfere the nitrogen balance and nitrogen retention.

**Key Words:** alternative foods, urinary nitrogen, fecal nitrogen

#### **W498 Ruminal parameters of sheep fed with fruit residue.**

Elenice Conceição dos Santos<sup>1</sup>, Darcilene Maria Figueiredo\*<sup>1</sup>, Dorismar David Alves<sup>2</sup>, Aldrin Vieira Pires<sup>1</sup>, Paulo Gustavo Macedo de Almeida Martins<sup>1</sup>, Janaína Leite Barbosa<sup>1</sup>, Mônica Lopes Paixão<sup>3</sup>, Adriano Cardoso dos Reis<sup>3</sup>, and Marianne Schorer<sup>1</sup>, <sup>1</sup>Universidade Federal dos Vales do Jequitinhonha e Mucuri, Diamantina, MG, Brazil, <sup>2</sup>UNIMONTES, Montes Claros, MG, Brazil, <sup>3</sup>Universidade Federal de Viçosa, Viçosa, MG, Brazil.

The objective of this study was to evaluate the digestibility of nutrients after the inclusion of pineapple (*Ananas comosus* L.), banana (*Musa* sp.), mango (*Mangifera indica*) and passion fruit (*Passiflora* sp.) residue, in diet for confined sheep, replacing 75% corn silage. Twenty-five Santa Ines crossbred sheep male, rumen fistulated, castrated were used with initial age of 24 mo and initial body weight of 45.4 ± 6.1 kg. Experiment was conducted in Latin square design (5 × 5), with 5 treatments, 5 animals, and 5 experimental periods with 13 d each. The forage: concentrate diet was 40:60 (% dry matter), and the forage in the control diet was composed of 100% corn silage. At 13th day of each experimental period, rumen fluid samples were collected via ruminal fistula, to determine pH, and ammonia nitrogen (NH<sub>3</sub>-N) in the following collections times: 0h (immediately prior the morning feeding), 2, 4, 6 h and 8 h after the morning feeding. Data collection was analyzed by regression, and to compare control diet with treatments, was performed by Dunnett test at 5% probability, using the Statistical Analysis System. There was no effect ( $P > 0.05$ ) on the ammonia concentration levels (N-NH<sub>3</sub>) in rumen fluid of animals fed with fruit residue, and obtained as an average value of 21.52 mg dL<sup>-1</sup>. In the different collections (0, 2, 4, 6 and 8 h) of ruminal fluid was observed a quadratic effect ( $P <$

0.05) for N-NH<sub>3</sub> (N-NH<sub>3</sub> = 19.64 + 3,11t - 0,44t<sup>2</sup>; R<sup>2</sup> = 0.63), where the highest concentration at this time was at 3.53 h post-treatment, which may be related to the lower diets fibrousness. Diet containing pineapple residue presented lowest ( $P < 0.05$ ) pH value, with an average between 6.13 collection times. Quadratic effect was observed ( $P < 0.05$ ) for pH (pH = 6.40 - 0,10t + 0,011t<sup>2</sup>; R<sup>2</sup> = 0.61) as rumen fluid collection time, with lowest value of pH occurred at 4.54 h post-treatment. We conclude that the use of all fruits residue do not decrease the amount of N-NH<sub>3</sub> available for rumen microorganisms, and not interfere with the pH of rumen fluid suitable for the microbiota.

**Key Words:** alternative foods, confinement, lamb

#### **W499 Effects of increasing dried distillers grains with solubles on intake, digestibility, blood and rumen metabolites of sheep consuming bermudagrass or eastern gamagrass hay.**

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In the southern United States, bermudagrass (*Cynodon dactylon*) is the predominantly used warm season forage. Eastern gamagrass (*Tripsacum dactyloides* L.) is being evaluated as an alternative to bermudagrass. The objective of this study was to determine intake and digestibility of bermudagrass or eastern gamagrass hay supplemented with increasing levels of dried distillers grains with solubles (DDGS) by sheep. In Experiment 1 (Exp. 1), bermudagrass hay (BG) was supplemented with 0, 10, 20, and 30% DDGS, and in Experiment 2 (Exp. 2), eastern gamagrass hay (EG) was also supplemented with the same levels of DDGS. In each experiment, a total of 4 crossbred wether sheep (Katahdin × Dorper; one year old, 33.2 ± 1.7 kg, BW) were used. The sheep were individually housed in pens and randomly assigned to each of the 4 diets. Each treatment consisted of 4 periods of 14 d/period (10 d adjustment and 4 d sample collection). Animals received hay ad libitum after the supplement was completely consumed. At the end of each period, a 7-mL blood sample was collected from each animal and analyzed for PUN, glucose, and NEFA concentrations and rumen fluid samples were also collected and analyzed for ruminal ammonia nitrogen (NH<sub>3</sub>-N) and volatile fatty acids (VFA) concentrations. Nutrient digestibility was estimated using the total fecal (bag) collection method. Data from each experiment were analyzed as a separate Latin square design using SAS. DDGS supplementation did not ( $P > 0.05$ ) affect BG hay DMI but increased ( $P < 0.05$ ) EG hay DMI. In both experiments, DM, OM, ash, NDF and ADF digestibilities were not affected ( $P > 0.05$ ) but CP and EE digestibilities increased ( $P < 0.05$ ) with DDGS supplementation. Although DDGS supplementation did not affect total VFA concentrations in Exp. 1, it tended ( $P = 0.09$ ) to decrease in Exp. 2. Plasma urea nitrogen (PUN) concentrations increased with DDGS supplementation in both experiments. Plasma glucose, NEFA, or rumen NH<sub>3</sub>-N, pH concentrations were not affected ( $P > 0.05$ ) by DDGS in both experiments.

**Key Words:** DDGS, digestibility, metabolites

#### **W500 Performance and feed efficiency of feedlot lambs fed different sources of non-protein nitrogen and carbohydrate.**

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Despite of the huge availability of agricultural lands and good weather conditions to forage growth in Brazil, feedlots have been an effective

way to increase animal production. The aim of this trial was to evaluate the performance and feeding efficiency of feedlot lambs fed diets with different non-nitrogen protein (NPN) and carbohydrates (CHO) sources in the concentrate. We used 20 non-castrated lambs with 4 mo of age and 26 kg of body weight. Lambs were fed ad libitum for 45 d with diets composed by 57% of grass hay (*Brachiaria* hybrid) and 43% of concentrate. The hay was harvested at late mature stage with high (72.4%) neutral detergent fiber and low (4.9%) crude protein. Four concentrates were formulated from the combination of 2 CHO (ground corn: starch; whey permeate: lactose) and 2 NPN (fast and slow release urea) sources. The diets were isoenergetic (2.32 Mcal/kg ME) and isoproteic (11% CP). Lambs were slaughtered at the end of trial after 45 d with a mean body weight of  $30.4 \pm 1.0$  kg. A completely randomized design was used with 4 diets and 5 replicates. Data were analyzed by ANOVA following a  $2 \times 2$  factorial scheme. Body weight gain (BWG), average daily gain (ADG), dry matter intake (DMI), gross feed efficiency (GFE) were analyzed. There was no interaction between CHO and NPN source ( $P > 0.05$ ) and NPN had no effect on any variables ( $P > 0.05$ ). Animals that were fed corn had higher BWG (5.28 vs. 3.54 kg), DMI (1.010 vs. 0.937 kg day<sup>-1</sup>), ADG (117.30 vs. 78.60 g day<sup>-1</sup>) and GFE (116.80 vs. 84.3 g kg<sup>-1</sup> DM) than those fed whey permeate. The best performance and high intake of diets with corn can be related to a possible microbial growth limitation and the depression of fiber degradation in diets containing whey permeate. Lactose provides less carbon than starch on a mass basis and has a higher potential for energy-spilling due to its faster fermentation. The source of NPN in the concentrate feed had no effect on performance of finishing lambs fed a low quality diet. Ground corn was a better ingredient for concentrate feed in diets containing low quality forage.

**Key Words:** energy, intake, urea

**W501 The use of Chambourcin grape extract as a natural anthelmintic in goat kids.** Kimberly A. Cash\*<sup>1</sup>, James D. Caldwell<sup>1</sup>, Bruce C. Shanks<sup>1</sup>, Amy L. Bax<sup>1</sup>, Luke S. Wilbers<sup>1</sup>, Heather L. Hilsenbeck<sup>1</sup>, Andrea T. McKnelly<sup>1</sup>, Taylor N. Drane<sup>1</sup>, Kelsey L. Basinger<sup>1</sup>, Jessica K. Clark<sup>1</sup>, Haley L. Bartimus<sup>1</sup>, and Harley D. Naumann<sup>2</sup>, <sup>1</sup>Lincoln University, Jefferson City, MO, <sup>2</sup>University of Missouri Columbia, Columbia, MO.

Gastrointestinal nematode parasitism is one of the greatest economic threats to goat production in the United States. With elevated incidences of anthelmintic resistance there is an increased interest in alternative natural dewormers, such as plants containing condensed tannins. Therefore, the objective of this study, supported by the NCR-SARE graduate grant program, was to evaluate the effects of fermented Chambourcin grape extract on parasite level and performance of goat kids. On October 14, 2014, a total of 45 mixed-breed goat kids (17.17 kg  $\pm$  0.79) were stratified by fecal egg count, weight, breed, and sex, and were allocated randomly to one of 3 treatments: 1) an oral dose (10 mL per 4.54 kg of BW) of fermented Chambourcin grape extract at 7-d (D7) intervals, 2) the same dose at 14-d (D14) intervals, or 3) control (30 mL oral dose of water at 14-d intervals). Condensed tannins were extracted, purified, and standardized by the Protein-Precipitable Phenolics method and found to have a concentration of 0.33 mg/mL. Goats were naturally exposed to gastrointestinal parasites on pasture consisting primarily of endophyte-infected tall fescue [*Lolium arundinaceum* (Schreb.) Darbysh] and mixed browse with access to 16% crude protein corn-soybean meal based creep feed for the duration of the 63-d study. Fecal egg counts, packed cell volumes, FAMACHA scores, weights, and body condition scores were measured every 7 d. Data were analyzed by the PROC MIXED procedure of SAS and treatments were reported as least squares means.

Two contrast statements were used to compare the mean of control versus D7 and D14 and the mean of D7 versus D14. Animal was the experimental unit. Start, final, and change from start to final fecal egg counts, packed cell volumes, FAMACHA scores, and body condition scores did not differ ( $P \geq 0.12$ ) across treatments. Average daily gain and total weight gain also did not differ ( $P \geq 0.42$ ) across treatments. Therefore, fermented Chambourcin grape extract may not be an effective natural anthelmintic for controlling nematodes in creep-fed goat kids.

**Key Words:** natural anthelmintic, condensed tannin, grape extract

**W502 Urushiol is not detected in blood or milk of Saanen dairy goats fed poison oak.** Massimo Bionaz\*, Claudia Ingham, Jennifer Belveal, Kristine Gomez, and Mark Keller, *Oregon State University, Corvallis, OR.*

Urushiol, the allergen present in *Toxicodendron diversilobum* (western poison oak), is responsible for contact dermatitis. More than 70% of adults have a reaction to urushiol, making this an important problem in the Northwest of US. Many anecdotal stories exist about desensitization to poison oak contact dermatitis acquired by drinking milk from goats fed poison oak. This has not been experimentally tested. In a previous study (Kouakou et al., 1992), urushiol was not detected in milk of goats fed 100% poison oak. To produce preliminary data for a large clinical trial, we have performed a pilot study in which 2 lactating Saanen goats were fed poison oak for 2 weeks. The experiment was carried out in the first 2 weeks of October 2014. The goats were at 80 d of lactation, weighed  $70 \pm 2$  kg, produced  $3 \pm 0.7$  kg/milk per day, and both were nursing triplets. The goats were kept in individual pens and fed twice a day with approx. 10% dry matter of fresh poison oak. Samples were obtained from each collection of poison oak. Feces, milk, and blood samples were collected at -1, 7, and 14 d after starting the experiment. Samples were spiked in with 2-dodecylphenol as internal standard and urushiol was extracted using 100% methanol. The extracted samples were purified using C-18 Solid phase Extraction columns before being injected into a Flexar HPLC (Perkin Elmer) equipped with a 5 $\mu$ m C18 HPLC column (Phenomenex). We were able to detect all 4 major congeners composing urushiol in the poison oak. We measured > 6 mg urushiol/g of fresh poison oak leaves (>14.0 mg/g of dry matter). Feces also contained urushiol (>1 mg urushiol/g of dry matter). We did not observe any urushiol in whole blood, plasma, or milk. The heptadecatrienylcatechol, the congener with the highest unsaturation and also the most irritant, was > 70% of the total extracted urushiol both in poison oak and feces. Our data confirmed the absence of detectable urushiol in milk; thus, milk from goats fed poison oak is safe to be consumed and handled by urushiol-sensitive people. Lack or minimal intestinal absorption and/or an extreme high clearance by the liver are inferred based on the absence of urushiol in blood.

**Key Words:** dairy goat, poison oak, milk

**W503 Performance and carcass parameters of lambs fed high grain diets with different fiber contents.** T. Brochado\*, S. B. Gallo, M. C. Freua, P. R. Leme, and R. A. Brandi, *Faculty of Animal Science and Food Engineering, University of Sao Paulo, Pirassununga, SP, Brazil.*

Although high concentrate diets are commonly used for finishing lambs, literature about the effect of minimum levels of NDF on performance is limited. The purpose of this study was to evaluate the effects of increasing dietary fiber levels on performance and carcass parameters of finishing lambs fed high concentrate diet. Twenty-four male lambs



(initial BW of  $24 \pm 3$  kg, 90 d old) were randomly assigned to 24 individual pens, in a complete randomized design with 3 treatments and 8 replicates. Treatments were dietary NDF levels of 12, 15 and 18% on DM basis. Diets were isoproteic, composed of corn, soybean meal, minerals and corn silage, and were offered twice a day with refusals daily measured to determine dry matter intake (DMI). Animals were weighed weekly and slaughtered after 50 d of feedlot, with average BW of  $40 \pm 4$  kg. Hot and cold carcass weight (HCW and CCW) as well as hot and cold carcass yield (HCY and CCY) were measured. Data were submitted to polynomial regression analysis, with initial BW as a covariate, using the statistical software R. There was a linear increase in DMI ( $P < 0.01$ ) from  $1.26 \pm 0.13$  to  $1.47 \pm 0.21$  kg/animal/d and a linear decrease on residual feed intake ( $P < 0.01$ ) from  $0.09 \pm 0.11$  kg/d to  $-0.06 \pm 0.19$  kg/d as dietary NDF increased. There was no difference in ADG ( $P > 0.05$ ,  $0.320 \pm 0.06$  kg/d) and feed conversion ( $P > 0.05$ ,  $4.27 \pm 0.64$  kg/kg) among treatments. There was a linear decrease ( $P < 0.05$ ) on HCW ( $21.08 \pm 1.10$  to  $19.65 \pm 2.20$  kg), HCY ( $51.93 \pm 1.99$  to  $49.71 \pm 2.47\%$ ) and CCY ( $50.04 \pm 2.02$  to  $47.98 \pm 2.64\%$ ) as dietary NDF increased. There was a trend of linear decrease ( $P = 0.06$ ) for CCW from  $20.31 \pm 1.08$  to  $18.97 \pm 2.15$  kg as dietary NDF increased. In conclusion, although ADG was not affected by treatments, carcass weight and carcass yield increased as dietary levels of NDF decreased.

**Key Words:** feedlot, lamb, neutral detergent fiber

**W504 Sheep performance under grazing supplemented with lime-hydrolyzed feather meal as a source of protein in their diet during the dry season in Mexico's Central Highlands.** Francisca Avilés Nova\*<sup>1</sup>, José M. Castro Salas<sup>3</sup>, Octavio A. Castelán Ortega<sup>2</sup>, Luis M. Ríos García<sup>1</sup>, and Anastacio García Martínez<sup>1</sup>, <sup>1</sup>*Centro Universitario Temascaltepec, Universidad Autónoma del Estado de México., Temascaltepec, Estado de México, México,* <sup>2</sup>*Facultad de Medicina Veterinaria y Zootecnia de la Universidad Autónoma del Estado de México, Toluca, Estado de México, México,* <sup>3</sup>*Unidad académica de Ciencias Agropecuarias y Ambientales, Universidad Autónoma de Guerrero, Iguala de la Independencia, Estado de Guerrero, México.*

The sheep continuous grazing on tropical forages during dry season requires supplementation with protein-based feed to improve production efficiency. The objective of this study was to evaluate the productive performance of grazing lambs supplemented with diets using 2 levels of inclusion of lime-hydrolyzed [ $\text{Ca}(\text{OH})_2$ ] feather meal. Experimental work was conducted at Rancho of the Temascaltepec University Center of the Autonomous University of the State of Mexico, from February to May. The treatments were: T1: grazing+diet with 6% feather meal; T2: grazing+diet with 9% feather meal; and T3: grazing+diet with 14% soybean meal. The diets were isoprotein (15.7%) and isoenergetic (4.6 Mcal/kg) and prepared with ground grain sorghum, wheat bran, molasses and minerals. The variables evaluated were daily weight gain, forage intake and feed conversion. Thirty lambs (Katahdin  $\times$  Pelibuey) (initial weight of  $26.1 \text{ kg} \pm 3.076$ ) were used in the study. All lambs were weighed at the beginning of trial and thereafter every week before grazing. The lambs were randomly distributed among the 3 treatments. They grazed for 6 h daily in 3 mixed paddock (1.0 ha) of *Panicum* (Tanzania and Mombasa) and *Brachiaria* (Insurgente and Mulato II). At the beginning of each week the forage supplied was measured, and at the end of the week, the remaining forage was measured. Grass was cut using grass shears utilizing the quadrat. After grazing each day, each lamb was fed 250 g of the corresponding supplement. The lambs were kept in individual pens. A totally random design was used, with 3 treatments and 10 repetitions (each lamb representing one repetition).

The Tukey's test was used ( $P < 0.05$ ). The highest daily weight gain ( $P < 0.05$ ) was obtained with the T2 ( $59.9 \text{ g d}^{-1}$ ) and T3 ( $67.6 \text{ g d}^{-1}$ ) treatments. No statistical difference ( $P > 0.05$ ) in feed conversion was found between the T2 and T3 treatments, obtaining relations of 11:1 and 10:1, respectively. We concluded that lime-hydrolyzed feather meal may be used as a protein supplement, when it represents 9% of the diet of grazing lambs during the dry season, and may thus replace soybean meal in their diet.

**Key Words:** lamb, supplemented, lime-hydrolyzed feather meal

**W505 Replacing corn with different levels of passion fruit by-product and its effects on feed intake, performance and digestibility in crossbred feedlot lambs.** Josemir S. Gonçalves\*<sup>1</sup>, Raimundo Neilson L. Amorim<sup>2</sup>, Raquel L. Salgado<sup>2</sup>, and Eric H. C. B. Van Cleef<sup>1</sup>, <sup>1</sup>*UNESP-Univ Estadual Paulista, Faculdade de Ciências Agrárias e Veterinárias, Jaboticabal, São Paulo, Brazil,* <sup>2</sup>*UFERSA-Federal University of Semi-arid, Mossoró, Rio Grande do Norte, Brazil.*

Two studies were conducted to evaluate effects of corn grain replacement with passion fruit by-product (PFB) in finishing lambs on feed intake, feedlot performance, carcass traits and digestibility. In study 1, 20 Morada Nova ram lambs ( $21.8 \pm 0.4$  kg BW) were used in a completely randomized design with 4 treatments: isonitrogenous (19% CP) experimental diets contained 30% Tifton-85 bermudagrass hay and 70% concentrate (soybean meal, wheat meal, corn and 0, 25, 50, or 75% PFB). Over a period of 21 d, lambs were housed in digestibility cages (14 d adaptation and 7 d data collection) and DM, CP and NDF digestibilities estimated by total fecal collection technique. In the study 2, 32 Morada Nova ram lambs ( $22.4 \pm 3.2$  kg BW) were housed in individual pens and assigned to a randomized block design (initial BW) and fed the same diets of study 1 for 78 d (14 d adaptation and 64 d data collection). DM and nutrient intakes, average daily gain (ADG) and gain to feed (G:F) were evaluated and after slaughter (32 kg), yields of hot carcass (HCY) and cold carcass (CCY) were calculated. Data from study 1 were analyzed using the GLM procedure of SAS and means were compared with Tukey test ( $P < 0.05$ ). For the study 2, data were analyzed with MIXED procedure with each animal as the experimental unit, and model effects included block and treatment. DM and CP digestibilities were not affected by PFB when it was added up to 50% ( $74.98 \pm 2.0$  and  $77.38 \pm 2.8\%$ , respectively;  $P > 0.05$ ). However, DM and CP digestibilities decreased in 75% PFB diet ( $69.7 \pm 0.7$  and  $72.5 \pm 1.1\%$ , respectively). NDF digestibility was unaffected by treatments ( $P > 0.05$ ), averaging  $49.4 \pm 3.4\%$ . There were no differences in DM and digestible nutrient intakes ( $P > 0.05$ ). No differences were evidenced ( $P > 0.05$ ) in ADG ( $152 \pm 0.03$  g), G:F ( $0.147 \pm 0.01$  kg), HCY ( $46.6 \pm 1.0\%$ ) and CCY ( $45.0 \pm 1.0\%$ ). The data indicate that although passion fruit by-product decreases diets DM and CP digestibility, it is a suitable replacement for corn grain in diets for feedlot lambs when used at up to 75%.

**Key Words:** by-product, intake, sheep

**W506 Effects of the utilization of increasing doses of aromatic plants on ruminal metabolism in Sarda dairy lactating ewes.** Roberta Boe, Oscar Boaventura Neto, Roberto Rubattu, Antonio Fenu, Antonio Mazza, and Antonello Cannas\*, *Dipartimento di Agraria, Sezione di Scienze Zootecniche, Università di Sassari, Sassari, Italy.*

The objective of this study was to investigate the effects of increasing doses of 3 aromatic plants *Carum* sp. (CAR), *Coriandrum* sp. (COR) and

*Satureja* sp. (SAT) as a natural dietary additive on ruminal metabolism in Sarda dairy sheep. Forty-four Sarda dairy ewes (BW = 45.11 ± 4.62 kg) were randomly divided into 4 groups (11 ewes per group) assigned to an aromatic plant or to the control (CON). The 3 plants were tested, for each group in chronological order, at 3 increasing doses (low = 25g, medium = 75g and high = 125g), each supplied individually for 21 d during the 2 daily machine milkings. The ewes also received during the day dehydrated alfalfa, beet pulp, corn and pea meals. Rumen samples were taken by using a stomach tube. Rumen pH (mean value 6.87) was influenced by type of aromatic plant and dose level ( $P < 0.004$ ), but not by their interaction. The ammonia content was higher in CON and COR groups (18.20 and 19.45 mg/dL, respectively) compared with CAR and SAT (14.79 and 15.87 mg/dL, respectively), and it was influenced by plant ( $P < 0.001$ ) but not by dose or their interaction. The molar proportions of acetate and propionate were affected by diet and dose level ( $P < 0.05$ ), with the higher values in CON and SAT (41.8% and 11.0%; 38.1% and 10.3%, respectively), while their ratio was influenced only by the dose level (increased from 3.19 to 4.34 for the low and high dose respectively). The molar proportion of butyrate was affected only by the type of plant, with the lowest value in COR and CAR (7.86% and 7.80%, respectively). A strong interaction ( $P < 0.001$ ) between type of aromatic plants and dose level was detected in the concentration of odd-branched-chain (OBCFA), saturated (SFA), monounsaturated (MUFA) and polyunsaturated (PUFA) fatty acids. In particular, CON and SAT groups had the higher concentration of OBCFA (7.93 and 7.86 g/100g FA, respectively) and PUFA (5.04 and 4.42 g/100g FA), while the concentration of SFA and MUFA were higher in CAR and COR groups. These results suggest that the aromatic plants used altered the FA profile of the rumen.

**Key Words:** aromatic plant, ruminal metabolism.

**W507 Effect of crude glycerin on in situ dry matter and neutral detergent fiber degradability in sheep.** E. H. C. B. van Cleef<sup>1,2</sup>, M. T. C. Almeida<sup>1</sup>, H. L. Perez<sup>1</sup>, H. B. Bussioli<sup>1</sup>, I. Monsignati<sup>1</sup>, J. R. Paschoaloto<sup>1</sup>, E. S. Castro Filho<sup>1</sup>, and J. M. B. Ezequiel<sup>1</sup>, <sup>1</sup>São Paulo State University, Jaboticabal, São Paulo, Brazil, <sup>2</sup>FAPESP, São Paulo, Brazil.

Eight ruminally cannulated crossbred male sheep (64.5 ± 8.5 kg) were used to evaluate the effect of inclusion of up to 30% crude glycerin, totally replacing corn, on dry matter intake, and DM and NDF in situ rumen degradability of rations. Animals were assigned into a replicated 4 × 4 Latin square design and, in pairs, were fed 1 of the 4 experimental treatments. Diets were composed of corn silage, soybean hulls, soybean meal, mineral premix, and crude glycerin (83% glycerol) replacing 0 (G0), 10 (G10), 20 (G20), or 30% (G30) corn cracked grain (DM basis), in a roughage:concentrate ratio of 40:60. Each experimental period lasted 21 d, composed of 14 d of adaptation and 7 d of data collection. Feed delivered and refused were weighed every morning to assess DMI. The 4 rations incubated in the rumen for 3, 6, 12, 24, 48, 72, and 96 h. The fraction “a” was determined by washing the bags in water and the residue after 96 h incubation was considered the fraction “c.” The potential degradability was calculated with the model:  $P = a + b(1 - e^{-kt})$ , and the effective degradability with the model:  $P = a + b * [k / (k + K_p)]$ . Treatments tented to promote a quadratic effect in DMI,

with increased values observed for G10 and G20 ( $P = 0.09$ ). Increasing crude glycerin linearly increased fraction “a” of DM and NDF of diets ( $P < 0.0001$ ), and linearly decreased its fraction “c” ( $P = 0.004$ ), increasing considerably the effective DM degradability ( $P < 0.0001$ ). However, degradation rate of DM was unaffected by treatments, while degradation rate of NDF decreased in glycerin treatments ( $P = 0.01$ ). No effects of crude glycerin was observed in effective degradability of NDF, regardless the passage rate evaluated (2, 5, and 8%/h). The addition of up to 20% crude glycerin seems to improve DMI. This by-product (up to 30% in dry matter basis) is a suitable energetic ingredient to replace corn in diets for sheep since it improves effective degradability of DM without affecting effective degradation of NDF.

**Key Words:** degradation kinetics, glycerol, sheep

**W508 Effect of increasing concentration of babassu meal in diets for feedlot crossbred lambs on apparent total-tract digestibility of dry matter and nutrients.** J. M. B. Ezequiel<sup>1</sup>, O. R. Serra<sup>1,2</sup>, J. R. S. T. Souza<sup>2</sup>, A. L. Lima<sup>2</sup>, and E. H. C. B. van Cleef<sup>1</sup>, <sup>1</sup>São Paulo State University, Jaboticabal, São Paulo, Brazil, <sup>2</sup>Maranhão State University, São Luiz, Maranhão, Brazil.

The objective of this study was to evaluate increasing concentrations of babassu (*Orbignya phalerata*) meal partially replacing Tifton-85 bermudagrass hay on dry matter and nutrients digestibility. Twenty-seven crossbred male lambs (90 d of age, 19.57 ± 0.41 kg BW) were randomly assigned to 1 of 3 experimental treatments. Isoenergetic (2.4 Mcal/kg ME) and isonitrogenous (19.6% CP) diets were formulated with 0.63% Ca, 0.36% P, and 2350 IU/kg vitamin A, to meet nutrient requirements for weight gain of 200g/d and were composed of ground corn, soybean meal, mineral premix, Tifton-85 bermudagrass hay, and 0 (T0), 15 (T15), or 30% (T30) babassu meal. Total fecal collection technique was used in this trial. Animals were adapted for 14 d to experimental diets, and between d 37 and d 41, samples of diets, feces and orts were collected. Samples were dried, ground (1 mm), and analyzed for dry matter, crude protein, neutral detergent fiber, acid detergent fiber, ether extract, and mineral matter contents. Digestibility coefficients (DC) were calculated by using the formula:  $DC (\%) = [(nutrient\ ingested - nutrient\ in\ feces) / nutrient\ ingested] \times 100$ . Data were analyzed as a completely randomized design by using mixed models. The animal was the experimental unit. Contrasts were used to determine linear and quadratic effect of babassu meal addition. The inclusion of babassu meal in the diets linearly increased digestibility of dry matter (T0 = 68.16, T15 = 74.38 and T30 = 79.63%), organic matter (T0 = 72.65, T15 = 78.06 and T30 = 82.44%), crude protein (T0 = 78.21, T15 = 83.19 and T30 = 87.13), ether extract (T0 = 79.62, T15 = 88.17 and T30 = 91.89), and neutral detergent fiber (T0 = 56.96, T15 = 62.91 and T30 = 68.30). Acid detergent fiber digestibility was unaffected by treatments and averaged 62.91%. In conclusion, babassu meal is a suitable feed ingredient to partially replace Tifton-85 bermudagrass in diets for finishing crossbred lambs and feasible for sheep producers, because this by-product is inexpensive when compared with grass hay, and improves dry matter and nutrient digestibility of diets when added up to 30% in dry matter basis.

**Key Words:** by-product, digestibility, sheep

## ASAS Early Career Awards

**874 Understanding the signaling pathways that regulate muscle mass in horses throughout the lifespan.** K. L. Urschel<sup>\*1</sup>, A. L. Wagner<sup>1</sup>, L. M. Mastro<sup>1</sup>, C. M. M. Loos<sup>1</sup>, A. A. Adams<sup>2</sup>, and K.M. Brennan<sup>3</sup>, <sup>1</sup>*Department of Animal and Food Sciences, University of Kentucky, Lexington, KY*, <sup>2</sup>*Department of Veterinary Science, University of Kentucky, Lexington, KY*, <sup>3</sup>*Alltech Inc., Nicholasville, KY*.

Skeletal muscle makes up ~50% of body weight in horses, with protein being the largest nonwater component. Muscle mass is largely determined by the balance of rates of muscle protein synthesis and breakdown. In an athletic species such as the horse, where muscle mass has been associated with performance, there is a need to understand how the underlying signaling pathways are regulated. In horses, similar to other species, activation of the downstream factors (4E-BP1, rpS6, S6K1) in the mechanistic target of rapamycin (mTOR) signaling pathway regulating protein synthesis is increased following feeding and insulin administration. The activation in response to feeding decreases with age in growing horses, with the muscle of yearlings being more responsive to feeding than that of 2 year olds. When horses are in an unfed state, there is no effect of age on the activation of mTOR signaling factors, either during growth or when comparing mature to old (> 20 yr old) horses. When comparing 2 populations of old horses: those that are relatively healthy and those with pituitary pars intermedia dysfunction, a condition that has been associated with a loss of muscle mass and insulin resistance, there were no differences between groups in the muscle activation or abundance of any of the mTOR signaling factors, myostatin (a factor related to muscle loss), or factors associated with the protein degradation signaling pathways (FoxO, atrogin-1, MuRF1), in response to either feeding or insulin infusion. Changes that occur in the muscle signaling pathways of old horses is an area where additional research is needed. In a dexamethasone-induced model of insulin resistance, where glucose uptake in response to insulin infusion was decreased by ~75%, there were profound decreases in the activation of mTOR signaling factors. These findings indicate that, in addition to being a risk factor for the development of laminitis and equine metabolic syndrome, insulin resistance may also impair the ability of the horse to maintain muscle mass, although additional research in other equine models of insulin resistance is needed. Understanding the

factors that regulate the signaling pathways of muscle protein synthesis and breakdown will allow the development of management and feeding strategies to promote muscle mass accretion and maintenance throughout the lifespan.

**875 The unexplored part of the rumen microbiome: Exploring the adaptive roles of bacteriophage auxiliary metabolism genes during dietary intervention in the rumen.** Christopher L. Anderson<sup>1,2</sup>, Galen Erickson<sup>1</sup>, and Samodha C. Fernando<sup>\*1</sup>, <sup>1</sup>*Department of Animal Science, University of Nebraska-Lincoln, Lincoln, NE*, <sup>2</sup>*School of Biological Science, University of Nebraska-Lincoln, Lincoln, NE*.

Viruses are the most abundant biological entity, yet the roles of viruses within ecosystems are poorly understood. Work from other environments demonstrates that bacteriophage populations contain auxiliary metabolism genes thought to increase host fitness by altering host metabolism. As an attempt to better understand the viral influence on host bacterial populations, we investigated viral and total microbial community structure and function using culture-independent metagenomic approaches under 4 different dietary conditions. Using a shared read approach based on median k-mer profiling, the structure of total microbial communities significantly differed based on diet and host, whereas enriched viral metagenomes differed only by diet. The majority of differences between viral populations from different diets were metabolic in nature. Using community level metabolic networks, we further explored why different diets enrich phage communities for specific metabolic pathways. Enzymes differentially abundant in the total metagenome and virome had significantly higher betweenness centrality and a lower average shortest path length compared with nondifferential genes in the network. In addition, differential viral genes had a significantly higher total degree and in-degree compared with nondifferential genes. This ongoing work begins to suggest that diet, rather than host factors, has a stronger influence on the structuring of rumen phage populations and that phages encode for an adaptive repository of central metabolic functions related to selection pressures driven by altering environmental conditions. Current efforts are focused on better understanding what governs why certain central metabolic genes are enriched and how this is related to the flow of information through metabolic networks.



# ASAS Graduate Student Symposium: Networking to achieve interdisciplinary research

**643 Networking beyond the animal sciences to facilitate interdisciplinary research.** Russell B. Muntifering\*, *Auburn University, Auburn, AL.*

Researchable problems in the animal sciences are often highly intricate, and progress toward their resolution is often best achieved through integration of several scientific disciplines. Interdisciplinary research involving animal scientists does not need to transcend departmental boundaries because our departments by definition comprise disciplinary specialization in nutrition, genetics, physiology, biochemistry, and so on. However, identifying and networking with disciplinarians in other departments and universities can be a daunting task, especially for new faculty members whose discipline-focused graduate training may not have exposed them to the scrutiny of persons who were not trained within the same scholarly limitations, culture and traditions. For example, a legitimate concern among social scientists is that they are often recruited to join a natural-sciences project too late to provide input toward experimental design and data collection that can often provide more productive and interesting opportunities for socioeconomic analyses. Involving external disciplinarians early in the planning stages of research, even if they do not ultimately collaborate, can be extremely useful in terms of exchanging ideas for collecting useful data and becoming familiar with each others' work. Another hurdle to interdisciplinary research is mutual ignorance or naiveté among disciplines that can feed unrealistic expectations of capabilities and deliverables. Such impediments can be overcome by dialog and working through graduate students; seating external disciplinarians on graduate student advisory committees is a good way for them to become familiar with new researchable topics through thesis/dissertation literature reviews. The unique governance system and academic culture of universities should ideally provide an environment where interdisciplinary research can flourish, but this is not always the case. Faculty need to be knowledgeable of and sensitive to norms of valuation and reward within disciplines other than their own, and new faculty especially need to be cognizant of evaluation and reward systems at their institutions pertaining to single-investigator vs. collaborative scholarship.

**Key Words:** networking, interdisciplinary research

**644 Effect of interdisciplinary research to animal science.** Janet R. Donaldson\*, *Mississippi State University, Mississippi State, MS.*

Interdisciplinary research is becoming increasingly more important to the progression of studies related to animal health. Broad training in interdisciplinary research, coupled with high quality specialized research, enhances job opportunities for students in government, academic, and industrial settings. Interdisciplinary training in animal science can entail, but is not limited to, associations with microbiologists, immunologists, and engineers. This symposium will focus on the need for people classically trained in microbiology, immunology, and so on, to network with animal scientists for the development of novel, translational therapeutic approaches to improve animal health. This symposium will be based on my personal experiences in multidisciplinary research

from the approach of a microbiologist with no training in animal science. Networking beyond my discipline began during my first year in a faculty position. Through networking beyond my discipline, my research program has expanded to include a multidisciplinary team, consisting of animal physiologists, immunologists, gastrointestinal physiologists, and veterinary microbiologists. Together with this team, we have made substantial advancements toward progressive therapies for circumventing both animal and human diseases.

**Key Words:** networking, interdisciplinary research

**645 Breaking the boundaries of animal science research through internationalization programs.** Luis O. Tedeschi\*<sup>1</sup> and James. P. Muir<sup>2</sup>, <sup>1</sup>*Texas A&M University, College Station, TX,* <sup>2</sup>*Texas A&M AgriLife Research, Stephenville, TX.*

Globalization has imposed many changes in economic and political arenas in many regions around the world, but scientific knowledge has no boundaries and multinational collaboration can increase scientific knowledge by fostering the discovery of techniques and resources that can assist current and foreseeable problems, including hunger and sustainability. Ruminant animals, for instance, convert human-inedible resources into animal products for human consumption, and humankind have benefited from this prehistoric symbiotic relationship because ruminants can adapt to diverse environment (climate, soil, and vegetation), making them apt to inhabit virtually anywhere in the world. Collectively, humans of all socio-economic and socio-cultural strata have relied on animals for energy and/or protein consumption, wool, and draft power. This relationship has sometimes led to extreme production systems (due to economy-of-scale factors in some countries) that may cause irreversible damage to the environment. Thus, appropriate management of animal production is essential to enable long-term, sustainable continuation of this human-livestock relationship within acceptable animal welfare criteria. Proper education is the first step. Study abroad courses expose students and faculty to new endeavors in animal science and provide effective communication skills for the advancement of science. Internationalization programs minimize technical and economic disparities among countries. The educational exchange among countries is imperative to prepare leaders and extend the frontiers of knowledge in sustainable livestock intensification. For university and agency researchers, international exchanges advance technical, socio-economic, and environmental differences and strategic possibilities among countries. International business partnerships are critical for major multinational companies based in the United States involved in animal products and genetics, seed, fertilizer, and agrochemicals. Therefore, the investment on international research collaboration increases our awareness of human resource needs and enhances the career opportunities of our students by preparing them not only with technical expertise but also with language capabilities and in-depth understanding of other cultures.

**Key Words:** networking, interdisciplinary research

## Breeding and Genetics: Beef and meat species

**646 Large-scale single-step genomic BLUP evaluation for American Angus.** Daniela A. L. Lourenco\*<sup>1</sup>, Shogo Tsuruta<sup>1</sup>, Breno O. Fragomeni<sup>1</sup>, Yutaka Masuda<sup>1</sup>, Ignacio Aguilar<sup>2</sup>, Andres Legarra<sup>3</sup>, Joseph K. Bertrand<sup>1</sup>, Tonya S. Amen<sup>4</sup>, Lizhen Wang<sup>4</sup>, Dan W. Moser<sup>4</sup>, and Ignacy Misztal<sup>1</sup>, <sup>1</sup>University of Georgia, Athens, GA, <sup>2</sup>INIA, Las Brujas, Uruguay, <sup>3</sup>INRA, Castanet-Tolosan, France, <sup>4</sup>Angus Genetics Inc., St. Joseph, MO.

This study aims to investigate the feasibility of single-step genomic BLUP (ssGBLUP) for American Angus evaluation. Over 6 million records were available on birth weight (BW) and weaning weight (WW), 3.4 million on post-weaning gain (PWG), and 1.3 million on calving ease (CE). Genomic information was available on 51,883 animals. Realized accuracies were based on a validation population of 18,721 young animals born in 2013. Traditional and genomic EBV were computed by BLUP and ssGBLUP, respectively, using a multiple-trait linear model for growth traits and a bivariate threshold-linear model for CE-BW. Additionally, 2 methods for handling a large number of genotyped animals were tested: indirect prediction (IND) based on SNP effects derived from ssGBLUP, and algorithm for proven and young (APY) that uses genomic recursions on a small subset of reference animals to invert the genomic relationship matrix (G). All ssGBLUP, IND\_ssGBLUP, and APY\_ssGBLUP were based on reference populations of about 2000 high accuracy sires and cows (2k), 2k + all genotyped ancestors of the validation population (8k), and 8k + all remaining genotyped individuals not in the validation (33k). With BLUP, realized accuracies were 0.48, 0.67, 0.52, and 0.29 for BW, WW, PWG, and CE, respectively. With ssGBLUP and the 2k (33k animals) reference population, the accuracies were 0.55, 0.71, 0.60, and 0.31 (0.62, 0.78, 0.65, and 0.31), respectively. Low accuracy for CE was due to many missing records and low incidence rate. With 8k reference population, index of indirect prediction with parent average was as accurate as prediction from regular ssGBLUP. With 33k reference population, indirect prediction alone was as accurate as prediction from regular ssGBLUP. APY with recursions on 4k (8k) animals reached 97% (99%) of regular ssGBLUP accuracy; the cost of APY inverse of G is 1% (4%) of the regular inverse. The genomic evaluation in beef cattle with ssGBLUP is feasible while keeping the same models already used in regular BLUP. Indirect predictions allow for low cost interim evaluations. Use of the APY allows for inclusion of large number of genotyped animals in the main evaluation.

**Key Words:** beef cattle, genomic selection

**647 Assignment of polled status using single nucleotide polymorphism genotypes and predicted gene content.** John B. Cole<sup>1</sup>, Daniel J. Null\*<sup>1</sup>, Chuanyu Sun<sup>2</sup>, and Paul M. VanRaden<sup>1</sup>, <sup>1</sup>Animal Genomics and Improvement Laboratory, ARS, USDA, Beltsville, MD, <sup>2</sup>Sexing Technologies, Navasota, TX.

There is growing interest in cattle that are naturally polled, but the polled allele has a very low frequency. The best way to increase its frequency is by index selection, which requires known polled status for all animals. Laboratory tests for polled are used as data, and US and Canadian bulls with  $\geq 500$  daughters and not designated as polled are assumed homozygous normal. Polled status is imputed for all other genotyped animals using these data. There are 2 mutations in the region around 1.7–1.9 Mb (UMD3.1) on BTA1 known to cause polled. The Celtic mutation is a deletion and an insertion, and the Friesian mutation a duplication. Both are located in a 75-marker window spanning 0.1–3.5 Mb on BTA1.

An animal is heterozygous if it has either mutation, and is homozygous if both haplotypes contain polled, regardless of the mutation. This is consistent with -P and -PP coding in all breeds. Brown Swiss, Holstein, and Jersey polled haplotypes have frequencies of 0.41%, 0.93%, and 2.22%, respectively. The National Dairy Database has genotypes for only 678,848 of 39 million cows with records. Gene content (GC) for non-genotyped animals, the number of polled haplotypes in an animal's genotype, was computed using records from genotyped relatives. The GC are real-valued and range between 0 and 2. Prediction accuracy was checked by comparing polled status from recessive codes and animal names to GC for 1,615 non-genotyped Jerseys with known status. 97% (n = 675) of horned animals were correctly assigned GC near 0, and 3% (n = 19) were assigned GC near 1. Heterozygous polled animals had GC near 0 (52%, n = 474) and near 1 (47%; n = 433), although 3 animals were assigned a GC near 2. The expectation for GC is near 1 for heterozygotes, but can be lower if many polled ancestors have unknown status or when pedigree is unknown. In those cases GC may be set to twice the allele frequency, which is low. Some with -P in the name may actually be PP. All homozygous polled animals (n = 11) were assigned GC near 2. Polled status for non-genotyped animals can be accurately determined, and this method can be extended to other genes of interest.

**Key Words:** gene content, imputation, polled

**648 Genetic analysis of hair coat shedding in beef cattle with data collection using a practical strategy.** Trent Smith\*<sup>1</sup>, Michael D. MacNeil<sup>2</sup>, and Joseph P. Cassady<sup>3</sup>, <sup>1</sup>Mississippi State University, Mississippi State, MS, <sup>2</sup>Delta G, Miles City, MT, <sup>3</sup>South Dakota State University, Brookings, SD.

Hair coat characteristics can affect adaptability of beef cattle and performance in various environments. Objectives of this study were to examine the usefulness of an annual hair coat shedding score (HCS) during the spring transition period and determine its relationship with maternal productivity as indicated by weaning weight (WW). Data were collected on 5,294 purebred Angus cows in May of 2011 and 2012 from various herds throughout the Southeastern US, Missouri, and Texas. Measurements included a HCS (1–5) and BCS (1–9) scored independently by 2 trained technicians and averaged. The WW of calves were obtained from the breed association database. The data included 2,225 cows that were observed in both years. Two bivariate analyses were conducted to examine the relationships of HCS with BCS and WW. The same model was used to analyze HCS and BCS. It included fixed effects of contemporary group and age, and random direct genetic and permanent environmental effects due to animals. The model for WW included fixed effects of contemporary group, age of dam and sex, and a linear covariate for age of calf at weaning. Random effects in the model for WW were direct and maternal genetic effects, and a permanent environmental effect due to dams. Phenotypic correlations of HCS with BCS and WW were 0.17 and approximately zero, respectively. Heritability estimates for HCS and BCS were  $0.42 \pm 0.03$  and  $0.12 \pm 0.03$ , with a genetic correlation of  $-0.25 \pm 0.10$ . For WW, heritability estimates were  $0.28 \pm 0.05$  for direct and  $0.05 \pm 0.04$  for maternal effects. The genetic correlation for WW direct and maternal was  $-0.34 \pm 0.24$ . Estimated genetic correlations of HCS with direct and maternal genetic effects on WW were  $0.17 \pm 0.22$  and  $-0.30 \pm 0.25$ , respectively. Results of this study suggest that HCS assessed once a year during a transitional period could be used in selection decisions if profitable in

certain environmental conditions. Associations of HCS with other traits of economic importance need to be explored further.

**Key Words:** genetic variances, hair shedding, beef cattle

#### 649 An application of MeSH enrichment analysis in livestock.

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It is an integral part of functional genomics studies to assess the enrichment of specific biological terms in gene lists found to be playing an important role in biological phenomena. Contrasting the observed frequency of annotated terms with those of the background is at the core of over-representation analysis (ORA). Gene Ontology (GO) is a means to consistently classify and annotate gene products and has become a mainstay in ORA. Alternatively, Medical Subject Headings (MeSH) offers a comprehensive life science vocabulary including additional categories that are not covered by GO. Although MeSH is predominantly applied in human and model organism research, its full potential in livestock genetics is yet to be explored. MeSH ORA was evaluated to discern biological properties of the identified genes and contrasted with the results obtained from GO enrichment analysis. Three published data sets were employed for this purpose representing a gene expression study in dairy cattle, the use of SNPs for genome-wide prediction in swine, and the identification of genomic regions targeted by selection in horses. We found that several over-represented MeSH annotations linked to these gene sets share similar concepts with those of GO terms. Moreover, MeSH yielded unique annotations, which are not directly provided by GO terms, suggesting that MeSH has the potential to refine the representation of biological knowledge. We demonstrated that MeSH can be regarded as another choice of annotation to draw biological inferences from genes identified via experimental analyses. When used in combination with GO terms, our results indicate that MeSH can enhance our functional interpretations for specific biological conditions or the genetic basis of complex traits in livestock species.

**Key Words:** MeSH, enrichment analysis, annotation

#### 650 Use of partial least squares regression to predict individual milk coagulation properties and cheese yield from Fourier transform infrared spectra in Sarda dairy sheep.

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Milk coagulation properties (MCP) are popular indicators of milk cheese making ability. They are measured as rennet coagulation time (RCT, min), curd firming time ( $k_{20}$ , min) and curd firmness ( $a_{30}$ , mm). The potential cheese yield of milk could be also be assessed by individual cheese micro-manufacturing experiments (ILCY). However, the routine measure of these traits appears to be rather problematic in terms of costs and logistics. In this work, partial least squares regression (PLSR) is used to predict individual MCP and cheese yield of 965 Sarda breed ewes located in 47 flocks. MCP were measured using the Formagraph, and cheese yield was assessed by ILCY. Mid infrared spectra was obtained by Milkoscan (Foss Electric). Animals were split into 2 data sets: (1) training (700 ewes) that was used to estimate the PLS model and (2)

validation (265 ewes), that was used to validate PLS predictions. One hundred replicates were performed, randomly assigning animals to training and validation sets. Goodness of predictions was assessed by calculating the determination coefficient ( $R^2$ ), the residual mean squared error of prediction (RMSEP), the regression slope ( $b_{obs,pred}$ ) and intercept ( $a_{obs,pred}$ ) (Table 1). The  $R^2$  indicates an accurate prediction for RCT and, to a lesser extent,  $a_{30}$ , and very poor for  $k_{20}$ . Also for ILCY the prediction was quite accurate. These figures were confirmed also by values of RMSEP and of regression parameters. The PLSR yielded prediction results of moderate accuracy for RCT and ILCY using mid-infrared spectral data as predictors and it could represent a valuable tool for the recording of these phenotypes for management and breeding purposes. Research supported by the regione Autonoma della Sardegna, project "Il latte Ovino della Sardegna".

**Table 1 (Abstr. 650).** Statistics of PLS prediction for MCP and ILCY in the validation data set

	Mean obs	Mean pred	$R^2$	RMSEP	$b_{obs,pred}$	$a_{obs,pred}$
RCT (min)	15.25 ± 6.62	15.20 ± 5.82	0.71 ± 0.05	3.57 ± 0.35	0.97 ± 0.08	0.57 ± 1.17
$k_{20}$ (min)	1.53 ± 0.85	1.55 ± 0.38	0.07 ± 0.03	0.83 ± 0.05	0.60 ± 0.17	0.60 ± 0.27
$a_{30}$ (mm)	49.8 ± 20.2	49.8 ± 15.9	0.55 ± 0.05	13.5 ± 0.8	0.95 ± 0.08	2.5 ± 4.4
ILCY (%)	36.3 ± 9.3	36.3 ± 7.7	0.63 ± 0.06	5.7 ± 0.5	0.95 ± 0.06	1.7 ± 1.93

**Key Words:** milk coagulation properties, sheep, partial least squares regression

#### 651 MUC1 gene polymorphism in Murrah water buffaloes and its association with milk production traits.

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MUC1 is a glycoprotein mucin expressed in apical mammalian epithelial tissues, such as mammalian gland, which the main function is protect the cell surface from the environment microorganisms. Hence, MUC1 gene is a candidate gene to mastitis resistance. The MUC1 molecule is well defined in bovines, which has been associated with a highly polymorphic variable number of tandem repeats region (VNTR), which are highly conserved. However, there is no information about MUC1 for water buffaloes (*Bubalis bubalis*). Thus, the aim of this study was to identify the VNTR polymorphism of MUC1 gene in water buffaloes of Murrah breed and evaluated the polymorphism associations with economical traits and mastitis resistance. 200 animals from the experimental farm of State University of Sao Paulo (Brazil) were used. Genotyping was performed by the polymerase chain reaction (PCR). The amplified fragments were separated by electrophoresis on 1.5% agarose gel. The length of the alleles was estimated using the GeneRuler plus molecular weight marker. The traits evaluated were: somatic cell count (SCC), fat percentage (%F), age at first calving (AFC), calving interval (CI), fat yield (FY), protein percentage (%P), protein yield (PY) and milk production (MP). The analyses were performed using the PROC MIXED procedure of the SAS program to evaluate the effect of the presence/absence of the MUC1 alleles on each trait. Differences with  $P < 0.05$  were considered significant. Three alleles of different lengths were amplified and named allele 1, 2 and 3. Allele 2 was the predominant with a frequency of 0.56. Genotype 2/3 was the most frequent in all animals with a frequency of 0.41. The statistical analysis considering the presence/absence of the MUC1 alleles revealed that the alleles 1 and 3 were associated with some economic traits. A significant effect for the



allele 1 was observed for higher means for SCC (7.52) and %P (4.39). The absence of allele 3 was significantly associated with higher means for PY (82.45) and AFC (862.12). The allele 1 effect for SCC persisted significant after the Bonferroni adjustment. Our data indicate MUC1 gene association with traits related to mastitis resistance like SCC and with economic traits like %P.

**Key Words:** buffaloes, mastitis, mucin1

**652 The effects of sire breed on reproductive and progeny performance in Kiko meat goats.** Henry J. Henderson\* and Chukwue-meka Okere, *Tuskegee University, Tuskegee Institute, AL.*

The aim of genetic selection in meat goat is to improve performance by incorporating the beneficial traits from a breed type. The primary objective of this study was to examine the effects of different sire types (Boer vs. Kiko) on reproduction and progeny performance of purebred Kiko does as well as growth and health performance of their offspring. Doe performance was analyzed by evaluating prolificacy (litter size), fecundity (fertility x prolificacy), and birth types (single, twins, or triplets). Doe and progeny performance were analyzed by evaluating prolificacy and pre-weaning growth and survival. A total number of 19 Kiko does were used in this study, 11 of which were bred to a Kiko buck and 8 to a Boer buck. Results revealed individual breed combination prolificacy values (1.9 and 1.75 kids/doe) for the Kiko Sired Group and Boer Sired group respectively. There were non-significant sire differences for gestation length ( $150 \pm 2.66$  vs.  $148 \pm 3.11$  d,  $P = 0.06$ ) for Boer and Kiko respectively. Weights of Kiko dams assigned to Boer and Kiko sires at breeding were similar ( $45.63 \pm 10.17$  vs.  $42.39 \pm 6.91$ , kg,  $P = 0.41$ ). At weaning, weights of dams bred to Boer and Kiko sire were not significantly different ( $52.77 \pm 14.64$  and  $42.76 \pm 7.13$ , kg,  $P = 0.10$ ). Litter size at birth and at weaning did not differ among sire breed ( $1.75 \pm 0.46$ ,  $P = 0.37$  vs.  $1.90 \pm 0.30$  and  $1.50 \pm 0.53$  vs.  $1.75 \pm 0.46$ ,  $P = 0.33$ ) respectively. Boers sired kids were significantly heavier at birth but not at weaning ( $3.41 \pm 0.48$  vs.  $2.78 \pm 0.53$  kg,  $P = 0.001$  and  $13.82 \pm 2.78$  vs.  $12.43 \pm 3.47$  kg,  $P = 0.26$ ). This suggests a growth-improvement potential for progeny when utilizing Boer sires. Non-significant differences were observed for ADG ( $0.15 \pm 0.03$  vs.  $0.13 \pm 0.03$  kg/d) for Boer and Kiko sired kids respectively. Overall,

results showed no consistent differences in reproduction and progeny performance traits between the main sire types used in this project.

**Key Words:** sire, prolificacy, progeny

**653 Comparison of zinc finger sequences of hybrid sterility gene *Prdm9* between yaks, cattle, and their sterile hybrids.**

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*Prdm9* (PR domain containing 9) is the first hybrid sterility gene reported in vertebrates, it is a meiosis-specific gene and possibly related to male infertility. The objective of this study was to compare the *Prdm9* zinc fingers of yaks (*Bos grunniens*, n = 33), Yellow cattle (*Bos taurus*, n = 6) and their sterile male hybrids (i.e., cattle-yaks, n = 7). Genomic DNA was extracted from muscle or testis tissues of the experimental animals, and PCR was performed to amplify zinc finger sequences of *Prdm9*. The PCR products were gel-purified and sequenced from both strands. The zinc finger sequence of yak *Prdm9* is highly conserved, the deduced *Prdm9* protein consists of 5 C<sub>2</sub>H<sub>2</sub> type zinc fingers, which share identical sequences among 3 yak breeds or individuals. However, Yellow cattle *Prdm9* showed variations in both sequence and numbers of zinc fingers among the 6 individuals, with zinc finger numbers of 5, 7 and 8 (5 in 4 cattle, 7 and 8 in 2 cattle). Amino acid sequence comparison of corresponding zinc fingers between yak and cattle revealed 6 polymorphisms and 9 mutations, and 7 of the mutations are located at the positive selection sites. These results indicate that the zinc fingers of *Prdm9* gene evolves much faster in cattle than in yak. Further analysis of the zinc finger sequences of *Prdm9* in sterile cattle-yaks showed that cattle-yaks carry heterozygous *Prdm9* alleles, 4 of which contain alleles with different numbers of zinc fingers. Cattle-yak exhibits higher body size, milk and meat yields than those of yak, but F1 to F3 male sterility. We propose that the differences of *Prdm9* zinc fingers at the positive selection sites as well as the numbers of zinc fingers between yak and cattle might be closely associated with the sterility of male cattle-yak.

**Key Words:** yak, *Prdm9* gene, hybrid sterility

## Dairy Foods: Processing and chemistry

**654 Effect of hydrodynamic cavitation on particle size of casein micelles, protein interactions and heat stability of skim milk.** Harsh Dahiya<sup>\*1</sup>, Hasmukh A. Patel<sup>1</sup>, and Thom Huppertz<sup>1,2</sup>, <sup>1</sup>South Dakota State University, Brookings, SD, <sup>2</sup>NIZO Food Research, Ede, the Netherlands.

Hydrodynamic cavitation (HC) is a process of vaporization, bubble generation followed by bubble collapse in a flowing liquid brought about by a decrease in pressure followed by a subsequent increase in pressure. We are currently applying this technology in the processing of milk and milk products. Therefore, the objective of this preliminary study was to investigate the effect of hydrodynamic cavitation (HC) on some important properties of milk such as particle size of casein micelles, interactions of proteins and heat stability of skim milk (SM). Pasteurized skim milk (3.5% protein and 9% total solids) was preheated to 50°C and then subjected to 2 sets of HC treatments, namely, HC at 20, 40, and 60 Hz at sufficiently high flow rate (950 L/h) to avoid any temperature increase during HC (T1) and HC at 60 Hz at low flow rates (200 L/h) to allow scale-free heating of skim milk increasing its temperature up to 90°C (T2) using APV Cavitator (supplied by SPX, Denmark) fitted with 4-row rotor in 6mm housing. The samples obtained from T1, T2 and untreated (control) skim milk samples were analyzed for changes in the particle size of casein micelles using Malvern Zetasizer Nano ZS and heat stability using the heat coagulation time (HCT) test at 140°C. The protein interactions in the whole sample and serum phase obtained from these samples were also studied using SDS-PAGE. SM subjected to T1 and T2 did not cause significant changes ( $P < 0.05$ ) to the casein micelle size and HCT (165–171 nm and  $>5$  min) compared with that in control sample (168 nm and  $>5$  min respectively). On the other hand, samples subjected to T2 exhibited significantly higher levels ( $P < 0.05$ ) of whey protein denaturation ( $P < 0.05$ ) compared with those subjected to T1, which was attributed to the heat generated due to cavitation during T2. The high molecular weight aggregates were also generated in samples subjected to T2 due to extensive denaturation of whey proteins and their interactions with casein micelles. The results of this study suggested that HC can be promising technique that can be potentially used as an alternative technology for scale-free heating of milk with minimal effect on important properties of milk.

**Key Words:** hydrodynamic cavitation, denaturation, scale-free

**655 Optimization of milk atomization by viscosity measurement.** Luc K. Belliere, Corentin Thierry<sup>\*</sup>, Valerie Lefevre, and Philippe Burg, *Sofraser, Villemandeur, France.*

Every year, very large quantities of powdered milk are produced worldwide (~5 billion tonnes). Optimization of the manufacturing is the key to ensure product yield and profitability. This can be achieved by optimizing product viscosity during the drying and atomization of the milk. During drying, water is removed to reduce the energy required in the atomization column. Good viscosity control leads to a better evaporation and therefore lower water content. During atomization, droplet size has a direct effect on the heat required for drying and therefore the energy required to produce powdered milk. One of the methods to optimize atomization is by controlling the viscosity of the fluid before spraying as viscosity has a direct effect on droplet size. The viscometer used during the trial (MIVI, Sofraser) is a viscometer at resonance frequency working at a high shear rate. The active part of the sensor is a vibrating rod held in oscillation at its resonance frequency where the amplitude

of the movement varies according to the viscosity of the product. The viscometer was rated for 500 bar and 200°C, with a full-scale range of 200 cP. The trials were performed at a Dutch milk processing company where the sensor was installed on a flow through-cell before the atomizer. The equipment was tested on 3 different products at the following operating conditions: atomization pressure between 150 and 225 bars, temperature in the cell ~75°C and flow rate of 2,800 L/h. The viscometer was used to control the viscosity of the incoming milk from the dryers and ensure the milk was within the optimal viscosity range for atomization. The setpoint viscosity for the 3 different formulations was 20, 50, and 180 cP. The viscometer allowed maintaining viscosity within  $\pm 1 \sigma$  for 85 to 90% of points and a maximum difference of 3% between setpoint and mean measurement. Depending on product  $1 \sigma$  is between 5 and 10% of setpoint. Higher casein to whey protein ratio gave more stable results, as well as higher carbohydrate to fat ratio. This will not be explained as it is not part of the study. Finally, during the trials, the use of a viscometer allowed 2% energy savings which represent about \$15,000 to \$20,000/year (based on \$0.07/kWh).

**Key Words:** process optimization, powdered milk, atomization

**656 Effect of membrane channel geometry on limiting flux and serum protein removal during skim milk microfiltration.** Michael C. Adams, Emily E. Hurt, and David M. Barbano<sup>\*</sup>, *Cornell University, Ithaca, NY.*

Our objectives were to determine the limiting fluxes (LF) and serum protein (SP) removal factors (SPR) of 2 100-nm ceramic microfiltration (MF) membranes: one with 4-mm round retentate flow channels (RFC) and one with 4-mm equivalent-diameter diamond-shaped retentate flow channels (DFC). Retentate and permeate were continuously recycled to the feed tank and a uniform transmembrane pressure (TMP) (TMP at the inlet minus TMP at the outlet) was maintained at  $25 \pm 3$  kPa using a permeate recirculation pump. The LF for each membrane was determined by increasing flux once per h from  $45 \text{ kg} \cdot \text{m}^{-2} \cdot \text{h}^{-1}$  until flux did not increase with increasing TMP. Temperature, average cross-flow velocity, and protein concentration in the retentate recirculation loop were maintained at 50°C,  $7 \text{ m} \cdot \text{s}^{-1}$ , and  $8.5 \pm 0.03\%$ , respectively. Experiments were replicated 3 times and the Proc GLM procedure of SAS was used for statistical analysis. The LF of the DFC membrane ( $71 \text{ kg} \cdot \text{m}^{-2} \cdot \text{h}^{-1}$ ) was lower ( $P < 0.05$ ) than the LF of RFC membrane ( $88 \text{ kg} \cdot \text{m}^{-2} \cdot \text{h}^{-1}$ ). Reynolds numbers based on the hydrodynamic diameter of the retentate flow channels were calculated for each membrane. Differences in Reynolds numbers between the membranes were proportional to the differences in LF. Permeate produced using the DFC membrane contained more ( $P < 0.05$ ) protein than the permeate produced using the RFC membrane due to additional casein passage through the DFC membrane. SPR was calculated by dividing true protein in the permeate by SP in the permeate portion of the feed to describe the ease of SP passage through each membrane. Higher SPR indicate higher rates of SP passage. After accounting for casein contamination in each of the permeates with SDS-PAGE, the DFC membrane SPR remained higher ( $P < 0.05$ ) than the RFC membrane SPR. Though DFC membrane LF was lower, DFC permeate removal on a modular basis was higher due to the increased module membrane surface area of the DFC membrane relative to the RFC membrane ( $2.07 \text{ m}^2$  vs.  $1.41 \text{ m}^2$ ). Depending on the

size of the system, using DFC membranes could reduce the capital cost of a MF system due to a reduction in the number of stainless steel modules.

**Key Words:** microfiltration, channel geometry, limiting flux

**657 Effect of soluble milk components on limiting flux and serum protein removal during skim milk microfiltration.** Michael C. Adams, Emily E. Hurt, and David M. Barbano\*, *Cornell University, Ithaca, NY.*

The tendency of calcium to promote microfiltration (MF) membrane fouling is well documented, but the role of lactose has not been studied. Milk protein concentrate that is 85% protein on a dry basis (MPC85) contains less calcium and lactose than skim milk (SM). Our objectives were to determine the limiting fluxes (LF) and serum protein (SP) removal factors (SPR) of 0.1  $\mu\text{m}$  ceramic graded permeability membranes that were fed with 3 different milks: SM, MPC85 that had been standardized to the protein content of SM with reverse osmosis (RO) water (MPC), and MPC85 that had been standardized to the protein and lactose contents of SM with RO water and lactose monohydrate (MPC+L). Retentate and permeate were continuously recycled to the feed tank. The LF for each feed was determined by increasing flux once per h from 55  $\text{kg}\cdot\text{m}^{-2}\cdot\text{h}^{-1}$  until flux did not increase with increasing transmembrane pressure. Temperature, pressure drop across the membrane length, and protein concentration in the retentate recirculation loop were maintained at 50°C, 220 kPa, and  $8.77 \pm 0.2\%$ , respectively. Experiments were replicated 3 times and the Proc GLM procedure of SAS was used for statistical analysis. The LF of SM (91  $\text{kg}\cdot\text{m}^{-2}\cdot\text{h}^{-1}$ ) was lower ( $P < 0.05$ ) than the LF of MPC+L (124  $\text{kg}\cdot\text{m}^{-2}\cdot\text{h}^{-1}$ ) due to the role of calcium in fouling. The LF of MPC+L was lower ( $P < 0.05$ ) than the LF of MPC (137  $\text{kg}\cdot\text{m}^{-2}\cdot\text{h}^{-1}$ ) due to the higher viscosity contributed by lactose. Permeates produced from the MPC and MPC+L contained more ( $P < 0.05$ ) protein than the SM permeate due to the transfer of micellar casein into the reduced-calcium sera of the MPC and MPC+L. SPR was calculated by dividing true protein in the permeate by SP in the permeate portion of the feed to describe the ease of SP passage through the membrane. After accounting for the nonmicellar casein with SDS-PAGE, no differences ( $P > 0.05$ ) in SPR were detected among the 3 feeds below the LF. As the fluxes approached the LF, SPR decreased ( $P < 0.05$ ) due to fouling. Feeding a MF system with MPC instead of SM will reduce the required membrane surface area, but the permeate protein composition will be different.

**Key Words:** microfiltration, calcium, lactose

**658 Comparison of 3 different variable selection strategies to improve the predictions of fatty acid profile in bovine milk by mid-infrared spectrometry.** Hélène Soyeurt\*<sup>1</sup>, Yves Brostaux<sup>1</sup>, Frédéric Dehareng<sup>2</sup>, Nicolas Gengler<sup>1</sup>, and Pierre Dardenne<sup>2</sup>, <sup>1</sup>*University of Liège-Gembloux Agro-Bio Tech, Gembloux, Belgium*, <sup>2</sup>*Walloon Agricultural Research Centre, Gembloux, Belgium.*

Mid-infrared (MIR) spectrometry is used to provide phenotypes related to the milk composition. Foss spectrum contains 1,060 datapoints. The number of reference values required to build a calibration equation is often lower than the spectral variables mainly due to the cost of chemical analysis. Problems of collinearity and overfitting appear when this high dimensional data set is used. This research will study the interest of using variable selection (VS) approach before the use of partial least square regression (PLS). The data set included 1,236 milk spectra related to their fatty acid (FA) contents. Saturated (SFA), monounsaturated (MUFA), polyunsaturated (PUFA), short chain (SCFA), medium chain

(MCFA), and long chain FA (LCFA) were studied. The data set was randomly divided in 3 groups which were used to create 3 calibration and validation data sets. Three different VS methods were compared. The first strategy was based on the part of trait variability explained by each considered variables (R2VS). The second method was based on the regression coefficient estimated after PLS procedure divided by the standard deviation of the considered spectral variable (BSVS). The third strategy permitted to underline the uninformative variables which were the ones having the lowest ratio of average regression coefficient to their corresponding standard deviation estimated after a leave-one out cross-validation (UVEVS). For UVEVS and BSVS, the cutoff was determined from the known uninformative region of MIR milk spectrum. The cutoff for R2VS was determined by testing different thresholds ranged between 5 and 40%. The most interesting cutoff for R2VS was 25%. The worst results in terms of validation root mean square error of prediction (RMSEPv) were obtained using a full PLS (i.e., without VS). The maximum difference (g/dl of milk) of RMSEPv obtained from the full PLS and from the PLS using selected variables were 0.156 for SFA, 0.139 for MUFA, 0.011 for PUFA, 0.025 for SCFA, 0.164 for MCFA, and 0.188 for LCFA. R2VS gave the best results for all studied traits followed by UVEVS and then BSVS. In conclusion, the use of VS improved significantly the performance of FA MIR equations.

**Key Words:** milk, fatty acid, infrared

**659 Factors influencing laboratory performance of oven drying total solids on whole milk.** David M. Barbano\* and Chassidy Coon, *Cornell University, Ithaca, NY.*

Milk analysis proficiency testing has been carried out monthly by a group 10 laboratories for about 10 years. Patterns of differences in total solids (TS) results among labs have been observed, but the causes of some of these differences (i.e., systematic increase or decrease in difference from the all-laboratory mean value as function of milk total solids level) have not been identified. Our objective was to identify specific sources that cause these variations in results among laboratories. The atmospheric force air oven method number 990.20 of the Association of Official Analytical Chemists was used. A set of 14 milks with a range of TS content from about 8.4 to 15.2% were tested in duplicate by 10 different labs and replicated in 4 mo. In mo 1 and 3, the 14 milks were tested on each of 3 different days by the same analyst and in mo 2 and 4, the 14 milks were tested on each of 3 different days by the different analyst within each lab. The differences from the all-lab mean with statistical outliers removed were analyzed using Proc GLM of SAS to test for effects of lab, day, sample, and their interactions. The ANOVA terms for lab and lab by sample interaction were significant ( $P < 0.05$ ) and explained most of the variation. The lab by sample interaction manifested itself as some labs having upward slopes, while other laboratories had downward slopes of the residual plot of 28 differences from the 14 sample all lab means as TS increased. The direction of the slope, when it occurred, was a function of laboratory or technician within laboratory. Follow-up work was conducted to identify causes of this behavior of TS results. Oven temperatures and oven temperature recovery rates were within method specifications for all labs. Attention was focused on the interaction of the analyst and balance. At the various weighing steps, the weights were recorded 3 times: immediately, after the balance indicated the weight was stable, and 7 s after the indication of stability. Based on these observations, a series of sensitivity analyses were done. The accuracy of TS results was most sensitive to variation (both random and systematic) in control of the balance zero during the steps of weighing the empty pan and the pan plus dry solids.

**Key Words:** milk, solids, oven drying



**660 Greek-style yogurt manufacture: A case study for eco-efficiency assessment in dairy processing.** Yves Pouliot\*<sup>1</sup>, Alain Doyen<sup>1</sup>, Catherine Houssard<sup>2</sup>, Adriana Paredes Valencia<sup>1</sup>, Scott Benoit<sup>1</sup>, Dominique Maxime<sup>2</sup>, and Manuele Margni<sup>2</sup>, <sup>1</sup>*STELA Dairy Research Center, Université Laval, Québec, QC, Canada*, <sup>2</sup>*CIRAIG, CIRODD, École Polytechnique de Montréal, Montréal, QC, Canada*.

Greek-style yogurt is characterized by a higher protein content (>10% w/v) compared with conventional products (4–5% w/v). The protein concentration is typically achieved by ultrafiltration (UF) or centrifugation processes after milk fermentation by yogurt starter cultures. However, this approach requires 3 times more milk than the traditional yogurt and it generates Greek yogurt co-product acid whey permeate for which the subsequent valorization represents a challenge. Although the concentration step increases the commercial value of yogurt, depending on the processing options selected, its manufacture affects the use of milk constituents and on the process environmental footprint. The concept of eco-efficiency (EE) provides an assessment of the environmental

performance of a product system in relation to its value. EE is a practical tool for managing environmental data (e.g., Life cycle analysis or LCA) and value aspects (e.g., financial, functional, sensory) in parallel. In a first part of our study, we have compared the environmental impact of Greek-style yogurt production from 2 different processing options, i.e., performing the UF concentration step before or after fermentation step. Pilot-scale experiments were performed to characterize both processes in terms of water use, energy consumption and waste generation such as permeate (acid or not). The relative environmental impact of both processes will be characterized by means of a comparative LCA. The value aspects of Greek-style yogurt manufacture in the EE equation will address its direct commercial value, but also the value (or costs) of co-products valorization (or disposal). Our research aims at developing an EE-based approach and provide a decision-support tool to help manufacturers identifying and selecting most eco-efficient processing options for the production of Greek-style yogurt.

**Key Words:** eco-efficiency, dairy processing, yogurt

# Dairy Foods Symposium: Advances in bacterial exopolysaccharides—From production to applications in dairy foods and health

**661 Advances in production of exopolysaccharides and simplified methods for their recovery and quantification.** Luc De Vuyst\* and Frédéric Leroy, *Vrije Universiteit Brussel*.

The capacity of strains to produce exopolysaccharides (EPS) is widespread among species of lactic acid bacteria, although the physiological role of these molecules is not clearly understood yet. When EPS are produced during milk fermentation, they confer technological benefits to the fermented end products, such as an improved rheology, a reduced syneresis, and an enhanced body and mouthfeel. In addition, some of these EPS may have beneficial effects on consumer health. This requires optimal and sufficient production of these molecules both in situ and ex situ not only to improve their yields but also to obtain a particular functionality. Therefore, appropriate methods of production and recovery should be established. One particular difficulty relates to the production and processing of the biomass of starter cultures when they produce high concentrations of EPS. All this requires a better understanding of EPS production and regulation mechanisms.

**Key Words:** lactic acid bacteria, exopolysaccharides, fermentation

**662 Chemical modification of EPS to improve its health functionalities.** Siqian Li and Nagendra Shah\*, *The University of Hong Kong, Pokfulam Road, Hong Kong*.

Exopolysaccharides (EPS) from *S. thermophilus* ASCC 1275 (ST1275) were isolated, purified and lyophilized and sulfated using SO<sub>3</sub>-pyridine complex and FT-IR was used to identify the characteristic bands in EPS and sulfated EPS. Changes in antioxidant activities after sulfate modification were examined by measuring FRAP, DPPH, superoxide anion and hydroxyl radical scavenging activities. Antibacterial activities and MIC tests of EPS and sulfated EPS were investigated against *S. aureus*, *E. coli* and *L. monocytogenes* by cylinder-plate diffusion method. Changes in anti-proliferative activities after sulfate modification were also examined on HepG2 and Caco-2 cells by MTT method, while changes in anti-inflammatory activities were examined in RAW264.7 macrophages. LPS was used to induce inflammation in RAW264.7 macrophages. Sulfate modification of EPS was achieved and the degree of sulfate modification, which was identified as the average number of oxygen-sulfate groups per residue in EPS, was 0.31. Additionally, bands in FT-IR spectra indicated that sulfate group was at the C6 position of the galactose skeleton in sulfated EPS. The results also revealed that the antioxidant activities of sulfated EPS were significantly ( $P < 0.05$ ) improved compared with those of unmodified EPS in all the 4 antioxidant activities assays. Sulfated EPS had larger inhibition zone on *S. aureus*, *E. coli* and *L. monocytogenes* compared with those of unmodified EPS which indicated that the antibacterial activities of EPS were significantly ( $P < 0.05$ ) improved after sulfate modification. Furthermore, sulfated EPS had lower MIC for *S. aureus*, *E. coli* and *L. monocytogenes*, which also suggested that the antibacterial activities of EPS improved after sulfate modification. Sulfated EPS had higher anti-proliferative activities on both HepG2 and Caco-2 cells. Also, pro-/anti-inflammatory (IL-6/IL-10; TNF- $\alpha$ /IL-10) cytokine secretion ratios of LPS-stimulated RAW264.7 macrophage were significantly ( $P < 0.05$ ) decreased after sulfated EPS treatments at non-cytotoxic doses. In conclusion, our results indicated that antioxidant, antibacterial, anti-proliferative and anti-inflammatory activities of ST1275 EPS were improved after sulfate modification

**Key Words:** EPS, sulfate modification, antimicrobial activity

**663 Advances in application of EPS in dairy foods, particularly in low-fat or fat-free yogurt, and low-fat mozzarella cheese.** Donald J. McMahon\*, *Western Dairy Center, Utah State University, Logan, UT*.

Exopolysaccharides (EPS) produced by bacterial cultures has a long history of usage in cultured dairy products such as yogurt for improving their texture and providing some of the unique attributes of these traditional foods. Depending of the organism that produces the EPS, the EPS material can be retained around the cell as a capsule or released into the surrounding medium. In some cases, the released EPS can form into clumps and strands of EPS and are these are considered to be ropey cultures. Investigations into use of capsular EPS+ cultures in cheese making became of interest 25 years ago as a means of improving the texture of low-fat cheeses and this still continues to be an area of research related to many different types of cheeses. A prime effect of using EPS+ cultures is the greater retention of moisture in low-fat cheeses either from using EPS+ starter cultures or by direct addition of an EPS mass into the cheese milk before renneting. Understanding the effect of EPS on both rennet and acid coagulated foods has depended on studies of the product's microstructure using electron microscopy and confocal laser scanning microscopy. The functional effects of inclusion of EPS into cheese and yogurt depends on the intrinsic chemical nature of the EPS such as sugar composition, extent of branching and charge. This presentation reviews recent findings related to biostructural analysis of fermented dairy foods containing EPS from a variety of EPS+ bacterial species.

**Key Words:** exopolysaccharides, microstructure, dairy

**664 Beneficial effects of EPS on human health and gut microbiota.** Hua Wei\*<sup>1,2</sup>, Zhihong Zhang<sup>1</sup>, Xueying Tao<sup>1,2</sup>, Feng Xu<sup>2</sup>, Hengyi Xu<sup>1</sup>, Cuixiang Wan<sup>2</sup>, Qinglong Wu<sup>3</sup>, and Nagendra P. Shah<sup>3</sup>, <sup>1</sup>State Key Laboratory of Food Science and Technology, Nanchang, Jiangxi, China, <sup>2</sup>Jiangxi-OAI Joint Research Institute, Nanchang, Jiangxi, China, <sup>3</sup>The University of Hong Kong, Hong Kong, China.

Recently bacterial exopolysaccharide (EPS) extracted from *Lactobacillus* spp. and *Bifidobacterium* spp. has received considerable attention, mainly due to the novel and unique physical characteristics and physiological functions. EPS contributes to biofilm formation and host healthy maintenance, it can be used as an instrument for probiotics to survive in harsh condition. In this review, we provide a fresh perspective and focus on the advancement of bioactivity and immune modulating capability of exopolysaccharide. Indeed, EPS from *Lactobacillus* spp. and *Bifidobacterium* spp. exhibit many beneficial bioactivity including anti-tumor activity, antiviral activity, radical scavenging activity and abrogate the cytotoxic effect. Therefore, it has great potential for further development as therapeutic agents or adjuvants for cancer. Furthermore, EPS binds to common surface receptors to elicit the immune response in host, it was able to reduce the secretion of the proinflammatory e.g., IL-6. Thus, the EPS seem to be critical in studying the physiology of *Lactobacillus* spp. and *Bifidobacterium* spp. and their interaction with the host.

**Key Words:** probiotics, exopolysaccharide, bioactivity

# Growth and Development Symposium: The mitochondrion—A powerhouse for the cell or a key to animal productivity?

## 665 Mitochondrial bioenergetics—Bringing the cell to life.

Darrell Neuffer\*, *East Carolina Diabetes and Obesity Institute, East Carolina University, Greenville, NC.*

This presentation will provide an overview of mitochondrial bioenergetics and redox systems biology—how life for cells is generated and maintained. Mitochondria are known as the engines of the cell, converting fuel derived from food into a chemical form of energy (ATP) used to support various cellular processes. A lesser-known but no less important role of the mitochondria is to generate and maintain an “electrical” charge throughout the cellular proteome. The respiratory system is composed of a series of proteins imbedded within the inner mitochondrial membrane that transport electrons by default, using oxygen as the final acceptor to produce water. The engine relies on a type of electrical circuit or redox system that is primed or “pressurized,” with energy demand decreasing and energy supply increasing that pressure. Normally the mitochondrial respiratory system operates as a demand based system, meaning that the rate of fuel utilization and oxygen consumption is determined by the rate at which energy is being utilized by cells, even at rest (i.e., idling). In fact, because skeletal muscle represents a high percentage of body mass, this idling of mitochondria represents a significant source of heat generation by animals. However, when energy supply exceeds energy demand, the redox pressure within the system increases, causing electrons to leak from the circuit to oxygen prematurely, leading to the formation superoxide and other reactive oxygen species (ROS) that can react with and damage proteins, lipids and DNA. Fortunately, antioxidant scavenging systems are extremely efficient and convert the majority of superoxide to the non-radical hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>). H<sub>2</sub>O<sub>2</sub> can freely diffuse out of the mitochondria and influence various signaling pathways. Thus, the mitochondrial respiratory system functions as a redox pressure gauge that senses and reflects cellular metabolic balance. When in positive balance, electron leak serves as a release valve, accelerating mitochondrial H<sub>2</sub>O<sub>2</sub> emission which has been implicated to play a significant role in numerous diseases related to disorders of metabolism.

**Key Words:** mitochondria, energy, redox

## 666 Mitochondrial bioenergetics and aging. Hazel H. Szeto\*, *Research Program in Mitochondrial Therapeutics, Department of Pharmacology, Weill Cornell Medical College, New York, NY.*

Mitochondria play a central role in energy generation in the cell, providing ATP to carry out essential biological functions. As energy output declines, the most energetic tissues are preferentially affected, including the skeletal muscles, heart and eyes. Age-related decline in function has been observed in these systems and is associated with a decline in mitochondrial function in both animals and humans. Furthermore, age-related decline in skeletal muscle and cardiac function results in immobility that further promotes loss of skeletal muscle mass (sarcopenia). This talk will summarize the current literature on the effects of age and immobility on mitochondrial function and ATP production in skeletal muscles, heart, and the retina. A novel compound (SS-31) that promotes mitochondrial bioenergetics has been shown to reverse skeletal and cardiac muscle dysfunction, and loss of visual acuity, associated with aging. The mechanism of action of SS-31 and its effects on aging will be presented.

## 667 Mitochondria function in Rendement Napole pig growth.

David E. Gerrard<sup>1</sup>, Samer W. El-Kadi<sup>1</sup>, and Tracy L. Scheffler\*<sup>2</sup>, <sup>1</sup>*Virginia Tech, Blacksburg, VA,* <sup>2</sup>*University of Florida, Gainesville, FL.*

The capacity for lean growth may be related to muscle fiber type characteristics. Previously, much emphasis has been placed on contractile phenotype and myosin heavy chain isoforms, yet metabolic properties (oxidative versus glycolytic) may play a more intimate role. Muscle oxidative capacity, largely determined by mitochondrial content and functional properties, has immense potential to fuel anabolic processes including protein synthesis. Yet, enhanced oxidative capacity is also associated with increased protein degradation and may restrict hypertrophy. Intriguingly, the relationships between metabolic phenotype, energy status, and protein accretion change during the lifetime of the animal. Thus, the influence of oxidative capacity on the potential for muscle growth likely depends on growth stage, along with input from various signaling pathways. These signaling pathways integrate cues related to energetic and nutritional status, hormones and other factors; this interplay coordinates adaptations in fiber metabolism, structure, and size to optimize function. In particular, AMP-activated protein kinase (AMPK) plays a key role in regulating energy producing and energy consuming pathways to maintain cellular energy homeostasis. Activated AMPK affects acute regulation of enzyme activity, as well as long-term adaptation through changes in gene and protein expression. Specifically, activated AMPK limits protein synthesis, while simultaneously promoting mitochondrial biogenesis and oxidative capacity. Pigs with the Rendement Napole (RN) mutation possesses a single nucleotide polymorphism in the  $\gamma 3$  subunit of AMPK, which results in increased AMPK activity. Therefore, the RN pig is a valuable model for investigating the effect of AMPK, energy status, and muscle metabolic phenotype on efficiency of lean gain during different stages of growth. Ultimately, understanding the relationship between muscle oxidative capacity and protein accretion and defining how this relationship evolves from birth to adult, is important for maximizing quantity and efficiency of meat production.

**Key Words:** mitochondria, AMP-activated protein kinase, skeletal muscle

## 668 Mitochondrial and cellular metabolism in response to selection for residual feed intake in pigs. S. M. Lonergan\*, S. M. Cruzen, J. K. Grubbs, E. Huff Lonergan, J. C. M. Dekkers, and N. K. Gabler, *Iowa State University.*

A primary goal of animal agriculture is to improve the efficiency of meat production by optimizing the growth performance of livestock. Divergent genetic selection of swine for improved residual feed intake (RFI) has resulted in a line of pigs selected for low RFI that reaches the same market weight while consuming 10 to 15% less feed than the high RFI line. Our long-term goal is to define the contribution of mitochondria function to this improved growth efficiency. Investigations using RFI selection have demonstrated that muscle mitochondria from pigs in the low RFI line exhibited less electron leakage from the electron transport chain. This is particularly evident in the mitochondria from the red portion of the semitendinosus (complex I and II) and the white portion of the semitendinosus (complex I, II, and III). Reactive oxygen species production from electron leakage in the mitochondria



was positively correlated with RFI, demonstrating a link between poorer mitochondria function and poorer growth efficiency. A parallel observation was that muscle from more efficient pigs had a greater capacity to decrease protein degradation and conserve muscle mass. Mitochondria from muscle of the more efficient low RFI pigs had a greater abundance of heat shock protein 70, heat shock protein 60, malate dehydrogenase, ERO1  $\alpha$ , and subunit 1 of the cytochrome *bc1* complex. Mitochondria from muscle of the less efficient high RFI pigs had a greater abundance of pyruvate kinase and glyceraldehyde 3-phosphate dehydrogenase. It is important to note that post-translational modification of proteins plays a central role in mitochondria function. Heat shock protein 70, heat shock protein 60, and ATP synthase are among the mitochondria proteins modified by phosphorylation. The combined evidence demonstrates that muscle growth and maintenance is directly influenced by mitochondria efficiency and cellular protein homeostasis. (Funded in part by the AFRI competitive grant number 2010–65206–20670 from USDA NIFA and by Iowa Pork Producers grant number 10–009).

**Key Words:** mitochondria, RFI, swine

**669 Browning of adipose tissue.** Shihuan Kuang\*, Pengpeng Bi, and Tizhong Shan, *Department of Animal Sciences, Purdue University, West Lafayette, IN.*

Adipose (fat) tissues mediate systemic energy homeostasis, and play an important role in animal growth, health and reproduction. In addition, adipose tissues contribute to meat marbling and are thus a key determinant of meat quality. Fat cells within adipose tissues can be classified into white, beige and brown adipocytes mainly based on their mitochondria content. White adipocytes contain few mitochondria and are the predominant cell type in various subcutaneous and visceral fat depots. Brown adipocytes contain numerous mitochondria and are mainly found in interscapular brown adipose tissues of many mammals. Beige adipocytes are a newly defined type of adipocytes containing intermediate abundance of mitochondria and are found to coexist with white adipocytes in subcutaneous fat depots. While white adipocytes are primarily involved in energy storage (storing lipids), brown and beige adipocytes are highly specialized in energy expenditure due to their higher mitochondria content and abundant expression of uncoupling protein 1 (UCP1), which detours mitochondrial proton gradient from regular ATP production to instead generate heat to warm up the body. Recent studies indicate that white and beige adipocytes are interconvertible, and the conversion of white to beige adipocytes is called browning. In this talk, I will present our latest results on the molecular regulation

of adipose browning and its implication in animal health, production and meat quality.

**Key Words:** white adipose tissue, brown adipose tissue, beige adipocyte

**670 Mitochondrial adaptations to physiological states in bovine adipose tissue.** Susanne Häussler\*, *University of Bonn, Institute of Animal Science, Physiology & Hygiene Group, Bonn, Germany.*

Mitochondria are key cellular components in energy metabolism. Their number and function varies depending on environmental, physiological or pathological conditions. The amount of mitochondria within a cell can be determined by measuring the mitochondrial DNA (mtDNA) copy number. In white adipose tissue (AT), mitochondria are important because they deliver energy for the differentiation of adipocytes. In addition, mitochondria provide key substrates in support of lipogenesis during adipogenesis. Therefore, the number of mitochondria is lower in mature adipocytes than in differentiating preadipocytes. Besides its role as an energy depot, AT is established as a major endocrine organ. Taking a focus on dairy cows, the extensive changes that AT undergoes during the course of lactation indicate alterations in mitochondrial content in that tissue. Differing needs for energy during lactation may lead to changes of mtDNA content. Being crucial for the lipogenic capacity of adipocytes, the number of mitochondria within bovine AT is thus important for the storage capacity for energy in dairy cows. In our laboratory, we investigated the number of mtDNA copies within AT by multiplex qPCR. Comparing different AT depots in dairy cows, more mtDNA copies were observed in visceral (vc) as compared with subcutaneous (sc) AT, which is in accordance with the higher metabolic activity of vcAT. Furthermore, increased mtDNA copy numbers were detected in scAT after excessive fat accretion in nonlactating, non-pregnant dairy cows. In that study, the number of mtDNA was positively associated with indicators of oxidative stress, thus pointing to an adaptive response to mtDNA damage caused by oxidative stress which resulted from increasing metabolic load. When compared with other tissues being of major importance in energy metabolism and lactation, such as liver and mammary gland, the mtDNA content in AT of high-yielding dairy cows was distinctively lower. Nevertheless, mitochondria are important for the cellular energy supply in AT of dairy cows and thus for the adequate adaptation by AT to the changes of energy balance and oxidative stress throughout lactation, that is important for animal health and efficiency.

**Key Words:** mitochondria, adipose tissue, cow

# Horse Species Symposium: Recent advances in the microbiome and physiology of the hindgut of the horse and dog

**671 Canine intestinal microbiology and metagenomics.** Jan S. Suchodolski\*, *Gastrointestinal Laboratory, Texas A&M University, College Station, TX.*

Recent molecular studies have revealed a complex microbiota in the dog intestine, consisting of thousand bacterial, fungal, and viral phylotypes. Convincing evidence has been reported linking changes in microbial communities to acute and chronic gastrointestinal inflammation, especially in canine inflammatory bowel disease (IBD). Most common microbial changes observed in intestinal inflammation are decreases in the bacterial phyla *Firmicutes* (i.e., *Lachnospiraceae*, *Ruminococcaceae*, *Fecalibacterium*) and *Bacteroidetes*, with concurrent increases in *Proteobacteria* (i.e., *E. coli*). Due to the important role of microbial-derived metabolites for host health, it is important to elucidate the metabolic consequences of gastrointestinal dysbiosis and a better understanding of the physiological pathways is helpful to potentially pinpoint specific diseases. Novel approaches such as shotgun sequencing of DNA allow characterizing functional changes in the bacterial metagenome in gastrointestinal disease. Furthermore, wide scale and untargeted measurements of metabolic products derived by the host and the microbiota in intestinal samples allow a better understanding of the functional alterations that occur in gastrointestinal disease. For example, changes in bile acid metabolism, short-chain fatty acid concentrations, and tryptophan pathways have recently been reported in humans and dogs. Also, metabolites associated with the pentose phosphate pathway were significantly altered in chronic gastrointestinal inflammation and indicate the presence of oxidative stress in dogs with IBD. Better understanding of the interactions of microbial-derived metabolites and the host will yield insights into the pathophysiology of gastrointestinal diseases. Furthermore, some of these metabolic pathways can be targeted to develop better treatment approaches.

**Key Words:** microbiome, metabolome

**672 The microbiome of the horse hindgut.** Véronique Julliard\*, *AgroSup Dijon, Dijon, France.*

The collective genomes of the different microbes (bacteria, fungi, protozoa, yeasts, archaea and bacteriophages) that live inside the horse hindgut compose its intestinal microbiome. This microbiome can be characterized by its microbial community structure (diversity and composition) and function (metabolic activities and resulting end products). It plays a vital role due to its capacity of hydrolysing and fermenting cell-walls into volatile fatty acid that represent an essential energy source for the host. Although not fully understood, it also completes important roles in health and several diseases pathophysiology. Traditionally, the microbes of the horse hindgut were studied using culture dependent techniques. More recently culture-independent methods have been used and have brought new insights. Regarding the ciliate protozoa community, 18S rRNA gene pyrosequencing allowed identifying 15 genera in the feces of pasture-fed horses. Most of these genera had been identified in previous microscopy studies. The 2 clades *Methanobrevibacter ruminantium* and *Methanocorpusculum* and their relatives were showed to dominate the fecal Archaea community using 16S rRNA gene pyrosequencing as it had been reported via cultural techniques. Regarding the bacteria community several studies have been done with molecular methods such as fingerprint techniques or more recently 16S rRNA gene pyrosequencing.

In the cecum and colon, *Firmicutes* and *Bacteroidetes* are the 2 predominant phyla. *Firmicutes* comprise *Clostridium* spp., *Ruminococcus* spp., *Butyrivibrio* spp., and *Eubacteria* spp. that play a major role in fiber degradation. The cellulolytic species *Fibrobacter succinogenes* was identified recently as the second most abundant species in the horse feces. Although a phylogenetic core bacteria community appeared to exist in all regions of the large intestine of healthy horses, strong inter-horse variations on bacteria populations are often reported. The microbiome can also be affected by several factors. The effect of the diet has been the most extensively studied on the microbial community structure and function. Understanding the microbiome changes and the factors in cause can help manipulating the microbiome to improve nutrition and health.

**Key Words:** horse, microbiome, hindgut

**673 Evaluation of the effectiveness of the establishment of an equine research herd with dual cannulation at the ileum and cecum.** Torea L. Bova\*, Cate Mochal-King, Brian J. Rude, Ben C. Weed, Lauren B. Hodge, and Molly Nicodemus, *Mississippi State University, Mississippi State University, MS.*

Cecal and ileal cannulations have been utilized to determine equine digestibility and gastrointestinal physiology, but these cannulations were limited to a single segment of the digestive tract. While these earlier cannulations were successful, the recovery process was difficult for the single cannulation; and thus, the objectives of this case study were to document the post-operative recovery of a dual cannulation surgery in equines and to determine the effectiveness of establishing a dual cannulated research herd. Eight Hackney/Shetland pony cross mares (BW: 200 to 250 kg) were selected for development of a cannulated herd. Initial incisions for placement of the ileal and cecal cannulas were done on the same day. A novel ileal cannula was constructed of a biologically compliant silicone, which included a flange to assist with surgical anchoring to improve stoma formation. For the cecal cannula, a rumen cannula (#7C; Bar Diamond, Parma, ID) was placed into the cecal fistula similar to previous studies. Ileal stomas adhered after 14 d and the cecal stoma at 10 to 14 d post-surgery. Full vital analysis occurred every 4 h for 4 wk post-surgery. Due to granulation at the cecal site, the most common complication for all ponies was increased rectal temperature of 38.9 to 40.9°C from 4 to 10 d post-surgery with a mean ranging from 37.8 to 38.5°C and a standard deviation ranging 0.59 to 1.4. Five ponies showed signs of colic approximately 1 to 2 times during recovery. After surgical recovery, 5 ponies remained healthy during 7 mo of observations, demonstrating vital signs within normal ranges and no colic signs. The 3 ponies that died had intestinal leakage into the abdominal cavity due to lack of adherence of ileum to the body wall resulting in peritonitis within 2 wk post-surgery. Compared with other tubing or polyvinyl chloride options, the current novel ileal cannula design was effective as the ileal cannula design formed a stoma and was easily maintained for the remaining 5 ponies. With a survival rate of 63% at 7 mo post-surgery, the dual cannulation of the ileum and cecum was considered successful.

**Key Words:** equine cannulation, ileum, cecum

**674 Can the microbiome of the horse be altered to improve digestion?** Josie A. Coverdale\*, *Texas A&M University, College Station, TX.*

Common management practices such as high concentrate diets, low forage quality, meal feeding, and confinement housing have an effect on intestinal function, specifically large intestinal fermentation. The microbiome of the equine large intestine is a complex and diverse ecosystem, and disruption of microflora and their environment can lead to increased incidence of gastrointestinal disorder. Digestion in the horse can be improved through a variety of mechanisms such as feedstuff selection, forage quality, feeding management, and inclusion of digestive aids. These digestive aids such as prebiotics and probiotics have been used to improve digestibility of equine diets and stabilize the microbiome of the large intestine. Probiotics, or direct fed microbials, have been widely used in horses for treatment and prevention of gastrointestinal disease. The introduction of these live, beneficial microorganisms orally into the intestinal tract has yielded variable results. However, it is difficult to compare data due to variations in choice of organism, dosage, and basal diet. While there are still many unanswered questions about the

mode of action of successful probiotics, evidence suggests competitive inhibition and enhanced immunity. Lactic acid bacteria such as *Lactobacillus*, *Bifidobacterium*, and enterococci and *Saccharomyces* yeast have all been successfully used in the horse. Use of these products has resulted in improved fiber digestibility in horses offered both high starch and high fiber diets. When high concentrate diets were fed, probiotic supplementation helped maintain cecal pH, decreased lactic acid concentrations, and enhanced populations of cellulolytic bacteria. Similarly, use of prebiotic preparations containing fructooligosaccharide (FOS) or mannanoligosaccharides (MOS) have improved DM, CP, and NDF digestibility when added to high fiber diets. Furthermore, use of FOS in horses reduced disruptions in colonic microbial populations after an abrupt change in diet and altered fecal VFA concentrations toward propionate and butyrate. Potential use of prebiotics and probiotics to create greater stability in the equine microbiome affects not only digestibility but also health of the horse.

**Key Words:** horse, microbiome, prebiotic



## Physiology and Endocrinology: Estrous synchronization and metabolism

**675 Hormonal manipulation of progesterone before initiation of an Ovsynch protocol to increase ovulatory response to the first GnRH treatment in Holstein cows.** P. D. Carvalho\*, M. C. Wiltbank, and P. M. Fricke, *Department of Dairy Science, University of Wisconsin-Madison, Madison, WI.*

Our objective was to evaluate the effect of decreasing progesterone (P4) before initiation of an Ovsynch protocol on ovulatory response to the first GnRH injection (G1) and pregnancies per AI (P/AI). Lactating Holstein cows ( $n = 800$ ) were synchronized using a Double Ovsynch protocol [Pre-Ovsynch protocol (GnRH; 7 d, PGF<sub>2α</sub>; 3 d, GnRH) followed 7 d later by an Ovsynch-56 protocol (G1; 7 d PGF<sub>2α</sub>; 24 h, PGF<sub>2α</sub>; 32 h, GnRH)] to receive first timed artificial insemination (TAI; 80 ± 3 DIM) 16 h after the last GnRH treatment. Cows were randomly assigned to receive a half-dose of PGF<sub>2α</sub> (12.5 mg dinoprost tromethamine) 2 d before G1 (TRT) or serve as untreated controls (CON). Data were analyzed by logistic regression using GLIMMIX and ANOVA with MIXED procedures of SAS. Overall, CON cows had greater ( $P < 0.01$ ) P4 than TRT cows at G1 (4.2 vs. 2.1 ng/mL). Ovulatory response to G1 was greater ( $P < 0.01$ ) for TRT vs. CON cows [81.9% (90/110) vs. 60.9% (70/115), respectively]. Luteal regression during the second Ovsynch protocol did not differ ( $P = 0.33$ ) between treatments [15.2% (15/99) vs. 10.6% (11/104); TRT vs. CON]. At 32 d after TAI, P/AI did not differ ( $P = 0.34$ ) between treatments [56.2% (223/397) vs. 52.8% (209/396); TRT vs. CON]. At 67 d after AI, P/AI also did not differ ( $P = 0.56$ ) between treatments [50.8% (190/374) vs. 48.6% (179/368); TRT vs. CON]. Pregnancy loss from 32 to 67 d after TAI did not differ [10.0% (21/211) vs. 9.6% (19/198); TRT vs. CON;  $P = 0.90$ ]. Overall, cows that ovulated to G1 had greater ( $P = 0.02$ ) P/AI compared with cows that did not ovulate [58.2% (89/153) vs. 41.5% (27/65), respectively]. The increase in P/AI in ovulating cows (16.7%) and observed increase in ovulation (21%; TRT – CON) produced an expected increase of 3.5% in P/AI in TRT vs. CON; similar to the observed 3.4% difference. Thus, administration of a half-dose of PGF<sub>2α</sub> 2 d before G1 during a Double Ovsynch protocol decreased P4 at G1 and increased ovulatory response. Larger studies are needed to determine if this modified protocol increases P/AI. Supported by USDA Hatch project 231440.

**Key Words:** fertility, timed AI, ovulation

**676 Progesterone concentration at initiation of Ovsynch and a second prostaglandin F<sub>2α</sub> treatment affect luteal regression and fertility to timed AI in lactating Holstein cows.** P. D. Carvalho\*, M. J. Fuenzalida, V. G. Santos, A. Ricci, M. C. Wiltbank, and P. M. Fricke, *Department of Dairy Science, University of Wisconsin-Madison, Madison, WI.*

Our objective was to compare luteal regression and pregnancies per AI (P/AI) after timed AI (TAI) for dairy cows receiving the first GnRH injection (G1) of an Ovsynch protocol in a low (<1.0 ng/mL) or a high (≥1.0 ng/mL) progesterone (P4) environment. Lactating Holstein cows ( $n = 851$ ) at first or second and greater AI service were randomized to receive an Ovsynch protocol (G1; 7d, PGF<sub>2α</sub>; 56h, GnRH; 16h, TAI) with 1PGF<sub>2α</sub> (PGF) treatment (1PG;  $n = 440$ ) or a modified Ovsynch protocol (G1; 7 d, PGF<sub>2α</sub>; 24 h, PGF<sub>2α</sub>; 32 h, GnRH; 16 h, TAI) with 2PGF treatments (2PG;  $n = 411$ ). Blood was collected at G1, the first PGF treatment, and the last GnRH treatment (G2) of the Ovsynch protocol for analysis of P4. To eliminate cows undergoing early luteal regression, cows ( $n = 153$ ) with P4 < 1.0 ng/mL at the first PGF treat-

ment were excluded from the analysis. Cows with P4 < 0.4 ng/mL at G2 were defined as undergoing complete luteal regression after PGF, whereas cows with P4 ≥ 0.4 ng/mL at G2 were defined as failing to undergo complete luteal regression after PGF. Data were analyzed by logistic regression using PROC GLIMMIX of SAS. Overall, more ( $P < 0.01$ ) cows with high P4 at G1 underwent luteal regression after PGF compared with cows with low P4 at G1 (92.2% vs. 82.8%, respectively). As expected, luteal regression after PGF was greater ( $P < 0.01$ ) for 2PG (96.3%) compared with 1PG (83.8%) cows. For cows with low P4 at G1, more ( $P < 0.01$ ) 2PG cows underwent luteal regression after PGF (69.6% vs. 96.2%, 1PG vs. 2PG). As a result, 2PG cows with low P4 at G1 had more ( $P < 0.01$ ) P/AI compared with 1PG cows with low P4 at G1 (35.3% vs. 57.3% for 1PG vs. 2PG cows). By contrast, for cows with high P4 at G1, although luteal regression was greater for 2PG (91% vs. 97%; 1PG vs. 2PG;  $P = 0.05$ ), P/AI did not differ (35.9% vs. 39.7%; 1PG vs. 2PG;  $P = 0.39$ ). We conclude that a second PGF injection 24 h after the first during an Ovsynch protocol increased luteal regression for cows with low or high P4 at G1 and increased P/AI for cows with low P4 at G1. Supported by USDA NIFA Hatch project 231440.

**Key Words:** Ovsynch, progesterone, fertility

**677 Effect of high or low P4 during ovulatory follicle development on fertility of dairy cows.** Joao Paulo N. Martins\*<sup>1</sup>, Dongliang Wang<sup>2</sup>, Nanheng Mu<sup>2</sup>, Guilherme F. Rossi<sup>3</sup>, Vinicius R. Martins<sup>1</sup>, Ana Paula Martini<sup>4</sup>, Gilson A. Pessoa<sup>4</sup>, and J. Richard Pursley<sup>1</sup>, <sup>1</sup>*Department of Animal Science, Michigan State University, East Lansing, MI*, <sup>2</sup>*Shuozhou Vocational and Technical College, Shuozhou City, Shanxi, China*, <sup>3</sup>*Department of Preventive Veterinary Medicine and Animal Reproduction, FCAV-UNESP, Jaboticabal, SP, Brazil*, <sup>4</sup>*Department of Large Animal Clinical Science, Universidad Federal de Santa Maria, Santa Maria, RS, Brazil.*

The objective was to determine the effect of high vs. low progesterone (P4) during early and late stages of the ovulatory follicle development in lactating dairy cows. Cows that ovulated a d 7 1st wave dominant follicle were assigned to treatments. Ovaries were manipulated to induce high (H) or low (L) circulating concentrations of P4 during 0 to 4 d (early stage) and/or 5 to 7 d (late stage) of the wave forming 4 treatments: H/H, L/L, L/H and H/L. Luteolysis was induced with PG on d 7 of the treatment period. Ovulation of the dominant follicle was induced with GnRH 56 h following PG. All cows received AI 16 h later ( $n = 559$ ). Pregnancy was determined 35 and 56 d after AI by ultrasonography and 117 d (±3) and 194 d (±3) by detection of pregnancy-specific protein B (PSPB) in milk. Data were analyzed with chi-squared analysis. Pregnancy/AI 35 d post AI was 43.2, 51.1, 53.6, and 60.4 for HH, HL, LH, and LL, respectively, and was greater for LL than HH cows. However, there was no difference on P/AI 56 d (HH = 42.4%, HL = 46%, LH = 51%, and LL = 51.1%), 117 d (HH = 38.3%, HL = 45.6%, LH = 48.3%, and LL = 48.9%), and 194 d post AI (HH = 36.8%, HL = 43.9%, LH = 44.4%, and LL = 46.3%). Pregnancy losses between 35 and 56 d post AI was 1.8, 10, 4.9 and 14.5% for HH, HL, LH, and LL, respectively, and was greater for LL than HH. There was no difference in pregnancy losses between 56 and 117 d post AI (HH = 5.8%, HL = 0%, LH = 2.8%, and LL = 2.9%) and between 117 and 194 d post AI (HH = 0%, HL = 0%, LH = 5.9%, and LL = 3.1%). Percentage of cows with double ovulations to the last GnRH was greater in LL (49%) compared with HH, HL and LH (12, 33, and 34% respectively). Cows that had double ovulation after the last GnRH had greater P/AI 35 d (45 vs. 66%) and 56 d post AI (43

vs. 57%) compared with cows with single ovulation. Pregnant losses between 35 and 56 d after AI was greater in cows with double ovulation compared with cows with single ovulation (4 vs. 14%) and tended to be greater if the ovulations were on the same ovary. In summary, low P4 during development of the ovulatory follicle increased the percentage of cows with double ovulations and P/AI 35 d post AI, but decreased embryonic survival between 35 and 56 d post AI.

**Key Words:** follicle, fertility, progesterone

**678 Effect of a second dose of prostaglandin F<sub>2a</sub> during Double-Ovsynch on successful luteolysis and fertility.** Giovanni M. Baez<sup>\*1</sup>, Rafael V. Barletta<sup>1</sup>, Alessandro Ricci<sup>1</sup>, Eduardo Trevisol<sup>1</sup>, Jerry N. Guenther<sup>1</sup>, Alvaro Garcia-Guerra<sup>1</sup>, Beatriz O. Cardoso<sup>1</sup>, Mateus Z. Toledo<sup>1</sup>, João P. Ferreira<sup>2</sup>, and Milo C. Wiltbank<sup>1</sup>, <sup>1</sup>University of Wisconsin-Madison, Madison, WI, <sup>2</sup>São Paulo State University, Botucatu, SP, Brazil.

Lack of complete regression of the corpus luteum (CL) after prostaglandin F<sub>2a</sub> (PGF) treatment may reduce fertility during timed AI (TAI) protocols. A total of 373 lactating Holstein cows (172 primiparous; 201 multiparous) were synchronized with Double-Ovsynch starting at 53 ± 3 DIM (GnRH-7d-PGF-3d-GnRH-7d-GnRH-7d-PGF-56h-GnRH-16h-TAI). At the final PGF, cows were randomized to 1 of 2 treatments: 1PGF = No additional PGF treatment; 2PGF = Second PGF treatment 24h after first PGF. Blood samples were collected at the final PGF and GnRH treatments (72 and 16 h before timed AI). Only cows with P4 above 2.0 ng/ml before PGF (n = 344) were further analyzed. Fisher's exact test and *t*-test were used to analyze categorical and continuous variables respectively, and logistic regression analysis was used to calculate probabilities of CL regression and P/AI. Primiparous cows had greater P4 than multiparous cows (7.7 ± 0.22 vs. 6.5 ± 0.18; *P* = 0.0001) before treatment. At 56 h after PGF, P4 was greater (*P* = 0.005) for 1PGF (0.4 ± 0.04) than 2PGF (0.2 ± 0.05) cows. The percentage of cows with complete CL regression (<0.5 ng/mL at 56 h after PGF) was increased by second PGF for primiparous (81.2% vs. 97.5%; 1PGF vs. 2 PGF; *P* = 0.001) or multiparous (84.4% vs. 96.7%; *P* = 0.006) cows. Cows with lower P4 at time of PGF had a reduced probability of complete CL regression with 1PGF by logistic regression (*P* = 0.02) or by comparing (*P* = 0.0016) quartile 1 (lowest P4; 2.0 to 4.8 ng/mL; 66.7%; 28/42) to the other 3 quartiles (88.1%; 118/134) which did not differ. In contrast, 2PGF cows had elevated CL regression (>95%) regardless of P4 at time of PGF (*P* = 0.60). Nonetheless, 2PGF increased CL regression for cows in both quartile 1 (66.7% vs. 95.1%; *P* = 0.0016) or quartiles 2–4 (88.1% vs. 97.6%; *P* = 0.0034). Interestingly, there was increasing P/AI with increasing P4 before PGF treatment for 2PGF (*P* = 0.02), but not 1PGF cows (*P* = 0.13). Cows with lower P4 at the time of PGF (Quartile 1) had similar P/AI for 1PGF (31.0%; 13/42) or 2PGF (31.7%; 13/41). In contrast, for quartiles 2–4 there was a tendency for decreased P/AI (*P* = 0.10) in 1PGF (44.0%; 59/134) vs. 2PGF (52.8%; 67/127) cows. Thus, treatment with a second PGF during Double-Ovsynch reduced inadequate CL regression and tended to increase P/AI, particularly in cows with greater P4 at the time of PGF treatment.

**Key Words:** prostaglandin F<sub>2a</sub>, fertility, luteolysis

**679 Differentially expressed genes in endometrium and corpus luteum of Holstein cows selected for high and low fertility are enriched for sequence variants associated with fertility.** Stephen G. Moore<sup>\*1,2</sup>, Jennie E. Pryce<sup>3</sup>, Ben J. Hayes<sup>3</sup>, Amanda J. Chamberlain<sup>3</sup>, Kathryn E. Kemper<sup>3</sup>, Donagh P. Berry<sup>1</sup>, Matthew McCabe<sup>4</sup>, Paul Cormican<sup>4</sup>, Patrick Lonergan<sup>2</sup>, Trudee Fair<sup>2</sup>, and Stephen T.

Butler<sup>1</sup>, <sup>1</sup>Teagasc, Animal and Grassland Research and Innovation Centre, Moorepark, Fermoy, Co. Cork, Ireland, <sup>2</sup>University College Dublin, School of Agriculture and Food Science, Dublin, Ireland, <sup>3</sup>Department of Economic Development, Jobs, Transport and Resources & Dairy Futures Cooperative Research Centre (CRC), Agribio, La Trobe University, Bundoora, Australia, <sup>4</sup>Teagasc, Animal and Grassland Research and Innovation Centre, Grange, Dunsany, Co. Meath, Ireland.

In this study, we combined RNA-sequence data, genome-wide association studies (GWAS) and imputed sequence data (Daetwyler et al., 2014; Nat. Genet. 46: 858–865) to identify variants associated with dairy cow fertility. We tested the hypothesis that genes differentially expressed in the endometrium and corpus luteum (CL) on d 13 of the estrous cycle between cows with either good (Fert+) or poor (Fert-) genetic merit for fertility, would identify quantitative trait loci (QTL) regions and sequence variants associated with fertility in cattle. After an adjustment for multiple testing (Benjamini and Hochberg, *P* ≤ 0.05), 9 and 560 genes were differentially expressed in the endometrium and CL, respectively, between Fert+ and Fert- cows. These differentially expressed genes (DEG) identified 93 QTL regions (*P* < 10<sup>-3</sup>) that were validated by fertility GWAS using high-density genotypes from independent dairy cattle populations in both Australia (16,794 bull and cow genotypes) and Ireland (2,660 bull genotypes), with 54% of the signals detected primarily on BTA18 (23%), 5 (9%), 7 (8%), 8 (8%) and 29 (6%). The QTL regions were primarily associated with genes involved in prostaglandin F<sub>2a</sub> (synthesis, secretion and action) and immune-related processes in the endometrium and CL. In addition, genes involved in steroidogenesis and mRNA processing were also identified in the CL. Seventeen sequence variants significantly associated with fertility (*P* ≤ 10<sup>-5</sup>) in the Australian population were identified within 2 kb upstream and downstream of DEG involved in mRNA processing or the immune system. One missense variant (SIFT value = 0.01; i.e., deleterious to protein function) significantly associated with fertility was identified in *EIF4EBP3*, a gene involved in translation initiation. The results of this study enhance our understanding of (1) the contribution of the endometrium and CL transcriptome to phenotypic reproductive performance; and (2) the genomic architecture influencing a complex trait such as dairy cow reproductive performance.

**Key Words:** fertility, corpus luteum, endometrium

**680 Liver metabolism in dairy cows during repeated short-term feed-restrictions and LPS induced systemic inflammation.** Josef J. Gross<sup>\*1</sup>, Emmanouil Kalaitzakis<sup>2</sup>, Olga Wellnitz<sup>1</sup>, Heiner Bollwein<sup>2</sup>, and Rupert M. Bruckmaier<sup>1</sup>, <sup>1</sup>Veterinary Physiology, Vetsuisse Faculty University of Bern, Bern, Switzerland, <sup>2</sup>Clinic of Reproductive Medicine, Vetsuisse Faculty University of Zurich, Zurich, Switzerland.

Body fat mobilization during a negative energy balance (NEB) requires metabolic adaptations on the part of the liver. We investigated if responses of hepatic mRNA abundancies of genes involved in metabolism and immune system to an energy deficiency differ between lactational stages. Holstein dairy cows (n = 14) (control (CON) and restricted (RES) group) were fed with grass and additional concentrate from wk 3 ap until wk 12 pp, except the RES group receiving only grass during 1-wk feed-restrictions in wk 2, 5, 8, and 11 pp. At the end of the first restriction period, LPS from *E. coli* was infused intravenously (0.5 µg/kg BW) to induce an inflammatory status. Energy balance (EB) was calculated on a weekly basis. Blood was obtained weekly and liver tissue was collected for biopsy before and after restriction periods and at 8h after the systemic LPS challenge. Blood samples were analyzed for

glucose, NEFA, and BHBA concentrations. Hepatic gene expression of 3-hydroxybutyrate dehydrogenase (BDH) 2, carnitine palmitoyltransferase (CPT) 1A and 2, mitochondrial glycerol-3-phosphate-acyltransferase (GPAM), 3-hydroxy-3-methylglutaryl-coenzyme A synthase (HMGCS) 2, cytosolic phosphoenolpyruvate carboxykinase (PEPCKc), growth hormone receptor (GHR) 1A, insulin-like growth factor (IGF) 1, Hp (haptoglobin), serum amyloid A (SAA), and tumor necrosis factor (TNF)  $\alpha$  were measured. Data were analyzed using a mixed model including group and wk as fixed effects. During restriction periods, RES had a more distinct negative EB (wk 2:  $\Delta 17$  MJ NEL/d; wk 5:  $\Delta 19$  MJ; wk 8: 6 MJ; wk 11  $\Delta 13$  MJ) and in wk 5 higher NEFA (0.64 mmol/L) and BHBA (0.65 mmol/L) concentrations compared with CON (0.39 and 0.37 mmol/L, resp.;  $P < 0.05$ ). Hepatic mRNA abundances showed expression patterns depending on the lactational stage. During feed-restriction periods, expression of IGF-1 was upregulated in RES compared with CON (based on ratios between RES and CON: +11.1 in wk 2, +14.7 in wk 5, +2.9 in wk 8, +7.3 in wk 11;  $P < 0.05$ ). At 8h after the initiation of the systemic LPS challenge, SAA, TNF $\alpha$  and Hp were downregulated in RES and CON, while GPAM, HMGCS2, GHR1A, CPT1A and 2, PEPCKc, and BDH2 were upregulated ( $P < 0.05$ ). In conclusion the experiment showed that hepatic metabolism was clearly affected by feed-restrictions and systemic LPS-challenge.

**Key Words:** feed restriction, metabolism, dairy cow

#### 681 Prediction of portal and hepatic blood flow in cattle.

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An integral part of linking a multi-organ post-absorptive model is the prediction of nutrient fluxes between organs via blood flow. This paper reports a multivariate meta-analysis approach to model portal vein blood flow (PORBF) and hepatic venous blood flow (HEPBF) simultaneously. The developmental database consisted of 296 measurements (pAH dilution) for growing and lactating cattle with 55 treatments from 17 studies, and a separate evaluation database with 31 treatment means from 8 studies. Both databases had information on feed intake, bodyweight and diet composition. Blood flows predicted with DMI or metabolizable energy intake (MEI) was tested with both linear and quadratic equations using the NLINMIX macro of SAS. Cow(study) and study were treated as random effects and blood flow location (PORBF or HEPBF) as a repeated effect. Equations based on DMI rather than MEI typically resulted in higher concordance correlation coefficient (CCC) values, indicating better predictions. Quadratic equations did not out-perform their linear counterparts (CCC analysis), and quadratic equation terms were frequently non-significant. The best predictive equations were: PORBF (L/d) =  $4855(\pm 1097) + 2007(\pm 74.8) \times \text{DMI (kg/d)}$  and HEPBF (L/d) =  $4463(\pm 1094) + 2492(\pm 74.5) \times \text{DMI (kg/d)}$ , with CCC values of 0.887 and 0.922, respectively. The residuals (predicted – observed)

for PORBF expressed as a fraction of HEPBF (PORBF/HEPBF), and hepatic arterial blood flow (ARTBF (L/d); = HEPBF – PORBF) were affected by the proportion of forage in the diet, and thus equations for PORBF and HEPBF based on forage and concentrate DMI were developed: PORBF (L/d) =  $5043(\pm 1186) + 1989(\pm 148.8) \times \text{Forage (kg DM/d)} + 1989(\pm 141.4) \times \text{Concentrate (kg DM/d)}$ , and HEPBF (L/d) =  $4416(\pm 1177) + 2223(\pm 145.5) \times \text{Forage (kg DM/d)} + 2741(\pm 137.5) \times \text{Concentrate (kg DM/d)}$ , where CCC values were 0.886 and 0.912, respectively. The CCC for ARTBF improved from 0.877 to 0.904 and PORBF/HEPBF from 0.115 to 0.447 with forage and concentrate DMI separation. Developed equations predicted blood flow well, and also suggest different sensitivity of PORBF and HEPBF to the downstream effects of DMI composition.

**Key Words:** blood flow, liver, meta-analysis

#### 682 Effects of reducing dietary cation-anion difference level on plasma Ca concentration and VDR expression level in gastrointestinal tract of transition mice. Wen-xuan Wu\*, Hai-liang Xin, Yi Yang, and Ruo-yu Liu, College of Animal Science, Guizhou Province, Guiyang, Guizhou Province, China.

The mechanism of reducing dietary cation-anion difference (DCAD; Na<sup>+</sup>K–Cl–S, mmol/kg DM) to prevent hypocalcemia is not completely known. Therefore, this study was conducted to clarify the mechanism why reducing DCAD was effective for increase in plasma Ca concentration and thus for hypocalcemia prevention by detecting plasma Ca concentration and vitamin D receptor (VDR) expression level in gastrointestinal tracts (GIT) of transition mice. One hundred twenty transition mice were randomly allocated to 3 blocks with each of 40 individuals and were fed 3 diets with varying DCAD level at +300 (treatment 1, HD), +150 (control, CON), and –150 (treatment 2, LD), respectively. Ten mice for each treatment were killed to collect blood sample for plasma Ca concentration analysis and harvest GIT tissues (stomach, duodenum, jejunum, ileum, cecum, and colon) samples for VDR expression level detection at 4 time points: day of 20 (–20) and 5 (–5) before kidding, day of kidding (0), and day of 3 after kidding (+3). Data on plasma Ca was analyzed using SAS 9.3 with Proc MIXED and VDR mRNA with Proc NPar1 Way. Plasma Ca concentration (mg/dL) in LD mice (6.37) was higher than that in HD (4.43) and CON (4.98) mice for the whole and individual blood sampling time ( $P < 0.05$ ). On d –20 diet LD resulted in higher VDR mRNA expression level in jejunum, ileum, and colon over diet HD ( $P < 0.05$ ). On d –5 mice fed LD diet had more VDR mRNA expression level in jejunum and colon relative to HD and CON ( $P < 0.05$ ). On d 0 feeding of treatment 2 had the highest VDR mRNA expression level in duodenum and colon, which was statistically higher than HD and CON ( $P < 0.05$ ). On d 3 duodenum, jejunum, and colon were observed to show increased VDR mRNA expression level for LD compared with HD and CON ( $P < 0.05$ ). These results indicated that reducing the DCAD upregulates VDR mRNA expression in GIT of transition mice and increases plasma Ca concentration, which explains some of the benefits of low DCAD on prevention of hypocalcemia.

**Key Words:** dietary cation-anion difference, plasma Ca, gastrointestinal VDR expression



## Production, Management, and the Environment III

**683 Analyzing the rear shape of dairy cows in 3D to better assess body condition score.** Amélie Fischer<sup>\*1,2</sup>, Thibault Luginbühl<sup>3</sup>, Laurent Delattre<sup>3</sup>, Jean-Michel Delouard<sup>3</sup>, and Philippe Faverdin<sup>1</sup>, <sup>1</sup>INRA/Agrocampus-Ouest UMR 1348 Pegase, St-Gilles, France, <sup>2</sup>Institut de l'élevage, Le Rheu, France, <sup>3</sup>D'Ouest, Lannion, France.

Body condition is an important trait in dairy cow management, mainly because it reflects the level and the use of body reserves and indirectly reproductive and health performance. Body condition score (BCS), which is done visually or by palpation, is the usual method on farm but is subjective and not very sensitive. The aim was here to develop and to validate 3DBCS which estimates BCS from 3D-shapes of dairy cows rear, the body area commonly used to assess BCS. For the calibration, a set of 57 3D-shapes from 56 Holstein cows with large BCS variability (0.5 to 4.75 on a 0–5 scale) were transformed with a principal component analysis (PCA). A multiple linear regression was fitted on the principal components to assess BCS. Four anatomical landmarks were extracted to normalize the 3D-shapes: the validation results of a manual labeling proved the concept. Then an automated labeling method was developed to extract them. Prior to the PCA, the 3D-shapes were either regularized to fill in the holes or not regularized. External validation was evaluated on 2 sets: one with cows used for calibration, but with a different lactation stage (valididem) and one with cows not used for calibration (validdiff). Repeatability was estimated with 6 cows scanned 8 times each the same day. The automated method performed slightly better than manual method for external validation (RMSE = 0.27 versus 0.34 for validdiff) and both were more repeatable than usual BCS ( $\sigma = 0.20$  for 3DBCS and 0.28 for BCS). Surprisingly, regularizing the 3D-shapes performed slightly less than without regularization. Nevertheless regularization should be an interesting process before BCS assessing, especially to avoid discarding too many 3D-shapes. The first results of 3D-BCS monitoring in dairy cows with a fully automated method show promising results in terms of phenotyping. The next step will try to reduce scanning time to decrease the number of bad 3D-shapes due to cow's movement without losing too much resolution.

**Key Words:** body condition score, 3D imaging, principal component analysis

**684 Modelling performance consequences on the probability of reproducing, and thereby on productive lifespan in dairy cows.** Ho N. Phuong<sup>1,3</sup>, Pierre Blavy<sup>1,3</sup>, Olivier Martin<sup>1,3</sup>, Luc Delaby<sup>2,4</sup>, Philippe Schmidely<sup>1,3</sup>, and Nic C. Friggens<sup>\*1,3</sup>, <sup>1</sup>INRA UMR MoSAR, Paris, France, <sup>2</sup>INRA UMR PEGASE, Rennes, France, <sup>3</sup>AgroParisTech, Paris, France, <sup>4</sup>AgroCampusOwest, Rennes, France.

Reproductive success is a key component of lifetime efficiency (ratio of total energy in milk to total energy intake over the lifespan) as failure to get in calf results in culling and thus has a negative effect on productive lifespan. At the animal level, breeding and feeding management can substantially affect milk yield, body condition, and energy balance of cows, which are all major contributors to reproductive failure in dairy cattle. This study developed a reproductive module that was incorporated into an existing lifetime performance model to enable prediction of the performance consequences of different breeding and feeding strategies on probability of reproducing, and thereby on productive lifespan. This then allows more realistic prediction of cow lifetime efficiency. The model is dynamic and stochastic with an individual cow being the

unit of modeling and one day being the unit of time. To evaluate the reproductive module, data from a French study including Holstein and Normande cows fed with high concentrate diet and data from a Scottish study including Holstein cows selected for high and average genetic merit for fat plus protein, fed with high versus low concentrate diets were used. On average, the model consistently simulated reproductive performance of various genotypes of cow across feeding systems. Relative to the French data, the model significantly under-predicted first service conception rate for Normande cows (48% vs. 58% for predicted vs. observed). On the Scottish data, simulated conception to first service was not significantly different from observed but interval traits (days to first service, days open) were under predicted, which was mainly due to the discrepancy between simulated and observed voluntary waiting periods. Simulation showed that genetic selection for greater milk production impaired reproductive performance and thus reproductive lifespan, but not lifetime efficiency. However, the definition of lifetime efficiency used did not include associated costs or consider herd-level effects, which should be included to allow more accurate simulation of lifetime profitability in different scenarios.

**Key Words:** dairy cow, lifetime efficiency, productive lifespan

**685 Modeling the effect of forage allowance, forage mass, and body condition on calf weaning weight and calving conception interval of primiparous cows grazing Campos grasslands.** Martín Claramunt<sup>\*1</sup>, Mariana Carriquiry<sup>2</sup>, and Pablo Soca<sup>3</sup>, <sup>1</sup>Facultad de Veterinaria, Universidad de la República, Paysandú, Paysandú, Uruguay, <sup>2</sup>Facultad de Agronomía, Universidad de la República, Montevideo, Montevideo, Uruguay, <sup>3</sup>Facultad de Agronomía, Universidad de la República, Paysandú, Paysandú, Uruguay.

The relationships among forage allowance (FA), forage mass (FM), and BCS during early (E) and middle gestation (M), calving (C) and lactation (L), and calf weight at weaning and calving conception interval (CCI) were studied employing records from an experiment that evaluated the effect 2 levels of FA on productivity of primiparous beef cows grazing Campos grassland. The study took place in Facultad de Agronomía, Uruguay (31°S 57°W). Eighty primiparous cows were assigned to a completely randomized experiment of 2 FA in spatial replication on 2 blocks during 2 years. The experiment started in autumn –150 d postpartum (dpp; early gestation [e]) and finished 190 dpp. Annual FA averaged 2.5 and 4 kg DM/kg liveweight (LW) for low (L) and high (H) FA, respectively. Cow LW and FM were measured monthly to adjust FA using the “put and take” method. The BCS was recorded (1–8 points scale). Calf birth weight (CBW) and weaning weight were recorded and calf weight adjusted at 205 d (CW) of age was estimated. Date of subsequent calving was recorded and CCI was calculated subtracting 285 d of gestation. Models were obtained by multiple regressions selected by Stepwise procedure (JMP 6.0). The BCSe, FAm, FMI, BCSe×FMI and CBW explain CW ( $CW = -8.6 + (13 \times BCSe) + (4.9 \times FAm) + (0.036 \times FMI) + (1.8 \times CBW) - [0.024 \times (BCSe - 5.6) \times (FMI - 1400)]$ ) ( $r^2 = 0.54$ ;  $P < 0.01$ ; Mean = 187; RMSE = 15). An increase in one unit of BCSe, FAe, and FMI increase CW in 13, 4.9 and 0.036 kg respectively. The interaction BCSe×FMI showed an increase in CW when BCSe increases, in FMI levels below 2000 kg DM/ha without effect on greater values. The CCI was affected by BCSe, Julian calving day (CD) and their interaction ( $CCI = 191 - (8.6 \times BCSe) - (0.6 \times CD) + \{0.4 \times [(BCSe - 4.5) \times (CD - 56)]\}$ ) ( $r^2 = 0.39$ ;  $P < 0.01$ ; Mean = 121; RMSE = 14). The BCSe mainly explains the CCI confirming his value to predict the reproductive response. Those

models contribute to the study of CW and CCI, and could be employed to predict the productive and reproductive response of primiparous beef cows grazing Campos grassland.

**Key Words:** rangeland, grazing management, cow production

**686 Associations between milk quality, type of bedding, and milking management on large Wisconsin dairy farms.** Robert F. Rowbotham\*<sup>1,2</sup> and Pamela L. Ruegg<sup>1</sup>, <sup>1</sup>University of Wisconsin-Madison, Madison, WI, <sup>2</sup>Grande Cheese Company, Brownsville, WI.

The objective of this study was to determine bedding and milking management practices associated with bulk tank (BT) quality (SCC and TBC), on large Wisconsin dairy farms. Ninety percent (325 of 360) of Wisconsin dairy farms producing in excess of 11,340 kg of milk daily participated in a personally administered survey consisting of 60 scripted questions. Milk quality test results were obtained from milk marketers for a 2-year period for 255 farms. Results were analyzed for 230 farms using the same bedding type (IB = Inorganic, MB = Manure solids, OB = Other organic) in all pens during the entire study period. Farms milked between 270 and 8,100 cows (mean = 908), selling an average of 33,714 kg daily. Farms which herd tested (n = 204) had an average RHA of 12,831 kg (IB), 11,746 kg (MB), or 11,973 kg (OB). The relationships between bulk tank somatic cell score (BTSCS), bedding type, and management practices were analyzed in a repeated measures model using PROC MIXED (SAS 9.4). Bulk tank SCS was least in the winter and spring, intermediate in the fall, and greatest in the summer with seasonal differences decreasing with increasing farm size ( $P < 0.001$ ). Farms using Iodine based postdip had greater BTSCS than those using other postdips ( $P = 0.011$ ) and BTSCS was lower on farms drying teats and wiping off predip than on those not drying teats ( $P < 0.001$ ). Farms with a WMP using MB had a greater BTSCS than those using IB or OB. The SCS for farms without a WMP was less for herds using IB as compared with herds using OB ( $P < 0.05$ ). Bulk TBC did not vary seasonally (Tukey adjusted  $P > 0.2$ ) or among bedding types (Tukey adjusted  $P > 0.75$ ).

**Table 1 (Abstr. 686).** Bulk tank SCS and SCC among farms with differing bedding types and presence of written milking protocols (WMP)

WMP	Bedding	n	SCS	SE SCS	SCC ( $\times 10^3$ )
Yes	Inorganic	122	4.06 <sup>b</sup>	0.08	210 <sup>b</sup>
Yes	Manure	17	4.55 <sup>c</sup>	0.14	293 <sup>c</sup>
Yes	Other organic	31	4.09 <sup>b</sup>	0.11	213 <sup>b</sup>
No	Inorganic	34	3.76 <sup>a</sup>	0.10	169 <sup>a</sup>
No	Manure	8	3.87 <sup>ab</sup>	0.16	183 <sup>ab</sup>
No	Other organic	18	4.30 <sup>bc</sup>	0.13	245 <sup>bc</sup>

<sup>abc</sup>Results with different superscripts within column differ (Tukey adjusted  $P < 0.05$ ).

**Key Words:** bedding, milk quality, SCC

**687 Using routinely recorded herd data to predict and benchmark herd and cow health status.** Kristen L. Parker Gaddis\*<sup>1</sup>,

John B. Cole<sup>2</sup>, John S. Clay<sup>3</sup>, and Christian Maltecca<sup>4</sup>, <sup>1</sup>Department of Animal Sciences, University of Florida, Gainesville, FL, <sup>2</sup>Animal Genomics and Improvement Laboratory, ARS, USDA, Beltsville, MD, <sup>3</sup>Dairy Records Management Systems, Raleigh, NC, <sup>4</sup>Department of Animal Science, North Carolina State University, Raleigh, NC.

Genetic improvement of dairy cattle health using producer-recorded data is feasible. Estimates of heritability are low, indicating that genetic

progress will be slow. Improvement of health traits may also be possible with the incorporation of environmental and managerial aspects into herd health programs. The objective of this study was to use the more than 1,100 herd characteristics that are regularly recorded on farm test days to benchmark herd and cow health status. Herd characteristics were combined with producer-recorded health event data. Parametric and non-parametric models were used to predict and benchmark health status. Models implemented included stepwise logistic regression, support vector machines, and random forests. At both the herd- and individual-level, random forest models attained the highest accuracy for predicting health status in all health event categories when evaluated by 10-fold cross validation. Accuracy of prediction (SD) ranged from 0.59 (0.04) to 0.61 (0.04) in logistic regression models, 0.55 (0.02) to 0.61 (0.04) in support vector machine models, and 0.61 (0.04) to 0.63 (0.04) with random forest models at the herd level. Accuracy of prediction (SD) at the cow level ranged from 0.69 (0.002) to 0.77 (0.01) for support vector machine models and 0.87 (0.06) to 0.93 (0.001) with random forest models. Results of these analyses indicate that machine-learning algorithms, specifically random forest, can be used to accurately identify herds and cows likely to experience a health event of interest. It was concluded that accurate prediction and benchmarking of health status using routinely collected herd data is feasible. Nonparametric models were better able to handle the large, complex data compared with traditional models. Further development and incorporation of predictive models into herd management programs will help to continue improvement of dairy herd health.

**Key Words:** health, machine learning, prediction

**688 Using parlor data to map liner performance.** John F. Penry\*<sup>1</sup>, Stefania Leonardi<sup>2</sup>, John Upton<sup>3,1</sup>, Paul D. Thompson<sup>1</sup>, and Douglas J. Reinemann<sup>1</sup>, <sup>1</sup>University of Wisconsin-Madison, Madison, WI, <sup>2</sup>Universita delgi Studi di Milano, Milan, Lombardia, Italy, <sup>3</sup>Animal & Grassland Research & Innovation Centre, Teagasc, Moorepark, Co. Cork, Ireland.

Liner performance can be described in terms of milking gentleness, speed and completeness of milk-out, with gentleness being the most important. It is widely accepted that peak milking speed will be increased as vacuum and the milking phase of pulsation are increased, but it is also known that raising the vacuum and b-phase duration increases teat end congestion. Increasing liner compression (LC) also results in higher milk flow rates while also elevating the risk of teat end hyperkeratosis. The aim of this experiment was to characterize the average milk flow rate of 8 liners, representing differing LC estimates, across a range of pulsation and vacuum settings. The 36-d trial involved an 80-cow herd milking 2 $\times$  at the UW-Madison Dairy Cattle Centre. The parlor was fitted with 8 commercial liners (round, triangular, vented and non-vented models), which were rotated through all stalls during the trial. Treatments were a combination of selected system vacuum and pulsation settings with a fixed 295ms d-phase. Nine treatments were used representing commercially applied settings for vacuum and pulsation applied over 3 equal periods in a central composite experimental design. Treatment settings for system vacuum level ranged from 36 to 49 kPa and pulsator ratios from 50:50 to 70:30. During the course of each 9 treatment cycle, the central point (42.3 kPa and 60:40 pulsator ratio) was applied every third day allowing for an estimate of within treatment variability. Parlor average milk flow (AMF) data were analyzed using a MIXED model in SAS 9.3. This model assessed the effect of liner, treatment, milking stall, milking time and milker. The SAS RSREG procedure was used to produce individual liner response surfaces. Liners with lower LC did not produce as high an AMF under high vacuum and long b-phase

compared with a high LC liner due to the effects of teat end congestion during each individual pulsation cycle. Knowledge of individual liner compression estimates and, where available a response surface, is highly useful for determining the optimum vacuum and pulsation settings without compromising gentleness of milking.

**Key Words:** liner, performance, compression

**689 A survey of management practices adopted by goat breeders in Azad Jammu and Kashmir (AJK), Pakistan.** Ghulam Bilal\*<sup>1</sup>, Muhammad Moaeen-ud-Din<sup>1</sup>, Muhammad Waseem<sup>1</sup>, Naveed Ullah<sup>1</sup>, James Reecy<sup>2</sup>, Muhammad Khan<sup>3</sup>, and Muhammad Yaqoob<sup>1</sup>, <sup>1</sup>Laboratories of Animal Breeding and Genetics, Faculty of Veterinary and Animal Sciences, PMAS-Arid Agriculture University, Rawalpindi, Pakistan, <sup>2</sup>Department of Animal Science, Iowa State University, Ames, IA, <sup>3</sup>Institute of Animal Sciences, Faculty of Animal Husbandry, University of Agriculture, Faisalabad, Pakistan.

A survey was carried out to gather information on current goat farming practices in 4 districts of Azad Jammu and Kashmir (Muzafarabad, Nelum Valley, Kotly, Mirpur). Survey included farmers (n = 50) of 5 goat breeds of AJK (Jattal, Buchi, Bairli, and Lambri and Kooti). Vaccination was never practiced by 42% farmers against prevailing diseases whereas

30% did vaccination occasionally and 28% on regular basis. Diseases for which vaccination was practiced were CCP (100%), enterotoxaemia (95.55%), FMD (31.03%) and HS and goat pox (3.45%). None of the farmer vaccinated against rabies and anthrax. As far as parasitic control is concerned, 30% of farmers did not deworm their flock while 44% of farmers dewormed their herds occasionally and 26% farmers did so on a regular basis. About 42% farmers were using traditional ways of treatment while other farmers got treatment for their goats from veterinary doctors on regular basis. Prevailing goat production system in the region was agro forestry (88%) followed by rangeland base system and crop livestock. Forty-eight percent of farmers had sheds for their herds, whereas 22% of farmers had only night confinement system and 40% confined their animals during winter season. Mostly farmer were relying on natural pasture (60%). Only 20% farmers used mineral mixture for their goats. Mating season of goats was April to June (84%). All respondents adopted ram selection and mating was not planned in most cases (98%). Animals were mainly marketed either to middlemen or local market (68%). The results from the present study could be used to formulate policies related to feeding, breeding and health management for overall improvement of goat production in the region.

**Key Words:** goat, Pakistan, management practices



## Production, Management, and the Environment IV

**690 Comparison of four beef production systems on carcass characteristics.** Jefferson McCutcheon\*, Steven Moeller, Henry Zerby, and Francis Fluharty, *The Ohio State University, Columbus, OH.*

The objective for the study was to compare novel beef production systems to determine the effects of 3 grass finishing regimens on carcass characteristics. This experiment was conducted at 3 research stations in Ohio over 5 yrs. The systems were: STOCK = calves born in March, weaned at 7 mo. of age, grazed in the fall, fed hay in the winter, and then grazed to 16 mo of age and slaughtered; DELAY = calves born in March, not weaned and slaughtered at 10 mo of age; FALL = calves born in September, fed hay when grazing was not possible and not weaned until slaughter in July; and FEED (control) = spring-born counterpart calves from each of the 3 research station locations weaned in October, transported to a feedlot, fed a 90% grain-10% forage diet until reaching 1.0 cm of backfat. All grass systems were conducted in all 3 locations in a randomized block experimental design (blocked by location). Each location had 12 calves and respective cows for each system. Statistics were run using SAS Proc Mixed with PDIFF for mean separation. Slaughter weight was greatest for FEED (505.0 kg, SEM 6.6 kg), followed by STOCK (421.2 kg), FALL (334.2 kg) and DELAY (312.8 kg;  $P < 0.05$ ). Hot carcass weight was greatest for FEED (299.9 kg, SEM 4.8 kg), followed by STOCK (218.4 kg), FALL (183.3 kg) and DELAY (163.7 kg;  $P < 0.05$ ). Ribeye area was greatest for FEED (71, SEM 1.7 cm<sup>2</sup>), followed by STOCK (57.5 cm<sup>2</sup>), and FALL (52.3 cm<sup>2</sup>;  $P < 0.05$ ) with no difference between DELAY (51.5 cm<sup>2</sup>) and FALL ribeye area ( $P > 0.05$ ). Backfat was greatest for FEED (1.52 cm, SEM 0.05 cm;  $P < 0.05$ ) while STOCK (0.58 cm) was not different from FALL (0.51 cm;  $P > 0.05$ ) and greater than DELAY (0.40 cm;  $P < 0.05$ ). Marbling scores were greatest for FEED (604 Modest, SEM 18;  $P < 0.05$ ) with DELAY (495 Slight) being greater than STOCKER (427 Slight) and FALL (419 Slight;  $P < 0.05$ ). Spring born calves weaned in the fall and placed on feed resulted in heavier carcasses, with greater muscling and back fat than grazing systems. Spring born calves, not weaned and slaughtered at 10 mo. produced a higher quality grade carcass than the other grazing programs. Leaner carcasses were produced from the grazing programs.

**Key Words:** beef, system, carcass

**691 BeefTracker mobile app for tracking and analysis of beef herd pasture use and location.** James W. Oltjen\*, Larry C. Forero, and Jeffrey W. Stackhouse, *University of California, Davis, CA.*

We have developed a web-based mapping platform named “BeefTracker” to provide beef cattle ranchers a tool to demonstrate that cattle production fits within sustainable ecosystems and to provide regional data to update beef sustainability lifecycle analysis. After initial identification and mapping of pastures, herd data (class and number of animals) are input on a mobile device in the field with a graphical pasture interface, stored in the cloud, and linked via the web to a personal computer for inventory tracking and analysis. Pasture use calculated on an animal basis provides quantifiable data regarding carrying capacity and subsequent beef production to provide more accurate data inputs for beef sustainability lifecycle analysis. After initial testing by university range scientists and ranchers, we have enhanced the BeefTracker application to improve automation for increased ease of use. The following have been added: ability to access and edit the BeefTracker livestock inventory while disconnected from Wi-Fi and cell service; ability to represent

portions of a pasture in BeefTracker as irrigated and non-irrigated; ability to report animal unit harvest (by pasture) calculated on an annual basis—this will provide quantifiable data regarding carrying capacity and subsequent beef production to provide more accurate data inputs for the beef sustainability lifecycle analysis; enhanced map synchronization; and improved security to allow a single individual to access multiple livestock operations without needing multiple user ids and passwords. We are now in the process of education to increase its use.

**Key Words:** cloud data storage, graphical interface, animal mapping

**692 Effect of yearling beef steer frame score, grazing sequence, and delayed feedlot entry on steer performance, carcass measurements, and system economics.** Songul Senturklu\*<sup>1,2</sup>, Douglas G. Landblom<sup>1</sup>, Robert J. Maddock<sup>3</sup>, and Steve I. Paisley<sup>4</sup>, <sup>1</sup>North Dakota State University, Dickinson Research Extension Center, Dickinson, ND, <sup>2</sup>Canakkale Onsekiz Mart Universitesi, Animal Science Department, Canakkale, Turkey, <sup>3</sup>North Dakota State University, Animal Science Department, Fargo, ND, <sup>4</sup>University of Wyoming, Animal Science Department, Laramie, WY.

Ninety-six yearling beef steers divided into 2 frame score groups and identified as small frame score (SF: n = 48, Avg. 3.40; Range 1.58–4.13; 1/2RA × 1/4LO × 1/4AN) and large frame score (LF: n = 48; Avg. 5.31; Range 4.48–6.65; 1/2SM or 1/2SH × 1/4AN × 1/4RA) were randomly assigned to replicated pens or fields (3) on May 1 to evaluate an extended grazing and delayed feedlot entry system (GRAZ) compared with a feedlot direct (FLOT) system. Growing and finishing for the FLOT treatment and final delayed finishing of the GRAZ treatment steers was at the University of Wyoming (UW), SAREC feedlot, Lingle, WY. FLOT steers were on feed 216 d and GRAZ steers were on feed 74 d. GRAZ steers grazed a sequence of native range (113 d), pea-barley intercrop (30 d), and unharvested corn (77 d) before being moved to the UW-SAREC feedlot. Grazing and feedlot total days was 294 d. Data were analyzed using the MIXED procedure of SAS. For the GRAZ-LF and GRAZ-SF steers, grazing gain, ADG, pasture grazing cost, annual forage crop expenses, and grazing cost/kg gain was 243 kg, 1.10 kg/d, \$285.05/steer, and \$1.17/kg; 211 kg, 0.96 kg/d, \$278.04, and \$1.32/kg, respectively. Compared with the LF steers in the feedlot, SF steers grew slower, consumed less feed, were equally efficient, but feed cost/kg of gain was higher. HCW for SF steers was lighter, and marbling score was greater for SF steers (Table 1). LF steer carcass value was greater and system net return was greater for the LF frame steers (Table 1). Managing grazing cost and delaying feedlot entry reduced DOF and enhanced LF and SF GRAZ system profitability.

*Contd.*

**Table 1 (Abstr. 692).** Performance data for beef steers

Item	FLOT-LF	FLOT-SF	GRAZ-LF	GRAZ-SF	SE	P-value
Days on feed	216	216	74	74		
Gain, kg	343	288	159	140	5.56	0.0001
ADG, kg	1.59	1.33	2.15	1.89	0.035	0.0001
DM feed/steer/d, kg	9.39	8.62	12.18	10.76	0.37	0.002
DM feed/kg gain, kg	5.90	6.46	5.70	5.70	0.30	0.52
Feed cost/kg gain, \$	1.88	2.07	1.38	1.39	0.018	0.001
HCW, kg	367	318	374	328	10.6	0.001
Fat depth, cm	0.89	0.99	0.64	0.82	0.074	0.05
REA, sq cm	82.6	74.8	81.3	74.8	1.35	0.004
YG	2.0	2.2	1.91	2.2	18.54	0.11
Marbling score	578	624	552	615		0.08
Carcass value, \$	1,728.55	1,515.66	2,004.38	1,763.68	57.25	0.0005
System net return, \$	67.95	-1.62	500.65	350.08		

**Key Words:** frame score, delayed feedlot entry, grazing system

**693 Housing and management practices on farms using automated calf feeders in the Midwestern United States.** Matthew Jorgensen<sup>\*1</sup>, Amber Adams Progar<sup>1</sup>, Kevin Janni<sup>1</sup>, Hugh Chester-Jones<sup>2</sup>, Jim Salfer<sup>3</sup>, and Marcia Endres<sup>1</sup>, <sup>1</sup>University of Minnesota, Saint Paul, MN, <sup>2</sup>University of Minnesota Southern Research and Outreach Center, Waseca, MN, <sup>3</sup>University of Minnesota Extension, Saint Cloud, MN.

Automated calf feeding systems are growing in popularity across the United States, yet information regarding feeder use and management is limited. This ongoing study is investigating housing and management practices on dairy farms with automated feeders. Thirty-eight Midwestern dairy farms were visited approximately every 60 d for 18 mo. Management practices data were collected using a questionnaire and calves (n = 10,185) and facilities were observed by research personnel. Of 38 calf feeding facilities, 39% were specifically constructed to house automated feeders and 61% were retrofitted; 53% were naturally ventilated barns, 39% were mechanically ventilated, and 8% were within a tunnel ventilated barn. A great majority of facilities (84%) supplemented ventilation systems with positive pressure tubes. Mean (±SD) pen size available to calves was 72.1 (±33.0) m<sup>2</sup>. Farms housed 17.6 (±7.8; range 2–63) calves per pen, allowing for 4.6 (±2.9; range 1.2–32.3) m<sup>2</sup> of space/calf and 16.7 (±6.6; range 2–38) calves per nipple station. Calves were introduced into group pens at 5.2 (±4.0; range 0–14) days of age. A total of 68% of farms fed calves reconstituted milk replacer, 24% whole milk plus replacer or protein balancer, and 8% unsupplemented whole milk. A medicated milk product was used by 76% of farms. Milk (or replacer) allowance per day at feeder introduction was 5.4 (±2.1; range 3–15) L rising to 8.3 (±2.0; range 5–15) L at its peak. Time from feeder introduction to peak milk allowance was 18.0 (±11.4; range 0–44) days. Weaning for calves on automated feeders started at 44.5 (±6.9; range 32–60) days of age and calves were fully weaned by 56.8 (±9.0; range 40–86) days. Management of automated feeding systems was highly varied among farms in this study and an investigation of the relationship between management factors and calf morbidity and mortality will provide an understanding of factors associated with improved calf performance and welfare. This project is supported by Agriculture and

Food Research Initiative competitive grant no. 2012–67021–19280 from the USDA National Institute of Food and Agriculture.

**Key Words:** automated feeder, calf management, calf housing

**694 Effect of calving interval on the economic results of dairy farms based on their typology.** Anne-Catherine Dalcq<sup>\*1</sup>, Yves Beckers<sup>1</sup>, Patrick Mayeres<sup>2</sup>, Benoit Wyzen<sup>2</sup>, and H el ene Soyeurt<sup>1</sup>, <sup>1</sup>Universit e de Li ege-Gembloux Agro-Bio Tech, Gembloux, Belgium, <sup>2</sup>Walloon Breeders Association, Ciney, Belgium.

The calving interval (CI) can influence the milk production (MP) and the economic results of a farm. This research aimed to highlight the most economically important CI, on the basis of the accounts of breeders. The data set contained 1,318 accounts spread between 2007 and 2012. Technical information such as mean CI of the herd, percent of cows with a CI of less than 380 d (m380), between 380 and 419 d (e380419), between 420 and 459 d (e420459) and more than 459 d (p459), mean MP of the herd; as well as typological information such as quantity of equivalent concentrate (CC), number of ares of grass (GR) and of corn silage (CS) per livestock unit (LU); and economic information such as mean gross margin per cow were available. The relation between CI and the gross margin showed that if a single economic optimum of CI cannot be determined, this optimum could depend on the typology of the farm. Therefore, 4 groups were created by using a multiple correspondence analysis, including quantity of equivalent CC, number of ares of GR and of CS per LU as variables. The first group was the most intensive one with a feeding based mostly on CC and CS; the second group was similar but less intensive. The third group was the most extensive with high GR consumption. The fourth group was characterized by a near absence of CS but more CC. Moreover, m380, e380420, e420459, p459 were transformed from quantitative to qualitative variables by using numerical classification. A qualitative variable CI profile was created as a summary of all these variables. In each group, MP was modeled using the different CI variables. The assumption behind this modeling was that for a typological profile, the breeder must have the highest MP to maximize the gross margin. These models showed that MP is maximized when p459 is lower than 26%, lower than 37%, above 27% for the group 1, 2, 3 respectively. For the group 4, the model with the variable CI profile suggested that the economic optimum of CI is intermediate. These results underlined that the economic optimum of CI is related to the typology of the considered farm. Studying individual data is a perspective to determine more precisely CI with the best economic results.

**Key Words:** calving interval, milk production, economic results

**695 Evaluation of winter housing systems for effects on production, udder health, BCS, hygiene, frostbite, and rumination of organic dairy cows.** Lucas S. Sjostrom<sup>\*1</sup>, Bradley J. Heins<sup>1</sup>, Marcia I. Endres<sup>2</sup>, Roger D. Moon<sup>2</sup>, and Ulrike S. Sorge<sup>2</sup>, <sup>1</sup>University of Minnesota, West Central Research and Outreach Center, Morris, MN, <sup>2</sup>University of Minnesota, Saint Paul, MN.

Certified-organic cows (n = 165), according to the USDA-National Organic Program rules, were used to evaluate the effect of 2 winter housing systems (December to April) on production, SCS, body weight, BCS, incidence of frostbite, and rumination. Cows were assigned to 2 treatments (2 replicates per group): (1) outdoor (straw pack, n = 81) or (2) indoor (3-sided compost-bedded pack barn, n = 84). There were 21 cows per replicate per year for the outdoor housing and 21 and 20 cows per replicate per year for the indoor housing. Cows calved during 2 seasons (spring or fall) at the University of Minnesota West Central

Research and Outreach Center, Morris, Minnesota, organic dairy. Milk, fat and protein production and SCS were recorded from monthly DHIA testing. Hygiene scores and BCS were recorded bi-weekly as cows exited the milking parlor. Frostbite incidence was collected monthly in year 2. The PROC MIXED of SAS was used for statistical analysis, and independent variables were fixed effects of year of study, season of calving (fall or spring), parity (1, 2, 3+), breed group, housing system, with replicate and cow nested within the interaction of housing system and season of calving as a random effect. Energy-corrected milk and SCS were not different for the indoor (15.8 kg/d, 2.75) and outdoor (15.5 kg/d, 2.88) housing systems. The BCS (3.38 for indoor cow vs. 3.08 for outdoor cows) within housing systems were not different. Indoor cows had greater ( $P < 0.05$ ) udder hygiene scores (1.73 vs. 1.45) and greater ( $P < 0.05$ ) abdomen hygiene scores (1.86 vs. 1.56) compared with outdoor cows. Incidence of clinical mastitis was greater ( $P < 0.05$ ) in indoor cows compared with outdoor cows (30% vs. 13%). Frostbite incidence was not different between indoor (34%) and outdoor (21%) cows ( $P = 0.14$ ). Daily rumination was 495 min/d for indoor cows and 474 min/d for the outdoor cows ( $P = 0.12$ ). In summary, cows housed outdoors on straw-bedded packs did not differ from cows housed in an indoor compost-bedded pack barn for production and SCS; however, the indoor cows were dirtier and had reduced udder health compared with outdoor cows.

**Key Words:** organic, outwintering, compost barn

**696 Risk factors for abnormal calf health scores on farms using automated feeders in the Midwest USA.** Matthew Jorgensen\*<sup>1</sup>, Amber Adams Progar<sup>1</sup>, Sandra Godden<sup>1</sup>, Hugh Chester-Jones<sup>2</sup>, Anne Marie de Passillé<sup>3</sup>, Jeff Rushen<sup>3</sup>, and Marcia Endres<sup>1</sup>, <sup>1</sup>University of Minnesota, Saint Paul, MN, <sup>2</sup>University of Minnesota Southern Research and Outreach Center, Waseca, MN, <sup>3</sup>University of British Columbia, Vancouver, BC, Canada.

Automated calf feeding systems are growing in popularity across the United States, yet research identifying risk factors that influence calf health is limited. This ongoing study is investigating associations between farm management, environment and housing with calf health outcomes. Thirty-eight Midwestern dairy farms were visited approximately every 60 d for 18 mo. During each visit calves ( $n = 10,185$ ) were scored by a single observer for health outcomes including attitude, secretions of the ears, eyes and nose, and cleanliness of the rear end as evidence of diarrhea (0 = normal, clean calf – 58.1% of the scores; 1 = moderate coverage of loose feces – 32.4%; 2 = significant coverage of watery fecal material – 9.4%). Risk factors for higher cleanliness score (diarrhea) were assessed using multilevel ordinal logistic regression. Variables in the analysis included pen size, group size and stocking density, farm's colostrum management, and milk diet type, amount, formulation, and bacterial count. Non-significant variables were removed using backward elimination. Variable retention was set at  $P < 0.05$ . The association between season and cleanliness score was found to be highly significant. Odds ratios indicated that winter 2012–13 (0.78), spring 2013 (0.43), summer 2013 (0.54), fall 2013 (0.72), and spring 2014 (0.47) were all associated with reduced likelihood of diarrhea compared with winter 2013–14. Each liter increase in peak milk allowance was associated with an 11.6% decrease in odds of higher

score or diarrhea (OR 0.88,  $P = 0.001$ ). The number of days taken to reach peak milk allowance was also associated with higher score (OR = 1.02,  $P = 0.006$ ). Increasing number of calves per group was associated with a small decrease in score or less diarrhea (OR = 0.99,  $P = 0.04$ ). The magnitude of association observed for greater peak milk allowance suggests that feeding calves on a higher plane of nutrition, and reaching that plane earlier, may be beneficial in reducing observed diarrhea. This project is supported by Agriculture and Food Research Initiative competitive grant no. 2012-67021-19280 from the USDA National Institute of Food and Agriculture.

**Key Words:** automated feeder, calf health, calf nutrition

**697 Management characteristics of cow-calf, stocker, and finishing operations in the North and South Plains.** Senorpe Asem-Hiablie\*<sup>1</sup>, C. Alan Rotz<sup>1</sup>, Robert C. Stout<sup>1</sup>, Jasmine A. Dillon<sup>2</sup>, and Kimberly R. Stackhouse-Lawson<sup>3</sup>, <sup>1</sup>USDA-ARS PSWMRU, University Park, PA, <sup>2</sup>The Pennsylvania State University, University Park, PA, <sup>3</sup>National Cattlemen's Beef Association, Centennial, CO.

Regional surveys of cow-calf, stocker, and finishing operations are being conducted nationwide to gather information on cattle, crop, and range management practices needed for a comprehensive life cycle assessment (LCA) of beef production in the United States. The South Plains (Kansas, Oklahoma, and Texas) and the North Plains (Nebraska, South Dakota, and North Dakota) make up 2 of 7 regions demarcated for the LCA and are the focus of the present study. A total of 633 ranch responses represented 1.4% of the beef cows maintained across both regions, with 0.9 and 2.8% represented in the South and North Plains, respectively. Notable differences in management practices among cow-calf and stocker ranches in both regions were observed. Mean stocking rates decreased from the wetter east to the drier, semi-arid west. In the South Plains, mean stocking rates decreased from 2.4 ha/cow (1.3 ha/stocker) in the east to 15.7 ha/cow (4.6 ha/stocker) in the west and the North Plains reported 2.9 ha/cow (1.9 ha/stocker) in the east and 6.7 ha/cow (4.3 ha/stocker) in the west. Differences in forage management influenced by varying soil morphology and rainfall patterns, showed decreasing fertilizer and lime use from east to west in both regions. A higher percentage of ranches in the North Plains (57%) produced a variety of feed crops including corn and alfalfa to feed cattle compared with 17% in the South Plains. Although the proportion of ranches that harvested rangeland as hay were similar at 42% and 47% in the South and North Plains, respectively, the portion of land harvested was lower in the South (2.5%) than in the North Plains (13%). Responses from 60 feedyards represented 9% of cattle finished in the South Plains, 4% in the North Plains, and 7.5% overall. The primary difference in feedyard management identified across regions and among states was size. Feedyard capacities increased from north to south with the largest located in Texas. The data collected are being used to develop representative operations in each state to serve as basis for a comprehensive national LCA studying the environmental, economic, and social impacts of beef production and to identify opportunities for improvement.

**Key Words:** beef production, sustainability, life cycle assessment



## Ruminant Nutrition: Lactation responses

**698 Effect of a phytogetic feed additive on feed intake and milk production in dairy cows.** Carina Schieder\*<sup>1</sup>, Annamaria Boczonadi<sup>1</sup>, and Bryan Miller<sup>2</sup>, <sup>1</sup>BIOMIN Holding GmbH, Herzogenburg, Austria, <sup>2</sup>BIOMIN USA Inc., San Antonio, TX.

Fifty-five Holstein Friesian cows were assigned according to previous milk performance and days-in-milk into control (n = 27) or experimental group (n = 28) on a commercial farm in Czech Republic. Animals were housed in groups including one milking robot in each building. Experimental cows received a partly mixed ration (PMR) with a phytogetic feed additive (PFA) including herbs, spices, essential oils and plant extracts [Digestaron Dairy (3 g/head/day), Biomin PhytoGENICS GmbH, Germany]. Control cows received PMR without any supplementation during the trial period of 90 d. Both groups received additional concentrates based on wheat, barley, corn and soybean extraction meal according to their milk performance in the milking robots. Feed intake was recorded per group. Feed refusals were evaluated once daily. Milk performance was recorded each time visiting the milking robot, while milk solids were determined 3 times during the trial period. Health status and medical treatments were recorded throughout the trial. Data of daily milk performance were statistically analyzed using the independent *t*-test (IBM SPSS 21.0). PFA addition in the PMR of dairy cows resulted in a slightly higher dry matter intake (44.23 vs. 43.88 kg/d). The PFA supplemented group produced higher quantities of milk fat (1.27 vs. 1.19 kg/d; *P* > 0.05), milk protein (1.20 vs. 1.18 kg/d; *P* > 0.05) and lactose (1.75 vs. 1.67 kg/d; *P* > 0.05). Animals receiving the PFA showed a significantly higher milk production (35.37 vs. 33.67 kg/d; *P* < 0.001). Increased daily milk yield and enhanced quantity of milk solids resulted in a superior amount of energy-corrected milk (33.01 vs. 31.89 kg/d; *P* > 0.05). In conclusion, the tested PFA was able to improve feed intake and milk performance, hence resulting in an increased farm benefit.

**Key Words:** dairy cow, performance, phytogetic feed additive

**699 Fractionated harvest and alkali treatment of whole-plant corn silage fed to lactating dairy cattle can increase intake and production.** David E. Cook, Robb W. Bender\*, Kevin J. Shinnors, and David K. Combs, *Department of Dairy Science, University of Wisconsin-Madison, Madison, WI.*

The objective of this study was to evaluate alkali treatment of whole plant corn and stalks fractionated from the whole plant at harvest as feeds for lactating dairy cows. A forage harvester with a 6-row modified ear-snapping head was setup to snap the corn ear from 2 rows and cut 4 rows at a height of 82 ± 5cm, resulting in a feed product called toplage. The toplage used in this study was 82% of the DM yield from the field and contained 44% starch. The remaining stalks were harvested with a forage harvester at a height of 12 ± 2 cm on a second pass. Stalks were treated with calcium hydroxide at a rate of 70 g·kg<sup>-1</sup> DM and ensiled. A second treatment was comprised of whole plant corn harvested at 28 ± 3cm and treated with calcium hydroxide at a rate of 70 g·kg<sup>-1</sup> DM, then ensiled. Conventional corn for the negative control diet (CON) was harvested at 26 ± 3cm from the same fields. BMR corn for the positive control diet was harvested from an adjacent field at a height of 26 ± 5cm. One hundred twenty-eight Holstein cows were stratified among 4 treatments in a randomized complete block design. Data were analyzed using the MIXED procedure of SAS, with treatment, week, and relevant interactions as fixed effects. Diets were TMR containing 40% corn silage, 20% alfalfa silage, and 40% concentrate. Treatments were

complete substitution of corn silage with a negative control (CON), positive control (BMR), treated whole plant (TRTCS), and a combination of toplage (82% of corn silage) and stalklage (18% of corn silage) (TOP). The TOP and BMR diet resulted in a similarly increased ECM over the CON diet (*P* < 0.01), and the TRTCS resulted in the highest ECM.

**Table 1 (Abstr. 699).** Effects of treatment on DMI, production, and rumination

Item	CON	BMR	TOP	TRTCS	SED	<i>P</i> -value
DMI	23.6 <sup>c</sup>	24.2 <sup>bc</sup>	24.8 <sup>ab</sup>	25.5 <sup>a</sup>	0.5	0.02
Milk, kg·d <sup>-1</sup>	43.1 <sup>b</sup>	47.1 <sup>a</sup>	45.6 <sup>a</sup>	48.0 <sup>a</sup>	1.0	<0.01
Fat, %	3.40	3.30	3.32	3.37	0.13	0.84
Fat, kg·d <sup>-1</sup>	1.47 <sup>b</sup>	1.55 <sup>b</sup>	1.54 <sup>b</sup>	1.66 <sup>a</sup>	0.05	<0.01
Protein, %	2.97 <sup>a</sup>	2.96 <sup>a</sup>	2.94 <sup>a</sup>	2.84 <sup>b</sup>	0.04	0.02
Protein, kg·d <sup>-1</sup>	1.29 <sup>b</sup>	1.40 <sup>a</sup>	1.38 <sup>a</sup>	1.39 <sup>a</sup>	0.03	<0.01
ECM	41.9 <sup>c</sup>	45.0 <sup>b</sup>	44.7 <sup>b</sup>	47.0 <sup>a</sup>	0.8	<0.01
ECM DMI <sup>-1</sup>	1.79	1.85	1.77	1.83	0.04	0.31
BW change, kg·wk <sup>-1</sup>	0.30	0.29	0.36	0.35	0.23	0.99
BCS	2.79	2.80	2.86	2.84	0.03	0.26
Rumination, min·d <sup>-1</sup>	510 <sup>a</sup>	474 <sup>bc</sup>	487 <sup>ab</sup>	459 <sup>c</sup>	10	0.01

**Key Words:** corn silage, alkali, fractionated

**700 Effect of abomasal infusions of saturated fatty acids differing in chain length on milk production, composition, and fatty acid profile in Holstein dairy cows.** Daniel E. Rico\*<sup>1</sup>, Jair E. Parales<sup>2</sup>, Ben A. Corl<sup>3</sup>, Andrea Lengi<sup>3</sup>, P. Yvan Chouinard<sup>1</sup>, and Rachel Gervais<sup>1</sup>, <sup>1</sup>Université Laval, Quebec, QC, Canada, <sup>2</sup>Universidad Nacional de Colombia, Bogotá, DC, Colombia, <sup>3</sup>Virginia Polytechnic Institute and State University, Blacksburg, VA.

Fat supplements are commonly fed to increase dietary energy density and improve animal performance. However, metabolic effects may vary depending on fatty acid (FA) chain length. Eleven multiparous Holstein dairy cows (150 ± 52 DIM; Mean ± SD) were randomly assigned to treatment sequence in a replicated 3 × 3 Latin Square design testing the effect of saturated fat supplements differing in chain length on milk production and composition, and on milk FA profile. Treatment periods were 7 d in length and were separated by 7 d of washout. Treatments were administered as abomasal infusions of emulsions providing 280 g/d of FA from: 1) free FA enriched in palmitic acid (>85% 16:0; PA), 2) free FA enriched in stearic acid (98% 18:0; SA), or 3) medium chain triglycerides (50% 8:0 and 50% 10:0; MCT). Milk yield was recorded and samples taken on the last 3 d of each period. The statistical model included the random effects of period, cow and sequence, and the fixed effect of treatment. Preplanned contrasts tested were PA vs. SA, and PA vs. MCT. Fat- and energy-corrected milk were not affected by treatments, and averaged 36.0 ± 2.0 kg/d and 35.1 ± 2.0 kg/d, respectively (*P* > 0.11). Milk fat content was higher in PA (4.49%) than in SA (4.18%, *P* < 0.001), but was not different between PA and MCT (4.37, *P* = 0.17). Similarly, milk fat yield was increased by PA (1.56 kg/d) relative to SA (1.46 kg/d, *P* = 0.01) and tended to be higher in PA than in MCT (1.48 kg/d; *P* = 0.06). De novo synthesized FA were higher in both SA and MCT (27.3 and 30.5% of FA, respectively) compared with PA (24.5% of FA; *P* < 0.001). The sum of 16:0 and cis-9 16:1 was higher in PA (43.7% of FA) relative to MCT and SA (38.0 and 35.6, respectively; *P* < 0.001). Lastly, the concentration of 18:0 plus cis-9 18:1 was increased by SA (31.2% of FA) relative to PA (26.6% of FA; *P* < 0.001), but

was not different between PA and MCT (25.8%,  $P = 0.42$ ). The chain length of fat supplements infused into the abomasum affected milk fat and fatty acid profile.

**Key Words:** dairy cow, chain length, fat supplement

### 701 Effect of *Bacillus pumilus* on early lactation performance of dairy cows fed low or high starch diets postpartum.

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Strains of *Bacillus pumilus*, a spore-forming gram-positive bacterial species can be highly resistant to environmental stresses and have the potential to produce xylanase. Xylanase increases fiber digestion and feed efficiency which might benefit cows during early lactation. This study aimed to investigate the effects of low (20%, LS) or high (27%, HS) dietary starch concentration and *Bacillus pumilus* 8G-134 (BP) supplementation during early lactation. We hypothesized that BP would increase DMI and milk yield of cows fed LS or HS diets postpartum. Forty-four ( $n = 11/\text{treatment}$ ) multiparous cows dried-off 42d before expected calving date were assigned to a common prepartum diet. At calving, cows were fed a LS or HS lactation diet and BP carrier or BP was top dressed on the TMR once daily to provide  $5 \times 10^9$  cfu/cow/day until 112 DIM. Factors combined produced 4 treatments: LS+carrier (LSCO); LS+BP (LSBP); HS+carrier (HSCO); HS+BP (HSBP). Blood samples collected on d 7, 14, 21 and 28 after calving were analyzed for NEFA and BHBA concentrations. Data were analyzed using the MIXED procedure of SAS. No changes were observed on postpartum DMI and 3.5%FCM yield; however, NEFA was lower and BHBA tended to be lower in serum of BP vs. Control. Results indicated that BP supplementation during early lactation decreased body lipid mobilization. We hypothesize that greater amounts of digestible energy available from fiber digestion might have caused lower lipid mobilization in cows fed BP. Future research should further investigate the use of BP as a feed additive because it appears to modify blood metabolite concentrations in early lactation.

**Table 1 (Abstr. 701).** Body condition, lactation performance, and blood metabolites in early lactation

Item	Treatment				SEM	P-value		
	LS		HS			S	BP	S × BP
	CO	BP	CO	BP				
DMI, kg/d	18.34	18.68	17.18	19.86	0.80	0.16	0.25	0.98
EB, Mcal/d	-10.66	-10.18	-9.29	-7.52	1.83	0.25	0.51	0.71
3.5% FCM, kg/d	39.98	39.87	40.72	42.25	1.78	0.35	0.67	0.62
NEFA, $\mu\text{Eq/L}$	363.95	311.66	475.85	314.84	68.80	0.29	0.05	0.33
BHBA, mg/dl	12.76	10.27	14.52	8.02	2.57	0.92	0.07	0.41

**Key Words:** *Bacillus pumilus*, dietary starch, early lactation

### 702 Effects of direct-fed *Bacillus pumilus* 8G-134 fed pre- and postpartum on feed intake, milk yield, milk composition, and feed efficiency of Holstein cows.

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The usage of direct-fed microbials (DFM) has become common in the dairy industry, but questions regarding their value and mode of action on dairy cows performance remain prevalent. The objective of this study was to evaluate the effects of DFM (*Bacillus pumilus* 8G-134) on pre-

and postpartum performance. Forty-three multiparous Holstein cows were fed a total mixed ration (TMR) according to NRC (2001) recommendations and assigned to 2 treatments in a randomized completely block design. Cows in the direct-fed microbial treatment (DFMt,  $n = 21$ ) received  $5.0 \times 10^9$  cfu of *B. pumilus* 8G-134 direct-fed microbial in 28g of maltodextrin carrier, whereas cows in the control treatment (CON,  $n = 22$ ) received 28g of maltodextrin carrier alone. Treatments were top-dressed on the TMR daily. Treatments were applied from 21 d before expected calving date to 154 d after calving. Intake from each cow was measured and dry matter intake was recorded daily. Milk weights were recorded daily, and milk samples were obtained weekly. Composite milk samples were analyzed for fat, protein, lactose, urea N (MUN), total solid and somatic cell count (SCC). Statistical analysis was performed using the MIXED procedures of SAS. Cows on DFMt had higher milk yield ( $41.2 \pm 1.34$  vs.  $37.8 \pm 1.33$  kg;  $P = 0.02$ ), fat corrected milk ( $48.5 \pm 1.48$  vs.  $42.0 \pm 1.49$  kg;  $P = 0.01$ ), energy-corrected milk ( $46.6 \pm 1.40$  vs.  $40.9 \pm 1.38$  kg;  $P = 0.01$ ), fat production ( $1.85 \pm 0.06$  vs.  $1.57 \pm 0.06$  kg;  $P = 0.03$ ), and protein production ( $1.27 \pm 0.04$  vs.  $1.18 \pm 0.04$  kg;  $P = 0.02$ ) on the second week of lactation than CON. There were no differences ( $P > 0.27$ ) between treatments for milk yield, fat, protein, lactose, total solids, MUN or SCC overall. Dry matter intake, BW, body condition score (BCS) were not affected ( $P > 0.35$ ) by DFMt supplementation. Cows on DFMt tended ( $P = 0.06$ ) to have higher feed conversion (ECM/DMI;  $2.02 \pm 0.04$ ) than cows fed CON ( $1.92 \pm 0.04$ ). In conclusion, cows fed DFMt tended to have higher feed conversion than CON with no detrimental consequences (i.e., BCS).

**Key Words:** direct-fed microbial, feed efficiency, *Bacillus pumilus*

### 703 Effects of fecal contents of aluminum (Al), iron (Fe), and manganese (Mn) on the apparent digestibility of phosphorus in dairy cows.

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To test if the apparent absorption of phosphorous (P) is reduced by aluminum (Al), iron (Fe), and manganese (Mn), relationships between the fecal contents of these minerals and the apparent digestibility (ADC) of P in dairy cows were determined. The study involved 10 commercial dairy farms in Manitoba, and 10 early and peak lactation, 10 mid lactation and 10 late lactation cows on each farm. Farms were visited once to collect diet, feed and fecal grab samples of individual cows. Feed, diet, and feces samples were analyzed by ICP for P and for the internal marker acid insoluble ash (AIA). Feces samples also were analyzed for Al, Fe, and Mn by ICP. Apparent digestibility coefficients (ADC) of P for individual cows were determined from the contents of P and AIA in the diet and the feces. Fecal Al contents of individual cows ranged from 2.7 to 36.8 ppm, with an average of 9.8 ppm (DM basis). Fecal Fe contents of individual cows ranged from 5.5 to 34.5 ppm, with an average of 11.6 ppm (DM basis). Fecal Mn contents of individual cows ranged from 0.6 to 5.6 ppm, with an average of 2.3 ppm (DM basis). These concentrations do not indicate that the fecal Al, Fe, and Mn contents were excessive. The ADC of P ranged from 15.9 to 59.7% among individual cows with an average of 35.1%. The fecal Al and Fe contents were correlated ( $r = 0.60$ ,  $P < 0.0001$ ), but the fecal contents of Mn were not correlated with those of Al ( $P = 0.91$ ) and Fe ( $P = 0.65$ ). The regression equation between the ADC of P and the fecal contents of Al, Fe, and Mn (in ppm) determined by SAS Mixed Procedure was  $\text{ADC P} = 27.51(\pm 2.79, \text{estimate} + \text{SE}) + 1.18(\pm 0.15) \times \text{Al} - 1.11(\pm 0.19) \times \text{Fe} + 1.73(\pm 0.93) \times \text{Mn}$  ( $R^2 = 0.31$ ). This shows that, as expected, the increases in fecal Fe contents were associated with decreases in the ADC of P, but that for the fecal Al and Mn contents the opposite occurred. These discrepancies may be

the result of the confounding of these fecal contents with other factors that affect the ADC of P and variation in the form that the dietary Al, Fe, and Mn contents were in. Nevertheless, results suggest that at the levels observed in our study the fecal contents of Al, Fe, and Mn may affect the digestibility of P.

**Key Words:** aluminum, phosphorus, iron

**704 Effects of feeding diets differing in dietary cation-anion difference (DCAD) and source of vitamin D on Ca status, health, and lactation performance in Holstein cows.** N. Martinez<sup>\*1</sup>, R. Rodney<sup>2</sup>, R. M. Santos<sup>1</sup>, L. F. Greco<sup>1</sup>, R. S. Bisinotto<sup>1</sup>, E. S. Ribeiro<sup>1</sup>, L. L. Hernandez<sup>3</sup>, C. D. Nelson<sup>1</sup>, E. Block<sup>4</sup>, I. J. Lean<sup>2</sup>, and J. E. P. Santos<sup>1</sup>, <sup>1</sup>University of Florida, Gainesville, FL, <sup>2</sup>SBSibus, Camden, NSW, Australia, <sup>3</sup>University of Wisconsin, Madison, WI, <sup>4</sup>Arm and Hammer Animal Nutrition, Princeton, NJ.

Objectives were to evaluate the effects of feeding diets differing in DCAD (PD = +130 vs. ND = -130 mEq/kg) and source of vitamin D3 prepartum (CH; cholecalciferol vs. CA; calcidiol; both fed at 0.27 mg/kg of diet DM) on Ca status, health, and lactation performance in dairy cows during the transition period. Seventy-nine Holstein cows, 51 parous and 28 nulliparous, at 255 d gestation were blocked by parity, and randomly assigned to 1 of 4 dietary treatments arranged as a 2 × 2 factorial that were fed for the last 21 d prepartum. Dry matter intake (DMI) was measured from 21 d pre- to 42 d in milk (DIM). Prevalence of subclinical hypocalcemia (SCH) was evaluated in the first 3 DIM and defined as blood ionized Ca <1.06 mM. Incidence of diseases was monitored daily for the first 30 DIM. Milk yield and composition were recorded for the first 49 DIM. Data were analyzed using PROC MIXED and PROC GENMOD of SAS. Feeding CA compared with CH increased ( $P < 0.01$ ) prepartum plasma concentrations of 25OH-D<sub>3</sub> (237.0 vs. 59.5 ± 6.4 ng/mL), 24,25(OH)<sub>2</sub>-D<sub>3</sub> (19.2 vs. 1.6 ± 1.0 ng/mL), and 1,25(OH)<sub>2</sub>-D<sub>3</sub> (55.3 vs. 48.4 ± 1.7 pg/mL), but decreased that of cholecalciferol (1.7 vs. 15.3 ± 0.5 ng/mL). Feeding ND increased prepartum concentration 1,25(OH)<sub>2</sub>D<sub>3</sub> (57.2 vs. 46.9 ± 1.7 pg/mL). The ND diet reduced ( $P < 0.01$ ) prepartum DMI in multiparous (11.5 vs. 13.7 ± 0.4 kg/d) but not in primiparous cows (11.3 vs. 11.0 ± 0.5 kg/d). Feeding the ND diet reduced ( $P < 0.05$ ) the prevalence of SCH at 0 and 1 DIM (20.0% and 34.3%) compared with PD (69.3% and 76.5%). Incidence of clinical hypocalcemia (milk fever) was 0% in ND compared with 23.1% in PD cows ( $P < 0.05$ ). Feeding CA compared with CH reduced ( $P < 0.05$ ) the incidences of retained placenta (2.5 vs. 30.8%) and metritis (23.1 vs. 46.2%). Cows fed prepartum ND and CA had reduced ( $P = 0.04$ ) morbidity compared with all other 3 treatments. Cows fed CA produced

3.70 ± 1.2 kg/d more ( $P < 0.04$ ) 3.5% fat- and energy-corrected milk than those fed CH. The use of prepartum ND in combination with CA improved Ca status, health, and lactation performance in dairy cows.

**Key Words:** dairy cow, vitamin D, DCAD

**705 Effects of the interaction between photoperiod and nutritional management on milk yield for dairy cows.** Oswaldo S. Espinoza\* and Masahito Oba, University of Alberta, Edmonton, Alberta, Canada.

The objective of this study was to evaluate the interaction effects of photoperiod management and nutritional management on milk production of lactating dairy cows. We hypothesized that feeding a high grain diet increases milk yield of cows exposed to long photoperiod to a greater extent compared with those exposed to short photoperiod. The study was conducted at a tie-stall barn with metal halide light fixtures. Thirty mid-lactating cows (109 ± 37 d in milk; mean ± SD) were exposed to long photoperiod (LP; 16 h/d light; n = 15) or short photoperiod (SP; 8 h/d light; n = 15). After 30 d of light adaptation, cows within each photoperiod treatment were fed 3 diets containing steam rolled barley grain at 15, 25 and 35% of dietary dry matter (LG, MG, and HG, respectively) in a 3 × 3 Latin square design. Dry matter intake was greater for cows fed the HG diet compared with those fed the LG diet (22.7 vs. 20.4 kg/d;  $P < 0.001$ ) whereas it was not affected by photoperiod treatment. The interaction effects between photoperiod and dietary treatments were significant for yields of milk ( $P = 0.03$ ), milk fat ( $P < 0.01$ ), and milk protein ( $P = 0.02$ ). Feeding the HG diet increased milk yield by 4.0 kg/d compared with the LG diet for cows exposed to SP treatment (30.5 vs. 26.5 kg/d,  $P < 0.05$ ) whereas the increase was 2.2 kg/d for cows on the LP treatment (30.4 vs. 28.2 kg/d,  $P < 0.05$ ). Likewise, the HG diet increased milk protein yield compared with the LG diet to a greater extent for cows on the SP treatment (1.03 vs. 0.84 kg/d,  $P < 0.05$ ) than those on the LP treatment (0.99 vs. 0.90 kg/d,  $P < 0.05$ ). However, dietary grain content did not affect milk fat yield for cows on the LP treatment while the HG diet increased milk fat yield compared with the LG diet for cows on the SP treatment (1.25 vs. 1.10 kg/d,  $P < 0.05$ ). These results indicate that milk production responses to high grain diets may be greater for cows exposed to short photoperiod, and that dietary grain content can be reduced without negatively affecting milk fat yield if lactating dairy cows are exposed to long photoperiod.

**Key Words:** photoperiod, grain, interaction.



## Ruminant Nutrition: Mineral nutrition

**706 Organic trace minerals during the transition period: 2. Supplemental Zn, Mn, and Cu from Availa Mins and Co from CoPro have a positive effect on systemic and hepatic indicators of inflammation and oxidative stress in Holstein cows.** Fernanda Batistel\*<sup>1</sup>, Johan S. Osorio<sup>1</sup>, Jin Ji<sup>1</sup>, Erminio Trevisi<sup>2</sup>, James K. Drackley<sup>1</sup>, Michael T. Socha<sup>3</sup>, and Juan J. Loo<sup>1</sup>, <sup>1</sup>University of Illinois, Urbana, IL, <sup>2</sup>Università Cattolica del Sacro Cuore, Piacenza, Italy, <sup>3</sup>Zinpro Corporation, Eden Prairie, MN.

The periparturient dairy cows undergo a state of reduced liver function linked with increased inflammation and oxidative stress. Objectives were to evaluate hepatic mRNA expression and systemic biomarkers of inflammation and oxidative stress in cows receiving supplementation of inorganic (INO) or organic (AAC) trace minerals. Twenty multiparous cows were assigned in a randomized complete block design to a common preparturient (1.5 Mcal/kg DM, 15% CP) and postparturient (1.76 Mcal/kg DM, 18% CP) diet. Both diets were partially supplemented with an inorganic mineral mix of Zn, Mn, and Cu to supply 35, 45, and 6 ppm, respectively, of the total diet DM. Treatments included cows receiving an oral bolus containing a mix of INO (n = 11) or AAC (n = 9) Zn, Mn, Cu, and Co to achieve 75, 65, 11, and 1 ppm, respectively, in total diet DM. Organic trace minerals were supplied via Availa Zn, Availa Mn, Availa Cu, and CoPro (Zinpro Corp., Eden Prairie, MN). Liver biopsies were collected at d -30, -15, 10 and 30, and blood samples at d -30, -14, 3, 15, and 30 relative to parturition. Although AAC did not affect biomarkers of liver function, an interaction (diet × time;  $P < 0.05$ ) for the concentration of albumin, glutamic-oxaloacetic transaminase (GOT), cholesterol (CHO) and  $\gamma$ -glutamyltransferase (GGT) was observed. The lower GGT at d 15 and 30, and GOT at d 15 in cows receiving AAC suggests a lower degree of liver cell damage. Furthermore, greater albumin concentration at d 15 and 30 ( $P < 0.05$ ), and greater gene expression of albumin (*ALB*;  $P < 0.01$ ) and interleukin 10 (*IL10*;  $P = 0.03$ ) in cows given AAC suggest a less pronounced inflammatory status. An interaction (diet × time;  $P < 0.05$ ) was observed for signal transducer and activator of transcription 3 (*STAT3*) and superoxide dismutase 2 (*SOD2*) because expression increased from d 10 to 30 in cows fed AAC compared with INO. Overall, these data provide some evidence that source of trace mineral during the periparturient period alters immunometabolism.

**Key Words:** immunometabolism, trace mineral

**707 Organic trace minerals during the transition period: 3. Favorable alterations in blood neutrophil (PMN) and endometrial inflammatory and oxidative status in Holstein cows supplemented with Zn, Mn, and Cu from Availa Mins and Co from CoPro.** Fernanda Batistel\*<sup>1</sup>, Johan S. Osorio<sup>1</sup>, Cong Li<sup>2</sup>, Ed F. Garrett<sup>1</sup>, Mohamed M. Elhanafy<sup>1</sup>, Jessica Caputo<sup>3</sup>, James K. Drackley<sup>1</sup>, Michael T. Socha<sup>4</sup>, and Juan J. Loo<sup>1</sup>, <sup>1</sup>University of Illinois, Urbana, IL, <sup>2</sup>China Agricultural University, Beijing, China, <sup>3</sup>University of Milan, Milan, Italy, <sup>4</sup>Zinpro Corporation, Eden Prairie, MN.

Objective was to evaluate mRNA expression of genes associated with inflammation, oxidative stress, eicosanoid synthesis and transcription regulation in PMN and endometrium in cows receiving inorganic (INO) or organic (AAC) trace minerals. Twenty multiparous cows were allocated in a randomized complete block design, and received the same preparturient (1.5 Mcal/kg DM, 15% CP) and postparturient (1.76 Mcal/kg DM, 18% CP) diet. The diets were partially supplemented with an inorganic mineral mix of Zn, Mn, and Cu to supply 35, 45,

and 6 ppm, respectively, of the total diet DM. Cows were assigned to an oral bolus administration containing a mix of either INO (n = 11) or AAC (n = 9) Zn, Mn, Cu, and Co to achieve 75, 65, 11, and 1 ppm, respectively, in total diet DM. Treatments began on d -30 and continued through d 30 relative to parturition. Organic trace minerals were supplied via Availa Zn, Availa Mn, Availa Cu, and CoPro (Zinpro Corp., Eden Prairie, MN). Endometrial biopsies were collected at d 14 and 30 postparturient. In PMN, expression of DEAD box polypeptide 58 (*DDX58*) was greater and Z-DNA binding protein 1 (*ZBP1*) was lower in cows supplemented with AAC. In addition, feeding AAC increased expression of genes related to oxidative stress (*MPO*) and eicosanoid metabolism (*PLA2G4A*, *ALOX5AP*). In endometrium, feeding AAC increased expression of genes related to oxidative stress (*SOD1*, *NRF2*), eicosanoid metabolism (*PTGES*) and inflammation (*MYD88*, *STAT3*, *IL10*). An interaction was observed for *IL6* (inflammation) because expression increased from d 14 to 30 in cows fed INO compared with AAC. The decrease in *IL1B*, *IL10*, *IL8* (inflammation) and *PTGES* (eicosanoids) expression on d 30 was more pronounced for cows fed AAC, whereas *PPARG* (an anti-inflammatory transcription regulator) increased on d 30. Overall, data demonstrate favorable alterations in PMN and endometrium inflammatory and oxidative status in cows supplemented with organic trace minerals.

**Key Words:** inflammation, trace mineral

**708 Parameterization of a ruminant model of phosphorus digestion and metabolism.** Xin Feng\*, Katharine F. Knowlton, and Mark D. Hanigan, Department of Dairy Science, Virginia Polytechnic Institute and State University, Blacksburg, VA.

The objective of the current work was to parameterize the digestive elements of the model of Hill et al. (2008) using data collected from animals that were ruminally, duodenally, and ileally cannulated, thereby providing a better understanding of the digestion and metabolism of phosphorus (P) fractions in growing and lactating cattle. The model of Hill et al. (2008) was fitted and evaluated for adequacy using the data from 6 animal studies (Feng et al., 2011, Ray et al., 2012b, Feng et al., 2013, Ray et al., 2013, Jarrett et al., 2014, Feng et al., 2015). It was hypothesized that sufficient data would be available to estimate P digestion and metabolism parameters and that these parameters would be sufficient to derive P bioavailabilities of a range of feed ingredients. Inputs to the model were DM intake; total feed P concentration ( $P_{PFd}$ ); phytate (Pp), organic (Po) and inorganic (Pi) P as proportions of total P ( $P_{PpPt}$ ,  $P_{PoPt}$ ,  $P_{PiPt}$ ); microbial growth; amount of Pi and Pp infused into omasum or ileum; milk yield; and BW. The available data were sufficient to derive all model parameters of interest. The final model predicted that given a 75 g/d total P input, the total-tract digestibility of P was 40.76%, Pp digestibility in the rumen was 92.4% and in the total-tract was 94.7%. A large proportion of Pi was absorbed from the small intestine (SI), however additional Pi was absorbed from the large intestine (3.15%). Absorption of Pi from the SI was regulated. Salivary recycling of blood P to the rumen was a major source of Pi flow into the SI, and the primary route of excretion. Milk synthesis used 16% of total absorbed P, and less than 1% was excreted in urine. The resulting model could be used to derive P bioavailabilities of commonly used feedstuffs in cattle production.

**Key Words:** model, phosphorus, digestion and absorption

### 709 Comparison of predicted ration phosphorus balance using bioavailabilities from the NRC (2001) and Virginia Tech models.

Xin Feng\*, Katharine F. Knowlton, Mark D. Hanigan, Jamie Jarrett, and Robert James, *Department of Dairy Science, Virginia Polytechnic Institute and State University, Blacksburg, VA.*

The objective of the current work was to use digestion coefficients from the Hill et al. (2008) model after parameterization by Feng et al. (2015; VT model) to calculate phosphorus (P) bioavailability ( $P_{VT}$ ) of common feeds used in dairy production. Compared with the bioavailability calculated by the VT model, using the NRC (2001) P absorption coefficient of 0.64 for forages would underestimate the bioavailabilities of alfalfa hay, alfalfa silage, corn silage, grass hay, mixed mainly legume silage and western hay. For concentrates, using the NRC (2001) P absorption coefficient of 0.70 would overestimate the bioavailabilities of canola, corn grain, dry barley, soybean meal and whole cottonseed but underestimate bioavailability of high moisture corn. Two dairy rations were formulated using nutrient values from the NRC (2001): a standard diet which includes minimal byproducts and a byproduct diet which has reduced corn and soybean meal that was replaced with corn gluten feed, distillers grains, hominy feed and wet brewers grains. For each diet, total bioavailable P was calculated using availability values from the NRC (2001) and the VT models. Comparison of P balance (the difference between required and bioavailable P) for each diet was made using the 2 sets of bioavailabilities for a reference cow weighing 682 kg, producing 38.6 kg of milk/d (3.5% fat and 3.0% true protein, 100 DIM) and consuming 23 kg of DM/d yielding an absorbable P requirement of 59.4 g/d. The standard diet supplied 56.7 g and 53.5 g of bioavailable P per day using bioavailabilities from the NRC (2001) and VT models, respectively, resulting in a P balance of -2.72 and -5.9 g/d. The byproduct diet provided 75.75 and 78.47 g/d of bioavailable P yielding P balances of 16.4 and 19.1 g per day respectively using the 2 sets of bioavailabilities. Thus using P bioavailabilities for individual ingredients that were based on the VT model resulted in different calculated P balances by diet and would result in differing P feeding levels if used in a field setting.

**Key Words:** phosphorus, balance, bioavailability

### 710 Effects of Se-fertilization on forage Se concentration and Se status of growing calves consuming these forages.

Juliana Ranches\*, Joao M. Vendramini, and John D. Arthington, *UF/IFAS Range Cattle Research and Education Center, Ona, FL.*

The objective of this study was to evaluate the effect of Se fertilization of 'Jiggs' bermudagrass (*Cynodon dactylon* L.) hayfields on the forage Se concentration and subsequent Se status of calves consuming the forage. Sodium selenate was dissolved into water (8.8 g/L) and sprayed onto bermudagrass hayfields at a rate of 257 g Se/ha and harvested 8 wk after Na selenate application. Forage fertilized with Na selenate had greater ( $P < 0.001$ ) Se concentration compared with the control forage without Se fertilization ( $7.7 \pm 1.81$  vs.  $0.1 \pm 0.04$  mg/kg DM). Control and high-Se hay were fed for 42d to weaned calves ( $n = 32$ ; initial BW =  $176 \pm 8.7$  kg) stratified by initial BW and randomly assigned to partially covered drylot pens (16 pens; 2 calves/pen). Treatments were randomly assigned to pens, including control hay + high-Se hay, control hay + supplemental Na selenite, or control hay without supplemental Se ( $n = 7, 7,$  and  $2$  pens, respectively). A pair-feeding design was utilized, whereas each pen receiving high-Se hay was paired to a pen receiving Na selenite. Ground, high-Se hay was offered to assigned pens for a 4 h period each morning. Hay intake was measured and total Se intake was estimated. The Na selenite paired-pen was then provided the same daily amount of Se via Na selenite hand-mixed into a limit-fed grain

supplement resulting in a range of daily Se intakes of 2.08 to 3.98 mg/d. Liver Se concentrations were greatest ( $P \leq 0.004$ ) on d 42 for calves consuming high-Se hay compared with calves receiving Na selenite or no supplemental Se (2.97, 2.49, and 0.60 mg/kg DM, respectively; SEM = 0.142). Amount of daily Se intake affected Se status, whereas, calves consuming  $< 3.0$  mg/d from high-Se hay had 54% greater ( $P = 0.02$ ) liver Se concentrations compared with calves consuming the same amount of Se from Na selenite. When consuming  $> 3.0$  mg/d, there were no differences ( $P = 0.71$ ) in liver Se concentrations due to Se source. These results imply that Se fertilization of bermudagrass hayfields results in increased Se concentration of harvested forage. Additionally, calves consuming this forage in amounts providing  $< 3.0$  mg Se daily have greater Se status compared with calves consuming the same amount of Se from Na selenite.

**Key Words:** calves, hay, selenium

### 711 Effect of Cu, Zn, and Mn source on preferential free-choice intake of salt-based supplements by beef calves and precipitation-induced metal loss.

Aline T. Wiebusch<sup>2</sup>, Maria L. Silveira<sup>1</sup>, Luana S. Caramalac<sup>2</sup>, Henrique J. Fernandes<sup>2</sup>, and John D. Arthington\*<sup>1</sup>, *<sup>1</sup>UF/IFAS Range Cattle Research and Education Center, Ona, FL, <sup>2</sup>Graduate Program in Animal Sciences, State University of Mato Grosso do Sul, Aquidauana, Brazil.*

Two studies were completed to evaluate the effects of Cu, Zn, and Mn source on preferential intake and precipitation-induced metal loss of salt-based mineral supplements. Complete free-choice, salt-based mineral supplements were formulated to contain 2,500, 5,500, and 4,000 mg/kg of Cu, Zn, and Mn, respectively. The supplements differed only by the source of Cu, Zn, and Mn, which were hydroxy, organic, or sulfate sources. In Exp. 1, the 3 formulations were offered simultaneously to preweaned beef calves (4 pasture replications; 17 calves/pasture) within separate stainless steel bowls inside covered cow-exclusion areas. Preferential intake was evaluated weekly for 18 wk. Consumption of mineral averaged  $21 \pm 2.4$  g/d (sum of all 3 sources), with a greater ( $P < 0.001$ ) percentage of the total intake coming from the formulation containing the hydroxy vs. organic or sulfate sources of Cu, Zn, and Mn (42.8, 30.2, and 27.0% of total intake, respectively; SEM = 1.03). In Exp. 2, each of the mineral formulations from Exp. 1, were exposed to a 10.2 cm precipitation event delivered in equal 3.4 cm applications on Monday, Wednesday, and Friday (4 replications/formulation). To accomplish this, 750 g of mineral was placed into Buchner funnels (177 cm<sup>2</sup>) on 20 to 25  $\mu$  pore filter paper. Deionized water (pH adjusted to 5.6) was poured over the mineral and the leachate collected into volumetric flasks and analyzed for Cu, Zn, and Mn concentration using inductively coupled plasma spectroscopy techniques. Total leaching losses of Cu, Zn, and Mn were less ( $P < 0.001$ ) for formulations containing hydroxy- vs. organic- and sulfate-sources (Cu losses = 0.8, 5.8, and 3.8%, SEM = 0.22; Zn losses = 18.0, 54.6, and 52.3%, SEM = 1.83; and Mn losses = 6.2, 50.1, and 54.8%, SEM = 1.83 for formulations containing hydroxy, organic, and sulfate sources, respectively). These results imply that calves preferentially consume free-choice salt-based mineral supplements formulated with hydroxy vs. sulfate or organic sources of Cu, Zn, and Mn. In addition, hydroxy sources of Cu, Zn, and Mn are less susceptible to precipitation-induced leaching losses compared with sulfate and organic sources.

**Key Words:** calves, supplementation, trace mineral

**712 Effects of trace mineral source on cow performance and mineral status during a production cycle.** Deborah M. Price\*<sup>1</sup>, Kaitlyn M. Havill<sup>1</sup>, Alex F. Swain<sup>1</sup>, Joseph M. Guevera<sup>2</sup>, Carley R. Trcalek<sup>2</sup>, Max Irsik<sup>2</sup>, Owen Rae<sup>2</sup>, Matthew J. Hersom<sup>1</sup>, and Joel V. Yelich<sup>1</sup>, <sup>1</sup>Department of Animal Sciences, University of Florida, Gainesville, FL, <sup>2</sup>College of Veterinary Medicine, University of Florida, Gainesville, FL.

Effect of trace mineral (TM) supplement source on cow performance and TM status over the production cycle were examined. Cows (n = 200, BW = 523 ± 6 kg, BCS = 5.1 ± 0.03) were divided in a factorial treatment arrangement, Angus (AN, n = 99) and Brangus, (BN, n = 101) and TM treatments (TRT; Inorganic = ING, n = 100, and Organic = ORG yeast source, n = 100). Supplementation (3 d/wk, 0.4 kg<sup>-1</sup>454 kg BW<sup>-1</sup>d in a pellet) of TM began ≥90 d before expected parturition and continued to weaning. A subset of cows had liver biopsies and serum collected, frozen and analyzed for TM content (Co, Cu, Fe, Mn, Mo, Se and Zn), by ICP-MS at 4 and 6 periods, respectively: start of TM (TMst), pre-calving (PreC), calving (C, serum only), post-calving (PostC, serum only), prebreeding (Pbrd) and weaning. Cow BW and BCS were recorded each period. Data were analyzed in SAS using Proc Mixed and repeated measures, with TRT, breed and time as fixed effects. No 3-way interactions were detected except for serum Cu, Mn and Mo. At PostC, BN were 0.17 BCS greater (P = 0.03) than AN. Cow BW and BCS did not differ (P > 0.21) by TRT at each period. The AN had greater (P ≤ 0.03) BW change from TMst to PreC (49 vs. 40 ± 2 kg) and from PostC to Pbrd (-44 ± 4 vs. -33 ± 3 kg) than BN. Change in BCS from PreC and Pbrd to weaning were greater (P < 0.05) in ING (-0.15 vs. -0.001 ± 0.05 and -0.36 vs. -0.10 ± 0.08, respectively) than ORG. Time (P ≤ 0.02) affected all serum and liver TM concentrations except for serum Fe (P = 0.13). Serum Co and liver Cu concentrations were greater (P < 0.05) in ING (1.7 vs. 1.3 ± 0.1 ng/mL and 266.4 ± 15.0 vs. 201.2 ± 14.1 µg/g, respectively), while serum Mo was greater (P = 0.006), in ORG (2.8 vs. 1.7 ± 0.3 vs. ng/mL). Serum Fe and liver Cu and Mn concentrations in BN (149.1 ± 5.5 µg/dL, 280.9 ± 15.2 and 11.7 ± 0.5 µg/g) were greater (P < 0.03) than AN (132.4 ± 5.3 µg/dL, 186.7 ± 13.9 and 9.5 ± 0.5 µg/g). Serum Se was greater (P = 0.003) in AN than BN (73.5 ± 1.7 vs. 65.4 ± 1.8 ng/mL). Cow TM status varies over time and is affected by breed and TM source; indicating development of nutritional management strategies can be based on cattle breed.

**Key Words:** trace mineral, liver, cattle

**713 Effect of prenatal trace mineral source on preweaning and weaning calf liver and serum mineral status.** Deborah M. Price\*<sup>1</sup>, Alex F. Swain<sup>1</sup>, Meaghan M. O'Neil<sup>1</sup>, Joseph M. Guevera<sup>2</sup>, Carley R. Trcalek<sup>2</sup>, Max Irsik<sup>2</sup>, Owen Rae<sup>1</sup>, Matthew J. Hersom<sup>1</sup>, and Joel V. Yelich<sup>1</sup>, <sup>1</sup>Department of Animal Sciences, University of Florida, Gainesville, FL, <sup>2</sup>College of Veterinary Medicine, University of Florida, Gainesville, FL.

The effect cow prenatal trace mineral (TM) supplement source has on calf serum and liver TM status from 115 ± 21 d of age through weaning (206 ± 21 d) was investigated. Factorial arrangements [Angus = AN, n = 95 and Brangus = BN, n = 96 cows; Inorganic = ING, n = 98, and Organic = ORG yeast, n = 93] utilized calves born to cows supplemented (3 d/wk at a rate of 0.4 kg<sup>-1</sup>454 kg BW<sup>-1</sup>d in a pellet) with TM ≥90 d before expected parturition. All calf (n = 190) BW were collected at d 115 and weaning, while a subset of calves (ING, n = 12, 6/breed, ORG, n = 14, 7/breed) had liver biopsies performed and serum collected by jugular vein puncture. Processed serum and liver samples were frozen at -20°C until analyzed for TM (Co, Cu, Fe, Mn, Mo, Se and Zn), by ICP-MS. Data were analyzed in SAS using Proc Mixed and repeated measures, with TRT, breed and day as fixed effects. With the exception of serum Co, Mn and Mo, there were no (P ≥ 0.08) 3-way interactions. Calf d 115 BW (149 ± 2 kg) were not affected (P = 0.65) by TRT. At weaning, ORG had greater (P ≤ 0.02) BW (226 vs. 216 ± 3 kg), 205 d adjusted BW (227 vs. 216 ± 3 kg), BW gain (190 vs. 181 ± 3 kg) and ADG (0.93 vs. 0.89 ± 0.01 kg) than ING. Serum Co and Zn were affected (P < 0.001) by day, while all liver TM except for Co were affected (P < 0.03) by day. The ORG calves had greater (P ≤ 0.01) serum Se (29.8 vs. 18.2 ± 1.1 ng/mL), liver Se (0.7 vs. 0.5 ± 0.1 µg/g) and Mn (9.3 vs. 7.8 ± 0.4 µg/g) than ING. The AN had greater (P ≤ 0.04) serum Co (0.8 vs. 0.5 ± 1.2 ng/mL), Se (27.9 vs. 19.4 ± 1.1 ng/mL) and liver Co (0.2 vs. 0.1 ± 0.02 µg/g), Fe (573.5 vs. 435.9 ± 1.1 µg/g) and Zn (128.1 vs. 116.7 ± 1.0 µg/g), while BN had greater (P < 0.05) serum Mo (5.0 vs. 2.5 ± 0.6 ng/mL) and liver Mn (9.1 vs. 8.0 ± 0.4 µg/g). Calf BW and ADG at weaning were increased by prenatal ORG TM. These data indicate calf TM status varies by day relative weaning, prenatal TM source and breed. Additional research is needed to determine how TM source affects calf immune function, performance, carcass traits and reproductive potential.

**Key Words:** trace mineral, prenatal nutrition, weaning



## Small Ruminant Symposium: Genetic improvement in small ruminants for the future

**714 National Sheep Improvement Program.** David Notter\*, *Virginia Tech, Blacksburg, VA.*

The US National Sheep Improvement Program (NSIP) was established in 1987 and is supported exclusively by US sheep breeders and their organizations. In 2010, limited options for sustainable involvement with US institutions led NSIP to establish a partnership with Meat and Livestock Australia to merge genetic evaluation activities with LAMBPLAN, the Australian sheep genetic evaluation system. Outsourcing genetic evaluation services to LAMBPLAN expanded NSIP resources and increased the frequency of updating of EBV. The operating philosophy of NSIP correspondingly evolved to become a proactive force for genetic improvement of US sheep. LAMBPLAN offers EBV for 85 different traits. NSIP provides EBV for a subset of these traits including body weights from birth through adulthood, reproductive and ewe productivity traits, ultrasonic predictions of fat and loin muscle depths, fleece weight and quality measurements, and worm resistance. LAMBPLAN offers selection indexes for various breeds and production objectives, and NSIP offers indexes specific to US breeds and conditions. NSIP serves all US sheep breeds, and also offers genetic evaluation services for meat goats, but is dominated by 4 groups: (1) Western range breeds (mainly Targhee, Rambouillet and Columbia), (2) terminal sire and other meat breeds (mainly Suffolk, Hampshire, Dorset, and Shropshire), (3) maternal wool breeds (mainly Polypay), and (4) hair sheep breeds (mainly Katahdin). In 2013, 123 flocks submitted data to NSIP. The 4 breed groups each submitted records on 2,700 to 3,200 lambs, and the 9 breeds listed above accounted for over 85% of NSIP flocks. Genetic trends in US breeds document the effectiveness of selection. Use of genomic strategies to increase accuracies of EBV is a major focus of LAMBPLAN research and development, providing opportunity for corresponding activities in NSIP. Detailed phenotypic records of animal performance are increasingly recognized as essential for discovery and validation of genomic markers, and NSIP is increasingly recognized as the most comprehensive repository of sheep data in the US. Storage of DNA from all progeny-tested NSIP sires and a sample of lambs in each major NSIP breed is intended to allow NSIP to make a larger contribution to meeting the research needs of the sheep industry.

**Key Words:** sheep, estimated breeding value, genetic evaluation

**715 Genomics and marker-assisted selection in small ruminants.** Brian L. Sayre\*, *Virginia State University, Petersburg, VA.*

As the world climate and population levels evolve, high demands are placed on livestock production systems. Small ruminants, especially goats, are critical to the small-scale producer as they are easy to acquire and maintain in sparse pasture and marginal crop regions that cannot easily sustain other livestock. Current small ruminant production systems around the world depend heavily on locally adapted breeds. While indigenous small ruminant genetics are relatively uncharacterized, most have undergone generations of adaptation and genetic isolation or bottlenecks, creating a large genetic reservoir of variation in genes and genetic markers. Identifying genes and genetic markers associated with disease susceptibility and resistance in locally adapted breeds is necessary to preserve local germplasm and to apply state-of-the-art genomic-based strategies to meet global food security challenges. Of particular interest are the genes or genetic markers involved in resistance to internal parasites and disease, and resilience to climate differences.

The aim of the current small ruminant genetics and genomics research is the development and utilization of state-of-the-art genomic-based evaluation tools for characterization of sheep and goat populations, development of sustainable strategies for improved production, and increased education and research opportunities. International collaborations have led to the development of de novo genome assembly projects, Illumina SNP panels and, for goats, a collaborative database for submission and access to goat SNP and re-sequencing data for breeds and populations worldwide (ADAPTmap). These tools establish the baseline necessary for the development and application of marker-based selection to the sheep and goat industry.

**Key Words:** genomics, marker-assisted selection, small ruminant

**716 Genetic improvement for internal parasite resistance.**

Daniel J. Brown\*, *Animal Genetics and Breeding Unit, University of New England, Armidale, NSW Australia.*

Internal parasites impose a huge financial burden for small ruminants across the world with recent estimates in Australia suggesting the cost could be as high as \$10AUD per breeding ewe per year. Genetic improvement has to be a key part of any integrated pest management plan for internal parasites. The number of worm eggs per gram of feces (WEC) is known to be a good indicator of internal parasite burden and is moderately heritable in both sheep and goats ( $h^2 = 0.2$  to  $0.3$ ). Breeding for internal parasite resistance is complicated by the fact that several different worm species can be present depending on the location, time of year, management practices and prevailing environmental conditions. In Australia, the national genetic evaluation system (Sheep Genetics) only uses data from *Haemonchus contortus* and *Trichostrongylus* spp. to reduce some of the complications caused by these multiple worm species. It is also known that WEC is genetically not the same trait across ages and thus there is benefit in breeders recording multiple assessments throughout the animal's life. Furthermore, research in Australia suggests that the genetic correlations between different sites across Australia may also be significantly less than one, and thus making it important for breeding animals to be assessed in similar environments to those in which their progeny are expected to exhibit their resistance. These issues make the estimation of breeding values and genetic improvement of this trait more challenging. Despite these challenges ram breeders in Australia have managed to make significant genetic improvements in WEC. Some breeders have reduced WEC by up to 5% per year for the last 15 years, while simultaneously improving other production traits. On-farm, these breeders have reported reduced requirements for drenching, less scouring and greater production. Genomic information has also been incorporated into the routine Sheep Genetics evaluations. Breeders now have the ability to obtain moderately accurate ASBV predictions for WEC (plus a range of other traits) via a simple blood test.

**Key Words:** internal parasite, worms, sheep

**717 Breeding strategies in sheep for low-input management systems.** Ronald M. Lewis\*<sup>1</sup>, Napoleón Vargas Jurado<sup>1</sup>, and Kreg A. Leymaster<sup>2</sup>, <sup>1</sup>*Department of Animal Science, University of Nebraska, Lincoln, NE,* <sup>2</sup>*Roman L. Hruska US Meat Animal Research Center, USDA-ARS, Clay Center, NE.*

Profitability of sheep enterprises increasingly depends on using low-input, pasture-based management systems where costs can be curtailed without sacrificing productivity. Their success rests on using maternal genetics well adapted to such systems yet with attributes to produce a quality product. In historically medium-wool industries, fleece has become a cost rather than a source of revenue. Therefore, hair sheep germplasm has been incorporated into maternal lines to facilitate wool shedding (WS), and to enhance health (e.g., internal parasite resistance) and fitness (e.g., vigor, aseasonality, longevity). Yet, there are caveats. Hair sheep are smaller in mature size and typically lighter muscled. Within the US, carcass weights are traditionally at least 30 kg, a challenge to achieve with smaller breeds. The Katahdin breed reflects that concern, where Caribbean hair sheep were combined with wool breeds and Wiltshire Horn, a larger mature size breed that sheds. Markets for hair sheep lambs of lighter weight have evolved, allowing greater flexibility. Crossing with terminal sires offers another option, although its impact on overall productivity needs attention. The definition of a maternal breed-type ideal for low-input systems is equivocal. At the US

Meat Animal Research Center, a 1/4 Katahdin, 1/4 White Dorper and 1/2 Romanov composite has been established to attain an intermediate optimum. The inclusion of Romanov increased prolificacy, allowing identification of ewes able to produce, nurture and rear twin and triplet lambs in a low-input system. The composite retains 62.5% of heterosis benefiting health and fitness. Data on WS, color, horns, major genes affecting disease, and rearing status have been collected, and used independently in selection. Genetic analyses of WS data have been conducted. Among 3345 autumn WS records on ewes, 64% had less than 25% wool coverage; WS was moderately heritable (0.26) and repeatable (0.40). This composite is early in development, and further generations of *inter se* mating would benefit its evaluation. A breeding program for low-input systems necessitates combining breeds. A strategy capturing key attributes of a maternal breed-type, as defined through a selection index, is needed to sustain its role in the sheep industry.

**Key Words:** sheep, low-input, maternal composite

# ADSA-ASAS Northeast Section Symposium: Bridging the gap between animal protein production and consumers, current and future

**718 University efforts to generate community support for a university farm.** Joe Hogan\*, *The Ohio State University, Wooster, OH.*

Maintaining the functional viability of university farms to assure availability of animals for teaching and research requires the cooperation of many entities both within and outside the university. The approach used by the Waterman Dairy on the Columbus campus of The Ohio State University to involve several groups outside the university will be presented as a successful example of community support. Key contributions from communities outside the university to be highlighted were provided by dairy producers, a breed association, local civic organization dedicated to water conservation, state governmental agencies and a dairy processor. The essential needs for cooperation from college and departmental administration will also be discussed. Specific outcomes from these private sector and university communal approaches for dairy farm support were the repopulation of the herd, renovation of facilities, employment of land and water conservation measures, and innovative marketing of products from the herd. Based on the experiences gained from revitalization of the Waterman Dairy, specific suggestions for involving community support for university farms will be presented.

**Key Words:** university farm, community support

**719 The New York State Junior DAIRY LEADER Program.** Deborah J. Grusenmeyer\*, *Cornell University, Ithaca, NY.*

The Junior DAIRY LEADER program prepares 16 to 19 year olds for successful dairy industry careers. The mission is to give teens an opportunity to build enthusiasm for the dairy industry through personal, professional and leadership development in a networking environment, while discovering the diversity of career options in agriculture. Exposure to different herd, crop and farm business management techniques gives participants the opportunity to experience diverse dairy management styles and ideas. The program also focuses on developing leadership skills necessary to make positive changes and contributions to the dairy industry and challenges participants to focus on personal and career development. Junior DAIRY LEADER is a yearlong program combines a series of 8 hands-on workshops focusing on specific facets of the dairy industry in veterinary science, dairy nutrition, production management, and on-farm production analysis. In addition, participants interact with dairy producers, industry professionals, and other dairy interested young people from across the state and region. Youth will gain advanced industry training for employment and furthering their education and career planning for successful dairy careers. Since 1999, 319 participants and graduates of the program have or will continue their education in dairy or industry related careers. Based on survey results from participants from 1999 to 2013 (using a scale of 1 to 5 and considering rankings of 3 and up to be considered significant influence), 96% of participants felt the program influenced their decision to pursue or enter into an agricultural

career. In the area of dairy knowledge and technical skills 98% had an increase in dairy knowledge and 77% learned technical skills directly applicable to production management. In addition participants responded to being better prepared in the following areas: communication skills (97%), working as a team (87%), choosing a career path (92%), problem solving (69%), and networking with other dairy industry people/leaders (85%). The Junior DAIRY LEADER survey results from the 1999 to 2013 show that 58% of participants who were undecided on a career path before the program, chose to a career path in agriculture after participating in the program

**Key Words:** dairy, education

**720 Using social media to increase consumer acceptance of animal agriculture.** Janeal W. Yancey\*, *University of Arkansas Division of Agriculture, Fayetteville, AR.*

Social media has fundamentally changed the way people communicate. As animal scientists, we have a story to tell and information to share about agriculture, and we need to be part of this new digital conversation. In today's environment, we see that science is constantly being denied, whether it concerns technology used in food production or the science behind vaccinations for our children. Consumers receive lots of information about their food and try to use that information to make decisions about feeding their families. Decisions about food are highly personal, and in the food industry, we must understand that facts alone do not drive the decisions consumers are making. To really reach consumers, we should be ready and willing to engage in dialog with them. Animal scientists have to embrace a listen-first mentality. We must understand a consumer's questions before we can answer them. Be ready to embrace skepticism; to connect with someone, you must let them know that you understand their concerns. Do not dismiss them. Research from the Center for Food Integrity reveals that a Mom Scientist is the most believable messenger for consumers concerning controversial topics about food production, such as antibiotics or GMO foods. Theodore Roosevelt said, "People don't care about how much you know until they know how much you care." On social media, scientists should be more than the researcher; you need to also be a mom, a dog lover, an artist, or a runner. Shared values carry more weight with consumers over time than a PhD. Last, remember that many voices are needed to fully engage the consumer population. Embrace the diversity among animal scientists and share animal agriculture with people that you really connect with. The ultimate goal is not to win scientific or social arguments, but to connect with people in ways that you become a resource to them (and them to you) for years to come.

**Key Words:** advocacy, consumer education, social media



## Animal Behavior and Well-Being II

**721 Effects of dietary fiber on the welfare of submissive multiparous sows.** Eridia Pacheco\*, Mayra Lopez, and Janeen Salak-Johnson, *University of Illinois at Urbana-Champaign, Urbana, IL.*

The effect of social rank, dietary fiber, and length of feeding stalls on the well-being of sows kept in groups was assessed on a sub population within the pens of the larger study. At gestational d 37, sows that were randomly assigned by BW and parity in a  $2 \times 2$  factorial arrangement to a dietary treatment of either (a) soy hulls-wheat middlings diet (WM) or (b) DDGS-corn germ meal diet (DDGS); and feeding stall length of either (c) 2 ft. (short) or (d) 6 ft. (long) in groups of 9 sows/treatment (36/block), were put in a feed competition test to determine social rank ( $n = 48$ ). Within each treatment pen, dominance value was calculated based on aggressive encounters among sows from the feeding test; with the 2 highest-ranking sows being dominant (DOM) and 2 lowest-ranking sows being submissive (SUB). Analysis of data was focused on this sub population of sows. Sow BW was recorded on gestational d 30, 70, 90, 104, and d 131 (end of lactation). Lesion scores and blood glucose levels were recorded every 3 d for 2-wks post mixing (Phase 1), and then bi-weekly until gestational d 104 (Phase 2). Live observations during feedings were performed at the first feeding, after mixing, and every 3-wk until gestational d 104, all aggressive encounters were recorded. Regardless of diet, SUB sows had greater ( $P < 0.01$ ) blood glucose than DOM sows. Overall, lesion scores decreased in severity from phase 1 to phase 2, but in phase 2, DOM-sows had less severe scores compared with SUB-sows ( $P < 0.01$ ). DOM-sows gained more ( $P < 0.01$ ) BW from d 30 till d 70 of gestation, and then again from d 90 till d 104. Total number of aggressive encounters were lowest among DOM-sows fed DDGS-diet and in pens with long feeding stalls and these same sows made less aggressive encounters toward SUB-sows ( $P < 0.01$ ). SUB-sows fed WM-diet had more ( $P < 0.05$ ) total piglets born alive than DOM-sows fed the same diet. SUB-sows fed WM-diet and kept in pens with short feeding stalls had more ( $P < 0.05$ ) total piglets born compared with SUB-sows on other treatments. These results indicate that dietary strategies and social status may serve as a management tool to improve sow well-being in small group pens.

**Key Words:** swine, social rank, welfare

**722 Behavioral changes in steers supplemented with tryptophan and vitamin E.** Paula R. Huenchullán<sup>2</sup> and Rafael E. Larrain<sup>1</sup>, <sup>1</sup>*Facultad de Agronomía e Ingeniería Forestal, Pontificia Universidad Católica de Chile, Santiago, Chile,* <sup>2</sup>*Facultad de Ciencias Veterinarias y Pecuarias, Universidad de Chile, Santiago, Chile.*

Tryptophan (TRP) and vitamin E (VITE) modulate the stress response in animals, so they may also induce changes in behavior. The goal of the study was to determine the effect of high TRP and VITE diets on steers behavior. Angus steers were used in 2 experiments (E1 and E2), with a 21-d washout period in-between. Steers were distributed in pairs in 16 pens and fed the experimental diets for 5 d before a stressful event (45 min in a single file chute for E1 and slaughter for E2). Diets were designed in a factorial arrangement, with TRP at 0 and 100 mg·kg BW<sup>-1</sup>·d<sup>-1</sup> and VITE at 0 and 2000 IU·hd<sup>-1</sup>·d<sup>-1</sup> ( $n = 4$  pens). Steers were randomly assigned to diets in E1, and E1 diet was used as blocking factor for randomization in E2. Video cameras were used to record 24 h·d<sup>-1</sup> during supplementation, and also one day after stress in E1. Behavior was evaluated at d 2, 4, 5 and 6 of starting supplementation in E1, and at d -1, 0, 2 and 4 in E2. Behavior was observed every 10

min and compared as proportion of observations where animals were in standing or lying posture; and doing one of the following activities: defecating, drinking, eating, interacting, no detectable activity, ruminating, self-grooming, and stereotypic movements (bar licking, tongue rolling, etc.). Statistical analysis was made using repeated measures, with TRP, VITE, time and their interaction in the model. In E2, diet in E1 was used as blocking factor and d -1 was used as covariate. When ANOVA  $P < 0.1$  for TRP, VITE or TRP  $\times$  VITE, comparisons with control were made using Dunnett adjustment. When ANOVA  $P < 0.1$  for any interaction with time, comparisons were made within day. In E1, none of the diets including TRP or VITE was different to the control in any variables. In E2, animals receiving TRP tended to spend a higher proportion of their time eating ( $P = 0.091$ ), while animals receiving VITE tended to spend a smaller part ( $P = 0.076$ ) than their controls. Animals receiving TRP and TRP+VITE presented less stereotypic movements ( $P = 0.008$  and  $0.017$ , respectively) than their controls, and animals receiving VITE spent a smaller proportion of their time interacting ( $P = 0.032$ ). We concluded that TRP and VITE produced small changes in behavior in steers and no changes the day after a stressful event.

**Key Words:** tryptophan, vitamin E, behavior

**723 Rumen and cecum expression of genes related to inflammation and behavior in Holstein bulls fed high-concentrate diets with different concentrate presentation forms and with or without straw supplementation.** Maria Devant<sup>\*1</sup>, Greg Penner<sup>2</sup>, Bruna Quintana<sup>1</sup>, Francesc Fábregas<sup>1</sup>, Alex Bach<sup>3,1</sup>, and Anna Arís<sup>1</sup>, <sup>1</sup>*IRTA-Ruminant Production, Animal Nutrition, Management, and Welfare Research Group, Caldes Montbui, Spain,* <sup>2</sup>*University of Saskatchewan, Saskatoon, Saskatchewan, Canada,* <sup>3</sup>*ICREA, Barcelona, Spain.*

Twenty-four individually housed Holstein bulls ( $395 \pm 7.3$  kg BW and  $252 \pm 3.1$  d age) were exposed to a  $2 \times 2$  factorial design (meal vs. pellet and with vs. without straw) to evaluate the effect of concentrate form and provision of straw in finishing diets. Concentrate and straw consumption were recorded monthly, and behavior by scan sampling biweekly. Bulls were slaughtered after 64 d. Rumen and liver lesions were recorded and tissue samples were collected. Data were analyzed using an ANOVA. Straw supplementation increased ( $P < 0.01$ ) percentage of rumination observations ( $9.4$  vs.  $3.1 \pm 1.02\%$ ), and decreased ( $P < 0.01$ ) oral non-nutritive behaviors ( $0.74$  vs.  $1.68 \pm 0.12$  times/15 min) relative to bulls not provided straw. Provision of straw increased rumen pH, but the magnitude was greater when meal was provided compared with pellet (interaction  $P < 0.05$ ). When straw was not supplemented, all rumens had papillae fusion, whereas only 16.7% of bulls fed pellet and straw had papillae fusions ( $P < 0.05$ ). In the rumen straw provision numerically increased expression of free fatty acid receptor 2 (stimulates PYY and serotonin secretion,  $P = 0.06$ ),  $\alpha$ -2C adrenergic receptor (modulates immune reactions and behavior,  $P = 0.09$ ), and increased occludin and claudin-4 (tight junction proteins,  $P < 0.05$ ), along with IL-1 $\beta$ , TNF $\alpha$  (pro-inflammatory cytokines,  $P < 0.01$ ) and toll-like receptor-4 ( $P < 0.01$ ); moreover, it also numerically increased  $\beta$ -defensin1 (antimicrobial peptide,  $P = 0.10$ ), and intestinal alkaline phosphatase ( $P = 0.10$ ), and increased CASP3 (pro-apoptotic gene,  $P < 0.05$ ) and numerically increase SPC25 (proliferative marker,  $P = 0.06$ ). Pellet decreased rumen gene expression of  $\alpha$ -2C adrenergic receptor ( $P < 0.05$ ) and increased occludin ( $P = 0.06$ ) and monocarboxylate transporter 1 ( $P < 0.05$ ). Bulls without straw had increased expression of IL-1 $\beta$  in the cecum. In conclusion, the lack of straw supplementation in bulls fed

high-concentrate diets modifies behavior, affects rumen macroscopic morphology, and expression of epithelial genes related to inflammation.

**Key Words:** bull, gut-brain axis, nutrition and behavior

**724 Immunological castration of gilts changes behavior, weight gain, and reproductive development.** Guilherme Picinin\* and John J. McGlone, *Texas Tech University, Lubbock, TX.*

Physical castration is a current practice on commercial farms used to reduce boar taint and aggressive behavior. Immunological castration (IC) using a GnRF conjugate is an alternative to physical castration. IC males eat more, grow faster and are easier to handle than intact males. The objective of this study was to evaluate the effect of IC on gilt behavior, handling during loading and unloading, weight gain, reproductive function, and physiological responses. Gilts were given a first immunization at wk 3 of the finishing period (about 12 weeks of age) followed by a second immunization at wk 7 of the finishing period. Gilts were marketed when they were 143.6 kg on average. Behaviors of immunized gilts (IG) (n = 48) and control gilts (CG) (n = 48) were assessed 2 times through the finishing period, one before (second week of grower-finisher phase) and one after effective immunization (eleventh week of grower-finisher phase). Handling during loading and unloading a livestock trailer was also recorded during a simulated transport experience. Weight gain was assessed weekly. Physiological response was assessed via heart rate, plasma cortisol and ovarian weights. IG spent more time feeding (4.17%) ( $P = 0.04$ ) than CG (3.43%). However, CG spent 1.41% more time standing ( $P = 0.002$ ) and 0.17% more time drinking ( $P = 0.01$ ) than IG. No difference was found in transport and handling measures between treatments. At the last week of grower-finisher phase IC were 5.15 kg heavier than CG ( $P = 0.04$ ). Control gilts had 225% heavier ovaries than IG ( $P < 0.001$ ). No differences were found in plasma cortisol and heart rate between CG and IG. Immunological castration does not induce a stress response or cause behavioral problems in the home pen or during handling and transport compared with intact gilts. IC is an effective tool to increased gilt feeding behavior, feed intake, and weight gain with no identified animal welfare concerns.

**Key Words:** GnRF immunization of gilts, heavy market weight, behavior-handling

**725 Behavioral responses of livestock exposed to unmanned aerial systems.** Phil Urso, Marcy Beverly\*, Stanley Kelley, John Wilson, Jessica Leatherwood, Mark Anderson, and Kyle Stutts, *Sam Houston State University, Huntsville, TX.*

Unmanned aerial systems (UAS) have been gaining popularity in several fields. Their uses in agriculture provide major management benefits while assessing production conditions. This study utilized an UAS to determine the flight zone upon approach and while hovering over different livestock species: cattle, goats, and horses. Barometric pressure, temperature, wind speed, and noise levels were all recorded to eliminate environmental variables affecting behavior. Selected animals were held in pens several days before the UAS flight to allow for the animals to acclimate and become familiar to the test area. Test areas were spacious so animals could flee and not feel confined during UAS approach and hover. Groups were tested with the UAS 3 times on separate days. Horses were tested in 2 independent groups: outdoor round pens and indoor arena. Cattle were split into 3 groups: cows with calves (CWC), cows without calves (COC), and weaned heifers (WH). Goats were tested as a group in their home pasture. With each test group, the UAS began at a height of 75 feet above the group and descended at 5 feet

increments when hovering. Environmental measurements were similar for each flight. All groups were acoustically signaled before seeing the UAS. The animal flight zone varied by test group at different hovering heights. The COC and WH allowed the system to hover at 15 feet above ground before they casually moved from the UAS. CWC, did not flee from under the drone, but huddled closer together around their offspring responding in a protective behavior. The goats allowed the UAS to hover at 10 feet before moving. Horses tested indoors expressed a startled, frightened behavior at 25 feet. The outdoor tested horses were calm and showed unstartled behaviors consistent with other test groups. The geldings were much more relaxed and calm with the approach of the UAS than the mares, which had a higher flight zone. Though foals were not present, this difference could be attributed to their protective instinct. Understanding these behavioral responses could help producers utilize UAS for herd observations without disturbing animals in their natural environments.

**Key Words:** unmanned aerial system (UAS), flight zone, behavior

**726 ADSA®-EAAP Speaker Exchange Presentation: REM sleep time varies during the lactation cycle.** Emma Ternman<sup>1</sup>, Emma Nilsson<sup>1</sup>, Per Peetz Nielsen<sup>1,2</sup>, Laura Hänninen<sup>3</sup>, and Sigrid Agenäs\*<sup>1</sup>, <sup>1</sup>*Department of Animal Nutrition and Management, Swedish University of Agricultural Sciences, Uppsala, Sweden,* <sup>2</sup>*Department of Large Animal Sciences, University of Copenhagen, Copenhagen, Denmark,* <sup>3</sup>*Department of Production Animal Medicine and Research Centre for Animal Welfare, University of Helsinki, Helsinki, Finland.*

Little is known about sleep in dairy cows. This study investigated the influence of stage of lactation on total lying time, rumination time and rapid eye movement (REM) sleep in dairy cows. Data on rumination and REM sleep was obtained by electrophysiological recordings in 19 high yielding dairy cows during 7 24-h recording sessions; late dry period, lactation week (wk) 2, 7, 13, 21, 37 and early in the next dry period. In addition, standing and lying was recorded using IceTag data loggers. During recordings cows were kept in individual pens with ad lib access to silage and water. Data analysis included total time and duration of bouts and was separated for daytime (05.00–21.00 h) and night (21.00–05.00 h). Effect of stage of lactation cycle and day/night distribution of activity was tested with a linear mixed model (SAS 9.4). In total, cows were lying down for  $50 \pm 4\%$  of the time in the study, distributed as lying half of the night in early lactation (wk 2 and 13) and up to 2/3rds of the night in other stages of the lactation cycle. During daytime more time was spent standing than lying in all stages of lactation. Overall, lying bouts were longer at night than at daytime ( $51.7 \pm 3$  vs.  $43.3 \pm 3$  min,  $P < 0.005$ ) and standing bouts were shorter at night than at daytime ( $33.1 \pm 3$  vs.  $58.8 \pm 3$  min,  $P < 0.001$ ). Rumination time was longest in wk 7 ( $573 \pm 23$  min) and shortest in late dry period ( $458 \pm 23$  min) ( $P < 0.01$ ) and bouts were longer at night than day ( $36.3 \pm 1.2$  vs.  $29.7 \pm 1.1$  min,  $P < 0.001$ ). REM sleep time was affected by stage of lactation ( $P < 0.05$ ) with the longest time ( $52 \pm 4$  min) in late lactation (wk 37) and shortest in early lactation ( $34 \pm 4$  min). The main part of REM sleep was found at night and REM sleep bouts were longer at night than day ( $4.6 \pm 0.1$  vs.  $4.1 \pm 0.2$  min,  $P < 0.001$ ). Total lying time and rumination time found in this study was in accordance with previous findings. Cows rest lying down and even if total lying time during 24 h does not vary during the lactation cycle there is a variation in the distribution of the lying time between day and night and in the amount of REM sleep.

**Key Words:** lying, REM, sleep

**727 Assessment of cognitive ability and memory retention in neonatal goats.** Isabelle C. Withrock\*, Paul J. Plummer, Timothy A. Shephard, John Stinn, Hongwei Xin, Johann F. Coetzee, and Suzanne T. Millman, *Iowa State University, Ames, IA.*

Our research team is exploring the suitability of inhalant methods for euthanasia of neonatal livestock. A key component of this project is examining the aversiveness of carbon dioxide and argon gases by preference tests and conditioned place avoidance. However, little is known about the cognitive ability of neonatal livestock. The objective of this study was to determine the problem-solving ability of goat kids by using a conditioned place preference paradigm. A secondary objective was to determine if the presence of a novel odor disrupts previous learning. A preference-testing box was custom built with 2 connected chambers divided by a sliding door. One chamber was vacant while the second chamber held a 32oz milk reward. Twelve mixed breed dairy kids were enrolled in the study. Kids were given a 5 min acclimatization period in the control chamber before the sliding door was opened. Kids were then given 5 min to travel through the doorway to access the milk, after which kids were physically assisted through the doorway. Kids were allotted 10 min in the treatment chamber to consume their milk meal. After 5 d of testing, kids were not tested for 3 to 6 d. Kids were then tested with a novel odor (<1 oz peppermint oil) present in the treatment chamber. The following behaviors were collected using live observation: frequency of vocalizations and defecation, latency to enter treatment chamber, latency to suckle and volume of milk consumed. A mixed effect Poisson regression model showed that vocalizations were reduced from d 1 to all other days ( $P < 0.0001$ ). Mixed effect logistic regression models showed defecation behavior did not differ ( $P = 0.3064$ ). Mixed effect Cox models showed that latency to enter the treatment chamber and latency to suckle also decreased from d 1 ( $P < 0.0001$ ). Milk consumption increased from d 1 to d 6 based on a linear mixed model ( $P < 0.0001$ ). These results suggest neonatal kids have the ability to problem solve, and a novel odor did not disrupt learning. Hence, approach-avoidance and conditioned place associations using a milk reward are relevant paradigms for asking kids about aversiveness of inhalants used for stunning and euthanasia.

**Key Words:** goat, learning, memory

**728 Development of a novel automated method of measuring tail-flick behavior in beef cattle.** Diego Moya\*<sup>1,3</sup>, Carollyne E. J. Kehler<sup>2</sup>, Sonia Marti<sup>3</sup>, Kim H. Ominski<sup>2</sup>, Christy Goldhawk<sup>1</sup>, and Karen S. Schwartzkopf-Genswein<sup>3</sup>, <sup>1</sup>*University of Calgary, Calgary, AB, Canada*, <sup>2</sup>*University of Manitoba, Winnipeg, MB, Canada*, <sup>3</sup>*Agriculture and Agri-Food Canada, Lethbridge, AB, Canada.*

Tail-flicking behavior is a reliable indicator of pain in cattle, however visual observation is challenging and time-consuming. Two trials were conducted to validate the use of tri-axial accelerometers to assess tail-flicking behavior in beef cattle. In Exp. 1 (proof of concept), accelerometers (HOBO, Onset Computer Corp., Pocasset, MA) were affixed with vet wrap to the tail of 5 Angus heifers at 3 different positions: 8 cm below the base, 26 cm below the base, and above the tail switch. The HOBOS were set to measure acceleration at 20 readings  $\times$  s<sup>-1</sup> for a 10-min period, with a total of 49 periods of data collection. The heifers were videotaped concurrently, and viewed by the same observer to determine the number of tail movement events. Data obtained from the vector sum of the acceleration of the X, Y and Z axes was cleaned using either 2 standard deviations, the 80th percentile or the mean as the threshold below which tail movement was considered unintentional, and therefore removed from the data set. Data were then pooled into movement events using either a 1- or 2-s criteria as the time between 2 tail

movements to be considered separate movement events. The resulting data sets were summarized for each 10-min period as total acceleration, maximum acceleration, average acceleration per movement event, and percentage of time in motion. Data from HOBOS located at the tail switch, and processed with the 80th percentile threshold and the 2-s criteria, resulted in the greatest ( $P < 0.01$ ) correlation with the number of tail movement events observed via video ( $r = 0.9217$ ;  $P < 0.01$ ). In Exp. 2 (biological validation), accelerometers were affixed above the tail switch of 14 bulls to measure movement of the tail during 2 20-min periods that took place from 3 to 6 h before and immediately after castration. Total acceleration was greater ( $P = 0.03$ ) after than before castration ( $805.0$  vs.  $409.7 \pm 18.27$  g-force, respectively), suggesting that HOBOS captured the tail-flicking behavior caused by the discomfort of the procedure. Accelerometers provide an objective and efficient method of measuring tail-flick behavior in beef cattle.

**Key Words:** welfare, behavior, pain

**729 Relationships between maternal defensiveness behavior and pre-calving flight speed in primiparous Nellore cows.** Mateus J. R. Paranhos da Costa\*<sup>1,2</sup>, Tiago S. Valente<sup>2</sup>, and Aline C. Sant'Anna<sup>1</sup>, <sup>1</sup>*Departamento de Zootecnia, Faculdade de Ciências Agrárias e Veterinárias, UNESP, Jaboticabal-SP, Brazil*, <sup>2</sup>*Programa de Pós-Graduação em Genética e Melhoramento Animal, Faculdade de Ciências Agrárias e Veterinárias, UNESP, Jaboticabal-SP, Brazil.*

Defensiveness is an important aspect of maternal temperament, which can be defined as the set of cow's behavior in response to disturbance of their relationship with newborn calf. The aim of this study was to evaluate the relationship between 2 temperament traits, flight speed (FS) measured 6 mo before calving and maternal defensiveness in primiparous Nellore beef cows. The FS was performed for 196 heifers measuring the speed (in m/s) at which each animal exited the cattle crush after weighting. The females calved around 24 mo of age and had their maternal temperament assessed 24h after birth, during the handling for calf ear-marking and weighing. We assessed the reactions of cows when their newborns were caught in a corral pen, using a maternal defensiveness score (MD), which ranged from 1 (cow exhibited no attention to the calf and remained indifferent to the procedure) to 6 (cow displayed aggressive behaviors toward the handler, trying to attack them). The latency to each cows touch the calf (LT, s) after finishing the handling procedure and the calf be released to the pen was also recorded. Pearson's coefficients of correlation were calculated among the MD, LT and FS. A significant correlation was found between MD and LT ( $r = -0.62$ ,  $P < 0.01$ ), indicating that the shortest latency was related to a more defensive behavior toward the calf. The FS had significant, but low correlation with LT ( $r = 0.16$ ,  $P = 0.04$ ), and it was not correlated with MD ( $r = -0.10$ ,  $P = 1.79$ ), suggesting that an indicator of fear/excitability is not associated with an indicator of MD. There is a belief among the Brazilian farmers that the calmer Nellore cows during handling are likely to be less defensive toward their calves. However, according to the present results the cows' temperament assessed during handling in the corral is not related with cows' MD, providing clear evidence that cows with more excitable yearling temperament (faster FS) would not present more defensive behavior toward their calves compared with the calmer ones.

**Key Words:** maternal behavior, temperament, beef cattle

**730 Relationship between quantitative measures of temperament and observed behaviors in receiving cattle.** Kelsey A. Bruno\*, Eric S. Vanzant, Alex W. Altman, Monoj Kudupoje, and Kyle R. McLeod, *University of Kentucky, Lexington, KY.*



Temperament has been related to gain and intake in beef cattle; more temperamental animals often have decreased gain and intake. Intake effects may be partially mediated through social interactions in that some temperament measures may directly relate to social dominance behavior. To examine the relationships between temperament, growth, and social dominance behavior, 32 crossbred steers were used in a 56-d RCBD experiment with a  $2 \times 2$  factorial treatment structure. Temperament treatments (assigned on d -7) were chute exit velocity (EV; slow vs. fast) and objective chute score (OCS; low vs. high), a novel temperament measure, the CV of weights collected at 5 Hz for 10 s while an animal's head was restrained in a chute. Electronic ear tags with accelerometers were used to continuously measure animal activity. Social dominance behaviors were quantified using video records of 2-h intervals directly following feeding for d 1–13. Dominance was calculated as average dominance index and David's score. Jugular blood samples were collected to analyze antibody response to leptospirosis vaccines. There were

no EV by OCS interactions ( $P \geq 0.23$ ) for average daily gain (ADG) or antibody titers. High OCS steers had higher ADG than low OCS steers ( $P < 0.01$ ) and slow EV steers had higher ADG than fast EV steers ( $P = 0.02$ ). Slow EV animals had an increased antibody response compared with fast EV animals ( $P = 0.05$ ). Ranking in the dominance hierarchy was influenced by an EV x OCS interaction ( $P \leq 0.02$ ), but there was not a linear relationship between dominance ranking and ADG. Low OCS steers tended to have higher daily activity counts than high OCS steers ( $P = 0.11$ ), and fast EV steers had higher ( $P = 0.08$ ) activity counts than slow EV steers. EV and OCS were independently related to differences in ADG. Although dominance ranking was significantly related to these 2 independent temperament measures, it was not linearly related to ADG. Alternatively, differences in accelerometer-based activity counts among temperament groups did correspond with trends in growth responses.

**Key Words:** temperament, dominance, beef cattle

# Animal Health Symposium: Maintaining animal health in organic dairy herds

## 731 Understanding animal welfare on organic dairy farms.

Kathryn Proudfoot\*, *The Ohio State University College of Veterinary Medicine, Columbus, OH.*

In response to an increasing demand for organic products, the number of certified organic dairy farms in the United States has increased dramatically over the last decade. The consumers and concerned citizens who are driving this demand report a desire to support agricultural systems that are perceived to be more sustainable, more natural, and have higher standards of animal welfare. Despite the perception that organic farms maintain high levels of animal welfare, the relationship between the 2 is not straightforward. The objective here is to review the current literature on the welfare of dairy animals in organic systems compared with conventional. The 2 systems clearly place different value on animal welfare concerns; conventional farms place greater emphasis on biological functioning and health, whereas organic farms place higher value on the animals' ability to perform natural behaviors and live in a natural environment. In the United States, organic farms are required to provide animals with year-round access to the outdoors, including at least 120 d of access to pasture during the grazing season. Moreover, synthetic chemicals are restricted, including antimicrobials commonly used to treat ill animals on conventional farms. A reliance on naturopathic remedies leads to challenges when it comes to improving welfare for the individual animal, as organic producers have fewer treatment options and must make difficult decisions when dealing with ill animals. There is also concern regarding the efficacy of these naturopathic treatments, and little research to support their usage. Despite efforts to create preventative disease control strategies, the limited research available indicates that there is little difference in disease prevalence between organic and conventional systems. There are clear gaps in the literature evaluating the welfare of animals on organic dairy farms. Continued efforts are still needed to identify effective ways of preventing disease, and to develop effective naturopathic therapies to help reduce the number of ill animals and the duration of time that each animal is ill. A better understanding of how some organic farms are currently able to maintain low disease incidence could also provide valuable insight into ways forward.

**Key Words:** well-being, organic, dairy

## 732 Nutrition and performance of lactating dairy cows in organic dairy herds.

Andre F. Brito\*, *University of New Hampshire, Durham, NH.*

The number of US organic dairy cows increased from 2,265 in 1992 to 254,771 in 2011. Despite this rapid growth, organic herds are smaller than conventional herds, and produce approximately 30% less milk/cow (McBride and Greene, 2009). These discrepancies are explained by geographical location as well as management and nutrition. First, about 86% of organic dairies are located in the Northeast and Upper Midwest, which are characterized by herd sizes smaller than those from the West (McBride and Greene, 2009). Second, organic dairies must comply with the National Organic Program Pasture Rules, which mandate ruminants to consume at least 30% of their total DMI from pasture for a minimum of 120 d during the grazing season. Even though 95% of organic dairies use rotational grazing, they feed approximately 45% less grain than conventional dairies (Stiglbauer et al., 2013). Moreover, only 49% of WI organic dairies use corn silage (Hardie et al., 2014), and this figure is likely lower in other regions (e.g., Northeast). Thus, higher reliance

on pasture and less grain and corn silage feeding limit metabolizable energy intake likely explaining the lower milk production in organic than conventional farms. Reduced milk production in organic dairies may be also associated with the low number of nonpasture feeds incorporated into lactation rations as well as the predominant use of component feeding in detriment of TMR. There is a growing interest among organic dairy farmers to feed lactating cows exclusively forage-based diets. In fact, organic milk processors (e.g., Organic Valley) are currently paying premiums for farmers shipping "grass-fed milk" in specific states (e.g., NY, PA, CA). In addition to reduced animal performance, the short- and long-term implications of no grain supplementation on animal health, BCS, and reproduction need to be addressed. Research should further address the effects on animal performance and health of feedstuffs commonly fed in organic dairies such as kelp meal, liquid molasses, and barley fodder, as well as the incorporation of alternative forage crops (e.g., small grains, millet, brassicas) as grazing options.

**Key Words:** organic dairy, nutrition, performance

## 733 Managing transition cows and reproduction in certified organic dairy herds.

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It is common to observe large between-herd variation in transition management practices and reproductive performance of dairy cows under certified organic management. Metabolic and infectious diseases increase the risk of voluntary and involuntary culling within 60 d in milk (DIM) and are a major animal welfare issue for the dairy industry in both organic and conventional herds. Reproductive performance of lactating dairy cows directly influences the annual frequency of calving; thus, the number of replacement heifers and milk yield (due to extended low productivity and dry period). Recent studies involving 4,111 pre- and postpartum cows from organic herds showed that cows experiencing hypocalcemia (HYPO) at calving had greater proportion ( $P < 0.05$ ) of metritis and culling rate within 60 DIM compared with non-HYPO cows. Furthermore, uterine diseases (primarily metritis and purulent vaginal discharge [PVD]) significantly increased ( $P < 0.05$ ) the risk of cysts at  $26 \pm 3$  DIM, reduced cyclicity (progesterone concentration  $\geq 1$  ng/mL) at  $40 \pm 3$  DIM as well as the hazard of pregnancy up to 300 DIM, and increased the risk for pregnancy loss. Regardless of parity, uterine diseases (RP, metritis and PVD) had an additive effect on milk loss and quality for at least one of the first 4 DHIA test-days ( $P < 0.05$ ), but was not different at later tests. Many factors such as feed management (i.e., diets, dry matter intake and water quality) and comfort (e.g., resting time) of transition cows determine the energy (e.g., BHBA, NEFA) and calcium balance. These factors also determine the status of the immune system of the animal; thus, the prevalence of infectious diseases in postpartum cows (e.g., metritis). Implementing a proactive transition cow program that focuses on preventing metabolic diseases by managing the leading risk factors (e.g., diets, resting time of cows per day, dry matter intake) will significantly reduce calving-related losses and improve uterine environment and ovarian structures (cyclicity); thus, improving the overall performance of lactating dairy cows and profitability of the herd, regardless of size.

**Key Words:** organic, dairy, reproduction

**734 Mastitis and milk quality in organic dairy herds.** Pamela Ruegg\*, *University of Wisconsin, Madison, WI.*

The purpose of this paper is to review research about mastitis and milk quality on organic dairy herds. Mastitis is consistently the most commonly reported disease on both ORG and conventional (CON) dairy farms. In the US, concern about udder health of cows on ORG herds is often related to the blanket prohibition of antimicrobial usage. Cows on ORG herds that develop mastitis are usually treated with a variety of alternative compounds such as whey-based products, herbals, and essential oils. Although none of these products have any proven efficacy, ORG dairy farmers are able to successfully manage mastitis. Several studies have compared bulk tank SCC and rate of clinical mastitis between herds managed using ORG or CON systems. Although most studies have shown no difference in bulk tank SCC between management systems, some studies have reported higher bulk tank SCC for ORG as compared with CON herds. ORG herds are typically smaller than CON herds and studies that have accounted for herd-size have not reported significant differences in bulk tank SCC or other measures of milk quality. Management practices used to achieve low bulk tank SCC have slightly varied. In one study a greater proportion of lactating cows in ORG herds had milk withheld from sale as compared with cows in CON herds. In contrast to bulk tank SCC, fewer cases of clinical mastitis are generally reported in cows on ORG as compared with CON farms. A recent study reported 0.21 versus 0.31 cases of clinical mastitis per 305-cow-days for cows in ORG versus CON herds. Within the framework provided by NOP regulations, the size and intensity of management of ORG dairy herds varies considerably. Milking practices used on ORG farms are very similar to practices used on similarly sized CON farms. Several management practices such as use of milking gloves and a quarantine unit to isolate infected cows have been shown to result in improved udder health and milk quality for ORG herds. The use of external consultants has also been associated with improved milk quality. In summary, both ORG and CON herds that use well-known mastitis prevention practices and consult with qualified advisors can minimize mastitis and produce high quality milk.

**Key Words:** organic, milk quality, mastitis

**735 Panel discussion: Best management practices to maintain animal health in organic dairy herds.** J. S. Velez\*<sup>1</sup>, P. Clutts<sup>2</sup>, G. Jodarski<sup>3</sup>, and G. M. Schuenemann<sup>4</sup>, <sup>1</sup>*Aurora Organic Farms, Boulder, CO*, <sup>2</sup>*Pleasantview Farm, Circleville, OH*, <sup>3</sup>*CROOP Cooperative/Organic Valley, La Farge, WI*, <sup>4</sup>*The Ohio State University, Columbus, OH.*

The organic dairy industry in the US has been growing steadily since 1990, and organic farmers have struggled to produce enough supply to meet the growing domestic demand. The whole certified organic process relies on several preventive management practices (e.g., transition cow program, vaccination, water quality, nutrition, reproduction) that ultimately will allow the animal to produce nutritious high quality milk. The basic physiological functions to prevent metabolic disorders in transition cows are dry matter intake and maintenance of calcium and energy balance (ketosis). These factors also determine the status of the immune system of the animal; thus, the prevalence of infectious diseases (e.g., metritis and mastitis), the risk of culling within 60 DIM, and performance of lactating dairy cows. Reproductive failure and mastitis are the top 2 reasons for early removal of lactating cows from dairy herds. This causes severe economic losses to organic dairy producers because of decreased milk yield and quality as well as increased replacement costs. Designing and implementing a proactive transition management program (i.e., protocols) that focuses on monitoring the leading risk factors (e.g., diets and feed management, water quality, comfort of pre- and postpartum cows [e.g., resting time], calving management, and personnel training) will significantly reduce calving-related diseases and the risk of culling while optimizing performance of lactating dairy cows. Therefore, best transition cow management practices during the weeks before and after calving are key determinants to avoid a drop in dry matter intake, hypocalcemia, and ketosis; and the subsequent negative effect on the immune system of animals in organic dairy herds.

**Key Words:** organic, dairy cattle, health



# Beef Species I

**736 Relationships between feed efficiency traits and indicators of energy expenditure in growing cattle.** Phillip A. Lancaster<sup>1</sup>, Gordon E. Carstens<sup>2</sup>, Luis O. Tedeschi<sup>2</sup>, Timothy P. Vining<sup>1</sup>, Nicolas DiLorenzo<sup>3</sup>, and G. Cliff Lamb<sup>3</sup>, <sup>1</sup>*Range Cattle Research and Education Center, University of Florida, Ona, FL*, <sup>2</sup>*Department of Animal Science, Texas A&M University, College Station, TX*, <sup>3</sup>*North Florida Research and Education Center, University of Florida, Marianna, FL*.

Recent research indicates that residual feed intake (RFI) may not be related to efficiency of energy use, but few results have been published for other feed efficiency traits. The current objective was to determine the relationships of different measures of feed efficiency with indicators of energy expenditure at constant feed intake. Five performance trials were conducted to measure feed intake, growth and heart rate (HR) or heat production in growing cattle. Feed efficiency traits included those where a lesser value is more efficient [feed conversion ratio (FCR), RFI, RFI adjusted for fat composition (RFI<sub>c</sub>), RFI from model predicted feed intake (RFI<sub>m</sub>)] and those where a greater value is more efficient [partial efficiency of growth, residual gain efficiency (RGE), and residual intake and gain (RIG), which is combination of RFI and RGE]. Heart rate alone or heat production (heart rate × oxygen pulse) was measured following the performance trial. Trials 1, 3, 4 and 5 included 39 Angus bulls and heifers, 119 Brangus heifers, 115 Brangus heifers, and 45 Angus heifers fed high-roughage diets with heart rate measured on 39 animals at fasting, 16 heifers at 1.1× expected maintenance requirement (MEM), 16 heifers restricted-fed at 120 g DM/kg<sup>75</sup>, and 16 heifers at 1× expected MEM, respectively. Trial 2 included 56 Angus bulls and heifers fed a high-concentrate diet with heart rate measured on 12 bulls and 12 heifers at 1.1× expected MEM. Phenotypic correlations of feed efficiency traits with DMI and ADG were similar to published studies. Phenotypic correlations of feed efficiency traits with fasting HR in Trial 1, maintenance HR in Trial 2 and 3, and restricted-intake HR in Trial 4 were not significant. In Trial 5, FCR (−0.62), RFI (−0.56), RGE (0.60), RFI<sub>c</sub> (−0.60), RFI<sub>m</sub> (−0.60), and RIG (0.65) had strong phenotypic correlations with maintenance heat production such that more efficient heifers had greater maintenance heat production. These data indicate that proposed traits to select for improved feed efficiency have no or an inverse relationship with energy expenditure when growing cattle are fed at constant feed intake.

**Key Words:** cattle, energy expenditure, feed efficiency

**737 Effect of post-weaning heifer development on pregnancy rates and subsequent feed efficiency as a pregnant first calf heifer.** Hazy R. Nielson\*, T. L. Meyer, and Rick N. Funston, *University of Nebraska, West Central Research and Extension Center, North Platte, NE*.

To determine the effect of heifer development system on pregnancy rates and feed efficiency as a pregnant first calf heifer a 3-yr study was conducted. In Yr 1, weaned heifers either grazed corn residue (CR) or were fed in a drylot (DLHI). In Yr 2 and 3 heifers either grazed CR, upland range (RANGE), or were fed diets differing in energy, high (DLHI) or low (DLLO), in a drylot setting. Percent of mature BW before the breeding season was similar among treatments except DLHI which was significantly greater ( $P = 0.04$ ) at 66.6% compared with 60.0, 61.0, and 61.7% for RANGE, CR, and DLLO treatments, respectively. Pregnancy

rates to AI were similar ( $P = 0.62$ ) among treatments (58.6, 66.3, 59.9,  $52.6 \pm 9.7\%$ ; RANGE, CR, DLHI, DLLO). A subset of AI-pregnant heifers from each development treatment were placed in a Calan gate system; they were allowed a 20 d acclimation and training period before beginning the 90 d ad libitum hay treatment period on approximately gestational d 170. Offerings were recorded daily and orts collected weekly. Initial BW was not different ( $P = 0.62$ ) among treatments (458, 468, 473,  $464 \pm 9$  kg; RANGE, CR, DLHI, DLLO). Body weight at the end of the treatment period was also not different ( $[P = 0.55]$  485, 497, 503,  $491 \pm 17$  kg; RANGE, CR, DLHI, DLLO). Intake did not differ among treatments, either as DMI ( $[P = 0.59]$  9.2, 9.4, 9.5,  $9.4 \pm 0.7$  kg; RANGE, CR, DLHI, DLLO) or as a percentage of BW ( $[P = 0.98]$  1.96, 1.95, 1.95,  $1.96 \pm 0.15\%$ ; RANGE, CR, DLHI, DLLO). There was no difference ( $P = 0.61$ ) in ADG (0.28, 0.33, 0.32,  $0.28 \pm 0.17$ ; RANGE, CR, DLHI, DLLO) or residual feed intake ( $P = 0.41$ ) (−0.095, −0.096, 0.144,  $0.113 \pm 0.156$ ; RANGE, CR, DLHI, DLLO) among treatments. Although there was no difference ( $P = 0.41$ ) in the 3-mo-development cost among treatments (\$166.06, 141.66, 160.63,  $171.80 \pm 12.52$ ; RANGE, CR, DLHI, DLLO), there was a \$30.14 numerical difference between the most expensive treatment, DLHI, and the least costly treatment, CR. Post-weaning heifer development system did not affect heifer pregnancy rate or feed conversion as pregnant first calf heifers.

**Key Words:** beef heifer, feed conversion, heifer development

**738 Effects of prepartum supplement level on growth performance, feed efficiency, and reproductive performance of female progeny.** Lindsay M. Shoup\*, Thomas B. Wilson, Dianelys González-Peña, Frank A. Ireland, Sandra Rodriguez-Zas, Tara L. Felix, and Daniel W. Shike, *University of Illinois, Urbana, IL*.

Objectives were to determine the effect of prepartum dam supplement level on growth performance, feed efficiency, and reproductive performance of female progeny (127 heifers in yr 1,  $n = 9$  dam pastures; 138 heifers in yr 2,  $n = 9$  dam pastures). Mature, multiparous, Angus × Simmental cows (BW =  $623 \pm 70$  kg; BCS =  $5.7 \pm 0.7$ ) were offered 1 of 3 supplement levels (supplement contained 70% dried distiller's grains plus solubles and 30% soybean hulls; fed 103 ± 11 d prepartum to 2 ± 11 d postpartum): no supplement (NS), 2.16 kg/d·hd<sup>−1</sup> (LS), or 8.61 kg/d·hd<sup>−1</sup> (HS) while grazing tall fescue pastures. Heifer progeny were weaned at  $187 \pm 11$  d of age and retained as replacements in a fall-calving system. Experimental unit was pasture (9 per year), and data were analyzed using PROC MIXED in SAS. Dam prepartum supplementation did not affect ( $P = 0.99$ ) heifer progeny BW at weaning. During a 70-d intake and feed efficiency evaluation, starting at  $305 \pm 31$  d of age, DMI, ADG, and RFI were similar ( $P \geq 0.14$ ) among heifer progeny, regardless of dam supplement level. Heifer body weight and BCS, at start of 1st breeding season, also did not differ ( $P \geq 0.60$ ). Body weight at pregnancy verification, percentage of heifers that conceived to AI in 1st breeding season, and overall pregnancy percentage in 1st breeding season were not different ( $P \geq 0.51$ ) among heifer progeny, regardless of dam supplement level. At first calving, calf birth BW and percentage of unassisted births were similar ( $P \geq 0.32$ ) among heifer progeny, regardless of dam supplement level. Milk production also did not differ ( $P = 0.48$ ) among heifer progeny, regardless of dam supplement level. In conclusion, these data suggest that within a fall-calving fescue-based production system, supplementing dams with 2.16 or 8.61 kg/d·hd<sup>−1</sup> of

a DDGS-based supplement does not affect growth performance, feed efficiency, and reproductive performance of subsequent female progeny.

**Key Words:** beef heifer development, fetal programming, maternal nutrition

**739 Stocking rate and feeder design affects hay waste .** Dexter J. Tomczak\*, Nick E. Mertz, Dylan L. Hamlin, and William J. Sexten, *University of Missouri-Columbia, Columbia, MO.*

Ninety-six mid-gestation spring-calving cows were stratified by BW ( $562.9 \pm 72.6$ ), BCS ( $4.9 \pm 0.7$ ), and age ( $4.5 \pm 2.0$ ) into 2 replicate groups of 8, 16, and 24 cows to evaluate stocking rate and feeder design effects on hay waste. Cone feeders were equipped with cradle-chains (cone), sheeting on upper (50 cm) and lower (60 cm) portion, and 16 feeding stations (45.2 cm wide) separated by vertical bars (230 cm diameter, 170 cm height). Open feeders (ring) had no sheeting and 17 feeding stations (44.4 cm wide) separated by angled bars (240 cm diameter, 120 cm height). Feeder type and stocking rate were arranged in a  $2 \times 3$  factorial, and randomly assigned to a  $6 \times 6$  Latin square. Tall fescue round hay bales (87.3% DM, 6.33% CP, 66% NDF, 152 cm width  $\times$  170 cm diameter) were offered on circular end and replaced every third d (8), every other d (16), or daily (24) to ensure ad libitum hay access. A single bale was offered to 8 and 16 each period, while 24 were offered 3 bales. Bale number was not significant, so bale measures were averaged within period. Waste was collected daily, and residual forage (ORTs) was collected before new bale offering. Estimated DMI was hypothesized to not differ, while waste was expected to decrease with increasing stocking rate in ring, but not differ in cone. Estimated DMI did not differ ( $P > 0.10$ ) as percent of BW ( $2.0 \pm 0.1\%$ ) or  $\text{kg} \cdot \text{hd}^{-1} \cdot \text{d}^{-1}$  ( $11.8 \pm 0.3 \text{ kg} \cdot \text{hd}^{-1} \cdot \text{d}^{-1}$ ). A stocking rate by feeder interaction was observed due to changes in magnitude of difference when waste was expressed as  $\text{kg} \cdot \text{hd}^{-1} \cdot \text{d}^{-1}$  ( $P = 0.05$ ), percent of disappearance (%disp) ( $P = 0.07$ ), and percent of intake (%intake) ( $P = 0.09$ ). Waste was greater in ring ( $P < 0.05$ ) than cone for 8 ( $2.9$  vs.  $2.0 \text{ kg} \cdot \text{hd}^{-1} \cdot \text{d}^{-1}$ ,  $18.8$  vs.  $14.0\%$ disp,  $23.4$  vs.  $16.4\%$ intake) and 24 ( $3.3$  vs.  $1.7 \text{ kg} \cdot \text{hd}^{-1} \cdot \text{d}^{-1}$ ,  $22.4$  vs.  $12.5\%$ disp,  $29.2$  vs.  $14.7\%$ intake), for 16 waste was increased ( $P = 0.09$ ) ( $2.7$  vs.  $2.1 \text{ kg} \cdot \text{hd}^{-1} \cdot \text{d}^{-1}$ ,  $18.8$  vs.  $15.4\%$ disp,  $23.6$  vs.  $18.3\%$ intake). Increasing stocking rate to greater than 1 cow per individual feeding space did not reduce estimated DMI. Stocking hay feeders greater than 1 cow per individual feeding space or having no defined feeding space within feeder increased hay waste.

**Key Words:** hay waste, stocking rate, feeder design

**740 Effects of feeding stockpiled tall fescue versus tall fescue hay to late gestation beef cows on circulating blood urea nitrogen and glucose concentrations.** Jill M. Larson\*, Katlyn N. Niederecker, and Allison M. Meyer, *Division of Animal Sciences, University of Missouri, Columbia, MO.*

We hypothesized that cows grazing stockpiled tall fescue (STF) during late gestation would have increased nutrient intake compared with cows fed summer-baled hay, which would result in increased nutrient availability for fetal growth and development. Forty-eight multiparous, spring-calving crossbred beef cows were allocated by BW ( $683 \pm 16$  [SE] kg), BCS ( $5.8 \pm 0.1$ ), age ( $5.6 \pm 0.4$  yr), and expected calving date (average = February 18) to 1 of 2 forage systems beginning on d  $188 \pm 2$  of gestation. Forage systems were: 1) strip-grazed endophyte-infected STF (59.7% NDF, 12.3% CP; DM basis; 80.4% utilization) in 4.05-ha pastures ( $n = 4$ ), or 2) ad libitum endophyte-infected tall fescue hay (64.9% NDF, 6.2% CP; DM basis) fed in uncovered drylots (18 m  $\times$

61 m,  $n = 4$ ). Cows remained on their respective treatments until calving. Jugular blood samples were obtained from cows on d 0 (baseline; d 188 of gestation), 35 (d 223 of gestation), and 77 (d 265 of gestation) for determination of serum blood urea nitrogen (BUN) and plasma glucose. Data were analyzed using a mixed model containing effects of treatment, sampling day, and their interaction, where sampling day was used as a repeated measure. Pasture or drylot was considered the experimental unit. During late gestation, there was a forage system  $\times$  day interaction ( $P < 0.001$ ) for serum BUN concentrations. Baseline serum BUN concentrations did not differ ( $P = 0.95$ ) between forage systems. On d 35 and 77 of the study, cows grazing STF had greater ( $P < 0.001$ ) serum BUN concentrations than cows fed hay only. There was no treatment  $\times$  day interaction ( $P = 0.57$ ) for plasma glucose, but cows grazing STF tended to have greater ( $P = 0.11$ ) glucose concentrations than cows consuming hay. Additionally, as gestation progressed, plasma glucose increased ( $P = 0.02$ ). We conclude that cows grazing stockpiled tall fescue had increased circulating BUN and glucose concentrations due to increased nutrient intake during late gestation, which may affect fetal development and subsequent calf performance.

**Key Words:** forage system, metabolite, pregnancy

**741 Preweaning calf circulating blood urea nitrogen and glucose concentrations in a tall fescue forage system model of developmental programming.** Katlyn N. Niederecker\*, Jill M. Larson<sup>1</sup>, Brian L. Vander Ley<sup>2</sup>, and Allison M. Meyer<sup>1</sup>, <sup>1</sup>*Division of Animal Sciences, University of Missouri, Columbia, MO,* <sup>2</sup>*Department of Veterinary Medicine and Surgery, University of Missouri, Columbia, MO.*

We hypothesized that cows grazing stockpiled tall fescue (STF) during late gestation have greater nutrient intake than cows fed summer-baled hay, which would result in increased prenatal nutrient supply and ultimately improved fetal development and subsequent postnatal performance. Forty-eight multiparous, spring-calving crossbred beef cows ( $683 + 16$  [SE] kg BW) were allocated by BW, BCS, age, and expected calving date to either strip-graze endophyte-infected STF (59.7% NDF, 12.3% CP; DM basis;  $n = 4$  pastures) or consume ad libitum endophyte-infected tall fescue hay (HAY; 64.9% NDF, 6.2% CP; DM basis) in uncovered drylots ( $n = 4$ ) beginning on d  $188 \pm 2$  of gestation. Cows remained on their respective forage systems until calving, but STF cows were moved to drylots at  $7.0 \pm 1.0$  d pre-calving and fed ryelage (58.6% NDF, 12.3% CP; DM basis). Within 1 wk postpartum, cow-calf pairs were moved to a single pasture for common pre-weaning management. At  $52 \pm 0.6$  h and  $83 \pm 1$  d postnatally, calf jugular blood samples were obtained for determination of serum blood urea nitrogen (BUN) and plasma glucose. Data were analyzed using a mixed model containing effects of forage system, sampling day, and their interaction, where sampling day was used as a repeated measure. Pasture or drylot was included as the experimental unit; calf date of birth and sex were included in the model when  $P < 0.25$ . We have previously reported that calves born to cows consuming STF tended to weigh more at birth and d 80. There was a tendency ( $P = 0.07$ ) for a forage system  $\times$  day interaction for serum BUN concentrations. At 52 h postnatally, calves born to cows consuming hay had decreased ( $P = 0.02$ ) serum BUN compared with STF, but BUN did not differ ( $P = 0.95$ ) between forage systems at 83 d of age. Treatment did not affect ( $P \geq 0.20$ ) plasma glucose concentrations. Despite this, calves had greater ( $P < 0.001$ ) glucose concentrations at 52 h then at 83 d. In conclusion, neonatal calves born to cows grazing

STF had increased circulating BUN, which may indicate increased perinatal nutrient supply.

**Key Words:** calf metabolite, developmental programming, pregnancy

**742 Effect of supplementing heifers on pasture with bambermycin or monensin on growth and development.** Paul Beck\*<sup>1</sup>, John Tucker<sup>2</sup>, William Galyen<sup>3</sup>, Shane Gadberry<sup>4</sup>, Don Hubbell<sup>2</sup>, Tom Hess<sup>2</sup>, Doug Galloway<sup>3</sup>, Michael Sims<sup>1</sup>, Beth Kegley<sup>3</sup>, and Matt Cravey<sup>5</sup>, <sup>1</sup>University of Arkansas SWREC, Hope, AR, <sup>2</sup>University of Arkansas LFRS, Batesville, AR, <sup>3</sup>University of Arkansas Department of Animal Science, Fayetteville, AR, <sup>4</sup>University of Arkansas Cooperative Extension Service, Little Rock, AR, <sup>5</sup>Huvepharma Inc., Amarillo, TX.

Spring calving (Block 1; n = 70 heifers; BW 208 ± 21.7 kg; age 231 ± 17.0 d) and fall calving (Block 2; n = 72 heifers; BW 225 ± 31.7 kg; age 276 ± 12.8 d) heifers were used to test the effects of feeding 20 mg of bambermycin (Gainpro; Huvepharma; Sofia, Bulgaria) or 150 mg of monensin (Rumensin; Elanco Animal Health, Greenfield, IN) in 0.91 kg/d corn gluten feed based supplements compared with non-medicated (Control) supplements on growth performance and development of grazing heifers. Heifers were allocated by breed, BW, and age to pasture groups (n = 5/group for Block 1 and n = 6/group for Block 2). Pasture groups were then randomly assigned to treatment (Control n = 4 pastures in Blocks 1 and 2; bambermycin and monensin n = 5 pastures in Block 1 and n = 4 pastures in Block 2). Block 1 began on 29 October 2013 and lasted 189-d, and Block 2 began on 24 June 2014 and lasted for 161-d. Heifers were weighed full on 2 consecutive days at beginning and end of each block. Reproductive tract scores (1 to 5 score) were assessed before breeding using ultrasound in Block 1 and rectal palpation in Block 2. In Block 1, heifers grazed non-toxic endophyte infected tall fescue pastures, while in Block 2 heifers grazed bermudagrass pastures until 14 October and then grazed non-toxic endophyte infected tall fescue pastures. Heifers were AI bred over 10-d, and natural service bulls were placed with heifers 14-d following final AI date. Data were analyzed as a randomized complete block design using the mixed procedure of SAS (SAS Inst. Inc., Cary, NC) least squares means were separated using contrasts: Control vs. Medicated and bambermycin vs. monensin. Ending BW and ADG of Control (323 ± 4.8 kg and 0.68 ± 0.0167 kg/d) was less ( $P \leq 0.04$ ) than medicated, yet monensin (346 ± 4.6 kg and 0.73 ± 0.0163 kg/d) and bambermycin (344 ± 4.6 kg and 0.74 ± 0.0163 kg/d) did not differ ( $P \geq 0.69$ ). Reproductive tract scores (3.5 ± 0.60), AI pregnancy rates (30 ± 12.3%) and total pregnancy rates (82 ± 11.5%, respectively) did not differ ( $P \geq 0.36$ ) among treatments. This experiment indicates that both bambermycin and monensin effectively increase performance of growing heifers, but did not affect reproduction.

**Key Words:** bambermycin, heifer development, monensin

**743 Effect of pre and postpartum herbage allowances of grasslands on metabolic and endocrine parameters of primiparous beef cows.** Alberto Casal\*<sup>1</sup>, Martin Clatamunt<sup>2</sup>, Ana L. Astessiano<sup>1</sup>, Pablo Soca<sup>3</sup>, and Mariana Carriquiry<sup>1</sup>, <sup>1</sup>Facultad de Agronomía, Universidad de la Republica, Montevideo, Uruguay, <sup>2</sup>Facultad de Vereterinaria, Universidad de la Republica, Paysandu, Uruguay, <sup>3</sup>Facultad de Agronomía EEMAC, Universidad de la Republica, Paysandu, Uruguay.

The aim of this study was to evaluate the effect of herbage allowance of grasslands during the prepartum and postpartum on metabolic and

endocrine parameters of beef cows. Forty Hereford heifers were used in a randomized block design and a factorial arrangement of prepartum (fall-winter; high vs. low, PREH vs. PREL) and postpartum (spring-summer, high vs. low, POSTH vs. POSTL) herbage allowance (4 vs. 2.5 kg of DM/kg of BW of annual mean for high vs. low). At the -90, -35, +80 and +160 ± 17 DPP (fall, winter, spring and summer, respectively), BW and BCS was recorded and blood samples were collected. Means from a repeated analysis using a mixed model were considered to differ when  $P \leq 0.05$ . During the prepartum (-90 to -35 DPP; fall and winter) all cows lost BW and BCS, increasing plasma urea, nonesterified fatty acids (NEFA) and  $\beta$ -hydroxybutyrate (BHB) while decreasing glucose, insulin and leptin concentrations. During this period, BCS and concentrations of urea were lower and NEFA tended ( $P = 0.09$ ) to be greater in PREL than PREH cows (4.6 vs. 5.0 ± 0.04 units, 7.3 vs. 8.1 ± 0.1 mmol/L and 1.1 vs. 0.9 ± 0.03 mmol/L for BCS, urea and NEFA, respectively). During the postpartum (spring and summer), BCS and metabolic-endocrine profiles were affected by POST but did not differ due to PRE or the PRE by POST interaction. Cow BCS and plasma glucose and leptin were greater whereas urea and NEFA were less in POSTH than POSTL cows (3.9 vs. 3.6 ± 0.04 units, 2.9 vs. 2.7 ± 0.05 mmol/L; 5.7 vs. 3.8 ± 0.1 ng/mL; 5.2 vs. 5.7 ± 0.1 mmol/L and 0.63 vs. 0.66 ± 0.03 mmol/L for BCS, glucose, leptin, urea and NEFA, respectively) during the lactation period. Plasma urea and NEFA decreased during spring to increase again toward mid-summer when insulin and BHB concentrations also decreased. Metabolic and endocrine profiles of grazing primiparous beef heifers reflected negative energy balance as herbage mass and/or quality decreased during winter gestation or during summer in the late lactation. Improve PRE and POST herbage allowance reduced negative energy balance during the pre and postpartum, respectively.

**Key Words:** beef cattle, endocrinology, rangeland

**744 The influence of tick loads of Senepol cows on calf tick loads and production traits in the tropics.** Robert W. Godfrey\*, Adam J. Weis, and Henry C. Nelthropp, *Agricultural Experiment Station, University of the Virgin Islands, St Croix, VI.*

This study was conducted to evaluate the relationship of tick load between multiparous Senepol cows (n = 127 observations) and their calves (n = 144 observations) born in fall 2010 and 2011 and spring 2011, 2012, 2013 and 2014. Cow tick load was evaluated using a visual score (clean, light, moderate or heavy) before monthly acaricide treatment at weaning. Calf BW and tick load were measured at weaning and yearling. Average daily gain (ADG) was calculated for birth to weaning and weaning to yearling. Data were analyzed using correlation and GLM procedures with calving season and cow tick load as the main effects and sire as a covariate due to unequal representation of sires across years. Calves born in the spring had greater 205-d adjusted weaning weights ( $P < 0.002$ ) than calves born in the fall (319.4 ± 10.7 vs. 236.9 ± 23.2 kg, respectively) but there was no effect of cow tick loads ( $P > 0.10$ ). There was no effect of calving season or cow tick load on 365-d adjusted weight of calves ( $P > 0.10$ ). Spring-born calves had a higher ADG from birth to weaning ( $P < 0.0001$ ) than fall-born calves (1.23 ± 0.04 vs. ± 0.93 ± 0.09 kg/d, respectively) but there was no effect of cow tick load ( $P > 0.10$ ). Calves of cows with light tick loads had lower ADG from weaning to yearling ( $P < 0.0001$ ) than calves of cows with clean, moderate or high tick loads (0.29 ± 0.02 vs. 0.49 ± 0.03 vs. 0.45 ± 0.03 vs. 0.44 ± 0.03 kg/d, respectively). Spring-born calves had greater tick loads at weaning ( $P < 0.001$ ) than fall-born calves (2.3 ± 0.1 vs. 1.5 ± 0.2, respectively) but there was no difference ( $P > 0.10$ ) as yearlings. Calves of cows with high tick loads at weaning had lower yearling tick loads ( $P < 0.02$ ) than calves from cows with clean, light or



moderate tick loads ( $1.4 \pm 0.3$  vs.  $2.1 \pm 0.3$  vs.  $2.5 \pm 0.2$  vs.  $2.1 \pm 0.3$ , respectively). Cow tick load at weaning had low correlations with calf tick load at weaning ( $r = 0.24$ ,  $P < 0.008$ ) and at yearling ( $r = -0.23$ ,  $P < 0.04$ ). Calf tick load at weaning was not correlated with tick load at yearling ( $r = -0.01$ ,  $P > 0.10$ ). Cow tick load does not affect calf tick load or pre-weaning growth. Because of the low correlation of tick load between cows and calves, and within calves, it may be difficult to select for this trait.

**Key Words:** cow, calves, tick load

**745 Comparison of the growth of F<sub>1</sub> Senepol x Brahman and Brahman steers in an Indonesian feedlot.** Tim J. Schatz\*, Northern Territory Department of Primary Industry and Fisheries, Darwin, Northern Territory, Australia.

Most cattle in northern Australia have a high *Bos indicus* (usually Brahman) content as they perform better in the harsh conditions than *Bos taurus* cattle. However, Brahmans from northern Australia often suffer price discrimination when they are sent to Australian domestic markets as they are regarded as having less tender meat than *Bos taurus*. Crossbreeding with Senepol bulls has been found to be a way of producing offspring from northern Australian herds that have higher growth rates and more tender meat than Brahmans. However there is some resistance to adoption of crossbreeding as there are concerns that crossbreds are discriminated against in the Indonesian live export market (which is a major destination for young northern Australian cattle) due to the perception that they do not perform as well as high grade Brahmans in the tropical environment. Therefore a study was conducted to compare the growth of Brahman (BRAH) and F<sub>1</sub> Senepol x Brahman (F<sub>1</sub> SEN) steers in an Indonesian feedlot. F<sub>1</sub> SEN (n = 54) and BRAH (n = 32) steers were exported from Darwin (Northern Territory, Australia) to Indonesia through the normal live export process and fed for 121 d in a commercial feedlot near Lampung (Sumatra, Indonesia). The average weights of the genotypes at feedlot induction were 312.3 kg (F<sub>1</sub> SEN) and 308.5 kg (BRAH). All the steers were fed in the same pen and received the normal feedlot management and rations. Their growth was studied and the average daily gain over the feeding period was 0.17 kg/d higher ( $P < 0.001$ ) in the F<sub>1</sub> SEN compared with BRAH (1.71 vs. 1.54 kg/d). As a result the F<sub>1</sub> SEN put on 21.6 kg more weight ( $P = 0.002$ ) over the feeding period. There was no significant difference between the genotypes in average fat depth at the P8 site at the end of the feeding period (F<sub>1</sub> SEN = 10.5 mm, BRAH = 10.6 mm) despite the F<sub>1</sub> SEN being 25.4 kg heavier on average. This study found that that F<sub>1</sub> SEN steers performed better than BRAH in an Indonesian feedlot, and so the results should allow live export cattle buyers to purchase these types of animals (Brahman x tropically adapted *Bos taurus*) with confidence that Indonesian feedlotters will be happy with their performance.

**Key Words:** Senepol, Brahman, feedlot

**746 Alternative weaning and finishing strategies affect finishing residual feed intake of beef steers.** Jason K. Smith\*<sup>1</sup>, Deidre D. Harmon<sup>1</sup>, Mark D. Hanigan<sup>2</sup>, Samer W. El-Kadi<sup>1</sup>, Sally E. Johnson<sup>1</sup>, Scott P. Greiner<sup>1</sup>, and Mark A. McCann<sup>1</sup>, <sup>1</sup>Department of Animal and Poultry Sciences, Virginia Tech, Blacksburg, VA, <sup>2</sup>Department of Dairy Science, Virginia Tech, Blacksburg, VA.

Recent evidence suggests that alternative early nutritional management strategies may metabolically imprint beef steers for reductions in finishing residual feed intake (RFI). To further evaluate this effect, as well as that of an alternative finishing nutritional management strategy, a 2 x 2

x 2 factorial experiment was conducted that included Angus (ANG) and Simmental (SIM) sired steers randomly assigned to one of 2 weaning treatments (early weaned [EW; weaned at 111 ± 23 d of age; n = 14] or conventionally weaned [CW; weaned at 233 ± 21 d of age; n = 14]) and one of 2 finishing treatments (high corn [HC; 68.7% of DM from steam-flaked corn; n = 14] or low corn [LC; 50% of DM from steam-flaked corn isoenergetically replaced with dried corn gluten feed; n = 14]). EW steers were fed a concentrate-based ration ad libitum for 122 d following weaning. All steers were then commingled and backgrounded on pasture for 190 d, finished in a feedlot for 154 ± 64 d, and harvested in groups upon reaching a common 12th-rib fat thickness of 1 cm. Feed intake was measured daily following a 42 d finishing ration adaptation period using a Calan-Broadbent system. Observed ADFI expressed in Mcal of NE<sub>g</sub> was regressed against average BW<sup>0.75</sup>, ADG and duration of the measurement period ( $R^2 = 0.73$ ;  $P < 0.0001$ ) via the Fit Model procedure of JMP Pro, and RFI was calculated as the difference between observed and predicted ADFI. Analysis of variance was conducted to determine the fixed main and interaction effects of sire breed, weaning treatment and finishing treatment on ADFI, ADG and RFI. Although no effects were observed for ADFI ( $P \geq 0.45$ ) or ADG ( $P \geq 0.36$ ), RFI of ANG HC steers was lower than ANG LC (-0.91 vs. 0.49 Mcal NE<sub>g</sub>; SEM = 0.22;  $P < 0.001$ ), SIM LC (-0.91 vs. 0.20 Mcal NE<sub>g</sub>; SEM = 0.22;  $P < 0.01$ ) and SIM HC (-0.91 vs. 0.10 Mcal NE<sub>g</sub>; SEM = 0.22;  $P < 0.05$ ), and was lower for EW than CW steers (-0.27 vs. 0.21 Mcal NE<sub>g</sub>; SEM = 0.15;  $P < 0.05$ ). These results provide additional evidence of metabolic imprinting of EW steers for reductions in RFI, and independent effects of finishing energy source on RFI of ANG steers.

**Key Words:** early weaning, metabolic imprinting, residual feed intake

**747 Alternative weaning and finishing strategies affect beef carcass marbling score.** Jason K. Smith\*<sup>1</sup>, Hailey S. Cassell<sup>1</sup>, Deidre D. Harmon<sup>1</sup>, Mark D. Hanigan<sup>2</sup>, Samer W. El-Kadi<sup>1</sup>, Sally E. Johnson<sup>1</sup>, Scott P. Greiner<sup>1</sup>, and Mark A. McCann<sup>1</sup>, <sup>1</sup>Department of Animal and Poultry Sciences, Virginia Tech, Blacksburg, VA, <sup>2</sup>Department of Dairy Science, Virginia Tech, Blacksburg, VA.

Increased volatility in climate, grain prices and grid-based carcass premiums have led beef producers to search for alternative weaning and finishing management options. An experiment was conducted to evaluate the effects of an alternative weaning strategy and finishing diet on carcass marbling score (MS), quality grade (QG) and Certified Angus Beef LLC (CAB) retail brand acceptance. Angus (ANG) and Simmental (SIM) sired steers were randomly assigned to one of 2 weaning treatments (early weaned [EW; weaned at 111 ± 23 d of age; n = 14] or conventionally weaned [CW; weaned at 233 ± 21 d of age; n = 14]) and 1 of 2 finishing treatments (high corn [HC; 68.7% of DM from steam-flaked corn; n = 14] or low corn [LC; 50% of DM from steam-flaked corn isoenergetically replaced with dried corn gluten feed; n = 14]) in a 2 x 2 x 2 factorial design. Following weaning, EW steers were fed a concentrate-based ration ad libitum for 122 d before commingling and pasture backgrounding with CW steers for 190 d. Steers were then finished for 154 ± 64 d and harvested in groups upon reaching a common ultrasound-estimated 12th-rib fat thickness of 1 cm. Chilled carcasses were evaluated to determine MS, QG and CAB acceptance, and ANOVA was conducted to determine the fixed main and interaction effects of sire breed, weaning treatment and finishing treatment using the Fit Model procedure of JMP Pro. All interaction effects were removed from the final ANOVA model due to lack of significance ( $P \geq 0.54$ ). MS was greater for carcasses of EW than CW steers (741 ± 14 vs. 680 ± 15;  $P < 0.01$ ), and HC than LC steers (775 ± 17 vs. 646 ± 15;  $P < 0.0001$ ),

however no main effects were evident for QG or CAB acceptance ( $\chi^2 \leq 2.32$ ;  $P \geq 0.13$ ). Although weaning and finishing treatments affected MS, the effects were not detectable in QG and CAB acceptance within this experiment, and further research is warranted to increase sample size and statistical power. Nonetheless, the management practices utilized in

this experiment affected MS independently, indicating that both early and late nutritional management strategies affect marbling development.

**Key Words:** early weaning, carcass, marbling

## Breeding and Genetics: Application and methods—Dairy II

### 748 The “it factor” for long-lived, high-producing dairy cows.

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Holstein cows that dairyman love live a long time and produce lots of milk. What is unique about these high-producing, long-lived cows? Obviously, these cows have received good management and avoided major health issues. Are the 50K genetics of these elite cows different from other Holsteins? Elite cows were defined phenotypically as having produced over 68,039 kg (150,000 lb) of milk during their lifetime and were classified as very good (VG) or excellent (EX). Elite cows were born in the decade before 2008. For a control, females born in the decade before 2008 with a 50K Holstein genome evaluation were chosen. Control females either had not produced 68,039 kg of milk during their lifetime or were not classified VG or EX. Genomes (50K or 77K) were available on 823 elite cows and 1,589 control females. Defining elite or control as binomial allowed detection of almost 200 markers that were different in allele frequency between elite cows and control females. The most significant chi-squared for differences in allele frequency between elite cows and control females identified a marker on chromosome 5, which had the largest difference in minor allele frequency of 0.17 between elite and control groups. Basing significance on chi-squared  $-\log_{10} P$  of 8.000 as a threshold, 199 markers were significant and were distributed across all bovine chromosomes. Minor allele frequencies of elite cows were greater for 140 of these markers and minor allele frequencies of control females were greater for 59 markers. As interpretation, minor allele frequencies define uniqueness. The uniqueness of elite cows was supported by more positive changes in minor allele frequencies for the elite cows. A single “it factor” is insufficient to identify uniqueness of elite cows, but many markers are candidates to contribute to the uniqueness. Because allele frequency differences were found across all chromosomes, balance continues to be important in striving to increase the number of elite cows in the breed.

**Key Words:** genome, milk production, longevity

### 749 Identification of gene networks underlying dystocia in dairy cattle.

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Dystocia is a trait with high impact in the dairy industry. Among its risk factors are calf weight, gestation length, breed and conformation. Biological networks have been proposed to capture the genetic architecture of complex traits, where GWAS show limitations. Our objective was to identify gene networks in Brown Swiss (BS), Holstein (HO) and Jersey (JE) cattle related to dystocia. De-regressed PTA (dPTA) for calving ease, gestation length, stature, strength and rump width of 8780 HO, 505 BS, and 1818 JE bulls were used in the analysis. A total of 45188 genotypes were available for all bulls. A single trait Bayes B GWAS was performed within breed with  $\pi = 0.9$ . The proportion of genetic variance ( $PV_g$ ) explained by each SNP was  $(2pq\tilde{a}^2)/\sum^{45188}(2pq\tilde{a}^2)$ , with  $\tilde{a}$  = posterior mean of the allelic effect. SNP with  $VP_g \geq 75$ th percentile of the sample were ruled significant. Relevant SNP (rSNP) were defined as: significant in all traits, significant in all functional traits, or significant in all type traits. An association weight matrix (AWM) was constructed with rSNP in rows and traits in columns. Cells of the AWM corresponded

to rSNP normalized effect size. These were mapped to genes with a 5' or 3' maximum distance of 2500 bp, rows in the AWM were indexed with genes. Genes were used to search for enriched functional annotation (FDR  $\leq 0.15$  HO, JE; FDR  $\leq 0.3$  BS). AWM row-wise partial correlations were computed. Significant correlations were interpreted as gene-gene interactions, resulting in a gene network. Networks included 1454 (BS), 1272 (HO) and 1455 (JE) genes. Their number of connections ranged between 1 and 15 (BS), 80 (HO), 13 (JE). A total of 13 (BS), 152 (HO), 108 (JE) genes in the networks were within reported dystocia QTL. Top enriched terms were cell adhesion (HO, JE), regulation of purine nucleotide metabolic process (BS). Most connected genes in the networks, enriching GO terms and within dystocia QTL were: FLOT1 (BS, 9 interactions), RASA1 (HO, 77) and ADRBK2 (JE, 12). Integrating knowledge from annotation tools to identify the functional biology of dystocia in dairy cattle can potentially improve genomic predictions that could result in increasing profitability of the dairy industry.

**Key Words:** dystocia, gene network, dairy cattle

### 750 Distribution of runs of homozygosity and its association with inbreeding depression in United States and Australia Jersey cattle.

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Differences in environment, management practices or selection objectives have led to a variety of choices being made in the use of dairy sires between countries. This may result in variation in selection intensity across the genome and could result in detectable differences in patterns of genome-level homozygosity between populations and consequently affect inbreeding depression differently across populations. The objective of the study was to characterize the frequency of homozygosity and its relationship with regions associated with inbreeding depression in Jersey dairy cattle from the United States (US) and Australia (AU). Genotyped cows with phenotypes on milk, fat and protein yield ( $n = 6,751$  US;  $n = 3,974$  AU) and calving interval ( $n = 5,816$  US;  $n = 3,905$  AU) were utilized in a 2-stage analysis. A run of homozygosity statistic (ROH4Mb), counting the frequency of a SNP being in a ROH of at least 4 Mb, was calculated across the genome. In the first stage residuals were obtained from a model that accounted for the additive genetic as well as fixed effects. In the second stage these residuals were regressed on ROH4Mb using a single marker regression model or a machine-learning tree based regression algorithm (gradient boosted machine). The relationship between ROH4Mb and the SNP effect of a region for each trait was further characterized based on sliding window (500kb) direct genomic value (DGV) derived from a Bayesian LASSO analysis. The ROH4Mb effects were estimated by regressing residuals from the 2-stage approach on ROH4Mb and SNP effects estimated by regressing residual deviations from a model including only fixed effects on SNP markers. Genomic regions across multiple traits were found to be associated with ROH4Mb on BTA13, BTA23 and BTA25 for the US population and BTA3, BTA7, BTA17 for the AU population. Furthermore, multiple potential epistatic interactions were characterized. Lastly, the covariance between ROH4Mb and the SNP effect of



a region depended on the genome region, with positive covariances in some regions

**Key Words:** run-of-homozygosity, genome-wide association study, inbreeding depression

**751 The effect of *DGATI* polymorphism on milk production and fat, protein, and mineral composition of dairy cattle.** Henk Bovenhuis\*<sup>1</sup>, Marleen Vikser<sup>1</sup>, Nina Poulsen<sup>2</sup>, Jakob Sehested<sup>3</sup>, Hein van Valenberg<sup>4</sup>, Johan van Arendonk<sup>1</sup>, Lotte Bach Larsen<sup>2</sup>, and Bart Buitenhuis<sup>5</sup>, <sup>1</sup>*Animal Breeding and Genomics Centre, Wageningen University, Wageningen, the Netherlands*, <sup>2</sup>*Department of Food Science, Aarhus University, Tjele, Denmark*, <sup>3</sup>*Department of Animal Science, Aarhus University, Tjele, Denmark*, <sup>4</sup>*Dairy Science and Technology Group, Wageningen University, Wageningen, the Netherlands*, <sup>5</sup>*Center for Quantitative Genetics and Genomics, Department of Molecular Biology and Genetics, Aarhus University, Tjele, Denmark*.

Since the identification of the diacylglycerol O-acyltransferase 1 (*DGATI*) K232A polymorphism, many studies reported associations between this polymorphism and milk production traits but only a few investigated effects on detailed milk composition. In the current study data collected as part of the Dutch and Danish-Swedish Milk Genomic Initiatives were used to estimate the effect of *DGATI* polymorphism on milk fat, protein, and mineral composition in Holstein and Jersey cattle. The use of data from different breeds allows identifying associations of *DGATI* that are consistent across populations. Results showed that effects of *DGATI* K232A polymorphism on especially milk yield, fat% and protein% are not constant during lactation. Effects of *DGATI* were small in early lactation and they gradually increased until d 50 to 150 in lactation. Analyses of the effects of *DGATI* polymorphism on detailed milk fat, milk protein, and mineral composition showed that *DGATI* has major effects on fat- and mineral composition of milk. There is also evidence for effects on milk protein composition but these effects seem to be more subtle. Part of the effects of the *DGATI* polymorphism on milk composition can be explained by effects on de novo fatty acid synthesis and on excretion of water (dilution effect). For example, the total amount of Ca, P, and Zn excreted in milk of cows with different *DGATI* K232A genotypes is identical; however, the milk volume differs and therefore contents of Ca, P, and Zn differed between *DGATI* genotypes. A similar “dilution effect” can be observed when studying effects on fatty acids: there were no significant effects of *DGATI* on the yields of C18:1 *cis*-9, CLA *cis*-9,*trans*-11, C18:2 *cis*-9,12 and C18:3 *cis*-9,12,15; however, there were highly significant effects of *DGATI* on these fatty acids when expressed as w/w%.

**Key Words:** *DGATI*, mineral, fatty acid

**752 Variation in milk fat globule size in Canadian dairy cattle and its prediction using mid-infrared spectroscopy.** Allison Fleming\*<sup>1</sup>, Astrid Koeck<sup>1</sup>, Flavio Schenkel<sup>1</sup>, Milena Corredig<sup>2,3</sup>, Mehdi Sargolzaei<sup>1,4</sup>, Bonnie Mallard<sup>5</sup>, R. Ayesha Ali<sup>6</sup>, Saranya Gunasegaram<sup>1</sup>, and Filippo Miglior<sup>1,7</sup>, <sup>1</sup>*CGIL, University of Guelph, Guelph, ON, Canada*, <sup>2</sup>*Gay Lea Foods, Mississauga ON, Canada*, <sup>3</sup>*Dept of Food Science, University of Guelph, Guelph, ON, Canada*, <sup>4</sup>*Semex, Guelph, ON, Canada*, <sup>5</sup>*Dept of Pathobiology, OVC, University of Guelph, Guelph, ON, Canada*, <sup>6</sup>*Dept of Mathematics and Statistics, University of Guelph, Guelph, ON, Canada*, <sup>7</sup>*Canadian Dairy Network, Guelph, ON, Canada*.

Milk fat globule (MFG) size in bovine milk is a trait of interest for selection due to its influence on the composition and nutritional quality of the milk as well as its technological properties. However, large-scale phenotyping is currently impractical. For a given fat content, milk with smaller MFG will have more membrane material, which has been proposed nutraceutical. The objective of this study was to examine the variation in MFG size and the potential of its prediction using mid-infrared spectral data. A total of 1,689 milk samples from 343 Canadian cows representing 4 dairy breeds from 44 herds across the provinces of Ontario, Alberta, and Quebec were collected during routine milk testing. Samples were divided and a certain portion was sent to a Canadian DHI laboratory and their spectral data were recorded using FOSS MIR machines. The MIR data for each sample contained 1,060 data points in the infrared range from 900 to 5,000 cm<sup>-1</sup>. The additional portion was analyzed for mean MFG size using integrated light scattering and reported as both a volume moment mean and surface moment mean. Mean values (±SD) for volume and surface moment means were 4.16 ± 0.51 μm and 3.49 ± 0.34 μm, respectively. The average MFG size was positively correlated with the percent fat in the sample (volume moment mean, r = 0.30; surface moment mean, r = 0.32). MFG size records were combined with their spectral data, and outliers and non-informative regions of the spectrum were removed. Records were randomly assigned to either the training or validation sets. Partial least squares regression method was utilized to predict MFG size. For volume moment mean, an R<sup>2</sup><sub>cv</sub> of 0.52 and R<sup>2</sup><sub>v</sub> of 0.41 were found. Surface moment mean equations had an R<sup>2</sup><sub>cv</sub> of 0.55 and R<sup>2</sup><sub>v</sub> of 0.52. At this time MFG size cannot be accurately quantified through MIR prediction, but it may be possible to identify milk samples with either small or large MFG.

**Key Words:** milk fat globule, dairy cattle, mid-infrared spectroscopy

**753 Including different groups of genotyped females for genomic prediction in the Nordic Jersey population.** Hongding Gao\*<sup>1</sup>, Per Madsen<sup>1</sup>, Ulrik S. Nielsen<sup>2</sup>, Gert P. Aamand<sup>3</sup>, and Just Jensen<sup>1</sup>, <sup>1</sup>*Center for Quantitative Genetics and Genomics, Department of Molecular Biology and Genetics, Aarhus University, Tjele, Denmark*, <sup>2</sup>*Knowledge Centre For Agriculture, Aarhus N, Denmark*, <sup>3</sup>*Nordic Cattle Genetic Evaluation, Aarhus N, Denmark*.

Including genotyped females in the reference population (RP) is an obvious way to increase RP but caution is needed because of potential preferential treatment of the genotyped cows and lower reliabilities of phenotypes compared with proven bulls. Denmark, Finland and Sweden have implemented a female genotyping project with voluntary genotyping of entire herds using low-density chip (LD project). The objective of the present study was to examine the effect of adding different sources of genotyped females to RP for Nordic Jersey. Five scenarios for building RP were considered: (1) bulls only; (2) bulls with females from LD project; (3) bulls with females from LD project plus non-LD project females genotyped before their first calving date; (4) bulls with females from LD project plus non-LD project females genotyped after their first calving date; (5) bulls with all genotyped females included. Genomically enhanced breeding values (GEBV) were predicted for 8 traits in the Nordic Total Merit (NTM) index through a genomic BLUP (GBLUP) model using deregressed proofs (DRP). The validation population (VP) was formed by a cut-off using birth year of 2005 based on the genotyped bulls with DRPs. Average gain in reliability over the 8 traits ranged from 1.8% to 4.5% points compared with the scenario with only bulls in RP (scenario 1). Adding all the genotyped females in the RP achieved highest gain in reliability (scenario 5), followed by scenario 3, scenario 2 and scenario 4. The mean reliability of scenario 3 was 0.5% points higher than scenario 2 due to a slightly larger size

of RP, and a decrease of 1.1% points in mean reliability were observed when including the extra 143 genotypes cows in scenario 4 compared with scenario 2. The mean reliabilities of scenario 2 and 3 were 1.6 and 1.1% points lower than of scenario 5. All scenarios led to inflated GEBVs since the regression coefficients are below 1. However, scenario 2 and scenario 3 led to less bias of genomic predictions than scenario 5 with the mean regression coefficients closer to 1. The results suggest adding unselected females in the RP significantly improve the reliabilities and tend to reduce the prediction bias compared with adding selectively genotyped females.

**Key Words:** genotyped female, reliability, prediction bias

**754 Estimation of genetic parameters for metabolic disease traits and their predictors in Canadian Holsteins.** Astrid Koeck\*<sup>1</sup>, Janusz Jamrozik<sup>1,2</sup>, Gerrit J. Kistemaker<sup>2</sup>, Flavio S. Schenkel<sup>1</sup>, Robert K. Moore<sup>4</sup>, Daniel M. Lefebvre<sup>4</sup>, David F. Kelton<sup>3</sup>, and Filippo Miglior<sup>1,2</sup>, <sup>1</sup>CGIL, Dept. of Animal and Poultry Science, Guelph, ON, Canada, <sup>2</sup>Canadian Dairy Network, Guelph, ON, Canada, <sup>3</sup>Department of Population Medicine, Ontario Veterinary College, Guelph, ON, Canada, <sup>4</sup>Valacta, Québec, QC, Canada.

The objective of this study was to estimate genetic parameters for metabolic diseases and their main predictors in Canadian Holsteins. Records from first to fifth lactation were considered for ketosis (KET), displaced abomasum (DA), milk fever (MF), fat to protein ratio (F:P) and milk  $\beta$ -hydroxybutyrate (BHBA), whereas for body condition score (BCS) only records from first lactation cows were available. Binary disease traits (0 = no case, 1 = at least one case), F:P and milk BHBA were treated as different traits in first and later lactations. Records for MF in first lactation were not considered in the present study as the disease frequency was near zero and a preliminary analysis revealed a heritability of zero. Bivariate and multivariate linear sire models were fitted using AI-REML. Heritability for metabolic disease traits ranged from 0.011 to 0.047. Higher heritabilities were found for BCS, F:P and milk BHBA, with estimates ranging from 0.10 to 0.22. First-lactation KET was strongly correlated with DA (0.76) and milk BHBA (0.75), whereas lower genetic correlations were found with BCS and F:P (-0.54 and 0.37, respectively). Displaced abomasum in first lactation was moderately correlated with BCS (-0.40) and F:P (0.19). Similar genetic correlation estimates were estimated in higher lactation cows. Milk fever, which was only evaluated in higher lactation cows, was moderately correlated with KET (0.39) and milk BHBA (0.33). Genetic correlations of disease traits between first and later lactations were relatively high (0.79 for KET and 0.86 for DA).

**Key Words:** metabolic disease, predictor, genetic correlation

**755 A genomic-wide association study on development of hyperketonemia in periparturient Holstein dairy cows.** Francisco A. Leal Yepes\*<sup>1</sup>, Heather J. Huson<sup>1</sup>, Sabine Mann<sup>2</sup>, Jessica A. A. McArt<sup>2</sup>, Luciano Caixeta<sup>1</sup>, Thomas R. Overton<sup>1</sup>, Joseph J. Wakshlag<sup>2</sup>, and Daryl V. Nysdam<sup>2</sup>, <sup>1</sup>College of Agriculture and Life Sciences, Cornell University, Ithaca, NY, <sup>2</sup>College of Veterinary Medicine, Cornell University, Ithaca, NY.

The objective was to detect cows with elevated postpartum nonesterified fatty acids (NEFA) and  $\beta$ -hydroxybutyrate (BHBA) concentrations, with or without concurrent hyperketonemia, and identify genomic regions associated with development of hyperketonemia in periparturient Holstein cows. The study population consisted of cows from 2 different trials: In the first study, 63 cows (parity  $\geq 2$ ) were enrolled and blood

was collected from 3 to 16 d in milk. In the second study, 84 cows (parity  $\geq 2$ ) were enrolled and sampled from -21 d to +21 d relative to calving. Blood samples were tested for NEFA and BHBA concentration. Hyperketonemia was defined as a BHBA concentration  $\geq 1.2$  mmol/L. All BHBA and NEFA measurements were grouped using incremental area under the curve (AUC) to identify individuals with the most variation. Holstein cows were genotyped on the Illumina Bovine High-density (777K) Beadchip. Quality control filtering produced ( $n = 522,231$ ) single-nucleotide polymorphism (SNP). A genomic wide association study was performed to establish correlation between low frequency SNP ( $< 5\%$ ) and development of hyperketonemia using Golden Helix software. The linear regression  $R^2 = 0.21$  suggested a low strength correlation between BHBA AUC and NEFA AUC concentration. Although a small sample size, given that these cows were managed under similar conditions, multiple SNP associated with high concentrations of BHBA were found (Table 1). These results might improve genetic selection criteria to identify high-risk animals and develop preventative measures to decrease hyperketonemia development.

**Table 1 (Abstr. 755).** Regions and candidate genes associated with development of hyperketonemia

Index	Chr.	Region start (bp)	Region end (bp)	$-\log_{10}$ (P-value)	Genes
1	2	12,122,028	12,128,393	5.086193951	<i>RRAGA</i> <i>PECR</i> ; <i>IGFBP2</i> and <i>IGFBP5</i>
2	2	104,958,840	104,958,840	5.410113588	<i>LRP8</i> ; <i>CPT2</i> and <i>SCP2</i>
3	3	93,634,769	93,634,769	5.031371132	<i>CHD2</i>
6	21	14,153,070	14,153,070	5.111968697	<i>SV2B</i>
7	21	16,112,290	17,017,717	6.451923226	

**Key Words:** hyperketonemia, SNP, gene

**756 Evaluation of survival in the first year after calving across years and seasons.** Mathijs L. van Pelt\*<sup>1,2</sup> and Roel F. Veerkamp<sup>1</sup>, <sup>1</sup>Wageningen UR Livestock Research, Animal Breeding and Genomics Centre, Wageningen, the Netherlands, <sup>2</sup>CRV BV, Arnhem, the Netherlands.

Longevity of dairy cattle is an important trait from an economic and welfare perspective. Dairy cows are culled for various reasons and therefore the trait definition of longevity might have changed over time. Also, culling is likely to be affected by the end of the quota year, and therefore a seasonal effect is expected. Therefore the objective was to evaluate if longevity is a different trait between years or seasons of first calving, utilizing genetic links through common bulls. Survival was defined as survival until 12 mo after first calving. The data set comprised 524,529 animals that could have lived until 12 mo after the first calving, with the first calving between 1988 and 2011. Multiple trait models were used with survival defined in 3 8-year blocks (1988–1995, 1996–2003, and 2004–2011) or 4 seasons of first calving (Jan-Mar, Apr-Jun, Jul-Sep, and Oct-Dec). The mean survival was 87.8% in the total data set, but increased with more recent calving years. The lowest survival rate was in 1991 with 82.9% surviving the first 12 mo and highest survival rate was in 2007 (92.2%). Survival rate also varied between calving months, with highest survival rate in September with 89.2% and lowest survival rate in March with 86.3%. Age of first calving showed an optimum for survival for heifers calved between 22 and 26 mo of age, with survival rates of 88.4% or higher. Survival rate decreases 0.75% for each extra month calving after 26 mo. Genetic correlations between 1988 and 1995 and 2004–200 was lowest (0.67  $\pm$  0.15), and suggests survival

has changed over years. Genetic correlations between all seasons were higher than 0.90, and although there appears a phenotypic effect of the end of the quota year on survival, genetically there is no effect of season of calving on survival until 12 mo after first calving.

**Key Words:** longevity, survival, genetic correlation

**757 Genetic and genomic analysis of superovulatory response in Canadian Holsteins.** Cindy Jaton\*<sup>1,2</sup>, Astrid Koeck<sup>1</sup>, Mehdi Sargolzaei<sup>1,3</sup>, Christopher A. Price<sup>4</sup>, Flavio S. Schenkel<sup>1</sup>, and Filippo Miglior<sup>1,5</sup>, <sup>1</sup>Centre for Genetic Improvement of Livestock, University of Guelph, Guelph, ON, Canada, <sup>2</sup>Centre d'insémination artificielle du Québec, St-Hyacinthe, QC, Canada, <sup>3</sup>Semex Alliance, Guelph, ON, Canada, <sup>4</sup>Université de Montréal, Faculté de Médecine Vétérinaire, St-Hyacinthe, QC, Canada, <sup>5</sup>Canadian Dairy Network, Guelph, ON, Canada.

Superovulation of dairy cattle is frequently used in Canada. The cost of this protocol is high, and so is the variability of the outcome. Knowing the superovulatory potential of a donor cow could influence the breeder's decision to superovulate it or not. The ultimate objective of this study was to identify chromosome regions associated with superovulatory response in Canadian Holsteins. Data were provided by Holstein Canada and contained the total number of embryos and the number of viable embryos from every successful flushing performed across Canada. After editing, 137,446 records of superovulation done between 1992 and 2014 were considered for the analysis. A univariate repeatability animal model analysis was performed for both total number of embryos and number of viable embryos, yielding heritability estimates (SE) of 0.18 (0.01) and 0.14 (0.01), respectively. Breeding values were estimated for 54,463 cows, and 3,513 sires. Only estimated breeding values of animals that were genotyped with a SNP panel denser than 3K and having a reliability higher than 40% were considered ( $n = 5,122$ ) for further analyses. All lower density genotypes were imputed to 50K using FImpute software. A genome-wide association study was carried out using a single SNP regression method, which also fits a polygenic background effect. Results were similar for the 2 highly genetically correlated ( $r_g = 0.94$ ) traits considered in this study. A major significant peak on chromosome 11 was detected, suggesting the presence of an important candidate gene in this region.

**Key Words:** superovulatory response, heritability, GWAS

**759 Genetic parameters of individual hoof lesions in Canadian Holsteins.** Francesca Malchiodi\*<sup>1</sup>, Astrid Koeck<sup>1</sup>, Núria Chapinal<sup>2</sup>, Mehdi Sargolzaei<sup>1,3</sup>, Allison Fleming<sup>1</sup>, David F. Kelton<sup>4</sup>, Flavio S. Schenkel<sup>1</sup>, and Filippo Miglior<sup>1,5</sup>, <sup>1</sup>Centre for Genetic Improvement of Livestock, University of Guelph, Guelph, ON, Canada, <sup>2</sup>Animal Welfare Group, University of British Columbia, Vancouver, BC, Canada, <sup>3</sup>Semex Alliance, Guelph, ON, Canada, <sup>4</sup>Department of Population Medicine, Ontario Veterinary College, Guelph, ON, Canada, <sup>5</sup>Canadian Dairy Network, Guelph, ON, Canada.

Hoof lesions in dairy herds represent a painful condition for the cow and pose a financial loss for farmers, due to the costs associated with treating lesions, as well as to decreased cow performance. The objective of this study was to estimate genetic parameters for individual hoof lesions in Canadian Holsteins. Data were recorded by 26 hoof trimmers serving 365 herds located in Alberta, British Columbia and Ontario, and trained to use a rugged touch-screen computerized lesion recording system. A total of 108,032 hoof-trimming records from 53,654 cows were collected between 2009 and 2012. Hoof lesions included in the analysis were digital dermatitis, interdigital dermatitis, interdigital hyperplasia, sole hemorrhage, sole ulcer, toe ulcer, and white line lesion. All variables were analyzed as binary traits, as the presence or the absence of the lesions, and as categorical variables, using a severity score from 1 to 3. Only the first hoof-trimming session of each lactation was included in the analyses. When considering the presence or absence of the lesions, heritabilities (SE) for digital dermatitis, interdigital dermatitis, interdigital hyperplasia, sole hemorrhage, sole ulcer, toe ulcer, and white line lesion were 0.053 (0.005), 0.011 (0.002), 0.025 (0.004), 0.012 (0.002), 0.031 (0.004), 0.004 (0.001), and 0.012 (0.002), respectively. Similar results were found when severity was considered. The estimated breeding values between these 2 set of variables showed correlations ranging from 0.88 to 0.97. The reliability of EBV was slightly higher when severity was analyzed. Infection lesions showed moderate genetic correlations with interdigital hyperplasia and low negative correlations with horn lesions. Among horn lesions, moderate to high genetic correlations were found.

**Key Words:** hoof lesion, dairy cattle



## Breeding and Genetics: Poultry and swine

**760 Comparison of traditional vs. genomic, and single vs. multiple trait analyses of broiler chicken mortality.** Xinyue Zhang\*<sup>1</sup>, Shogo Tsuruta<sup>1</sup>, Daniela A. L. Lourenco<sup>1</sup>, Robyn L. Sapp<sup>2</sup>, and Rachel J. Hawken<sup>2</sup>, <sup>1</sup>University of Georgia, Athens, GA, <sup>2</sup>Cobb-Vantress Inc., Siloam Springs, GA.

The purpose of this study was to determine whether broiler mortality is influenced by selection on correlated traits, determine the accuracy of genetic evaluation for mortality with traditional and genomic evaluations, and determine whether the use of the multi-trait model increases the accuracy of predictions. Phenotypes were available on 181,022 broilers for up to 8 traits, 4 linear and 4 categorical. Pedigree was available for 186,596 broilers and genotypes were available on 18,047 animals. For linear traits the model included the fixed effects of sex and contemporary group, random direct genetic, and maternal genetic effect for body weight. Contemporary group is the grouping of source, mini-generation and hatch. For categorical traits, the contemporary group was treated as random and a fixed effect of generation was added. (Co) variance components were estimated with a Gibbs sampling program THRGIBBS1F90 for threshold-linear models. A traditional BLUP using a pedigree relationship matrix and a genomic BLUP (ssGBLUP) using a combined pedigree-genomic relationship matrix were used to predict EBV and genomic EBV, respectively. Because few dead animals were genotyped, the traditional validation techniques did not apply. Subsequently models were compared using a data splitting technique based on the correlation of EBV from 2 non-overlapping samples, each one with one-half of the phenotypes selected across contemporary groups. The correlations were computed only for genotyped animals in the last generation and are measures of realized accuracy. The genetic correlation between mortality and the other traits were generally small (absolute value < 0.15), with notable differences for maternal body weight (-0.50) and Ascites (0.77); heritability for mortality was 13%. The correlations between independent samples in a univariate model for mortality were 0.59 for the non-genomic and 0.64 for the genomic model. In a multiple trait genomic model, the correlations increased by 0.09 over single trait genomic model. Results indicate mortality may be more affected by ascites and maternal growth compared with other traits under selection. Use of the genomic data increases the accuracy of genetic evaluation for mortality.

**Key Words:** mortality, broiler, threshold model

**761 Polymorphisms in *CAST*, *TNNI1*, and *MYOG* genes and their relationship with pig carcass traits at different weight groups.** Andrea Nyisalovits\*<sup>1</sup>, János Posta<sup>2</sup>, Levente Czeglédi<sup>2</sup>, Márta Horváth<sup>1</sup>, and László Babinszky<sup>1</sup>, <sup>1</sup>Department of Feed- and Food Biotechnology, University of Debrecen, Debrecen, Hungary, <sup>2</sup>Department of Animal Breeding, University of Debrecen, Debrecen, Hungary.

The aim of this study is to determine the relationship between 3 previously described polymorphisms (SNPs) located at 3 candidate genes (*CAST*, *TNNI1*, *MYOG*) and slaughter traits in different weight groups of commercially housed hybrid pigs. A total of 402 pigs [Large White × (Landrace × Duroc) sows line × Pietrain boar line] were tested using PCR-RFLP method. After 111 and 118 d in fattening (DF) their carcasses were qualified in a slaughterhouse where the hot weight (HW), backfat thickness (BF) and loin diameter (LD) were measured, lean meat % (LM %), live weight (LW) and average daily gain (ADG) were

calculated. Because of the wide range of LW (82–144 kg) heterogeneity of experimental population had to be reduced, therefore animals were divided into 3 groups based on mean (116.1 kg) and SD (10.2 kg) of LW as follows: low (L) (<mean-1 SD, n = 63), medium (M) (mean ± 1 SD, n = 276) and high (H) (>mean+1 SD, n = 63). The effects of SNPs on carcass quality traits were analyzed in the 3 classes separately using the least square analysis of the GLM method with Tukey-Kramer correction (SAS 9.1) including sex and DF as fixed effects and LW as covariance factor. According to our results the examined SNPs show different effects on carcass quality at different weight. Pigs with the GG genotype of *CAST* (Ser66Asn) gene show the worst BF and LM % ( $P < 0.05$ ) values in the L group, but no significant results were found in the other 2 groups. The *TNNI1* gene has no significant effect on traits. The SNP in 3' UTR region of *MYOG* gene influenced BF and LM % ( $P < 0.05$ ) values, but only in the H group. The marker-assisted selection plays crucial role in animal husbandry, despite economically important traits show multifactorial inheritance with a strong environmental influence. Our results prove that the effect of polymorphisms depends on LW, moreover, the LW strongly influences the carcass quality traits, therefore if the test population shows a high degree of variability the use of weight categories should be considered.

**Key Words:** carcass quality, pig, RFLP

**762 Estimates of variance components for gilt retention traits.** Matthew D. A. Morrison\*<sup>1</sup>, Kent A. Gray<sup>2</sup>, Miles T. See<sup>1</sup>, and Mark T. Knauer<sup>1</sup>, <sup>1</sup>North Carolina State University, Raleigh, NC, <sup>2</sup>Smithfield Premium Genetics, Rose Hill, NC.

Numerous studies have been conducted on sow retention and longevity; however, less research has been completed on gilt retention. Increasing the percentage of gilts that farrow a litter would reduce gilt development costs, improve sow longevity, and enhance farmer profitability. Hence the purpose of this study was to estimate variance components for gilt retention traits. Data consisted of 6,282 commercial gilts from Large White dams and Landrace sires (Smithfield Premium Genetics, Rose Hill, NC). During development, gilts were reared in environmentally controlled facilities with slatted concrete flooring, natural ventilation and ad libitum access to feed and water. At an average age of 211 d (SD ± 41.5) females were moved to one of 11 sow farms in eastern North Carolina. Traits included successful gilt farrow event (STAY; success = 1, failure = 2) gilt culled for reproductive (REPRO; Yes = 1, no = 2), or non-reproductive reasons (OTHER, yes = 1, no = 2), age at first service (AFS), and age at first farrowing (AFF). Variance components were estimated using an animal model with THRGIBBS1F90 for categorical traits and AIREMLF90 for linear traits. All models contained a fixed effect of contemporary group (farm × year × month) and a random effect of animal. Of the gilts entering sow farms, 15.4% did not farrow a litter, 7.3% were culled for REPRO, and 8.1% were culled for OTHER. Average AFS and AFF were 262 d (SD ± 24.7) and 377 d (SD ± 24.9), respectively. Heritability estimates for STAY, REPRO, OTHER, AFS, and AFF were 0.15, 0.19, 0.05, 0.27, and 0.21, respectively. Phenotypic variance estimates for STAY, REPRO, OTHER, AFS, and AFF were 1.18, 1.24, 1.06, 356.2, and 386.2, respectively. Results suggest selection for increased gilt retention is possible.

**Key Words:** gilt, heritability, retention

**763 Estimates of genetic parameters for sow body weight loss during lactation.** Cassandra L. Ferring<sup>\*1</sup>, Dale Hentges<sup>2</sup>, Clint Schwab<sup>2</sup>, and Mark T. Knauer<sup>1</sup>, <sup>1</sup>North Carolina State University, Raleigh, NC, <sup>2</sup>The Maschhoffs, Carlyle, IL.

The purpose of this study was to estimate genetic parameters for sow BW loss during lactation. Data and pedigree information were available for Landrace (n = 3,310), York (n = 827) and Landrace × York F<sub>1</sub> sows (n = 354) from The Maschhoffs (Carlyle, IL). Sows were housed in environmentally controlled facilities with slatted concrete flooring and had ad libitum access to water. Females were restricted fed during gestation based on a visual body condition score of 1 to 5 (1 = thin, 5 = fat). During lactation sows were fed ad libitum. Sow traits analyzed included body condition score at farrowing (FBCS), total number born (TNB), number born alive (NBA), litter birth weight (LBW), litter weaning weight (LWW), number weaned (NW), body condition score at weaning (WBCS), and sow BW loss during lactation (WTD). Variance components were estimated using ASReml. All models included fixed effects of genetic line, parity, and contemporary group and random effects of animal (sow) and permanent environment. Covariates were included for LBW (NBA), LWW (piglet age at weaning and number of piglets fostered), NW (number of piglets fostered), WBCS (lactation length) and WTD (lactation length). Heritability estimates for FBCS, TNB, NBA, LBW, LWW, NW, WBCS and WTD were 0.17, 0.15, 0.15, 0.25, 0.15, 0.11, 0.16, and 0.13 respectively. Permanent environment variance estimates for FBCS, TNB, NBA, LBW, LWW, NW, WBCS and WTD were 0.0007, 0.26, 0.20, 0.49, 2.65, 0.04, 0.0004 and 1.29 respectively. Phenotypic variance estimates for FBCS, TNB, NBA, LBW, LWW, NW, WBCS and WTD were 0.015, 11.50, 10.49, 5.66, 112.4, 4.35, 0.014 and 251.5 respectively. Genetic correlations between WTD with FBCS, TNB, NBA, LBW, LWW, NW and WBCS were -0.05, 0.01, -0.02, 0.50, 0.28, 0.05 and -0.19 respectively. Phenotypic correlations between WTD with FBCS, TNB, NBA, LBW, LWW, NW and WBCS were 0.10, 0.09, 0.08, 0.15, 0.20, 0.15 and -0.03 respectively. Selection for reduced BW loss during lactation appears possible and would have minimal effect on litter size.

**Key Words:** body weight, lactation, reproduction

**764 Genetic selection for feed efficiency in crossbred animals.** Rob Bergsma\* and Egbert F. Knol, *Topigs Norsvin Research Centre B.V., Beuningen, the Netherlands.*

Worldwide, the majority of the commercial slaughter pigs are a cross of 3 or even 4 purebred lines. Although genetic selection takes place in purebred lines, the breeding goal should be directed toward crossbred performance. Applying a crossbred breeding goal is important if purebred traits show a genetic correlation significantly lower than 1.0 with their crossbred counterparts and/or when the genetic variation of purebred traits is different from those of crossbred traits. Recent research showed that this especially applies to feed intake and feed efficiency, while carcass composition in purebreds and crossbreds appeared to be more similar traits. The percentage of the genetic trend in purebred lines, expressed in crossbreds can be calculated as the ratio of the crossbred genetic standard deviation multiplied by the genetic correlation and the genetic purebred standard deviation. Our results show that of the genetic progress in purebreds 62, 52, 111 and 85% is expressed in crossbreds for daily gain, feed intake, back fat and loin depth, respectively. For feed efficiency this ratio depends on the trait considered. For feed conversion ratio, 60% of the genetic trend in purebreds was expressed in crossbreds, whereas for residual feed intake this ratio was only 41%. Feed intake (and thus feed efficiency) are traits that are not widely available on individual crossbred animals. The limited availability may also

indicate that the crossbred/purebred phenotypes are confounded with their environment, given rise to a possible genotype by environment interaction. Another possible explanation is that dominance plays a larger role in feed intake and feed efficiency compared with other traits. And finally, genomic selection has been implemented for purebreds but when using crossbred animals as reference population we have to take into account that a QTL in breed A can behave different than in breed B. Current research focuses on partitioning the possible causes in explaining the lower expression of the genetic trend of purebreds in crossbred. Results will facilitate decisions on the most appropriate genetic model to include crossbred information in a combined crossbred purebred breeding program.

**Key Words:** pig breeding, crossbred performance, feed efficiency

**765 Genomic regions associated with response to PRRSV and PCV2b co-infection in nursery pigs.** Jenelle R. Dunkelberger<sup>\*1</sup>, Nick V. L. Serão<sup>1</sup>, Maureen A. Kerrigan<sup>2</sup>, Joan K. Lunney<sup>3</sup>, Raymond R. Rowland<sup>2</sup>, and Jack C. M. Dekkers<sup>1</sup>, <sup>1</sup>Department of Animal Science, Iowa State University, Ames, IA, <sup>2</sup>College of Veterinary Medicine, Kansas State University, Manhattan, KS, <sup>3</sup>USDA, ARS, BARC, APDL, Beltsville, MD.

The objective of this study was to identify genomic regions associated with porcine reproductive and respiratory syndrome (PRRS) viral load (VL), porcine circovirus type-2b (PCV2b) VL, and weight gain (WG) in nursery pigs co-infected with PRRS virus (PRRSV) and PCV2b. Two PRRS Host Genetics Consortium trials were conducted using commercial crossbred pigs (n = 400) pre-selected (50% AA and 50% AB) for the WUR single nucleotide polymorphism (SNP) on chromosome (SSC) 4 that is associated with response to PRRS. Half of the pigs received a PRRS modified live vaccine and all pigs were co-infected with PRRSV and PCV2b 28 d later. For the genome-wide association studies (GWAS), 61,730 SNPs were fitted simultaneously using the BayesB method with  $\pi = 0.994$ . For analysis of PCV2b VL, trial, WUR, vaccination (Vx), WUR×Vx, weight at Vx, age at Vx, PRRS viremia at 0 dpi, PCV2b viremia at 0 dpi, and pen were fitted as fixed effects. For analysis of WG and PRRS VL, trial×Vx replaced WUR×Vx. Marker-based heritability estimates were 0.33, 0.34, and 0.14 for WG, PCV2b VL, and PRRS VL, respectively. Compared with AA pigs, AB pigs did not significantly differ ( $P = 0.70$ ) in WG but had lower PRRS VL ( $P < 0.0001$ ) and, in the vaccinated group, lower PCV2b VL ( $P = 0.09$ ). Sizable genetic associations were identified for the GWAS of WG and PCV2b VL, but not for PRRS VL. A 2-Mb window on SSC9, at 27–28 Mb, explained 7.7% of genetic variance in WG, which has been associated with growth of nursery pigs across multiple breeds. For PCV2b VL, 1-Mb windows on SSC1 (162 Mb), SSC5 (102 Mb), and SSC7 (41 Mb) explained 1.4, 1.1, and 1.0% of genetic variance, respectively. Thus, the AB WUR genotype was associated with reduced PRRS VL and with reduced PCV2b VL in vaccinated pigs, following co-infection with PRRSV and PCV2b. Genomic regions associated with WG and PCV2b VL were identified, regions for the latter trait being novel. These results suggest the possibility of selectively breeding pigs, based on these regions, for improved host response to PRRSV/PCV2b co-infection. This work was supported by the USDA ARS NIFA awards 2012–38420–19286 and 2013–68004–20362.

**Key Words:** genetic susceptibility, GWAS, swine

**766 Genetic and economic effects of incorporating genomic predictions on health in swine breeding schemes.** Chandraratne M. B. Dematawewa\*<sup>1</sup>, Anna Grosse Holthaus<sup>2</sup>, Henner Simianer<sup>2</sup>, and Jack C. M. Dekkers<sup>1</sup>, <sup>1</sup>Iowa State University, Ames, IA, <sup>2</sup>University of Göttingen, Göttingen, Germany.

A study was conducted to determine the effect of inclusion of genomic evaluations for health (GE-health) on improvements in health, the overall breeding goal ( $\Delta H$ ) and discounted profit ( $\Delta\Omega$ ) in commercial cross breeding schemes. A 3-way cross breeding scheme with 2 maternal lines [female: Yorkshire (YS); male: Landrace (LR)] and a terminal sire line (Duroc, DU) was deterministically simulated using the ZPLAN+ software. The YS nucleus consisted of 50 boars and 1000 sows, while LR and DU nuclei had 100 boars and 800 sows each. Productive life of nucleus and multiplier ( $n = 2400$ ) animals was 1 year, while  $F_1$  crossbred sows were kept for 2 years to produce 532,400 commercial piglets/year. Days to market (DY), backfat thickness (BF), and litter weight at 21 d (LW; for YS and LR only) were considered with heritabilities ( $h^2$ ) of 0.4, 0.5, and 0.09. The traits were standardized to genetic SD = 1. A health trait (HL) was simulated with  $h^2 = 0.05$  and genetic SD = 1, with positive genetic (0.2) and phenotypic (0.3) correlations with DY and BF, and zero correlations with LW. HL was recorded from 80 commercial halfsibs/animal. Economic weights (\$) for DY, BF, LW and HL were -1.86, -1.82, 5.35, and 1.86 per genetic SD, respectively. Planning horizon was 10 years (discount rate = 0.05). Costs for high and low density genotyping and health recording were \$100, \$40 and \$10 per animal, respectively. Accuracy of GE-health ( $r_{MG-HEALTH}$ ) was varied from 0 to 1.0 and 0.7 for the other traits. Inclusion of GE-health for both sires and dams increased both  $\Delta H$  and  $\Delta\Omega$  as  $r_{MG-HEALTH}$  increased, mainly due to greater genetic gain in HEALTH. At  $r_{MG-HEALTH} = 0.8$ , extra response in  $\Delta H$  and  $\Delta\Omega$  for were 6.25% and \$0.55 per animal in the breeding program, compared with having no GE-health. The corresponding values were higher (7.06% and \$1.34, respectively) when HEALTH phenotypes were not recorded, partly due to lower initial response. The benefit of health recording diminished when  $r_{MG-HEALTH}$  increased. These results show economic feasibility for implementing GE-health in commercial swine breeding. Funded by Genome Canada.

**Key Words:** genomic selection, health, swine breeding

**767 Exploring causal networks underlying fat deposition and muscularity in pigs through the integration of phenotypic, genotypic and transcriptomic data.** Francisco Peñagaricano\*<sup>1,2</sup>, Bruno D. Valente<sup>1</sup>, Juan P. Steibel<sup>3</sup>, Ronald O. Bates<sup>3</sup>, Cathy W. Ernst<sup>3</sup>, Hasan Khatib<sup>1</sup>, and Guilherme J. M. Rosa<sup>1</sup>, <sup>1</sup>University of Wisconsin-Madison, Madison, WI, <sup>2</sup>University of Florida, Gainesville, FL, <sup>3</sup>Michigan State University, East Lansing, MI.

Joint modeling and analysis of phenotypic, genotypic and transcriptomic data have the potential to uncover the genetic control of gene activity and phenotypic variation, as well as shed light on the manner and extent of connectedness among these variables. Current studies mainly report associations; that is, undirected connections among variables without causal interpretation. Knowledge regarding causal relationships among genes and phenotypes can be used to predict the behavior of complex systems, as well as to optimize management practices and selection strategies. Here, we performed a multistep procedure for inferring causal networks underlying carcass fat deposition and muscularity in pigs using multi-omics data obtained from an  $F_2$  Duroc  $\times$  Pietrain resource pig population. The data set included several carcass and meat quality phenotypes, genotypic information spanning the whole swine genome, and gene expression data from loin muscle for a total of 171  $F_2$  individuals. We initially explored marginal associations between

genotypes and phenotypic and expression traits through whole-genome scans, and then, in genomic regions with multiple significant hits, we assessed gene-phenotype network reconstruction using causal structural learning algorithms. One genomic region in SSC6 showed significant associations with 3 relevant phenotypes, midline 10th-rib backfat thickness, loin muscle weight, and average intramuscular fat percentage, and also with the expression of 7 genes, including *ZNF24*, *SSX2IP*, and *AKR7A2*. The inferred network indicated that the genotype affects the 3 phenotypes mainly through the expression of several genes. Among the phenotypes, fat deposition traits negatively affected loin muscle weight. Overall, our findings shed light on the antagonist relationship between carcass fat deposition and meat lean content in pigs. In addition, the procedure described in this study has the potential to unravel gene-phenotype networks underlying complex phenotypes.

**Key Words:** causal inference, complex trait, systems genetics

**768 Genomic prediction accuracy of porcine respiratory and reproductive syndrome (PRRS) antibody response in commercial gilts and sows.** Nick V. L. Serão\*<sup>1</sup>, Robert A. Kemp<sup>2</sup>, Benny E. Mote<sup>3</sup>, John C. S. Harding<sup>4</sup>, Philip Willson<sup>4</sup>, Stephen C. Bishop<sup>5</sup>, Graham S. Plastow<sup>6</sup>, and Jack C. M. Dekkers<sup>1</sup>, <sup>1</sup>Iowa State University, Ames, IA, <sup>2</sup>Genesus, Oakville, ON, Canada, <sup>3</sup>Fast Genetics, Saskatoon, SK, Canada, <sup>4</sup>University of Saskatchewan, Saskatoon, SK, Canada, <sup>5</sup>The Roslin Institute, Easter Bush, Midlothian, UK, <sup>6</sup>University of Alberta, Edmonton, AB, Canada.

The objective of this study was to assess the genomic prediction accuracy (GPA) of antibody response to PRRS in purebred sows and crossbred gilts. Data on 2,180 commercial crossbred gilts (CrossData), from 7 breeding companies, and 512 purebred multiplier Landrace sows (PureData), from one breeding company, were used to assess the ability to predict PRRS antibody response, measured as sample-to-positive (S/P) ratio, using 38,191 single nucleotide polymorphisms (SNPs). S/P ratio was measured in the CrossData at 40.7  $\pm$  16 d after gilts entered commercial herds with a history of health problems, whereas in the PureData, S/P ratio was measured 46 d after a natural PRRS outbreak. Two prediction strategies were used: 1) CrossData was used for training and PureData for validation; 2) 7-fold cross-validation using the CrossData. Previous results showed that S/P ratio is mainly controlled by 2 regions on chromosome 7 (SSC7), at 24–31 Mb and 128–131 Mb. Therefore, different sets of SNPs were used for prediction: all SNPs (All\_SNP), SNPs on SSC7 24–31 Mb (QTL1\_SNP), SNPs on SSC7 128–131 Mb (QTL2\_SNP), SNPs on both regions (SSC7\_SNP), all SNPs except those on the SSC7 regions (Not7\_SNP). GPA was measured as the correlation between genomic estimated breeding values and pre-adjusted phenotypes, divided by square root of heritability. Heritability estimates were 0.46 (PureData) and 0.31 (CrossData). When training using the CrossData and validating on PureData, GPAs were 0.49 (All\_SNP), 0.55 (QTL1\_SNP), 0.30 (QTL2\_SNP), 0.63 (SSC7\_SNP), and 0.15 (Not7\_SNP). GPAs were slightly lower using from cross-validation in the CrossData: when averaging across folds: 0.32 (All\_SNP), 0.30 (QTL1\_SNP), 0.27 (QTL2\_SNP), 0.39 (SSC7\_SNP), and 0.1 (Not7\_SNP). The highest and lowest GPA from individual folds were -0.14 (Not7\_SNP) and 0.60 (SSC7\_All). These results show that S/P ratio can be accurately predicted using SNPs in pure and crossbred female pigs. In addition, greater accuracy can be obtained using the 2 QTL on SSC7 than the whole genome. Financial support from Genome Canada, the Canadian Swine Health Board, and PigGen Canada.

**Key Words:** SNP, host genetics, cross-validation



**769 Genetic analysis and whole-genome wide association for feeding behavior traits in Duroc pigs.** Shihui Jiao\*<sup>1</sup>, Christian Maltecca<sup>1</sup>, Yijian Huang<sup>2</sup>, and Kent A. Gray<sup>2</sup>, <sup>1</sup>North Carolina State University, Raleigh, NC, <sup>2</sup>Smithfield Premium Genetics, Rose Hill, NC.

This study was aimed at estimating genetic parameters for feeding behavior and production traits and identifying genomic regions influencing those behavior traits in a Duroc population. Feed intake and related measures were collected from 2004 to 2013 for 14,869 boars with 4,940,348 visits. Several 7,319 pigs were genotyped with 35,700 SNP available for analysis. Traits included 4 production traits; average daily feed intake, average daily gain, off-test body weight and backfat thickness and 12 feeding behavior traits include; daily occupation time (OTD), number of visits (NVD), feeding rate (DFR), feed intake per visit (FIV), occupation time per visit (OTV), standard deviation (SD) of feed intake per visit (ASDFIV), SD of occupation time per visit (ASDOTV), SD of feeding rate per visit (ASDFRV), SD of daily feed intake (SDFI), SD of daily occupation time (SOTD), SD of number of visits (SDNVD) and SD of feeding rate (SDFR). Except for SDFI and SDFR, all behavior traits were moderately heritable with estimates ranging from  $0.32 \pm 0.03$  (SOTD) to  $0.68 \pm 0.03$  (FIV). Genomic heritability estimates using single-step methods incorporating genotypes were slightly lower than those obtained using pedigree by  $0.10 \pm 0.09$  averaged across all behavior traits. Genetic correlation of behavior traits with production traits varied (ranging from  $-0.481$  to  $0.618$ ). Single trait association analyses for behavior traits for 1,541 boars were performed using mixed linear models. Significance of marker was declared with a p-value less than  $1.40 \times 10^{-6}$  (Bonferroni correction). A total of 35 significant markers on SSC 2, 7, 8, 9 and 18 associated with NVD, OTV, ASDFRV and SOTD were identified and putative genes included *NCR3* (involving in natural killer cell mediated cytotoxicity and associated with obesity in human), *CLPS* (activating in digestion of dietary lipid and absorption) and *PPARD* (regulator of lipid metabolism). In conclusion, feeding behaviors were moderately heritable and we have identified several genomic regions associated with traits affecting feeding behaviors that may help us understand the genetic mechanism controlling the traits.

**Key Words:** genomics, feeding behavior

**770 Genomic mitigation of seasonality effect on carcass weight in commercial pigs.** Breno D. Fragomeni\*<sup>1</sup>, Shogo Tsuruta<sup>1</sup>, Daniela A.L. Lourenco<sup>1</sup>, Kent Gray<sup>2</sup>, Yijian Huang<sup>2</sup>, and Ignacy Misztal<sup>1</sup>, <sup>1</sup>Department of Animal and Dairy Science, University of Georgia, Athens, GA, <sup>2</sup>Smithfield Premium Genetics, Rose Hill, NC.

The purpose of this study was to determine the effect of heat stress on carcass weight in a commercial pig population. Phenotypes included hot carcass weight records on 227,043 commercial pigs collected from 2 packing plants located in the states of North Carolina and Missouri; the pigs were a cross between purebred Duroc sires and F1 Landrace x Large White dams. The pedigree file included 553,442 animals, and 60k SNP genotypes were available for 8,232 sires. Weather information was collected from airport weather stations within 100 miles distance from the finishing farms. Average temperature humidity index (THI) was calculated for a period of 70 d before each HCW was collected. The THI measurement was classified as heat stress (THI > 78, n = 32,783) or comfort conditions (THI = < 78, n = 194,260). Analyses were done with an animal model as either a single-trait or 2-trait model using records identified as heat stress and comfort conditions treated as

separate traits. Variance components were estimated with AIREML, and traditional and genomic (G) EBV were computed either with BLUP or single-step genomic BLUP (ssGBLUP). Validations were computed for 94 animals from the last generation using the forward prediction method, and reliability of (G)EBV was calculated as  $R^2$  of predictions based on the training population (all except the last generation) on progeny yield deviations of the last generation. The heritability estimate for hot carcass weight in the single-trait model was 0.20. In the multiple trait model, the heritability estimate was 0.20 under comfort conditions and 0.25 under heat stress, with a genetic correlation of 0.62. Under comfort conditions and heat stress, reliabilities in traditional EBV were 0.22 and 0.14 whereas reliabilities in GEBV were 0.38 and 0.19, respectively. The heritability of carcass weight is higher under heat stress. Use of ssGBLUP increases reliabilities of carcass weight under both heat stress and comfort conditions. Effects of seasonality on carcass weight can be mitigated by genetic selection, especially with the genomic information.

**Key Words:** heat stress, genotype  $\times$  environment interaction, genomic selection

**771 Effect of divergent selection for residual feed intake in finishing pigs on juvenile IGF-I.** Emily D. Mauch\*<sup>1</sup>, Nick V. L. Serão<sup>1</sup>, Joel R. Steckelberg<sup>1</sup>, Anna Wolc<sup>1,2</sup>, and Jack C. M. Dekkers<sup>1</sup>, <sup>1</sup>Department of Animal Science, Iowa State University, Ames, IA, <sup>2</sup>Hy-Line International, Dallas Center, IA.

Since 2001, purebred Yorkshire pigs at Iowa State University have been divergently selected for increased (Low RFI) and decreased (High RFI) feed efficiency based on single trait selection for residual feed intake (RFI). Using data from generations 2–5 (G2–5), Bunter et al. (2010) found that serum insulin-like growth factor-I (IGF-I) concentration in young pigs (33–42 d) was lower in the Low RFI line, and had a high genetic correlation (0.63) with RFI, suggesting that it could be used as an indicator trait for RFI. The objective here was to validate these findings in G10 of the RFI lines. IGF-I concentration was measured by Rivalea Ltd. (Australia) using the Primegro assay on blood samples collected from 35 to 42 d of age on 377 piglets from G10. IGF-I data were analyzed in ASREML with a univariate sire model and a bivariate animal model with RFI data on 2,370 pigs from G1–10. Both models for IGF-I included the fixed effects of line, sex and contemporary group, covariates of age at blood collection and interval from weaning to blood collection, and random effects of litter, and sire (no pedigree) or animal (with complete pedigree). The concentration of IGF-I (ng/mL) was estimated to be  $55.7 (\pm 19.1)$  and  $53.4 (\pm 44.2)$  lower in the Low RFI line compared with the High RFI line based on the sire ( $P = 0.01$ ) and animal ( $P = 0.3$ ) models, respectively. Bunter et al. (2010) reported a similar line difference of 47.0 ng/mL in G5. The estimated genetic correlation between RFI and IGF-I was  $0.68 (\pm 0.38)$ , similar to Bunter et al. (2010). In conclusion, lower juvenile IGF-I concentration was validated to be associated with pigs selected for increased feed efficiency based on RFI and, therefore, may serve as an early blood indicator to select for feed efficiency. However, when considering the full pedigree, the line difference was not significant. Funding provided by AFRI-NIFA grant #2011–68004–30336 and by Rivalea Ltd. (Australia).

**Key Words:** swine, genetic parameter, residual feed intake

# Companion Animal Symposium: Comparative nutrition—Protein and energy across species

**772 Thinking comparatively allows flexibility: The legacy of Duane Ullrey.** Mark S. Edwards\*, *California Polytechnic State University, San Luis Obispo, CA.*

A Michigan native, Duane Ullrey completed a BS degree in animal husbandry (1950) and an MS degree in animal pathology (1951) from Michigan State College. Duane earned a PhD in animal nutrition with minors in physiology and biochemistry (University of Illinois, 1954). Upon graduation, he taught at Oklahoma A&M University. In 1956, Duane was hired as an assistant professor, becoming a full professor in 1968, of animal nutrition with a joint appointment in Departments of Animal Science and Fisheries and Wildlife at Michigan State University. He taught and conducted research in swine and white tailed deer nutrition, as well as taught Animal Nutrition in the College of Veterinary Medicine, ultimately developing a Comparative Animal Nutrition program. Duane influenced many (34 graduate students, 140 graduate committees) in formal and informal learning environments. Duane established new care standards in the field of zoo animal nutrition. His comparative approach facilitated service on prominent committees, including the National Academy of Sciences, Committee on Animal Nutrition (1981; Chair, 1985), Lab Animal Nutrition (1978), Mineral Toxicity (1980), Horse Nutrition (1989), and Primate Nutrition (Chair, 2003). Duane was recognized by his colleagues with honors, including the MSU Distinguished Faculty Award (1983), ASAS Morrison Award (1988), ASAS Fellow (1990), the first Comparative Nutrition Society honorary life member (1996), and the first award recipient of the Duane E. Ullrey Achievement Award by the American Association of Zoo Veterinarians (1999). Duane was a mentor, friend and colleague. He cultivated sound scientific principles in species husbandry, emphasized a strong foundation in nutrition, and guided our comparative discovery of relevant similarities and differences among novel species with known models. A consummate life-long learner, Duane embodied integrity, thoughtfulness, humor, honesty, and perseverance in his personal and professional life. Through his students, he continues to make remarkable contributions in academia, industry, and regulatory agencies in areas of animal agriculture, wildlife and natural resource management, veterinary medicine, and human nutrition and health.

**Key Words:** comparative animal nutrition

**773 Protein and energy metabolism in the development and management of obesity and chronic diseases in humans.** David J. Baer\*, *USDA, ARS, Beltsville, MD.*

Obesity and several obesity-related diseases, such as cardiovascular disease and diabetes, are epidemic not only in the United States but around the globe. In addition to their impact on premature death and loss of quality of life, the combined economic burden of obesity and diabetes is nearing one-half trillion dollars in the United States. Furthermore, in many countries, including the United States, there is significant growth in the proportion of the older population, and optimal dietary protein is necessary for healthy aging to minimize age-associated loss of muscle (sarcopenia). Protein is an energy yielding macronutrient, and consumption of protein or any energy yielding macronutrient in excess of need will contribute to unhealthy weight gain. However, some studies have demonstrated that consumption of protein in excess of the Recommended Dietary Allowance for adults (0.8 g/kg body weight/day) is associated with improved health outcomes, including improved body composition

and improved insulin sensitivity without detrimental effects on bone or renal function. The effect of higher protein diets on reducing food intake is one mechanism by which protein may influence body composition. In addition to the amount of dietary protein, the quality of that protein can affect health outcomes. Increased consumption of dairy protein, and whey protein in particular, has been associated with decreased body fat without a loss of lean mass. The relatively high branch-chain amino acid concentrations of whey protein may contribute to its physiological effects. Leucine, and perhaps other branched chain amino acids, might mediate changes in body composition through modulation of the mTOR or other key regulatory pathways. With the global public health challenges of obesity and its comorbidities, dietary recommendations to improve health continue to be evaluated. Higher protein diets and high quality proteins can play an important role in improving body composition and supporting healthy aging.

**Key Words:** obesity, protein, humans

**774 Brain and cognitive development: Assessing the impact of nutrition in a neonatal piglet model.** Rodney W. Johnson\*, *University of Illinois, Urbana, IL.*

Prenatal and early postnatal insults increase the likelihood for neurobehavioral problems later in life. One hypothesis suggests that the pre- and postnatal periods represent sensitive periods during which exposure to adversity programs biological systems in a manner that persists and accentuates vulnerability to behavioral problems later in life. This hypothesis is predicated on pre- and/or postnatal stress influencing structural and functional plasticity of the brain and subsequent stress resilience. The piglet is an excellent model for perinatal developmental studies because it is a gyrencephalic species with brain growth and morphology similar to humans. Its gastrointestinal anatomy and physiology are comparable and in a recent study that examined the similarity of immune-related genes in humans, pigs, and rodents, the overall mean similarity to human immune-related proteins was significantly higher for pigs compared with mouse. In the piglet model we have reported profound effects of perinatal infection, postnatal nutrition, and being born small for gestational age on brain development and behavior. The later issue is particularly relevant to this symposium because protein-energy malnutrition during pregnancy is a leading cause of intrauterine growth restriction and small for gestational age births.

**Key Words:** low birth weight, learning and memory, stress resilience

**775 Comparative gastrointestinal utilization of nitrogen, lysine, and glucose in equids and suids.** Nathalie Trottier\*, *Michigan State University, East Lansing, MI.*

Equids are obligate hindgut fermenters and thus are equipped with a large cecum and voluminous proximal colon for microbial fermentation of plant cell wall fiber components. Suids are also hindgut fermenters; however, because they have evolved consuming highly diverse diets, they are also classified as omnivores. Suids share similar gastrointestinal anatomical features with equids, and therefore may be a useful model of gastrointestinal nitrogen and carbohydrate metabolism. Important differences exist in the kinetics of glucose transport between equine and porcine small and large intestinal tissues, and these differences

shed light on the divergent ability of these 2 species to absorb dietary carbohydrates. In contrast, the 2 species share distinct similarities in intestinal Lys transport kinetics. In both species, the large colon apical membrane is capable of Lys transport via a high capacity and lower affinity transport process that is higher than that of the small intestine. Lys transport across the pig and the pony jejunum is 1.14 and 1.16 pmol/mg tissue ( $V_{\max}$ ), respectively, and uptake by the pig and pony colon is 0.84 and 0.91 pmol/mg tissue ( $V_{\max}$ ), respectively. In both species, the large colon has a lower affinity for Lys compared with the small intestine (i.e., 0.89 and 1.05  $K_M$ , pig and pony, respectively). The broader capacity of Lys transport in the large colon is indicative of the potential role it plays in the uptake of Lys. It is proposed that the porcine model may be useful to predict amino acid requirements for maintenance and digestibility in equids.

**Key Words:** suid, equid, nutrient

**776 Carnivores, omnivores, and herbivores—Concepts of ideal protein formulation for ornamental and commercial aquaculture feeds.** Delbert M. Gatlin III\* and Sergio Castillo, *Texas A&M University System, College Station, TX.*

Aquacultural production of various fish and crustacean species is continuing to grow throughout the world at over 6% per year, and projected

to continue expanding as the demand for seafood increases and harvests from wild stocks remain at or beyond maximum sustainable levels. The efficiency of producing various aquatic species is generally considered to be greater than that of terrestrial livestock, primarily due to efficient feed utilization and protein accretion per unit of energy consumed. There is considerable variation in dietary protein requirements of various fish and crustacean species as well as their ability to use protein and lipid for energy. This variation appears to be primarily related to natural feeding habits in which aquatic species are generally categorized as being herbivorous, omnivorous or carnivorous. This presentation will provide an overview of the protein and amino acid requirements of prominent freshwater omnivorous and carnivorous fish species such as the channel catfish (*Ictalurus punctatus*) and rainbow trout (*Oncorhynchus mykiss*). Those requirements will be compared and contrasted with that of the red drum (*Sciaenops ocellatus*), a marine carnivore, and the omnivorous common carp (*Cyprinus carpio*) of which colored varieties are produced for ornamental purposes. Specific emphasis will be placed on application of the ideal protein concept in formulating diets for these various species.

**Key Words:** aquatic animals, amino acid requirement, comparative nutrition



## Dairy Foods Symposium: Processing and ingredient innovations to grow fluid milk sales

**777 Market implications and opportunities in the fluid milk and beverage sector.** Alan Reed\*<sup>1</sup>, Madlyn Daley<sup>1</sup>, Marla Buerk<sup>1</sup>, and Chris Costagli<sup>2</sup>, <sup>1</sup>*Dairy Management Inc., Rosemont, IL*, <sup>2</sup>*Information Resources Inc., Chicago, IL*.

Changing consumer demographics, habits, practices, and attitudes create both challenges and opportunities for fluid milk in the current beverage marketplace. This presentation will explore data from a variety of sources detailing these consumer trends, and suggest possible opportunities and strategies for growth in the fluid milk category. Data demonstrating the market and consumer impacts of competitive entrants, including non-dairy alternatives made of soy, almond, coconut and rice, will also be explored.

**Key Words:** milk, seminar, consumer

**778 Implications of microbial quality of incoming milk and plant sanitation on shelf life and acceptance.** Nicole H. Martin\*, Rachel A. Miller, Stephanie N. Masiello, David J. Kent, Kathryn J. Boor, and Martin Wiedmann, *Cornell University, Ithaca, NY*.

Microbial growth in fluid milk is one of the primary drivers of consumer liking. In an age of declining fluid milk consumption, control of microbial growth is critical. The 2 primary mechanisms of microbial spoilage in fluid milk are (1) post-pasteurization contamination (PPC) in the processing facility with psychrotolerant gram-negative bacteria (i.e., *Pseudomonas*); and (2) entry of psychrotolerant sporeforming bacteria (i.e., *Paenibacillus*) via raw milk contamination. Historically, the majority of fluid milk was spoiled by psychrotolerant gram-negative bacteria introduced into the product post-pasteurization. This type of contamination leads to drastically reduced shelf-life and significant flavor, body and odor defects. Results of recent work indicate that over 50% of fluid milk samples show signs of PPC, and of those samples, only 40% are contaminated with coliform bacteria, a commonly used indicator of PPC. These results indicate that PPC is still an issue that needs to be addressed by the dairy industry to offer consumers the highest quality fluid milk. Additionally, traditionally used hygiene indicators (i.e., coliform) need to be reexamined to provide relevant, actionable information to dairy processors regarding PPC. In the absence of PPC, psychrotolerant sporeforming bacteria become the limiting biological factor in fluid milk shelf-life. Addressing fluid milk spoilage from psychrotolerant sporeforming bacteria includes understanding and reducing the transmission of these organisms through identification of key management practices associated with their presence in raw milk, as well as sources of these organisms in the farm environment. Recent work indicates that udder hygiene and farm size are associated with the presence of psychrotolerant spores in raw bulk tank milk. Further, these organisms have been identified in diverse environmental samples (e.g., bedding, feed, manure and soil), indicating their abundance in the dairy farm environment. Ultimately, providing consumers with high quality, long lasting fluid milk requires a systems approach to preventing contamination of fluid milk throughout the grass to glass continuum.

**Key Words:** quality, sporeformer, sanitation

**779 Effect of milk composition and processing conditions on consumer liking.** M. A. Drake\*, *Southeast Dairy Foods Research Center, North Carolina State University, Raleigh, NC*.

Fluid milk consumption has decreased in the past 20 years while the beverage market continues to expand and diversify. Understanding consumer desires and perceptions for milk and milk flavor are crucial to identify opportunities. A combination of consumer and product research is required. Surveys and interviews demonstrate that flavor and nutrition are the key desires for consumers, and fluid milk holds unique associations with comfort and family. Variety and enhanced protein are 2 opportunities for milk/milk beverage expansion. Appearance plays a critical role in fluid milk preferences, especially for skim milk, and fat, protein and heat treatment also influence the sensory properties. A thorough knowledge of the role of processing conditions and composition on sensory properties and consumer perception of fluid milks will enable milk and milk beverage innovation.

**Key Words:** milk, consumer liking, processing

**780 Shedding light on consumer acceptability of fluid milk: The partnership of retail dairy case lighting systems and packaging.** Susan Duncan\*, *Virginia Tech, Blacksburg, VA*.

Declining fluid milk purchasing and consumption trends suggest that fluid milk is not competing well in the beverage market. Contributors to this trend include a lack of established packaging selection criteria as well as storage conditions during the early stages of shelf-life before purchase. Fluid milk quality changes rapidly during light exposure, especially in retail lighted display cases. However, lighting in retail refrigerated cases enhances product marketing by showcasing product and package color. Traditionally, fluorescent light is used in retail systems; however, LED lighting systems are increasing because of new federal mandates for energy conservation as well as brighter, more efficient, more attractive display capabilities. It is well established that translucent or clear packaging with no light protective additives does not protect milk flavor and nutrient quality. However, only limited information is available on how fluorescent or LED light exposure influences milk acceptability. Changes in milk flavor are detectable within 2 to 3 h of lighted exposure and acceptability decreases significantly within 8 h of storage under fluorescent lighting when packaged in a translucent high-density polyethylene package. Acceptability does not tell the complete story though. In a recent study from our laboratory, consumers reported more negative emotion terms, specifically disgust, when light-exposed milk was evaluated. Lower hedonic scores were directly related to negative emotions. Milk that was packaged for light protection retained high acceptability scores and positive emotional terminology through 7 d of lighted retail storage conditions. Processors should select packaging that protects milk quality during retail dairy case display lighting to positively influence fluid milk purchasing and consumption behavior. Packaging criteria for protecting fresh fluid milk flavor and nutrients from processing to purchase will be described and justified.

**Key Words:** consumer, milk, packaging

**781 Characterizing factors that influence milk flavor quality.**

Smaro Kokkinidou, David Potts, and Devin Peterson\*, *University of Minnesota, St. Paul, MN.*

Flavor is known to be a key attribute that determines the acceptability of milk. This presentation will cover a range of off-flavor development sources, from farm to processing, focusing on spontaneous oxidized flavor (SOF) milk as well as aseptically processed milk. Spontaneous oxidized flavor (SOF) milk has been a problem for the dairy industry for many years, causing significant economic losses. After collecting raw milk labeled as SOF milk from the Midwest, causative off-flavor compounds were identified as microbial taints (i.e., 2-methylisoborneol) and disproved the hypothesis that the off-flavor was of oxidative origin. Identifying the source of the off-flavor marker provided a basis to improve farming practices to produce higher quality milk. Additionally

it demonstrated the likelihood off-flavors in milk of the misclassification as SOF milk, which would inhibit developing solutions. In the second part of this presentation, a more in-depth understanding of the off-flavors generated during aseptic thermal treatment and subsequent storage will be discussed. Milk undergoes aseptic processing to enhance product stability and shelf life but the high temperature short time treatment can also be detrimental to flavor quality. Key off-flavor markers in aseptic milk were identified and pathways of generation were defined providing information for the development of successful flavor improvement strategies. The application of common food phenolic compounds (i.e., in cocoa, tea, and soybeans) to suppress off-flavor development and improve consumer acceptability in UHT products will also be discussed.

**Key Words:** milk flavor, quality, off-flavor

# Milk Protein and Enzymes

**782 Dietary whey and casein produce differential effects on energy balance, gut hormones, glucose metabolism, and taste preference in diet-induced obese rats.** Adel Pezeshki\*, Andrew Fahim, and Prasanth Chelikani, *University of Calgary, Calgary, AB, Canada.*

Milk proteins decrease food intake, body weight, and improve glycemic control; however, little is known about the underlying mechanisms of improvements in energy balance and glucose metabolism. The objectives of this study were to determine the effects of dietary whey, casein, and a combination of the 2, on food intake, energy expenditure, body composition, gut hormones, glucose tolerance, metabolic markers, and taste preference in diet-induced obese rats. In experiment 1, obese rats were randomly assigned to isocaloric high-fat diets ( $n = 12/\text{group}$ ; 33% calories from fat) containing egg white (control; 14% protein calories), whey (WH; 40% protein calories), casein (CA; 40% protein calories) or whey + casein (WHCA; 40% protein calories). Measurements included various behavioral and metabolic parameters. In experiment 2, following an 8-d training period, preference for WH, CA, WHCA or control diets was assessed on consecutive days. Data were analyzed by MIXED procedure. WH, CA and WHCA decreased food intake, body weight and fat content with WH and CA producing pronounced effects. The hypophagic effects of WH were likely due to reduced dietary preference. WH and CA but not WHCA improved glucose tolerance, with WH being more effective. WH reduced energy expenditure at early stages, whereas CA and WHCA increased energy expenditure at later stages of the study. WH, CA and WHCA decreased plasma leptin, glucose-dependent insulinotropic polypeptide and interleukin-6, whereas WH increased glucagon-like peptide-1 concentrations. The CA and WHCA diets increased plasma membrane glucose transporter-4 (GLUT4) to total GLUT4 ratio in skeletal muscle; protein abundance of other markers of glucose and energy metabolism (total Akt, AMPK $\alpha$ , COX-IV, HADH, SIRT3) in the adipose and cardiac tissues did not differ. Overall, our data demonstrate that dietary whey, casein, and their combination, improve energy balance through divergent effects on food intake, energy expenditure, glucose tolerance and gut hormone secretion, with whey being more efficacious. These findings have potential significance for developing whey-based functional foods and nutraceuticals for weight and diabetic control.

**Key Words:** whey, casein, energy balance

**783 Characterization of the bovine milk proteome produced by Holstein and Jersey breeds of dairy cows.** Rink Tacoma\*, Lam Ying Wai, Julia Ganister Fields, and Sabrina Greenwood, *University of Vermont, Burlington, VT.*

Low-abundance milk proteins are of interest because of their diverse bioactivity. The objective of this study was to characterize the low-abundance protein profile within the whey fraction of milk produced by 2 dairy cattle breeds. A 7-d trial was conducted with 6 Jersey ( $80 \pm 49$  DIM) and 6 Holstein ( $75 \pm 21$  DIM) cows paired by DIM and parity, housed in the same tie-stall barn. Cows were all maintained on the same TMR diet and were fed individually to determine daily DMI. Milk samples were collected at a.m. and p.m. milking during the experiment. Milk composition (protein, fat, milk urea nitrogen (MUN), SCC) was determined in all samples collected and subsamples were collected and stored at  $-80^\circ\text{C}$  for low-abundance protein analysis. Milk samples for low-abundance protein analysis were thawed, pooled within animal

and a mammalian protease inhibitor was added to each sample before centrifugation to isolate the whey fraction. Samples were depleted of casein and separated by SDS-PAGE. Samples were excised and subjected to a tryptic digest followed by LC-MS/MS analysis on a linear ion trap (LTQ)-Orbitrap Mass Spectrometer (MS). Product ion spectra were searched using SEQUEST on Proteome Discoverer 1.4 against a curated Bovine database. A linear mixed model was used to perform a repeated measures analysis on milk parameters and DMI in SAS (9.4). DMI ( $P = 0.0108$ ), milk protein % ( $P = 0.0021$ ), protein yield (kg/d;  $P = 0.0068$ ) and fat % ( $P = < 0.001$ ) were different between breeds whereas MUN ( $P = 0.86$ ) and SCC ( $P = 0.59$ ) were not different between breeds. MS analysis identified 947 proteins including over 45 proteins present at significantly different peptide counts between breeds ( $P < 0.05$ ) with fold differences in peptide counts ranging from 0.2 to 6.5. Some known bioactive proteins were present at significantly different levels, including lactotransferrin ( $P = 0.0026$ ) and complement C2 ( $P = 0.0001$ ), whereas other known bioactive proteins including osteopontin ( $P = 0.17$ ) and lactoperoxidase ( $P = 0.29$ ) were present at similar levels in both breeds. This work provides insight into the low-abundance protein composition of milk produced by 2 dairy breeds.

**Key Words:** bioactive, whey

**784 Lactoferrin and lactalbumin are more effective than whey protein in improving energy balance and glucose tolerance in diet-induced obese rats.** Rizaldy Zapata\*<sup>1</sup>, Adel Pezeshki<sup>1</sup>, Arash-deep Singh<sup>1</sup>, Mary Chou<sup>2</sup>, and Prasanth Chelikani<sup>1</sup>, <sup>1</sup>*University of Calgary, Calgary, AB, Canada*, <sup>2</sup>*Advanced Orthomolecular Research Inc., Calgary, AB, Canada.*

Whey protein is often reported to promote satiety, aid in weight loss and stimulate energy expenditure. However, there is limited information on the relative efficacies of the individual components of whey – lactalbumin and lactoferrin – on energy balance and diabetic control. Our objectives were to compare the effects of whey with lactalbumin and lactoferrin on weight, body composition, food intake, energy expenditure, glucose tolerance and meal-induced hormone responses in diet-induced obese (DIO) rats. The DIO rats were randomized to receive one of 3 isocaloric and isonitrogenous high-fat diets ( $n = 8/\text{group}$ ; 40% calories from fat, 30% protein calories): (1) whey (WH), (2) lactalbumin (LA), or (3) lactoferrin (LF) and were followed for 65 d (d). Food intake, energy expenditure, body composition, glucose tolerance and plasma satiety hormone concentrations were measured. Data were analyzed by repeated measures ANOVA or ANCOVA. Compared with WH, LF decreased food intake, and decreased energy expenditure during the dark period for the first 14 d of treatment. LA did not alter food intake, but increased energy expenditure during the first 3 h of the dark period after ~34 d of treatment interventions. LF reduced body weight by reducing adipose mass after 7 d whereas LA did not alter body weight and composition. When food intake was used as a covariate, LF reduced blood glucose and plasma leptin, but neither LA nor LF altered plasma concentrations of satiety hormones (GLP-1, GIP, PYY, insulin, amylin). Though glucose tolerance did not differ between WH, LA and LF by 30 d, LA and LF improved glucose tolerance by 60 d. In summary, lactoferrin is more effective than whey in inducing hypophagia, promoting fat loss, improving glucose tolerance and in decreasing plasma leptin, whereas lactalbumin is effective in increasing energy expenditure and improving



glucose tolerance. These components seem to be more beneficial than just whey itself in improving energy balance and glucose tolerance.

**Key Words:** whey, lactoferrin, lactalbumin

**785 Partial hydrolysis of whey protein using immobilized enzymes and conjugation of these hydrolyzates with the aim of lowering whey protein allergenicity.** Yuansheng Gong\*<sup>1</sup>, Lei Xu<sup>1</sup>, and John A. Lucey<sup>1,2</sup>, <sup>1</sup>*Department of Food Science, University of Wisconsin-Madison, Madison, WI*, <sup>2</sup>*Center for Dairy Research, University of Wisconsin-Madison, Madison, WI*.

Our previous research indicated that conjugation of whey protein isolates (WPI) with dextran (DX) via the Maillard reaction could provide an alternative approach to decrease the immunogenicity of whey protein. It has been well established that partial enzymatic hydrolysis of proteins also reduces their antigenicity. We want to explore a combination of partial protein hydrolysis and conjugation to decrease whey protein immunogenicity. In this study, we partially hydrolyzed whey protein by immobilized trypsin and chymotrypsin. All the hydrolyzed proteins were then conjugated with dextran (molecular weight 40kDa). Trypsin and chymotrypsin from bovine were immobilized onto (aldehyde) agarose (6% gels). WPI was partially hydrolyzed by these enzymes at 40, 45 and 50°C. Samples of hydrolyzed whey protein at 30, 60, 120, 180 and 240 min of hydrolysis were analyzed by SDS-PAGE and HPLC. The molecular weight of hydrolyzates was measured by Size-exclusion chromatography coupled with multi-angle laser light scattering detector. The degree of hydrolysis (DH) was 5 and 12% after 30 and 180 min respectively of hydrolysis at 50°C. Molecular weights of most hydrolyzates were between 1 to 5 kDa after 180 min hydrolysis at 50°C. Two hydrolyzates with DH values of 5 and 12% were conjugated with 40 kDa molecular weight dextran via our novel aqueous Maillard reaction method. Conjugates were separated and purified by chromatography. The IgE binding capacity was determined by ImmunoCap method using blood serum from cow's milk protein patient. The IgE binding capacity of conjugated whey protein hydrolyzates is currently being investigated and will be reported. Partial hydrolysis of whey protein plus conjugation with dextran may provide another option to reduce whey protein allergenicity.

**Key Words:** hydrolysis, conjugation, allergenic

**786 Effect of Maillard modification on reducing immunogenicity of whey protein isolate.** Lei Xu\*<sup>1</sup>, Yuansheng Gong<sup>1</sup>, and John A. Lucey<sup>1,2</sup>, <sup>1</sup>*University of Wisconsin-Madison, Department of Food Science, Madison, WI*, <sup>2</sup>*Wisconsin Center for Dairy Research, Madison, WI*.

A growing concern around the world is the number of people that are suffering from food protein allergies, especially among infants and young children. One potential approach is to block IgE binding epitopes of the protein allergen via the Maillard reaction with polysaccharides to decrease the allergy potential. Dairy infant formula is often formulated to a high proportion of whey proteins, and if infants are sensitive to whey proteins then hydrolysis of whey proteins is often used. The goal of this research was to reduce the immunogenicity of whey protein isolate (WPI) by conjugating WPI with dextran (DX). During this study, the effect of the molecular weight ( $M_w$ ) of DX, ranging from 1 to 2000 kDa, on the immunogenicity of WPI-DX were explored. Our data indicated that the WPI to DX molar ratios in the conjugates made from DX with  $M_w$  values of 1 (G1), 3.5 (G3.5), 10 (G10), 150 (G150), 500 (G500), and 2000 kDa (G2000) were 1:4, 1:3, 1:2, 1:1.5, 1:1, and 1:1, respectively. With the increase in the  $M_w$  of DX, the  $M_w$  values of the corresponding conjugates were also increased, as determined by size exclusion chromatography with multiangle laser light scattering. The immunogenicity of conjugates were evaluated by IgE binding capacity of conjugates incubated with serum from blood samples obtained from patients with cow's milk protein allergy. Our results showed that WPI-DX conjugates have a lower WPI-specific IgE binding capacity than native WPI, with the lowest IgE binding capacity obtained in G10 conjugate, demonstrating that glycation via Maillard reaction did significantly reduce the immunogenicity of WPI. Furthermore, atomic force microscopy images suggested that conjugation of WPI with small  $M_w$  dextran resulted in greater surface coverage on the protein compared with large dextran conjugates, hence significantly reducing protein immunogenicity by creating steric hindrance that limited IgE binding.

**Key Words:** whey protein isolate, dextran, immunogenicity

## Nonruminant Nutrition: Immune support

**787 Evaluating the efficacy of chemicals to mitigate *Salmonella* cross contamination in rendered protein meals.** Roger A. Cochrane\*, Anne R. Huss, and Cassandra K. Jones, *Kansas State University, Manhattan, KS.*

*Salmonella* is a potential feed safety hazard that is destroyed during rendering, but this does not eliminate the potential for cross contamination. The objective of this experiment was to evaluate the effectiveness of chemicals to mitigate *Salmonella* cross-contamination in rendered proteins. Treatments were arranged in a 6 × 4 factorial with 6 chemical treatments: (1) chemically negative control, (2) 0.3% commercial formaldehyde, (3) 2% essential oils (1:1 garlic, turmeric, capsicum, oregano, and rosemary), (4) 2% medium chain fatty acids (1:1 caproic, caprylic, and capric acid 1:1), (5) 3% organic acids (1:1 lactic, propionic, formic, and benzoic), (6) 1% sodium bisulfate in 4 rendered proteins: (a) feather meal, (b) blood meal, (c) meat and bone meal, and (d) poultry by-product meal. Matrices were chemically treated, then inoculated with *Salmonella* Typhimurium, and enumerated on d 0, 1, 3, 7, 14, 21, and 42 post-inoculation. Analyzed values represent colony forming units per gram (cfu/g). All main effects and interactions were significant ( $P < 0.05$ ). *Salmonella* concentration in ingredients treated with medium chain fatty acid and commercial formaldehyde were similar to one another ( $P = 0.23$ ), but 2 logs lower than the control ( $P < 0.05$ ;  $3.20 \times 10^0$  and  $4.45 \times 10^0$  vs.  $3.65 \times 10^2$  cfu/g, respectively). Ingredients treated with organic acid and essential oils had less *Salmonella* than the control ( $P < 0.05$ ;  $1.58 \times 10^1$  and  $1.26 \times 10^2$  cfu/g, respectively). However, treating ingredients with sodium bisulfate resulted in similar *Salmonella* concentrations as the control ( $P < 0.05$ ;  $2.42 \times 10^2$  cfu/g). Time played a role in *Salmonella* mitigation as all days ( $P < 0.05$ ) except d 14 and 21 ( $P = 0.92$ ) were different from one another ( $3.14 \times 10^4$ ,  $4.48 \times 10^2$ ,  $5.57 \times 10^1$ ,  $8.98 \times 10^0$ ,  $3.07 \times 10^0$ ,  $3.16 \times 10^0$ ,  $1.35 \times 10^0$  cfu/g for d 0, 1, 3, 7, 14, 21, and 42, respectively). Matrix also affected *Salmonella* stability as concentrations in meat and bone meal and blood meal and were similar to one another ( $P = 0.36$ ;  $6.54 \times 10^1$  and  $5.32 \times 10^1$  cfu/g, respectively), but greater than ( $P < 0.05$ ) levels in feather meal and poultry by-product meal ( $2.29 \times 10^1$  and  $2.31 \times 10^1$  cfu/g, respectively). In summary, chemical treatment and time both mitigated *Salmonella*, but their effectiveness was matrix dependent. Chemical treatment with medium chain fatty acids or a commercial formaldehyde product was most effective at mitigating *Salmonella* in rendered protein meals.

**Key Words:** *Salmonella*, chemical treatment, feed safety

**788 Supplemental effects of herbal additive on growth performance, health status, and carcass quality in finishing pigs as alternatives to the use of antibiotics.** W. Parnsen\*, S. H. Zhang, and S. W. Kim, *Department of Animal Science, North Carolina State University, Raleigh, NC.*

This study was conducted to determine the effect of herbal additive (AV/AGP/10, Ayurved Limited, Baddi, H.P., India, based on extracts of garlic, ginger, menthol, cinnamon, and thyme) on the growth performances, health status, and carcass quality of finishing pigs fed diets. Ninety-six pigs (48 barrows and 48 gilts) at 84 d of age ( $41.5 \pm 2.7$  kg BW) were allotted to 4 treatments (2 × 2 factorial arrangement with 0 and 0.05% Tylan40 (Elanco) and herbal additive (0 and 0.05% AV/AGP/10) based on a randomized complete block design with 8 pens (3 pigs per pen, 4 barrow and 4 gilt pens) per treatment. Body weight and feed intake were measured weekly. Blood samples were taken on d 37 to obtain

plasma to measure tumor necrosis factor  $\alpha$  (TNF- $\alpha$ ), protein carbonyl, and immunoglobulins. On d 40, 32 pigs (1 pig from each pen and 8 pens per treatment) were euthanized to collect duodenal and jejunal mucosa layers for TNF- $\alpha$ , protein carbonyl, IgA, and IgG concentrations. Gut tissues from middle part of jejunum were obtained to measure villus height (VH), crypt depth (CD), villus width and VH/CD. Carcass traits were measured including backfat thickness (10th rib), loin pH, marbling score, loin color, and drip loss. Data were analyzed using the MIXED procedure in SAS with pen as the experimental unit (treatment and sex as fixed effects and BW as a random effect). Overall, antibiotic supplementation increased ( $P < 0.05$ ) final BW (88.0 to 90.4 kg), ADG (1.176 to 1.225 kg/d), hot carcass weight (62.0 to 63.8 kg), and cold carcass weight (60.0 to 61.7 kg). Herbal supplementation tended to reduce ( $P = 0.079$ ) ADFI (2.829 to 2.785 kg/d), tended to enhance ( $P = 0.060$ ) G:F at wk 5 (0.430 to 0.460), and enhanced ( $P < 0.05$ ) G:F at wk 6 (0.612 to 0.643). Backfat thickness, loin weight, loin color, marbling score, plasma TNF- $\alpha$ , and tissue TNF- $\alpha$  were not affected by factors. There were no interactions between 2 factors in all measurements. Collectively, antibiotic supplementation improves growth performance and carcass weight whereas herbal additive can potentially enhance feed efficiency if it is fed to finisher pigs longer than 4 wk regardless of the use of antibiotics.

**Key Words:** carcass quality, gut health, herbal additive

**789 Effect of additives on survival of swine delta coronavirus (SDCoV) in complete feed.** Katie M. Cottingim\*<sup>1</sup>, Harsha Verma<sup>2</sup>, Fernando Sampedro<sup>2</sup>, Pedro E. Urriola<sup>1</sup>, Gerald C. Shurson<sup>1</sup>, and Sagar M. Goyal<sup>2</sup>, <sup>1</sup>Department of Animal Science, University of Minnesota, St. Paul, MN, <sup>2</sup>Veterinary Population Medicine, University of Minnesota, St. Paul, MN.

Swine delta coronavirus was first discovered in 2012 in China, and the first confirmed case in the US was in Ohio in 2014. Feed contaminated with feces from infected pigs is a potential route of transmission of SDCoV. Organic acids (OA) and feed additives are included in diets to improve nutritional value. The objectives of our study were to determine if OA or additives at recommended or twice the recommended concentrations are effective in reducing the survival of SDCoV in feed. Five g aliquots of complete feed were placed in scintillation vials followed by addition of 1 of 8 additives at recommended concentrations of 150mg Ultracid P (orthophosphoric, citric, fumaric, and malic acid; Nutriad), 20mg Activate DA (organic acids and 2-hydroxy-4-methylthio butanoic acid; Novus Intl.), 10mg KEM-GEST (phosphoric, fumaric, lactic, and citric acid; Kemin Agrifoods), 10mg Acid Booster (phosphoric, citric, and lactic acid; Agri-Nutrition), 56 $\mu$ L Luprosil (propionic acid; BASF), 46 $\mu$ L formic acid (formic acid; BASF), 20mg salt, or 20mg sugar. Virus was grown and titrated in swine testicular (ST) cells and was added to all vials at 1mL/vial (initial virus titer =  $3.2 \times 10^5$  TCID<sub>50</sub>/mL) followed by vortexing and storage of vials at room temperature (25°C) for 5 wk. Exp. 2, twice the recommended concentration of additives was added to aliquots and experimental conditions were identical to those used in Exp. 1 except that virus survival was evaluated on d 0, 1, 3, 7, and 10 of storage. Samples were eluted with 3% beef extract-0.05M glycine solution and inoculated in ST cells to calculate TCID<sub>50</sub>/mL. Data were analyzed using Weibull inactivation kinetics model. In Exp. 1, we observed that it takes 0.86 d to reduce the concentration of virus by 1 log. None of the additives tested reduced the inactivation kinetics of the virus at recommended doses. However, adding twice the recommended concentration reduced the time for 1 log inactivation in feeds with Acid

**Table 1 (Abstr. 791).** Serum CK, AST, and LDH (U/L) concentrations for weeks 1 to 11 (wk 0 = baseline)

Item <sup>1</sup>	Adaptation period				Exercise				Rest		No supplementation	
	0	1	2	3	4	5	6	7	8	9	10	11
CK	180.5 <sup>bc</sup>	164.4 <sup>c</sup>	182.5 <sup>bc</sup>	224.8 <sup>a</sup>	168.7 <sup>bc</sup>	186.3 <sup>bc</sup>	227.9 <sup>a</sup>	199.5 <sup>bc</sup>	194.8 <sup>b</sup>	159.4 <sup>c</sup>	197.1 <sup>bc</sup>	217.5 <sup>ab</sup>
AST	319.8 <sup>ab</sup>	261.8 <sup>c-f</sup>	226.6 <sup>f</sup>	330.4 <sup>a</sup>	284.0 <sup>b-e</sup>	301.0 <sup>a-c</sup>	332.3 <sup>a</sup>	322.5 <sup>ab</sup>	276.7 <sup>c-e</sup>	250.4 <sup>ef</sup>	256.9 <sup>d-f</sup>	292.2 <sup>a-d</sup>
LDH	469.6 <sup>de</sup>	469.5 <sup>de</sup>	504.5 <sup>c-e</sup>	606.8 <sup>a</sup>	514.5 <sup>b-d</sup>	506.5 <sup>cd</sup>	524.3 <sup>bc</sup>	490.0 <sup>c-e</sup>	424.2 <sup>e</sup>	391.8 <sup>f</sup>	473.7 <sup>c-e</sup>	563.3 <sup>ab</sup>

<sup>a-f</sup>Mean values within a line with different letters are different ( $P < 0.05$ ).

<sup>1</sup>SEM: CK, 10.4; AST, 13.3; LDH, 24.48.

Booster (0.28 d) ( $r^2 = 0.93$ ), Activate DA (0.12 d) ( $r^2 = 0.72$ ), and salt (0.09 d) ( $r^2 = 0.91$ ). Thus, using these feed additives may reduce virus survival in complete feed faster.

**Key Words:** feed additive, organic acid, swine delta coronavirus

**790 Infection with porcine reproductive and respiratory syndrome virus (PRRSV) affects body protein deposition and alters amino acid metabolism in growing pigs.** Whitney D. Stuart<sup>\*1</sup>, Thomas E. Burkey<sup>2</sup>, Nicholas K. Gabler<sup>3</sup>, Kent Schwartz<sup>3</sup>, Thu Dinh<sup>4</sup>, Cornelius F. M. de Lange<sup>5</sup>, David Klein<sup>1</sup>, John A. Dawson<sup>1</sup>, and Anoosh Rakhshandeh<sup>1</sup>, <sup>1</sup>Texas Tech University, Lubbock, TX, <sup>2</sup>University of Nebraska-Lincoln, Lincoln, NE, <sup>3</sup>Iowa State University, Ames, IA, <sup>4</sup>Mississippi State University, Mississippi State, MS, <sup>5</sup>University of Guelph, Guelph, ON, Canada.

Changes in plasma free amino acid (AA) kinetics reflect modification of AA metabolism in different metabolic states. Infectious diseases in growing pigs redistribute AA from body protein deposition (PD) toward processes involved in immune response. The aim of the current study was to quantify the effects of PRRSV infection on PD and AA metabolism. Twenty PRRSV-negative gilts (BW 9.4 ± 0.9 kg) were blocked by time, surgically catheterized, housed in metabolism crates, fed a corn-SBM based diet (ME 14 MJ/kg, SID Lys 11.5 g/kg), feed-restricted (550 g/d), and then inoculated intramuscularly with a live PRRSV. Blood samples were collected via the catheters at 0, 2, 4, 6, 8, and 10 d post inoculation, and assayed for blood chemistry, hematology, and serum viral load. Body temperature (BT) was monitored on a daily basis. N-balances were determined during a 3 d pre inoculation period and a 3 d post inoculation period. At the end of each N-balance period a single dose of [U-13C, U-15N] AA mixture (Lys, Met, Thr, Trp, Ile, Leu, Val, Phe, Gln) was infused intravenously to study plasma AA kinetics. For each pig and AA, an irreversible loss rate (ILR; disappearance of AA from plasma pool) was determined. Data were analyzed using a randomized complete block design (PROC MIXED in SAS). Blood chemistry, hematology, BT, and serum viral load results indicated that PRRSV injection induced effective immune system stimulation in pigs ( $P < 0.05$ ). The PRRSV challenge reduced PD from 59.4 to 38.1 g/d, SE 4.56, but increased the ILR (μmol/kg BW/h) for Met (from 108 to 228, SE 26.7) and Thr (from 83 to 129, SE 11.5;  $P < 0.05$ ). The ILR for other AA was not affected by PRRSV. These results suggest that PRRSV infection reduces PD and alters metabolism of Met and Thr in growing pigs. The increased ILR for Met and Thr in PRRSV challenged pigs could be associated with enhanced utilization of Met and Thr for synthesis of immune system metabolites and increased catabolism of these AA. This may increase dietary Met and Thr requirements of health challenged pigs, relative to requirements for other AA.

**Key Words:** amino acid, kinetic, PRRS

**791 Effect of selenium and vitamin E supplementation on muscular damage enzymes in horses under moderate exercise.**

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Twenty-four horses (450 kg, 5–15 yr) were involved in an 11-wk trial to study the effect of both selenium (Se, Se-yeast) and vitamin E (E, α-tocopheryl) supplements on serum concentration of muscle damage enzymes (creatinine phosphokinase, CK; aspartate aminotransferase, AST and lactate dehydrogenase, LDH) in horses under moderate exercise. Horses were not exercised for a month before this study; they were housed in individual stables and assigned into a factorial trial arrangement (2 × 2; Se, E levels; n = 6) with repeated measures. Basal ration was deficient in Se and vitamin E (<2 μg; 14.4, IU/kg DM). The supplementation levels met NRC 2007 requirements: low (L: Se, 0.1 mg/kg DM; E, 1.6 IU/kg BW) and high (H: Se, 0.3 mg/kg DM; E, 2 IU/kg BW) for horses under moderate or intense exercise, respectively. Hence, treatments were: LSeLE, HSeLE, LSeHE and HSeHE. Exercise was done from wk 4 to wk 7: 3 consecutive days for 30 min (5–20–5), including: warm up-moderate gallop-cool down. Serum samples were taken every week (wk 4-wk 7, at the end of last exercise day). Creatine phosphokinase, AST and LDH were quantified by spectrophotometry. Data were analyzed using the SAS PROC MIXED procedure (SAS Inst. Inc., Cary, NC) for the design described above. Fixed effects were Se, E, wk, and their interactions, while horse nested within treatment was the random effect. Tukey-Kramer test was used to compare LSM. Neither Se nor vitamin E levels affected enzymes concentrations ( $P < 0.01$ ). An effect of wk was observed for CK and AST at wk 6, exercise explained this fact; CK, AST and LDH increased at wk 3 and wk 11 (un-supplemented horses)

**Key Words:** exercise, antioxidant, horse

**792 Effect of Sal CURB on digestibility of energy and nutrients by growing pigs.** Yanhong Liu\* and Hans H. Stein, *Department of Animal Sciences, University of Illinois, Urbana, IL.*

Sal CURB brand ASF liquid antimicrobial is a blend of aqueous formaldehyde 37% solution and propionic acid. An experiment was conducted to determine the effects of Sal CURB on apparent total-tract digestibility (ATTD) of energy, Ca, and P, and apparent ileal digestibility (AID) of CP and AA in a diet fed to growing pigs. Eight barrows (initial BW: 26.81 ± 1.77 kg) had a T-cannula installed in the distal ileum and were allotted to a 2-period switch back design with 2 diets and 8 replicates per diet. A saline treated base mix or a Sal CURB treated base mix was added to the diet that were based on corn, soybean meal, distillers dried grains with solubles, and oats (10.0%). Each period lasted 7 d and fecal samples were collected on d 5 and 6 and ileal digesta were collected



on d 6 and 7 of each period. Three random samples from each of the 2 base mixes were analyzed for phytase and vitamins on d 0, 7, 28, and 49 after sampling to determine stability of vitamins and phytase in the base mixes. Data were analyzed by ANOVA using PROC MIXED of SAS in a randomized complete block design with diet as fixed effect and pig and period as random effects. Pigs fed Sal CURB diet had greater ( $P < 0.05$ ) ATTD of Ca (69.33%) and P (61.72%) than pigs fed the control diet (63.80 and 57.53%, respectively). No differences were observed in the ATTD of DM and GE and the AID of CP and all AA between the control diet and the Sal CURB diet, with the exception that the AID of Met was greater ( $P < 0.05$ ) in the Sal CURB diet (89.56%) than in the control diet (88.41%). The Sal CURB treated base mix contained less ( $P < 0.05$ ) phytase than the saline treated base mix on d 0, 7, 28, and 49. The Sal CURB treated base mix contained more ( $P < 0.05$ ) vitamin B3 and B9 on d 0, more ( $P < 0.05$ ) vitamin B1 on d 28, and more ( $P < 0.05$ ) vitamin B1, B2, and D3 on d 49, compared with the saline treated base mix. In conclusion, supplementation of Sal CURB to diets for growing pigs does not affect the digestibility of energy and AA, but increases the digestibility of Ca and P in phytase containing diets. However, Sal CURB treatment reduces the concentration in the base mix of phytase by approximately 20%, but concentrations of vitamins are not reduced.

**Key Words:** digestibility, pigs, Sal CURB

**793 Effect of a 3-strain *Bacillus*-based direct-fed microbial on growth performance, volatile fatty acid production, carcass characteristics, and gastrointestinal tract weights in growing-finishing pigs fed low or high fiber diets.** Neil W. Jaworski<sup>\*1</sup>, Augustine Owusu-Asiedu<sup>2</sup>, Ajay Awati<sup>2</sup>, Alastair Thomas<sup>2</sup>, and Hans H. Stein<sup>1</sup>, <sup>1</sup>University of Illinois at Urbana-Champaign, Urbana, IL, <sup>2</sup>Danisco Animal Nutrition, DuPont Industrial Biosciences, Marlborough, UK.

The effect of a 3-strain *Bacillus*-based direct-fed microbial (DFM) on growth performance, VFA production, carcass characteristics, and gastrointestinal tract weights in growing-finishing pigs was evaluated. A total of 160 pigs (initial BW: 26.61 ± 2.17 kg) were randomly allotted to a 2 × 2 factorial design with 2 diet types [low fiber (LF) or high fiber (HF)] and 2 levels of DFM [0 or 60 g DFM (2.4 × 10<sup>12</sup> cfu/kg feed)/t of feed] and 4 pigs per pen. Grower and early finisher diets were fed for 5 weeks and late finisher diets were fed for 24 d. The LF diets contained corn and soybean meal as main ingredients and HF diets contained corn, soybean meal, corn distillers dried grains with solubles (30%) and wheat middlings (10%). One pig/pen was harvested at the conclusion of the experiment. Cecum and rectum contents were analyzed for VFA, and carcass characteristics and gastrointestinal tract weights were measured. Data were analyzed as a 2 × 2 factorial with DFM level, diet type, and the interaction of DFM and diet type as fixed effects and rep as a random effect using the Mixed procedure of SAS. Results indicated that for the overall growing-finishing period, there was no difference in ADG between pigs fed LF and HF diets, but pigs fed HF diets had greater ( $P < 0.01$ ) ADFI (2.5 vs. 2.4 kg/d), reduced ( $P < 0.01$ ) G:F (0.36 vs. 0.37 kg/kg), reduced ( $P < 0.05$ ) dressing percentage (77.5 vs. 78.2%), and increased ( $P < 0.05$ ) weight of empty intestinal tract ( $P < 0.05$ ) compared with pigs fed LF diets. Pigs fed LF diets had a greater ( $P < 0.05$ ) concentration of propionate in cecum contents and greater ( $P < 0.05$ ) concentrations of all VFA in rectal contents compared with pigs fed HF diets. Pigs fed diets supplemented with DFM had greater ( $P \leq 0.05$ ) HCW (88.5 vs. 84.3 kg) and backfat thickness than pigs fed diets with no DFM, but fat-free lean percentage was reduced ( $P < 0.05$ ). In conclusion, pigs fed HF diets had greater overall ADFI, lower G:F, and a lower dressing percentage than pigs fed LF. The addition of

this DFM to LF or HF diets had no effect on growth performance, but increased HCW.

**Key Words:** direct-fed microbial, fiber, pig

**794 Effect of a 3-strain *Bacillus*-based direct-fed microbial on growth performance and volatile fatty acid production in nursery pigs fed low or high fiber diets.** Neil W. Jaworski<sup>\*1</sup>, Augustine Owusu-Asiedu<sup>2</sup>, Ajay Awati<sup>2</sup>, Alastair Thomas<sup>2</sup>, and Hans H. Stein<sup>1</sup>, <sup>1</sup>University of Illinois at Urbana-Champaign, Urbana, IL, <sup>2</sup>Danisco Animal Nutrition, DuPont Industrial Biosciences, Marlborough, UK.

The effect of a 3-strain *Bacillus*-based direct-fed microbial (DFM) on growth performance and VFA production in weaning pigs fed low or high fiber diets was evaluated. Two hundred pigs (initial BW: 6.31 ± 0.73 kg) were allotted to 1 of 4 dietary treatments with 5 pigs/pen. Treatments were arranged in a 2 × 2 factorial design with 2 diet types [low fiber (LF) or high fiber (HF)] and 2 levels of DFM [0 or 60 g DFM (2.4 × 10<sup>12</sup> cfu/kg)/t of feed]. Phase 1 diets were fed for 2 weeks post-weaning and phase 2 diets for the following 29 d. The LF diets contained corn and soybean meal as main ingredients and HF diets contained corn, soybean meal, corn distillers dried grains with solubles (7.5 and 15.0% in phase 1 and 2, respectively) and wheat middlings (10.0%). The NE in phase 1 LF and HF diets was 2,525 and 2,463 kcal/kg, respectively. The NE in phase 2 LF and HF diets was 2,483 and 2,414 kcal/kg, respectively. No diets contained antibiotic growth promoters. Pigs and feed were weighed at the start and at the end of each phase, and ADG, ADFI, and G:F were calculated. At the conclusion of phase 2, 1 pig/pen was killed. Cecum and rectum contents were collected and analyzed for VFA. Data were analyzed as a 2 × 2 factorial with DFM, diet type, and the interaction of DFM and diet type as fixed effects and rep as a random effect using the Mixed procedure of SAS. During phase 1, a reduction ( $P < 0.05$ ) in ADFI of pigs fed HF diets and a tendency for pigs fed diets supplemented with DFM to have an improved ( $P = 0.10$ ) G:F were observed. Pigs fed LF diets had a greater ( $P = 0.05$ ) BW (27.06 kg) at the end of phase 2 compared with pigs fed HF diets (26.31 kg). The concentration of acetate, propionate, and isovalerate in rectum contents were greater ( $P < 0.05$ ) in pigs fed LF diets, and there was a tendency for addition of this DFM to increase ( $P = 0.10$ ) isovalerate concentration in cecum content. For the entire experimental period, HF diets decreased ( $P = 0.05$ ) ADFI and ADG of pigs by 32.5 and 17.5 g/d, respectively, and the addition of this DFM improved ( $P < 0.05$ ) G:F. In conclusion, the 3 strain *Bacillus*-based DFM improved overall G:F by 0.8 and 7.3% in LF and HF diets, respectively.

**Key Words:** direct-fed microbial, fiber, pig

**795 Effect of L-DOPA on performance and serum cholesterol of broiler.** Babatunde R. Omidwura<sup>\*</sup>, Adebisi F. Agboola, and Eustace A. Iyayi, University of Ibadan, Ibadan, Oyo, Nigeria.

The L-DOPA found in mucuna seed, when present with other antinutritional factors, has been implicated in nutritional disorders of monogastric animals. It is also reputed to influence muscular development. However, information on its effect on the hydrolysis of triglycerides is scanty. Therefore, the effect of L-DOPA on growth performance and serum cholesterol in broiler chickens was investigated. Two hundred forty 1-d-old chicks were allotted to 6 treatments containing positive control (PC) with normal energy, negative control (NC) with high energy, NC+0.1, NC+0.2, NC+0.3 and NC+0.4% L-DOPA. All treatments had 4 replicates in a completely randomized design. Feed intake (FI), final weight (FW), body weight gain (BWG), feed conversion ratio (FCR),

dressed weight (DW), serum total cholesterol (TC), triglyceride (Tg), high density lipoprotein (HDL), low density lipoprotein (LDL) and very low density lipoprotein (VLDL), were determined following standard procedure. Data were analyzed using descriptive statistics and ANOVA at  $\alpha_{0.05}$ . The body weight gain and final weight of birds fed PC were 1791.0 and 1830.0 g, NC+0.1% L-DOPA were 1827.7 and 1866.7 g and NC+0.2% L-DOPA were 1871.9 and 1910.9 g, respectively, and the feed intake of PC (3231.5 g), were better than other treatments. The serum total cholesterol were 125.0, 129.4, and 131.5 mg/dL of PC, NC and NC+0.4% L-DOPA, respectively, and LDL were 75.1 and 75.7 mg/dL of PC and NC, respectively. The values observed were similar but higher than other treatments with lowest LDL observed in birds fed NC+0.2% (61.2 mg/dL) and NC+0.3% L-DOPA (62.4 mg/dL). The VLDL values observed, 19.8, 22.0, 22.3 and 22.7 mg/dL of birds fed NC+0.1%, NC+0.2%, NC+0.3% and NC+0.4% L-DOPA, respectively, were similar, lower than 26.8 mg/dL of NC but higher than 15.7 mg/dL of PC. The dressed weight at 1375.0 g and 1357.1 g of NC+0.1% and NC+0.2% L-DOPA, respectively, were similar but better than other treatments. Triglyceride and HDL were not affected across treatments. L-DOPA extract, at levels tested, had no detrimental effect on broilers; rather, the serum cholesterol profile was improved and better bird performance was observed especially at 0.1 and 0.2% L-DOPA inclusion rates. Therefore, 0.2% inclusion is recommended in diets of broiler chickens for improved performance, carcass characteristics and cholesterol profile.

**Key Words:** L-DOPA, serum cholesterol, broiler

**796 Effect of high caloric diet enriched in medium-chain triglycerides and arginine supplementation on cholesterol and lipid metabolism in finishing pigs.** Z. H. Zhou, D. W. Chen, P. Zheng, G. Tian, B. Yu\*, and Y. Yao, *Animal Nutrition Institute, Sichuan Agricultural University, Chengdu, Sichuan, China.*

A total of 60 DLY pigs ( $88.16 \pm 8.03$  kg) were used in a 28-d study to determine the effects of high caloric diet enriched in medium-chain triglycerides (HCD-MCT) and L-arginine (Arg) supplementation on cholesterol and lipid metabolism in a  $2 \times 2$  factorial design. Data were analyzed by ANOVA using the GLM procedures of SPSS with the main effects of diet (10% corn starch vs. 10% coconut oil, coconut oil is enriched in MCT, dietary DE were 3.40 Mcal/kg and 3.87 Mcal/kg), supplemented Arg (0 vs. 1%) and their interaction. HCD-MCT increased serum triacylglycerol (+35.48%), total cholesterol (+34.80%) and HDL-C (+66.05%) levels ( $P < 0.05$ ), decreased the ratio of LDL-C to HDL-C (-35.24%,  $P < 0.05$ ). Moreover, HCD-MCT upregulated mRNA expression of 3-hydroxy-3-methylglutaryl coenzyme A reductase (+30.69%) and lecithin cholesterol acyltransferase (+53.06%) in liver ( $P < 0.05$ ), downregulated mRNA expression of fatty acid synthase (FAS) in backfat (-50.72%,  $P < 0.05$ ) and peroxisome proliferator-activated receptor- $\gamma$  (PPAR- $\gamma$ , -23.93%) in liver ( $P < 0.05$ ), accompanied by decreased backfat thickness and fat content in liver (-11.91% and -12.76%,  $P < 0.05$ ). Arg supplementation suppressed gene expression of FAS in backfat (-47.41%,  $P < 0.05$ ) and PPAR- $\gamma$  in liver (-19.50%,  $P < 0.05$ ), elevated gene expression of hormone-sensitive lipase (HSL) and carnitine palmitoyltransferase 1 (CPT-1) in backfat (+78.20% and +30.53%,  $P < 0.05$ ), while reduced backfat thickness (-11.48%,  $P < 0.05$ ) and fat content in liver (-7.18%,  $P < 0.05$ ). There was a diet  $\times$  supplemented Arg interaction observed for FAS, CPT-1 and PPAR- $\gamma$  mRNA abundance in backfat and liver ( $P < 0.05$ ), in which pigs fed control diet had less mRNA expression of FAS (-62.00% in backfat, -59.00% in liver) and PPAR- $\gamma$  (-37.00% in liver), and greater CPT-1 expression (+66.00% in liver) compared with pigs fed HCD-MCT diet. In conclusion, our results indicated that HCD-MCT was helpful to

reduce fat deposition in backfat and liver, and improve serum HDL-C concentration. Arg supplementation also prevented fat deposition by regulating lipid metabolism without any effects on cholesterol profile.

**Key Words:** medium-chain triglyceride, arginine, metabolism

**797 Effects of lactulose on growth, carcass characteristics, fecal microbiota, and blood constituents in broilers.** M. Mohammedi\*, S. Shanmugam, H. Y. Shin, S. Mohana Devi, and I. H. Kim, *Department of Animal Resource & Science, Dankook University, Cheonan, Chungnam, South Korea.*

A total of 324 one-day-old Ross 308 mixed-sex broiler chicks with an average initial body weight of 38 g were used in a 35-d growth assay to determine the effect of supplementing diets with lactulose on growth performance, carcass characteristics, fecal microbiota, and blood constituents. There were 18 birds/pen and 6 pens/treatment with food and water consumed ad libitum. Treatments were a corn-soybean-meal-based diet with none, 0.25 and 0.5% of lactulose. Data were analyzed as a completely randomized design using the mixed procedures of SAS (SAS Institute, 1996), orthogonal polynomial contrasts were used to look for linear and quadratic effects of Lactulose dose in the diet. Results indicated that gain of body weight (BWG; 1,715 vs. 1,765 vs. 1,843 g/d) and feed conversion ratio (FCR; 1.63 vs. 1.58 vs. 1.54) were improved linearly ( $P < 0.05$ ). As for results increasing concentration of lactulose in the diet from none to 0.5% increased ( $P < 0.05$ ) BWG and decreased FCR for d 8 to 21, 21 to 35, and 0 to 35. Also chickens fed supplemented diet with 0.5% lactulose showed higher relative weight of breast meat compared with other groups (7.26 vs. 8.50 vs. 8.64%). Inclusion of lactulose decreased the count of *Salmonella* (2.73 vs. 2.50 vs. 2.47 log<sub>10</sub>cfu/g) and *E. coli* (6.57 vs. 6.39 vs. 6.40 log<sub>10</sub>cfu/g) of feces in chickens fed diets containing 0.25 and 0.5% lactulose ( $P < 0.05$ ), but the count of *Lactobacillus* was not affected. Addition of 0.5% lactulose decreased drip loss percentage on d 1 ( $P < 0.05$ ; 3.98 vs. 2.94 vs. 2.35%) but there wasn't any significant effect on meat color. Blood characteristics were not influenced. Thus it was concluded that inclusion of lactulose improved growth performance and altered fecal microbial populations with no negative effect meat quality and blood profile.

**Key Words:** broiler, carcass characteristic, lactulose

**798 Effect of supplying a nucleotide product (Ascogen) to sow diets on growth performance of the offspring in the nursery.** I-Fen Hung\*<sup>1</sup>, Klaus Hoffmann<sup>2</sup>, Peter Koeppel<sup>2</sup>, and Merlin D. Lindemann<sup>1</sup>, <sup>1</sup>University of Kentucky, Lexington, KY, <sup>2</sup>Chemoforma Ltd., Augst, Switzerland.

Nucleotides are not classified as essential nutrients as animals can synthesize them to a limited extent. However, studies have demonstrated their ability to improve biological responses including growth, immunity, and stress resistance. This experiment was conducted with a subset of sows in a commercial farm in China to evaluate the effects of late gestation and lactation supplementation with nucleotides and the performances of pigs post weaning. Sows ( $n = 40$ ) around d 87 of gestation were randomly assigned to 2 dietary treatments: 1) the farm standard gestation and lactation diets (corn and soybean meal based,) or 2) the standard diets blended with 0.05% Ascogen (NT; Chemoforma, Switzerland) during gestation and 0.1% during lactation. At weaning, piglets ( $n = 112$ ) from sows on each diet were split into 2 groups and fed diets that contained either 0 or 0.1% NT in a 42-d study which resulted in a  $2 \times 2$  factorial arrangement. The data were analyzed by ANOVA using GLM program of SAS. Supplying NT to sow diets tended to

reduce piglet mortality at birth (2.4 vs. 9.5%,  $P = 0.10$ ) and the days to rebreeding (5.4 vs. 9.6d,  $P = 0.10$ ). The individual piglet weight at birth and weaning as well as milk and serum immunoglobulin profile (IgA, IgG, IgM by ELISA) were not affected by NT. However, pigs from NT sows were heavier than those from control sows at Week 3 (13 vs. 12kg,  $P = 0.02$ ) and Week 6 post weaning (22 vs. 20 kg,  $P = 0.01$ ). They also had increased ADG (353 vs. 320 g,  $P = 0.01$ ) and ADFI (592 vs. 539 g,  $P = 0.04$ ) throughout the nursery period. Nursery treatments had no effect on growth performance. Piglets from NT sows had higher serum

IgA levels at weaning (0.17 vs. 0.13 mg/mL,  $P = 0.01$ ) and 1 week post weaning (0.20 vs. 0.17 mg/mL,  $P = 0.05$ ), but no effect on serum IgG and IgM levels was observed. In conclusion, adding NT to the sow diets had beneficial effects on mortality of birth, days to rebreeding, and the growth of piglets during nursery in the commercial environment of this study. However, dietary NT supplementation in nursery may have no effect on growth.

**Key Words:** nucleotide, sow, nursery



# Physiology and Endocrinology Symposium: Insulin revisited

**799 Insulin receptor signaling in normal and insulin-resistant states.** Brian O'Neill\*<sup>1,2</sup>, <sup>1</sup>Joslin Diabetes Center, Boston, MA, <sup>2</sup>Harvard Medical School, Boston, MA.

As type 2 diabetes has reached pandemic levels affecting nearly 370 million people worldwide, it is critical to understand the cellular processes that influence this disease. Insulin resistance, or the inability of normal levels of insulin to achieve the normal effect, is a hallmark of type 2 diabetes and metabolic syndrome. When going from the fasted to the fed state, insulin binds to the ubiquitously expressed insulin receptor to activate downstream signaling, which regulates many cellular actions such as glucose, lipid, and protein metabolism. At the level of the organism, insulin's effect on glucose and lipid metabolism occurs primarily through actions on liver, skeletal muscle, and adipose tissue. The various proteins and isoforms that positively and negatively modulate the insulin-signaling cascade ensure a proper response to feeding. However, these regulators of insulin signaling can be disrupted in a variety of ways in response to disease states such as obesity, inflammation, or even during the aging process and contribute to insulin resistance. Understanding the processes by which insulin signaling is affected in response to these disease states is critical to the discovery of new treatments to prevent diabetes, metabolic syndrome, and their complications.

**Key Words:** diabetes, insulin resistance

**800 Roles for insulin-supported skeletal muscle growth.** Robert P. Rhoads\*<sup>1</sup> and Lance H. Baumgard<sup>2</sup>, <sup>1</sup>Virginia Tech, Blacksburg, VA, <sup>2</sup>Iowa State University, Ames, IA.

Basic principles governing skeletal muscle growth and development, from a cellular point of view, have been realized for several decades. Skeletal muscle is marked by the capacity for rapid hypertrophy and increases in protein content. Ultimately, skeletal muscle growth is controlled by 2 basic means; myonuclear accumulation stemming from myoblast proliferation and the protein synthesis and degradation balance. Each process underlies the rapid changes in lean tissue accretion evident during fetal and neonatal growth and are particularly sensitive to nutritional manipulation. Although multiple signals converge to alter skeletal muscle mass, postprandial changes in the anabolic hormone, insulin, link feed intake with enhanced rates of protein synthesis in the neonate. Indeed, a consequence of insulin-deficient states such as diabetes or malnutrition is reduced myoblast activity and a net loss of body protein. A well-characterized mechanism mediating the anabolic effect of insulin involves the phosphatidylinositol 3-kinase (PI3K) mammalian target of rapamycin (mTOR) signaling pathway. Activation of mTOR leads to translation initiation control via the phosphorylation of downstream targets. Modulation of this pathway by insulin, as well as other hormones and nutrients, accounts for enhanced protein synthesis leading to efficient lean tissue accretion and rapid skeletal muscle gain in the growing animal. Dysfunctional insulin activity during fetal and neonatal life stages likely alters growth through cellular and protein synthetic capacities.

**Key Words:** insulin, skeletal muscle, myoblast

**801 The biology of hyperinsulinemia induction of polycystic ovarian syndrome and its complications.** Jean-Patrice Bailargeon\*<sup>1,2</sup>, <sup>1</sup>Université de Sherbrooke, Sherbrooke, QC, Canada,

<sup>2</sup>Research Center of Centre Hospitalier Universitaire de Sherbrooke, Sherbrooke, QC, Canada.

Polycystic ovarian syndrome (PCOS) is a common condition affecting 6 to 10% of women of childbearing age. It is the most frequent endocrine disorder among young women in North America. In addition of being the most frequent cause of female infertility, PCOS is the commonest cause of hyperandrogenism in women, thus leading to esthetical concerns such as excessive hair growth, acne and alopecia. Accordingly, PCOS women are more likely to have impaired quality of life, including depression, anxiety and increased risk of social phobia and suicide attempts. In addition, PCOS is currently considered as a paradigm of cardiometabolic disease, because the prevalence of metabolic syndrome, dyslipidemia and type 2 diabetes (T2D) are much higher in PCOS than in normal age- and BMI-matched women. Indeed, PCOS women demonstrate metabolic insulin resistance (IR) and compensatory hyperinsulinemia, which play a critical role in the syndrome's development. Yet, many questions remain unanswered regarding the mechanisms by which metabolic IR and hyperinsulinemia leads to hyperandrogenemia and PCOS infertility. The existence of women developing PCOS without IR suggests that it is not a requisite for the development of this syndrome. Nevertheless, several evidences suggest that impairment in insulin signaling may be implicated in androgen overproduction at the level of androgen producing tissues. This talk will therefore review potential metabolic mechanisms in the development of PCOS in predisposed women. This talk's objectives are to (1) review briefly the PCOS and some characteristics of insulin resistance in PCOS; (2) describe the mechanisms of insulin action on androgen biosynthesis in PCOS; and (3) discuss the implication of nonesterified fatty acids (NEFAs) in PCOS hyperandrogenemia and its metabolic complications. This talk will discuss the hypothesis that lipotoxicity (the cellular adverse consequences of NEFAs) may cause both the hyperandrogenemia and insulin resistance that characterize PCOS women. Lipotoxicity could therefore explain PCOS symptoms, mainly due to hyperandrogenism, and long-term metabolic consequences.

**Key Words:** polycystic ovary syndrome, insulin, lipotoxicity

**802 Insulin effects on mammary gland extraction and milk synthesis.** Wendie S. Cohick\*, Rutgers University, New Brunswick NJ.

During lactation there is an increased demand for nutrients required for milk protein and lipid synthesis as the mammary gland undergoes functional differentiation from late pregnancy to early lactation. Insulin resistance in insulin-sensitive peripheral tissues channels nutrients to the mammary gland for milk synthesis in early lactation. Glucose uptake by the mammary gland is insulin-independent, with insulin showing little effect on mRNA expression of glucose transporters. In contrast, it has long been known that the lactogenic hormones hydrocortisone, insulin and prolactin are required for maximum expression of milk protein genes in the mammary gland. Recently it has been recognized that the regulation of protein translation may play a central role in determining milk protein production. The mammalian target of rapamycin (mTOR) signal transduction pathway has been identified as a master regulator of protein translation. Data indicating that hormones (i.e., insulin and IGF-I), nutrients (i.e., amino acids) and intracellular energy status interact

to regulate the mTOR signaling pathway and thus protein synthesis in the mammary gland will be presented.

**Key Words:** insulin, milk synthesis, differentiation

**803 Effects of insulin and heat stress on mTOR signaling cascade in bovine mammary epithelial cells.** Kimberly R. Kassube\*, Jeffrey D. Kaufman, and Agustin G. Rius, *The University of Tennessee, Knoxville, TN*.

Insulin increases protein synthesis by activating the signaling pathway that regulates protein translation in mammary tissue. Lactating cows exposed to heat stress (HS) have increased basal levels of insulin but exhibit reduction in milk protein synthesis. The activity of mammalian target of rapamycin (mTOR) signaling cascade is mediated upon phosphorylation and dephosphorylation of protein kinase B (Akt), P70 S6 kinase (S6K1), ribosomal protein S6 (rpS6), and eukaryotic elongation factor 2 (eEF2). The objective of this study was to determine the effects of insulin and HS on phosphorylating activity in Akt, S6K1, rpS6, and eEF2 factors in immortalized bovine mammary cell line (MAC-T). Cells were cultured in 15 mL of Dulbecco's Modified Eagle Medium with 10% fetal bovine serum and 1 µg/mL of insulin at 37°C and 5% CO<sub>2</sub> before the treatments were imposed. The experimental design consisted of a 2 × 2 factorial arrangement of treatments with 2 temperature environments, 37°C thermoneutral or 41°C HS, and 2 insulin concentrations, 0 µg/mL and 1 µg/mL for 12 h. Cell lysates were separated by gel electrophoresis and transferred onto a polyvinylidene fluoride membrane. Western blotting was conducted to identify total and site-specific phosphorylated forms of Akt (Thr308/Ser473), S6K1 (Thr389), rpS6 (Ser235/236) and eEF2 (Thr56). The relative densities for phosphorylated and total forms of Akt, S6K1, rpS6 and eEF2 were quantified and expressed as phosphorylated to total ratio. ANOVA was conducted with SAS 9.4 using mixed models. Preliminary results indicate a significant HS by insulin interaction for rpS6 ( $P < 0.05$ ). There was an increase in phosphorylated to total ratio from  $0.26 \pm 0.09$  to  $0.6 \pm 0.09$  in response to insulin when cells were exposed to HS. However, there was a reduction of this ratio from  $0.38 \pm 0.09$  to  $0.2 \pm 0.09$  in response to insulin when cells were exposed to thermoneutral conditions. The remaining protein factors were not affected by treatments. These results would indicate that the response of mTOR signaling cascade to insulin was altered in MAC-T cells exposed to HS.

**Key Words:** heat stress, insulin, protein synthesis

**804 Heat stress reduces the phosphorylation activity of mTOR signaling cascade in bovine mammary cells.** Jeffrey D. Kaufman\*<sup>1</sup>, Kimberly R. Kassube<sup>1</sup>, Celina Baravalle<sup>2</sup>, and Agustin G. Rius<sup>1</sup>, <sup>1</sup>*The University of Tennessee, Knoxville, TN*, <sup>2</sup>*Universidad Nacional del Litoral, Esperanza, Santa Fe, Argentina*.

Heat stress (HS) alters metabolism of amino acids and reduces synthesis of caseins in bovine mammary glands. The mammalian target of rapamycin (mTOR) cascade regulates the initiation of the translation of protein synthesis and is mediated by protein factors that are activated or inhibited upon phosphorylation. It has been reported that essential amino acids increased protein synthesis by activating the mTOR cascade. Our objective was to determine the effect of HS in phosphorylating mTOR protein factors in immortalized bovine mammary cells line (MAC-T). It was hypothesized that the phosphorylation activity of mTOR signaling factors would be altered in MAC-T cells exposed to HS. Cells were cultured in 15 mL of Dulbecco's Modified Eagle Medium with 10% fetal

bovine serum at 37°C and 5% CO<sub>2</sub>. Cells were subjected to one of 2 treatments: 1) 37°C (control) and 2) 41.5°C (HS) for 12 h. The treatments were repeated 5 times in 5 different days. Cell proteins were harvested and separated by gel electrophoresis and transferred to a polyvinylidene fluoride membrane. Western blotting was conducted to identify total and site-specific phosphorylated forms of protein kinase B (Akt; Thr308/Ser473), P70 S6 kinase (S6K1; Thr389), ribosomal protein S6 (rpS6; Ser235/236), and eukaryotic elongation factor 2 (eEF2; Thr56). Relative densities for phosphorylated and total forms of Akt, S6K1, rpS6 and eEF2 were quantified and expressed as phosphorylated to total ratio. ANOVA was conducted using a mixed model. Compared with control, cells exposed to HS decreased phosphorylation to total ratio of Akt (0.41 vs. 0.29;  $P < 0.001$ ), S6K1 (1.65 vs. 0.97;  $P = 0.042$ ), and rpS6 (1.45 vs. 1.07;  $P < 0.001$ ). However, preliminary results indicated that HS did not affect the ratio of eEF2. These results indicate that HS impaired the translation of proteins by altering the phosphorylation activity of mTOR signaling factors in MAC-T cells.

**Key Words:** heat stress, mammary cell, translation of protein

**805 Proteome of adipose tissue in periparturient dairy cows related to insulin resistance.** Maya Zachut\*, *Department of Ruminant Science, ARO, Volcani, Bet Dagan, Israel*.

Adipose tissue serves as a major endocrine organ with a profound influence on metabolism by secreting and regulating numerous molecules, hormones and adipokines. Many proteins activate intracellular pathways that promote the development of insulin resistance (IR); however, the role of specific proteins in adipose tissue dysfunction is not well defined. The objective was to identify proteins in adipose that are linked to IR and to cows' metabolic status. Adipose tissue biopsies were obtained from 8 multiparous cows at -17 and +4 d relative to parturition. Proteins were analyzed by intensity based, label-free quantitative shotgun proteomics at Weizmann Institute of Science (Rehovot, Israel). Proteins were extracted and subjected to in-solution tryptic digestion. This was followed by nanoflow liquid chromatography coupled to high-resolution tandem mass spectrometry (nanoLC-MS/MS). Quantitative data were extracted using the Genedata Expressionist data analysis package and proteins identified using the Mascot search engine. Cows were previously divided to those with IR or insulin-sensitive (IS) adipose based on phosphorylation of protein kinase B (Akt) in response to insulin stimulation. Proteomics data, after logarithmic transformation, were analyzed by 2-way ANOVA to measure the effects of time (prepartum vs. postpartum), subgroup (IR vs. IS) and their interaction. Body weight (BW) differences were analyzed with GLM of SAS. It was found that cows with IR adipose lost more BW postpartum compared with IS cows. Proteomic analysis revealed 586 proteins in adipose tissues. Comparing IR to IS adipose showed that 18.9% of proteins were differentially expressed (fold change (FC) > 1.5 and  $P < 0.05$ ). The expression of 106 proteins were increased, whereas only 5 were decreased, in IR adipose compared with IS. The abundance of several proteins related to lipolysis was increased in IR adipose compared with IS: hormone-sensitive lipase (FC = 6.8,  $P < 0.03$ ), perilipin (FC = 1.5,  $P < 0.05$ ), and monoglycerol-lipase (FC = 8.2,  $P < 0.0003$ ). This is in accordance with the elevated lipolysis in IR adipose. These proteins could be used as novel biomarkers to identify IR cows, which may indicate of the metabolic status of the peripartum dairy cow.

**Key Words:** proteomics, adipose, insulin

# Production, Management, and the Environment V

**806 Metabolizable energy intake effects on carcass quality of steers finished in southern Chile during summer time.** Rodrigo A. Arias\*<sup>1</sup>, Christian Alvarado-Gilis<sup>1</sup>, Tami Brown-Brandl<sup>2</sup>, and Terry L. Mader<sup>3</sup>, <sup>1</sup>*Instituto de Producción Animal, Universidad Austral de Chile, Valdivia Chile*, <sup>2</sup>*MARC-ARS-USDA, Clay Center, NE*, <sup>3</sup>*Mader Consulting LLC, Gretna, NE*.

A total of 24 red Angus steers (BW = 431.2 ± 10.4) were sorted by BW and allocated in 4 pens (6/pen) equipped with a Calan Broadbent Feeding System to assess the effect of metabolizable energy intake (MEI) on carcass quality during the summer in southern Chile, and then allocated into one of 2 treatments 1.85 × or 2.72 × MEI for maintenance. Animals were fed once per day with the same diet, and treatments were applied by controlling DMI. Climatic data were obtained from a weather station located at 5 km southeast to assess animal thermal comfort by mean of 2 thermal indices (THI and the Comprehensive Climate Index). The study considered a period of 84 d on feed plus 19 d of pre-experimental period to allow the animals get used to the facilities and feeders. Finally, steers were slaughtered on 04/28/2014 in Temuco, where carcass data including hot and cold carcass weight, KPH, muscle pH, rib eye area, marbling score, and back fat were collected between 9th and 10th rib, after 48 h post slaughtering. All data were analyzed under a randomized complete block design arrangement ( $\alpha = 0.05$ ), with each steer an experimental and observational unit. The pH values in both treatments were considered suitable, 5.57 and 5.52 ± 0.05 for T1 and T2, respectively ( $P = 0.42$ ). Values of back fat observed did not show differences (3.92 and 3.87 ± 0.77 for T1 and T2, respectively;  $P = 0.96$ ). In the case of average values for rib eye area and KPH were also similar between treatments ( $P > 0.05$ ). Finally, hot carcass weight was also similar between treatments ( $P = 0.15$ ). Therefore, no differences between T1 and T2 were observed for carcass quality of steers finished during summer time in any of the variables measured. In addition, based on the thermal comfort indices assessed, animals did not suffer heat stress, being able to deal in good shape the summer season. The latter mainly due to the large temperature drop as resulting of cold nights and the availability of water. On the other hand, there were no differences in ADG (0.95 vs. 1.25 ± 0.15 kg/day for T1 and T2, respectively;  $P = 0.18$ ). Based on the data herein collected we can conclude that for southern Chile conditions during summer time, MEI had no effect on beef carcass quality

**Key Words:** beef, summer, heat stress

**807 Metabolizable energy intake effects on tympanic temperature and ADG of steers finished in southern Chile during summer.** Rodrigo A. Arias\*<sup>1</sup>, Christian Alvarado-Gilis<sup>1</sup>, Tami Brown-Brandl<sup>2</sup>, and Terry L. Mader<sup>3</sup>, <sup>1</sup>*Instituto de Producción Animal, Universidad Austral de Chile, Valdivia, Chile*, <sup>2</sup>*MARC-ARS-USDA, Clay Center, NE*, <sup>3</sup>*Mader Consulting LLC, Gretna, NE*.

A total of 24 red Angus steers (BW = 431.2 ± 10.4) were used to assess the effect of metabolizable energy intake (MEI) on ADG and tympanic temperature (TT) during summer time in southern Chile. Steers were sorted by BW and allocated in 4 pens (6/pen) equipped with a Calan Broadbent Feeding System, and then allocated into one of 2 treatments 1.85 × or 2.72 × MEI for maintenance. Animals were fed once per day with the same diet, and treatments were applied by controlling DMI. The TT of each steer was collected during 10 d in the summer with an ibutton device. Ambient temperature (AT), relative humidity, wind speed, and solar radiation data were obtained from a weather station

near to the farm and used to estimate 2 thermal indices (THI and the Comprehensive Climate Index; CCI). All data were analyzed under a randomized complete block design arrangement ( $\alpha = 0.05$ ). The mean AT for all the period was 17.2°C. However, there was a great fall of AT (24°C) between day and night. The low temperatures at night allowed for the dissipation of heat accumulated during the day. The steers fed with 1.85 × MEI showed higher TT than those fed with 2.72 × MEI (38.07 vs. 38.02 ± 0.001°C;  $P < 0.01$ ). However, this difference does not represent a significant change from the biological standpoint. None of the observed thermal comfort indices outperformed the thresholds established in the literature (THI = 68 and CCI = 25). However, maximum values of thermal indices THI, THI<sub>adj</sub> and CCI were 75.5, 79.97, and 34.66, respectively. Animals showed signs of thermal discomfort, due to the increase in respiration rate, which was higher by 20 points during the afternoon ( $P < 0.01$ ). This could be associated with the largest solar radiation received during the afternoon. On the other hand, there were no differences in ADG (1.25 vs. 0.95 ± 0.15 kg/day for 2.72 × and 1.85 × MEI, respectively;  $P = 0.18$ ). In addition, both values are in line with those projected by the NRC Beef model, with superior value predicted in the case of 1.85 × MEI. Based on the data herein collected we can conclude that for southern Chile, the summer weather conditions and MEI had no effect on productive and physiological responses of animals

**Key Words:** thermal index, environment, climate

**808 Effect of maternal heat stress on calf growth performance and metabolism.** J-D. Liu\*<sup>1</sup>, A. P. A. Monteiro<sup>1</sup>, B. M. Ahmed<sup>2</sup>, T. O. Asar<sup>2</sup>, Z. Wu<sup>1</sup>, M. J. Hayen<sup>2</sup>, G. E. Dahl<sup>2</sup>, J. K. Bernard<sup>1</sup>, and S. Tao<sup>1</sup>, <sup>1</sup>*University of Georgia, Tifton, GA*, <sup>2</sup>*University of Florida, Gainesville, FL*.

Preliminary study suggests that maternal heat stress (HS) during late gestation exerts carryover effects on calf's insulin response after weaning, but comprehensive evaluation of how maternal HS affects calf feed intake and metabolic response from birth to weaning is still lacking. Our objective was to evaluate the effects of maternal HS during the dry period on calf feed intake, growth and metabolism from birth to weaning. After birth, 20 heifers born to either HS (n = 10) or cooled (CL, n = 10) dry cows were immediately separated from their dams and fed 3.8 L of pooled colostrum within 4 h. Then, all heifers were managed identically and weaned at 49 d of age (DOA). Calf starter intake was recorded daily, and body weight and withers height were assessed twice a week from birth to 56 DOA. Blood samples were collected at birth (before colostrum feeding), 24 h after birth, and then twice weekly until 56 DOA to assess hematocrit, plasma total protein, and concentrations of insulin and metabolites. To evaluate metabolic responses to maternal HS, a glucose tolerance test, insulin and epinephrine challenges were performed on 3 consecutive days for all heifers at 8, 29, and 57 DOA. Maternal HS during the dry period did not affect heifer birth weight. Compared with HS, CL calves consumed more starter (0.34 vs. 0.53 kg/d,  $P < 0.06$ ) from birth to 56 DOA, were heavier (61.4 vs. 71.7 kg,  $P < 0.01$ ) and tended to be taller (82.2 vs. 84.5 cm,  $P = 0.15$ ) at 56 DOA. Although there was no treatment effect on hematocrit, a treatment by time ( $P < 0.05$ ) interaction was observed, such that CL calves had higher hematocrit at birth (35.7 vs. 32.3%,  $P = 0.03$ ) and 24 h after birth (31.9 vs. 28.4%,  $P < 0.05$ ) compared with HS. Further, there were no differences between treatments on plasma total protein and concentrations of insulin and glucose. In conclusion, maternal HS during late gestation reduces calf starter intake and negatively affects calf growth. The



lower hematocrit of HS calves relative to CL at birth may indicate fetal hypoxia due to maternal HS.

**Key Words:** calf, heat stress, growth performance

**809 Cows exposed to heat stress in utero exhibit improved thermal tolerance.** Bahroz M. S. Ahmed\*<sup>1</sup>, Umair Younas<sup>1</sup>, Turkey O. Asar<sup>1</sup>, Serdal Dikmen<sup>2</sup>, Peter J. Hansen<sup>1</sup>, and Geoffrey E. Dahl<sup>1</sup>, <sup>1</sup>University of Florida, Gainesville, FL, <sup>2</sup>University of Uludag, Bursa, Turkey.

Maternal heat stress during the dry period affects calf performance during postnatal life. The objective was to evaluate whether calves that experienced heat stress in utero have altered thermoregulatory responses to acute heat stress later in life. Cows used in the study were born to dams exposed to heat stress (HT) or cooled (CL) during the dry period preceding their birth. All animals were raised postnatally under identical management. Eight HT (173 ± 132 DIM; 28.9 ± 5.2 kg/d milk) and 8 CL (167 ± 124 DIM; 30.1 ± 8.0 kg/d milk) lactating Holstein cows were used for the study. A heat stress challenge was conducted in 2 blocks using 4 HT and 4 CL cows matched according to milk yield, stage of lactation, and parity. Each challenge, which consisted of transfer from a barn with shade and evaporative cooling to one with shade but no additional cooling for a period of 48 h, was replicated twice for each block. Sweating rate, respiration rate, rectal temperature (RT), and skin surface temperature were measured on each cow at 0900, 1100, 1300, 1500 and 1700 h for 2 consecutive days. Mean ambient temperature across 4 challenge days was 23.8 ± 3.8°C. Sweating rate and skin temperature were measured on the right side of the cow on both shaved (5 × 5 cm) and un-shaved areas on the rump of the cow. During the challenge, differences were observed between treatments for RT (CL: 39.2 ± 0.06; HT: 39.0 ± 0.06°C; *P* = 0.02) and respiration rate (CL: 68.1 ± 1.6; HT: 62.2 ± 1.6 breath/min; *P* = 0.01). There were also tendencies for differences in sweating rate for shaved skin (CL: 30.7 ± 1.6; HT: 27.1 ± 1.6 g/m<sup>2</sup>h; *P* = 0.12) and for unshaved skin (CL: 23.95 ± 1.18; HT: 21.44 ± 1.18 g/m<sup>2</sup>h; *P* = 0.13). There was no effect on skin temperature at the shaved area (CL: 35.28 ± 0.12; HT: 35.32 ± 0.12°C; *P* = 0.81) or the un-shaved area (CL: 34.1 ± 0.14; HT: 34.0 ± 0.14°C; *P* = 0.55). The results support the hypothesis that heat stress in utero in late gestation increases heat tolerance at maturity by increasing capacity to dissipate heat to maintain core body temperature.

**Key Words:** heat stress, heat tolerance, cow

**810 Feeding slow fermentable grains has the potential to ameliorate heat stress in sheep.** Paula A. Gonzalez-Rivas\*, Kristy DiGiacomo, Brian J. Leury, Jeremy J. Cottrell, and Frank R. Dun-shea, Faculty of Veterinary and Agricultural Sciences, The University of Melbourne, Parkville, Victoria, Australia.

Rapid rumen starch fermentation of wheat increases the heat of fermentation and may increase the risk of heat stress (HS). Therefore, feeding slowly fermentable grains such as maize and reducing metabolic heat load may reduce HS in grain-fed sheep. Twenty-two Merino x Poll Dorset wethers were housed in 2 climate-controlled rooms and were fed either maize grain plus forage (39% starch) (MF, *n* = 11) or wheat grain plus forage (37% starch) (WF, *n* = 11) during 3 experimental periods: P1) 7 d of thermoneutral conditions (18–21°C and 26–30% relative humidity (RH)) and restricted feed intake (85% of unrestricted feed intake); P2) 7 d of HS (28–38°C and 40–50% RH) and restricted feed intake; and P3) 7 d of HS as P2 with unrestricted feed intake (1.5 times maintenance) in a complete randomized block design. Water was offered

ad libitum. Physiological parameters—rectal temperature (RT), heart rate (HR), respiration rate (RR) and left and right flank skin temperature (LST, RST)—were measured at 0900, 1300, 1700, and 2100 h. Blood samples were collected on d 2 and 7 of each period for pH, blood gas and hematocrit (HcT). Data were analyzed using restricted maximum likelihood (REML) in GenStat v14. All physiological parameters were elevated (*P* < 0.001) during HS especially during P3. RR, RT, LST, RST and HR were lower (*P* < 0.05) in sheep fed MF, particularly during HS. LST was higher (*P* < 0.001) than RST and sheep feed WF had a larger (*P* < 0.001) difference between LST and RST. During HS total CO<sub>2</sub> (cTCO<sub>2</sub>), hemoglobin (cHgb), bicarbonate (cHCO<sup>-3</sup>), HcT, lactate, glucose and base excess were lower than P1 (*P* < 0.05). In sheep fed MF, the partial pressure of CO<sub>2</sub>, cTCO<sub>2</sub> and cHCO<sup>-3</sup> were higher (*P* < 0.05), while cHgb, pH and HcT were lower (*P* < 0.05) than WF. In conclusion, dietary maize, a slowly fermentable grain, reduced the metabolic heat load from feeding, expressed as reduced physiological parameters at thermoneutral conditions and ameliorated some of the physiological responses negatively affected by HS compared with dietary wheat.

**Key Words:** heat stress, sheep, grain

**811 Heat stress at conception affects lifetime fertility, milk yield, and survival of Holstein cows.** Pablo Pinedo\*<sup>1,2</sup> and Albert De Vries<sup>3</sup>, <sup>1</sup>Texas A&M AgriLife Research, Amarillo, TX, <sup>2</sup>Department of Veterinary Pathobiology, College of Veterinary Medicine & Biomedical Sciences, Texas A&M University System, College Station, TX, <sup>3</sup>Department of Animal Sciences, University of Florida, Gainesville, FL.

The objective of this study was to analyze the association between month of conception (associated with heat stress) and subsequent milk yield, fertility, and survival in cows maintained in FL dairy farms. The hypothesis was that stress conditions at conception and during the initial stages of embryonic development would have a lifetime impact on performance and survival after birth. Initial data consisted of 667,104 DHI lactation records from cows calving between 2000 and 2012 in 152 FL herds. The magnitude of heat stress in each herd was evaluated after fitting Wood's lactation curves and comparing residuals from predicted and actual test-day milk yields by month as the direct effect of season. A sine function was fitted on the 12 residuals per farm using PROC NLIN (SAS). The difference between the highest and lowest points on the sine function was used as a seasonality index (SI). Only herds within the highest quartile for SI were included in the analyses. Cows were grouped according to the date when they were conceived: Summer (SU: Jul-Sept) and winter (WI: Dec-Feb) and comparisons were performed by parity using logistic regression and ANOVA. Control variables included calving month and year, age at first calving, and herd. Age at first calving (d) were 781 vs. 796 (*P* < 0.001) for WI and SU. The odds (95%CI) of survival to a second calving for WI cows were 1.19 (1.09–1.28) times the odds of survival for SU cows. Days from calving to first breeding were 138 vs. 143 (*P* = 0.070); 126 vs. 132 (*P* = 0.030); and 124 vs. 131 (*P* = 0.004) for WI and SU in parities 1, 2, and ≥ 3. Days to conception were 172 vs. 180 (*P* = 0.003); 182 vs. 187 (*P* = 0.070); and 179 vs. 184 (*P* = 0.010) for WI and SU in parities 1, 2, and ≥ 3. Milk yields (305d; kg) were 7,612 vs. 7,099 (*P* < 0.001); 8,011 vs. 7,665 (*P* < 0.001); and 7,814.6 vs. 7,532 (*P* < 0.001) for WI and SU in parities 1, 2, and ≥ 3. Milk yield by 70 DIM (kg) were 30.0 vs. 27.3 (*P* < 0.001); 35.5 vs. 33.1 (*P* < 0.001); and 36.8 vs. 35.2 (*P* < 0.001) for WI and SU in 1, 2, and ≥ 3 parity. In conclusion, cows that were conceived in the winter had better subsequent performance and survival than cows that were conceived in the summer.

**Key Words:** heat stress, conception, performance

**812 Effect of mild and moderate heat stress on milk yield and bovine milk composition in the tropical region.** Vivian Fischer<sup>\*1</sup>, Alexandre Sausenbach Abreu<sup>3</sup>, Marcelo Tempel Stumpf<sup>4</sup>, André Thaler Neto<sup>2</sup>, Daise Werneke<sup>1</sup>, and Fernando André Schmidt<sup>2</sup>, <sup>1</sup>Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil, <sup>2</sup>Universidade Estadual de Santa Catarina, Lages, SC, Brazil, <sup>3</sup>Universidade do Sul de Santa Catarina, Tubarão, SC, Brazil, <sup>4</sup>Universidade de Rio Grande, São Lourenço, RS, Brazil.

In hot climates, the combination of high temperature, humidity and intense solar radiation impose stress to the animals, especially if temperature is not sufficiently reduced during the night or if animals do not have protection against high temperatures or radiation, which is very frequent in the tropics. Variation in environmental variables between and within days affects the negative impact on dairy production. The present trials aimed to evaluate the effect of mild and moderate heat stress on dairy production, stability and physicochemical characteristics of milk. The temperature and humidity index (THI) was used to classify the heat stress as mild (72 to 78) or moderate (79–88). Two experiments were conducted in the tropical region, from January to March, on Tifton pasture. The first was conducted with 16 lactating Holstein cows, 520 ± 74 kg BW, BCS of 3.0 ± 0.3, MY of 21.5 ± 4.2 kg/day, 120 ± 61.2 DIM. The second trial was carried out with 14 lactating Holstein cows, 537.6 ± 93.5 kg of BW, BCS of 2.8 ± 0.3, MY of 20.6 ± 6.5 kg/day, 149.3 ± 48.9 DIM. In both trials the cows were allocated into 2 groups: paddocks without shade (NSH) and paddocks with natural shade (SH) - more than 10m<sup>2</sup>/head of shade. At 15:00 h, under mild stress, paddocks with shade and without shade presented THI values of 75.2 and 77.7, respectively. Under moderate stress, THI values in paddocks with and without shade were 77.7 and 83.8, respectively. Under mild stress, cows in NSH and SH did not present any differences in MY and composition. Under moderate stress, cows in NSH decreased MY by 50%, ethanol stability from 75% to 68%v/v, coagulation time from 190 s to 49 s, crude protein by 15%, lactose by 4% while they increased titratable acidity by 43%, fat by 13% and MUN by 12% when compared with cows in SH. After 1 week of regaining access to shade, all differences disappeared. Shade kept MY and milk composition within normal range under moderate stress. Under mild stress shade did not provide beneficial effects on milk yield and composition.

**Key Words:** heat stress, milk composition, severity of stress

**813 A cow cooling investment decision support tool for dairy farms in low and high humidity regions.** Bettie S. Kawonga<sup>\*</sup> and Jeffrey M. Bewley, *Department of Animal and Food Sciences, University of Kentucky, Lexington, KY.*

The objective of this study was to develop an interactive cow cooling investment decision support tool for dairy farms in low and high humidity regions, and to demonstrate the economic benefits of investing in cow cooling. The low humidity region in the study was specific to semi-arid areas of sub-Saharan Africa (e.g., Malawi and Zambia) with THI range of 68 to 72. The high humidity region represented countries in the temperate regions with a climate distinctly different from that of sub-Saharan Africa in terms of humidity and temperature. The user inputs included total herd size, lactating herd size, milk yield, feed required per kg of added milk, feed price per ton, and milk price. Cow cooling inputs included fan and soaker costs, water and electricity cost, annual number of days cooling system was used, annual interest rate, discount rate, tax rate, predicted daily increase in milk production due to cooling and predicted yearly increase in pregnancy rate for the herd. The investment analysis outputs included net present value (NPV) and benefit: cost ratio (BCR). Assumptions used in calculating NPV and BCR

included economic benefits of reduced heat stress extend to periods of low heat stress, no cooling existed, fans installed in stalls and holding area, and soakers installed in feed line and exit area from parlor. The assumptions were sourced from published literature and actual market prices of farm inputs and utilities. Using a herd size of 100 cows and a 10-year investment horizon, cow cooling in a high humidity region had a greater NPV (\$54,653) compared with a low humidity region (\$26,520). This could be attributed to high operating and initial investment cost for low humidity region. The BCR was greater than 1 for all the regions, indicating a good investment scenario. Further studies should be conducted to compare the current cow cooling investment analysis and other heat abatement strategies such as feed adjustment for small herd sizes in low humidity regions and semi-arid environments.

**Key Words:** cow cooling, net present value, cost:benefit ratio

**814 Effect of feeding phytogenic compounds on preweaned dairy calves in a commercial setting.** Bryan G. Miller<sup>\*1</sup> and Nicolas D. Bettencourt<sup>2</sup>, <sup>1</sup>Biomim USA, San Antonio, TX, <sup>2</sup>A.L. Gilbert, Oakdale, CA.

Feeding programs and feed ingredients that maximize calf growth rate add to a dairy's profitability through both reduced feed costs and lifetime days open. Phytogenic feed additives, derived from plant extracts, offer a potential means of supporting feed intake and calf growth. A field trial was conducted at a commercial "calf ranch" near Atwater California in the summer and fall of 2014. Calf growth was compared between those that consumed a standard calf starter with those that received the same diet with a phytogenic product, Digestrom Calf (Biomim Inc.) Holstein heifer calves were individually housed. To accommodate farm labor, calves were fed calf starter with or without Digestrom Calf as a group based upon calving date. Calves were fed a calf milk replacer (26% CP, 11% CF) and a calf starter (18% CP, 4.4% CF) either containing or not containing Digestrom Calf. Calves were fed between 72 and 78 d. Weight and height were measured at weaning. During the trial, morbidity, both in the form of scours and respiratory issues, was recorded for each set of calves. There were no apparent starting date effects and data from all groups were combined and a simple *t*-test for differences was conducted. Treatments for scours or respiratory diseases were analyzed using Chi-squared differences; results are in Table 1. Those calves supplemented with the phytogenic product, Digestrom, had greater average daily gain and hip height.

**Table 1 (Abstr. 814).** Growth and recorded health incidences among preweaned calves in a commercial setting

Treatment	Average		Initial wt (kg)	Final wt (kg)	ADG (kg)	Hip height (cm)	Scours (%)	Pneumonia (%)
	Calves (no.)	days fed (no.)						
Control	50	73.7	38.5	80.3 <sup>a</sup>	0.57 <sup>a</sup>	92.5 <sup>a</sup>	60.0 <sup>c</sup>	28.6 <sup>e</sup>
Digestrom	115	72.0	38.2	84.0 <sup>b</sup>	0.64 <sup>b</sup>	95.0 <sup>b</sup>	42.5 <sup>d</sup>	18.4 <sup>f</sup>

<sup>ab</sup>Means in the same column differ ( $P = 0.0002$ ).

<sup>cd</sup>Means in the same column did not differ ( $P = 0.1352$ ).

<sup>ef</sup>Means in the same column differ ( $P = 0.0732$ ).

**Key Words:** calves, phytogenic, growth

**815 Effect of condensed tannin extract supplementation on beef cattle performance and nitrogen balance: II. Finishing phase.** Pake J. Ebert<sup>\*1</sup>, Adam L. Shreck<sup>2</sup>, Jenny S. Jennings<sup>3</sup>, Noel A. Cole<sup>2</sup>, and Eric A. Bailey<sup>1</sup>, <sup>1</sup>West Texas A&M University, Canyon,

TX, <sup>2</sup>USDA-Agricultural Research Service, Bushland, TX, <sup>3</sup>Texas A&M AgriLife Research, Amarillo, TX.

Nitrogen emissions from concentrated animal feeding operations are of increasing concern to regulatory agencies. As such, we evaluated the effect of top-dressing a finishing diet (14.4% CP) for beef cattle with a commercially available condensed tannin extract (CT) at 3 levels (0, 0.5, and 1.0% of diet, DM basis). British-cross steers ( $n = 27$ ; initial BW =  $350 \pm 32$  kg) were fed individually via a Calan gate system for 126 d. Diet digestibility and N balance were estimated approximately 30 d after the experiment began (EARLY) and 30 d before the animals were harvested (LATE), using  $\text{TiO}_2$  as a marker of fecal output and creatinine:BW ratio as a marker for urine output, respectively. Inclusion of CT in the diet did not affect ( $P \geq 0.21$ ) ADG or DMI over the entire finishing period. Hot carcass weight was not different ( $P = 0.83$ ) among treatments, but fat thickness and LM area tended to decrease ( $P \leq 0.08$ ) when CT was included in the diet. Organic matter intake tended ( $P = 0.10$ ) to increase when CT was fed during EARLY. Apparent total-tract starch digestibility during EARLY was lesser ( $P = 0.03$ ) for 1% CT than either 0 or 0.5% CT. Intakes of OM and starch were similar ( $P \geq 0.31$ ) among treatments during LATE; similarly, apparent total-tract digestibility of OM and starch were similar ( $P \geq 0.31$ ) during LATE. Nitrogen intake did not differ ( $P \geq 0.11$ ) among treatments during EARLY and LATE, but fecal N excretion was greater ( $P = 0.05$ ) for 1.0% CT than 0% CT during EARLY. Urinary N excretion was not different ( $P \geq 0.43$ ) among treatments during EARLY and LATE, but urine N:total N excretion decreased when CT was included in the diet during EARLY. Retention of N was not different ( $P = 0.40$ ) among treatments during EARLY, but tended to decrease ( $P = 0.07$ ) when CT was included in the diet during LATE. Under the conditions of this experiment, supplementation of a finishing diet with condensed tannins had minor effects on performance and nutrient digestibility of beef steers fed a finishing diet, but did alter the site of N excretion.

**Key Words:** condensed tannin, feedlot, nitrogen balance

**816 Evaluation of an extended release anthelmintic on performance and fecal parasite load of beef cattle grazing cool season pastures.** J. D. Rivera\*, M. L. Gipson, R. G. Gipson, and P. J. Slusher, *Mississippi Agriculture and Forestry Exp. Station White Sand Branch Unit, Poplarville, MS.*

Seventy-four crossbred beef steers (average BW =  $239.2 + 10.7$  kg) were used in a randomized complete block to evaluate the use of an extended-release eprinomectin (ER) on performance and fecal worm count during grazing. Steers were stratified and blocked by BW into sixteen 1.2-ha pastures of annual ryegrass, and pasture was randomly assigned to one of 2 treatments: a single injection of an extended release eprinomectin (ER) or 2 injections (d 0 and d 64) of a typical anthelmintic used in stocker operations (doramectin; DOR). Steers were individually weighed and fecal samples collected on d 0 and 114. Steers in the DOR group were re-worked on d 64 for a second dose of DOR. Data were analyzed as a mixed model with block considered random and treatment considered fixed, LSMEANS were separated using PDIF. Pasture was the experimental unit. No differences ( $P = 0.36$ ) were noted in initial BW (238.9 vs. 239.6 kg for DOR and ER, respectively), however at final BW, steers treated with ER (366.4 kg) were heavier ( $P = 0.04$ ) than steers treated with DOR (357.8 kg). Moreover, steers treated with ER had greater ADG than DOR ( $P = 0.05$ ; 1.11 vs. 1.04 kg/d for ER and DOR, respectively). As a result, steers treated with ER had greater

gain per ha compared with steers administered DOR ( $P = 0.05$ ). Due to the greater cost of ER and despite reworking costs of DOR, overall working cattle cost was greater for ER compared with DOR ( $P = 0.009$ ; \$41.44/hd vs. \$39.83/hd for ER and DOR respectively). Total pasture cost per ha was greater ( $P = 0.009$ ) for ER (\$99.82/ha) compared with DOR (\$99.37/ha), however increased weight gain resulted in decreased cost of gain ( $P = 0.04$ ) for ER (\$1.13/kg) compared with DOR (\$1.19/kg). No differences among treatments ( $P = 0.66$ ) were noted for fecal egg count at d 0, nor were any differences detected for fecal egg count at d 114 ( $P = 0.32$ ), with a tendency ( $P = 0.13$ ) for decreased percentage of fecal egg count associated with ER (88.9%) when compared with DOR (76.1%). Despite the greater costs associated with ER and similar fecal egg control compared with DOR, ER resulted in greater performance and decreased cost of gain.

**Key Words:** beef cattle, anthelmintic, grazing performance

**817 Effect of yeast culture plus enzymatically hydrolyzed yeast supplementation during transition period on milk production and metabolic profile of dairy cows.** Claudia Faccio Demarco<sup>1</sup>, Vanessa Oliveira Freitas<sup>1</sup>, Tatiele Mumbach<sup>1</sup>, Eduardo Xavier<sup>2</sup>, Raquel Fraga e Silva Raimondo<sup>3</sup>, Fernanda Medeiros Gonçalves<sup>1</sup>, Francisco Augusto Del Pino<sup>1</sup>, Viviane Rohrig Rabassa\*<sup>1</sup>, Sangita Jalukar<sup>4</sup>, Marcio Nunes Corrêa<sup>1</sup>, and Cássio Cassal Brauner<sup>1</sup>, <sup>1</sup>Universidade Federal de Pelotas, NUPEEC, Pelotas, RS, Brazil, <sup>2</sup>Granjas 4 Irmãos S/A, Rio Grande, RS, Brazil, <sup>3</sup>Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brasil, <sup>4</sup>Arm & Hammer Animal Nutrition, Princeton, NJ.

The aim of this study was to evaluate the effect of yeast culture plus enzymatically hydrolyzed yeast on milk production and metabolic profile during transition period of dairy cows. Twenty-nine multiparous Holstein cows were blocked by milk production on the previous lactation and randomly assigned into 2 groups: one received 28 g/d top-dressed yeast culture plus enzymatically hydrolyzed yeast (YC-EHY; Celmanax, Arm & Hammer Animal Nutrition, Princeton, NJ), while the control group did not receive the supplement. The experiment was conducted from d -35 relative to calving until 150 of lactation. During this period cows were weighed weekly and their body condition score (BCS) evaluated. Cows were milked twice daily and were kept in a semi-extensive production system. Milk yield and milk composition were evaluated weekly from calving to 150 DIM. Plasma samples collected on -21, -14, -7, 0, 3, 7, 14, 21, 28, 35, and 42 d relative to calving were analyzed for  $\beta$ -hydroxybutyrate (BHBA), Nonesterified fatty acids (NEFA), cholesterol and plasma urea nitrogen (PUN). Data were analyzed using mixed models with repeated measures over time. The YC-EHY group had higher milk yield (27.75 vs.  $24.98 \pm 0.5$  kg/d,  $P = 0.05$ ) versus the control group. However, the control group had more milk protein concentration (2.97 vs.  $2.87 \pm 0.03\%$ /d,  $P = 0.03$ ) in comparison with YC-EHY group. Other milk compounds were not affected ( $P > 0.05$ ) by supplementation. No differences ( $P > 0.05$ ) were observed for BHBA, NEFA, cholesterol, and PUN concentrations during the transition period. Also body weight and BCS were not affected ( $P > 0.05$ ) by supplementation. In conclusion, yeast culture plus enzymatically hydrolyzed yeast supplementation increased milk production during the first half of lactation, however, metabolic profile are not affected by supplementation during the transition period in dairy cows.

**Key Words:** yeast, bovine, metabolism



# Production, Management and the Environment Symposium: Environmental footprint of livestock production—Greenhouse gas emissions and climate change

**818 Environmental footprint of livestock production: A global perspective.** Frank M. Mitloehner\*, *University of California, Davis, Davis, CA.*

Global livestock production is projected to double by 2050 and the majority of this growth will be occurring in the developing world. Much of the growth in the global livestock sector will occur in areas that are currently forested (i.e., parts of South America and South East Asia). It has been well established that significant reductions of carbon sequestering forests will have large effects on global climate change. Livestock production in most countries of the developed world (e.g., United States and Europe) has a relatively small greenhouse gas (GHG) contribution within the countries' overall carbon portfolios, dwarfed by large transportation, energy, and other industry sectors. In contrast, livestock production in the developing world can be a dominant contributor to a country's GHG portfolio, due to the developing world's significantly smaller transportation and energy sectors. The fact that land-use changes associated with livestock (i.e., forested land converted to pasture or cropland used for feed production) are a significant source of anthropogenic GHGs in Latin America and other parts of the developing world is apparent. The Food and Agriculture Organization attributes almost half of the climate-change impact associated with livestock to the change of land-use patterns. The United States and most other developed countries have not experienced significant land-use change practices around livestock production within the last few decades, sometimes centuries. Intensification of livestock production provides large opportunities for climate change mitigation and can reduce greenhouse gas emissions from deforestation, thus becoming a long-term solution to a more sustainable livestock production. Overall, growing demands for animal protein could strongly increase GHG emissions from agriculture. However, knowledge exists to improve efficiencies in livestock production, which dramatically reduces GHG per unit of production. What is called for is a sustainable intensification in animal agriculture, coupled with technology transfers from developed to developing countries, to supply a growing demand for animal protein using sustainable and modern production practices.

**Key Words:** environmental footprint, livestock

**819 Environmental impact reduction strategies for pig farms.** Richard Ulrich\*<sup>1</sup>, Greg Thoma<sup>1</sup>, Jennie Popp<sup>1</sup>, and Mark Hanigan<sup>2</sup>, <sup>1</sup>*University of Arkansas, Fayetteville, AR,* <sup>2</sup>*Virginia Tech, Blacksburg, VA.*

The purpose of this project was to determine what design and operational factors have a significant influence with minimal cost on the GHG, water and land usage impacts of a pork production facility. The Pig Production Environmental Footprint Calculator provides a detailed comparison of impacts as a function of animal feed practices, manure treatment strategies, barn heating/cooling settings and additives. The model utilizes the NRC nutrition equations to estimate feed intake and manure production as a function of barn temperature, animal gender, immunocastration, ractopamine use, and crowding. The model's economic code calculates the dollars per kg of avoided equivalent carbon dioxide emissions. The results confirm that feed production and manure management are the leading causes of environmental impacts from pig farms but differen-

tial improvements can come from other areas. Decreasing treatment temperatures can lessen GHG emissions from manure systems through managing barn and tank conditions. Digesters can recover energy from produced methane while converting the methane to lower-impact carbon dioxide. Barn temperatures have an impact on animal feed intake and subsequent growth rates. Common feeds are evaluated for their environmental impacts per calorie or gram protein provided.

**Key Words:** environmental impact, modeling, pig farms

**820 Quantifying greenhouse gas fluxes in animal production.** Wendy Powers\* and Matheus Capelari, *Michigan State University, East Lansing, MI.*

Direct and indirect sources of CO<sub>2</sub>, CH<sub>4</sub>, and N<sub>2</sub>O emissions in animal production systems includes the animals, feed storage areas, manure deposition and storage areas, and feed and forage production fields. These 3 gases comprise the primary greenhouse gas (GHG) emissions from animal feeding operations. Each GHG may be more or less prominent from each emitting source. Similarly, species dictates importance of enteric CH<sub>4</sub> emissions. Measures of GHG flux from animals are often made using respiration chambers for measurement of concentration and flux, head boxes or halters allowing for measures of concentration directly and flux indirectly (tracer gas techniques), or in vitro gas production techniques. Concentration measures are made using gas chromatography, photoacoustic, open path Fourier transform infrared (FTIR) or non-dispersive infrared (NDIR) spectroscopy. The prominent methods for measuring GHG emissions from housing include tracer gas techniques (indirect ventilation measures) or direct ventilation measures, each coupled with concentration measures of gases of interest. Methods for collecting and measuring GHG emissions from manure storage and/or production lots include use of downwind concentration measures combined with modeling techniques, static chambers or flux hood methods. Similar methods can be deployed for determining GHG emissions from fields. Each method identified has its own benefits and challenges to use for the stated application. Considerations for use include intended goal, compatibility with production system, equipment investment and maintenance, frequency and duration of sampling needed to achieve desired representativeness of emissions over time, accuracy and precision of the method, and environmental influences on the method. In the absence of a perfect method for all situations, full knowledge of the advantages and disadvantages of each method is extremely important during the development of the experimental design and interpretation of results. Attributes of the various options for measuring GHG flux from different sources within a farming system will be discussed including methods to measure both concentration of gas and flux from the various emission sources.

**Key Words:** emissions, flux, greenhouse gas

**821 Greenhouse gas emissions and nitrogen cycling from beef production systems: Effects of climate, season, production system, and diet.** Galen E. Erickson\*, Samodha C. Fernando, Terry J. Klopfenstein, Andrea K. Watson, James C. MacDonald, Anna C.

Pesta, Allison L. Knoell, and Henry Paz, *University of Nebraska-Lincoln, Lincoln, NE*.

Greenhouse gas (GHG) and nitrogen emissions from beef cattle production systems are receiving greater attention. Emissions of methane and nitrous oxide in grazing and confined feedlot systems are microbial-mediated, whether from the rumen (methane) or soil surfaces (methane and nitrous oxide). Additionally, emissions of ammonia are microbial-mediated as well as dependent on ammonium concentration. Factors that increase microbial activity, such as temperature and season, are positively correlated with emissions from soil surfaces, whereas enteric methane emissions are not affected by ambient temperature and therefore not affected by climate or season. Previous research illustrates that diet, season, and type of production system dramatically affect N emissions as ammonia. However, the effect of season, diet, and type of production system on nitrous oxide emissions from cattle production systems and soils are poorly understood. Additionally, methane emissions data from production systems are lacking and needed. Enteric methane emissions have received the greatest attention across a wide array of climates, season, and diets. Forage quality has been shown to have a large impact on methane emissions, with lower quality forage increasing methane per unit of energy intake. Likewise, feeding finishing diets results in less enteric methane per unit of energy intake compared with forages. However, both high quality forages, and finishing diets increase energy intake and thus absolute amount of methane produced per day. Expressing enteric emissions as amount per day will lead to different mitigation strategies compared with decreasing methane emissions per unit of energy intake or per unit of productive function (i.e., gain). Many benefits exist to utilize forages in beef production systems, thus converting existing forage-based systems to intensive (i.e., grain-based) beef production as a mitigation strategy for methane emissions is not logical. However, greater opportunity likely exists to mitigate methane emissions within forage-based production systems as compared with grain-based systems, which will require understanding microbial mediated processes to dramatically decrease enteric methane.

**Key Words:** emissions, methane, nitrogen

**822 Forage utilization to mitigate greenhouse gas emissions by ruminants.** Karen A. Beauchemin\*, *Agriculture and Agri-Food Canada, Lethbridge Research Centre, Lethbridge, AB, Canada*.

Meat and dairy products account for almost half of food-generated greenhouse gases (GHG), and as global consumption of livestock products continues to grow there is pressure on the livestock industry to lower its emissions. Poultry, pig, dairy, and feedlot production rely on the use of grains and oilseeds, increasing the demand for limited resources. With continued expansion of livestock production to meet global demand for protein, ruminant production will need to increase its use of forages. Unlike pigs and poultry, ruminants can utilize cellulosic materials to produce high quality protein for human consumption. Furthermore, perennial forages, forage-cropping rotations, and well-managed grazing lands provide numerous environmental benefits. However, high-fiber diets increase enteric CH<sub>4</sub> emissions from ruminants, so increasing forage use by ruminants may at first seem counterproductive in terms of reducing GHG emissions. However, there is increasing knowledge of mitigation practices that lower CH<sub>4</sub> emissions from cattle. Some strategies are (1) technologies to reduce methanogenesis in the rumen (nitrate, inhibitors), (2) improving fiber digestibility (pre-treatment, genetic selection of forages, harvest management), and (3) targeted supplementation and management of animals to improve productivity. When examining the total GHG emissions from a system, all emissions and removals of CH<sub>4</sub>, N<sub>2</sub>O, and CO<sub>2</sub> on the farm and from purchased inputs must be considered using a life cycle approach. Forage-based ruminant systems tend to be relatively low input with less fossil fuel CO<sub>2</sub> emissions and fertilizer-based N<sub>2</sub>O emissions than grain-based systems. Use of N-fixing legume forages further displaces the use of fertilizers. With grazing systems, the nutrient cycle is relatively closed with excreted nutrients returned directly to the land. Well managed grasslands subject to moderate grazing can augment soil carbon reserves. Such lands also provide many other ecosystem services including conservation of biodiversity, water quality, and wildlife habitat. Thus, continued development and adoption of mitigation strategies will allow ruminant production systems to rely more extensively on forages while lowering GHG emissions and providing enhanced ecosystem services.

**Key Words:** forage, methane, environmental sustainability

# Ruminant Nutrition: Modifying rumen microbial populations

**823 Protozoa reduction in the rumen of grazing cattle fed mixtures of coconut and palm kernel oils.** Oludotun O. Adelusi, Adebayo O. Oni\*, Oluwaseun J. Idowu, Victoria O. Ojo, and Christopher F. Onwuka, *Federal University of Agriculture, Abeokuta, Ogun State, Nigeria.*

A 12-wk study was conducted to investigate the effect of feeding ratios of coconut and palm kernel oils (CNO-PKO) on the rumen microbial population of grazing cattle. The experiment was carried out at Cattle Production Venture Farm, Federal University of Agriculture Abeokuta, Ogun State, Nigeria. Twelve (12) White Fulani cattle with average weight of  $164 \pm 2.81$  kg were allotted into 4 treatments of CNO-PKO administration (0 g/day (control), 100:50 g/day (HCLP), 50:100 g/day (LCHP) and 75:75 g/day (ECP) in a completely randomized design. Rumen fluid samples were obtained from each cattle after oil administration period and microbial population was determined by total direct count of protozoa and fungal zoospores while bacteria analysis was carried out using anaerogen packs. Data were analyzed using one-way ANOVA (SPSS Statistics 20). The results showed that rumen bacteria in the genus *Bacteroides* spp. was not significantly ( $P > 0.05$ ) affected by the treatments. *Clostridium* spp. count of those maintained in the control group recorded the greatest ( $P < 0.05$ ) number with  $6.08 \times 10^6$  cfu/mL while the least ( $P < 0.05$ ) was recorded from cattle maintained on HCLP administration with  $2.35 \times 10^6$  cfu/mL. *Lactobacillus* spp. count of cattle maintained in the control group and those on ECP administration recorded greater ( $P < 0.05$ ) counts of  $0.20 \times 10^6$  cfu/mL and  $0.15 \times 10^6$  cfu/mL respectively while lesser ( $P < 0.05$ ) counts were obtained from cattle maintained on HCLP and LCHP with both recording  $0.05 \times 10^6$  cfu/mL. Rumen fungi count increased ( $P < 0.05$ ) with LCHP administration recording  $0.60 \times 10^6$  cfu/mL while the least ( $P < 0.05$ ) was recorded with HCLP administration ( $0.10 \times 10^6$  cfu/mL). Rumen protozoa population of cattle was significantly ( $P < 0.05$ ) reduced with the greatest ( $P < 0.05$ ) count obtained from cattle maintained in the control group recording 755.99 FEC/g while the least ( $P < 0.05$ ) was obtained from cattle maintained on ECP administration recording 0.00 FEC/g. Feeding 75 g/d each of coconut and palm kernel oils to grazing cattle reduced the protozoa population of the rumen and might therefore be employed as rumen modifier for methane mitigation.

**Key Words:** cattle, rumen, protozoa

**824 Total-tract fatty acid digestibility responses to increasing levels of palmitic acid supplementation of dairy cows receiving low- and high-fat diets.** Jonas De Souza\*, J. Eduardo Rico, Courtney L. Preseault, Michael S. Allen, and Adam L. Lock, *Michigan State University, East Lansing, MI.*

Dose-dependent effects of a palmitic acid-enriched fat supplement (PA; 87% C16:0; Bergafat F-100) on total-tract digestibility responses of dairy cows were evaluated. Low and high basal dietary fatty acid (FA) diets (LF: 2.2% DM and HF: 3.5% DM) were used as a split-plot to determine relationships between basal dietary FA content and PA dose. Sixteen Holstein cows ( $149 \pm 56$  DIM) were assigned randomly to treatment sequence within basal FA groups. PA was supplemented at 0, 0.75, 1.50, or 2.25% of ration DM in a  $4 \times 4$  Latin Square design within each basal FA group. Periods were 14 d with the final 4 d used for data collection. FA content of LF and HF diets was achieved by altering the proportion of soyhulls and cottonseed in diets. The statistical model included the random effect of cow and the fixed effects of basal FA group, PA dose,

period, and their interactions. Linear, quadratic, and cubic contrasts were used to determine effects of PA dose. Compared with HF (with cottonseed), LF diets (with soyhulls) increased NDF digestibility (50 vs. 48%;  $P < 0.01$ ). PA dose also increased NDF digestibility (46, 50, 51, and 52%; linear  $P < 0.01$ ). There was a tendency for an interaction of treatments ( $P = 0.12$ ) as NDF digestibility increased more for HF with increasing PA than for LF. Compared with HF, LF diets decreased 16-carbon FA digestibility (65 vs. 71%  $P < 0.01$ ) and tended to increase 18-carbon FA digestibility (85 vs. 82%;  $P = 0.07$ ). PA dose decreased 16-carbon FA digestibility (76, 67, 64 and 64%; quadratic  $P < 0.01$ ) and increased 18-carbon FA digestibility (82, 83, 85 and 85%; linear  $P < 0.05$ ) for 0, 0.75, 1.50, and 2.25% PA, respectively. PA dose linearly decreased the digestibility of total FA in LF diets (81, 76, 73, and 71%) but did not in HF diets (77, 76, 76, and 76%; interaction  $P < 0.05$ ) for 0, 0.75, 1.50, and 2.25% PA, respectively. HF diets increased total FA absorbed compared with LF diets (0.94 vs. 0.71 kg/d;  $P < 0.05$ ). Additionally, PA dose increased total FA absorbed (0.64, 0.77, 0.90 and 0.97 kg/d; linear  $P < 0.01$ ) for 0, 0.75, 1.50, and 2.25% PA, respectively. In conclusion, the total-tract FA digestibility responses to PA dose were affected by the FA content of the basal diet.

**Key Words:** dietary fat, digestibility, palmitic acid

**825 Daily patterns of hydrogen and volatile fatty acid concentrations in relation to thermodynamic control on fermentation in the bovine rumen.** Hendrikus J. van Lingen\*<sup>1,2</sup>, Jueeli D. Vaidya<sup>1,3</sup>, Sanne van Gastelen<sup>1,2</sup>, Bartholomeus van den Bogert<sup>1,3</sup>, André Bannink<sup>4</sup>, Caroline M. Plugge<sup>3</sup>, Hauke Smidt<sup>3</sup>, and Jan Dijkstra<sup>2</sup>, <sup>1</sup>*Top Institute Food and Nutrition, Wageningen, Gelderland, the Netherlands*, <sup>2</sup>*Animal Nutrition Group, Wageningen University, Wageningen, Gelderland, the Netherlands*, <sup>3</sup>*Laboratory of Microbiology, Wageningen University, Wageningen, Gelderland, the Netherlands*, <sup>4</sup>*Animal Nutrition, Wageningen UR Livestock Research, Wageningen, Gelderland, the Netherlands.*

Elevated levels of both hydrogen and molar proportions of propionate to acetate and butyrate are found in the bovine rumen right after meals. Hydrogen is believed to thermodynamically control fermentation pathways in the rumen in favor of propionate production. Elevated levels of hydrogen inhibit reoxidation of NADH to NAD<sup>+</sup>. Propionate production, however, enables this reoxidation and explains why elevated levels of hydrogen and propionate are hypothesized to occur simultaneously. Nonetheless, hydrogen partial pressure in the rumen headspace may not be high enough to inhibit NADH reoxidation. Furthermore, studies reporting diurnal patterns of volatile fatty acids (VFA) concentration and hydrogen pressure simultaneously, if any, are limited. The aim of this study is to monitor daily patterns of hydrogen pressure, pH, and VFA concentration in the bovine rumen and to calculate thermodynamic inhibition of specific fermentation pathways. Four rumen fistulated multiparous lactating cows were used in a crossover design with 2 17-d experimental periods and a control and treatment diet. Both diets consisted of 40% corn silage, 30% grass silage and 30% concentrates on DM basis. The treatment diet had a 2.5% higher fat content by supplementing the concentrate with linseed oil. On d 11, rumen headspace gas and fluid samples were taken at 0, 0.5, 1, 1.5, 2, 3, 4, 5, 6, 8, 10 h after morning feeding using a custom fistula lid enabling rumen gas and fluid sampling. Gas samples were analyzed for hydrogen pressure, and fluid samples for pH and concentrations of dissolved hydrogen as well as VFA. Fluid samples were also taken for transcriptome analysis



to monitor microbiome gene expression. From d 13 to 17 cows were housed in respiration chambers to relate rumen headspace pressure to emissions of hydrogen. An increase of hydrogen partial pressure by 2 orders of magnitude up to 30 mbar was observed following feeding. This finding might indicate hydrogen partial pressure to be high enough to inhibit NADH reoxidation.

**Key Words:** rumen fermentation, thermodynamics, hydrogen

**826 Effects of chitosan on ruminal metabolism and in situ degradability of beef cattle.** Darren D. Henry\*, Francine M. Ciriaco, Vitor R. G. Mercadante, Tessa M. Schulmeister, Martin Ruiz-Moreno, G. Cliff Lamb, and Nicolas DiLorenzo, *North Florida Research and Education Center, University of Florida, Marianna, FL.*

We determined the effects of supplementing chitosan, a natural biopolymer, on ruminal metabolism and in situ degradability in beef steers consuming hay. Eight ruminally cannulated crossbred steers ( $345 \pm 81$  kg BW) were used in a crossover design. Steers were stratified by weight and randomly assigned to 1 of 2 treatments: control (CTRL; no chitosan supplementation) or treatment (TRT; 80 g/d of chitosan). Chitosan was dosed daily via ruminal cannula and steers had ad libitum access to Tifton 85 bermudagrass hay and water throughout the study. Ruminal fluid was collected before dosing of chitosan (0 h) and every 3 h post-dosing for 24 h. Immediately after each collection, ruminal pH was measured. Ruminal fluid was analyzed for VFA and  $\text{NH}_3\text{-N}$  concentrations. Data were analyzed as repeated measures and the model included the fixed effects of treatment, time, and treatment  $\times$  time interactions. In situ degradability of DM, NDF, and ADF was determined by incubating Tifton 85 bermudagrass hay in nylon bags within the rumen for 24, 48 or 72 h. Supplementing with 80 g/d chitosan had no effect ( $P > 0.05$ ) on ruminal in situ degradability of DM, NDF or ADF. Chitosan did not affect ( $P = 0.97$ )  $\text{NH}_3\text{-N}$  concentrations ( $7.4 \pm 0.58$  mM) within the rumen. A treatment  $\times$  time interaction was observed ( $P < 0.001$ ) on ruminal pH. A treatment  $\times$  time interaction ( $P = 0.016$ ) was observed for acetate-to-propionate ratio (A:P), being decreased ( $P < 0.05$ ) at h 21 and 24 for TRT vs. CTRL. Molar proportions of acetate and propionate had a treatment  $\times$  time interaction ( $P < 0.05$ ). Molar proportions of propionate were increased at h 18, 21 and 24 for TRT when compared with CTRL. Supplementing 80 g/d of chitosan to steers consuming ad libitum hay had no effect on in situ ruminal degradability; however, VFA molar proportions were shifted to an energetically more efficient composition by increasing propionate and decreasing A:P.

**Key Words:** chitosan, ruminal metabolism, in situ degradability

**827 Ellipsoid equation improves accuracy and efficiency of estimating protozoal volume.** Benjamin A. Wenner\*, Brooklyn K. Wagner, and Jeffrey L. Firkins, *Department of Animal Sciences, The Ohio State University, Columbus, OH.*

Previous observations of protozoa in cultures treated with essential oils or ionophores indicated possible cell shrinkage due to deleterious effects on cell function. Cell volume reduction by formaldehyde preservation, combined with visually flattened or tapered morphology of rumen protozoa limited our ability to detect volume differences using common cylindrical derivations for protozoa. The advent of affordable, high definition imaging equipment enables recording of live protozoa from cultures treated with various additives that potentially shrink cells. We hypothesized that using still frames from video of protozoa swimming would improve accuracy of volume predictions by optimizing an approach to measure one maximal longitudinal measurement and

both minimum and maximum diameter measurements perpendicular to the longitudinal axis, thus yielding a 3-dimensional estimation of protozoal volume. An ellipsoid formula ( $E, \frac{4}{3}\pi abc$ ) was compared with previously published estimations using cylindrical [ $C, \pi(\frac{W}{2})^2$ ] or species coefficient (SP,  $XLW^2$ ) calculations. Testing this method on objects shaped similarly to protozoa demonstrated the ellipsoid is more accurate in predicting volume as measured by displacement. True displacement was 11.8 mL for 10 large particles, and estimated volumes were 12.7 to 27.1, 7.7 to 16.6, and 12.3 mL for C, SP, and E, respectively. For smaller particles with more surface area, true displacement was 4.5 mL, and estimated volumes were 5.6 to 13.1, 3.2 to 7.5, and 5.6 for C, SP, and E, respectively. Ruminal fluid sampled from 2 lactating Jersey cows was flocculated and wet-mounted on a microscope fitted with an HD (1080p) camera. Residuals (SP - E) were plotted against predicted (E) centered to the mean ( $X - \text{mean}$ ) to evaluate for both mean and slope bias. For entodinia (ENTO),  $Y = 1.97 \times 10^4 (\pm 1.48 \times 10^3) + 0.248(\pm 0.0371)(X - 7.98 \times 10^4) \mu\text{m}^3$ , with significant slope ( $P < 0.01$ ) and mean ( $P < 0.01$ ) bias. For isotrichids (ISO),  $Y = -1.21 \times 10^4 (\pm 4.86 \times 10^4) - 0.124(\pm 0.0685)(X - 2.54 \times 10^6) \mu\text{m}^3$ , where slope trended toward significance ( $P = 0.08$ ) with no mean ( $P > 0.10$ ) bias. For epidinia (EPI),  $Y = 1.02 \times 10^5 (\pm 1.46 \times 10^4) + 0.372(\pm 0.219)(X - 1.45 \times 10^5) \mu\text{m}^3$ , with no slope ( $P > 0.10$ ) and significant mean ( $P < 0.01$ ) bias. This demonstrates that SP more likely overestimates volume for ENTO or EPI than for teardrop-shaped ISO. This ellipsoid method offers potential to advance prediction of treatment effects on protozoal viability and volume.

**Key Words:** protozoa, imaging, rumen

**828 Effect of monensin inclusion on ruminal fermentation parameters in *Bos indicus* and *Bos taurus* steers consuming bermudagrass hay.** Natasha L. Bell<sup>\*1,2</sup>, Robin C. Anderson<sup>3</sup>, Todd R. Callaway<sup>3</sup>, Marcia O. Franco<sup>4</sup>, and Tryon A. Wickersham<sup>1</sup>, <sup>1</sup>Texas A&M University, College Station, TX, <sup>2</sup>Texas A&M University-Kingsville, Kingsville, TX, <sup>3</sup>Southern Plains Agricultural Research Center, Agricultural Research Service, USDA, College Station, TX, <sup>4</sup>Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.

Effects of monensin inclusion and the subspecies of cattle on utilization of bermudagrass hay (13.7% CP) were evaluated using ruminally cannulated steers (5 *Bos indicus*, BI and 5 *Bos taurus*, BT; 398 kg BW). Subspecies were concurrently subjected to a 2 period, 2 treatment crossover design. Treatments consisted of 0 (CON) or 200 (MON) mg·hd<sup>-1</sup> monensin (Rumensin 90; Elanco Animal Health, Greenfield, IN) fed daily in 0.91 kg DDGS. Steers were group housed during adaptation periods and moved to individual covered pens to facilitate sampling. Periods were 70 d in length allowing 42 d adaptation, and 28 d withdrawal between periods. Ruminal fluid was collected with a suction-strainer 0, 2, 4, 8, and 12 h after feeding on d 42 for analysis of pH, VFA, and ruminal ammonia-N. Ruminal contents were squeezed through 4 layers of cheesecloth into insulated containers 2 h after feeding on d 42 for determination of rate of ammonia production and CH<sub>4</sub>-producing activity. No subspecies  $\times$  treatment interactions were observed ( $P \geq 0.14$ ). There was an effect of time after feeding ( $P \leq 0.01$ ) on pH, ruminal ammonia-N, total VFA, acetate:propionate, and molar percentages of acetate and propionate. Total VFA concentration was greater ( $P = 0.01$ ) in CON vs. MON steers (66.5 vs. 62.0 mM). Total VFA concentration was similar ( $P = 0.28$ ) for BI and BT subspecies (62.7 and 65.8 mM, respectively). Monensin decreased the molar percentage of acetate ( $P = 0.02$ ) from 72.5% to 71.2% and increased the molar percentage of propionate ( $P < 0.01$ ) from 16.9% to 18.7%, reducing ( $P < 0.01$ ) the acetate:propionate ratio from 4.34 to 3.85. Although CH<sub>4</sub>-producing

activity was not significantly different ( $P = 0.19$ ) between CON and MON, monensin feeding resulted in a 15.8% reduction in  $\text{CH}_4$ -producing activity. *Bos indicus* steers had greater ( $P = 0.07$ )  $\text{CH}_4$ -producing activity than BT steers ( $21.37$  vs.  $16.62 \mu\text{mol CH}_4 \cdot \text{mL}^{-1} \cdot \text{h}^{-1}$ ). Monensin had no effect ( $P \geq 0.19$ ) on pH, ruminal ammonia-N, or rate of ammonia production. Overall, monensin decreased the acetate:propionate ratio by decreasing acetate and increasing propionate and numerically reduced  $\text{CH}_4$ -producing activity. *Bos indicus* had greater  $\text{CH}_4$ -producing activity compared with BT steers.

**Key Words:** cattle subspecies, ionophore, VFA

**829 Effect of monensin withdrawal on ruminal fermentation parameters in *Bos indicus* and *Bos taurus* steers consuming bermudagrass hay.** Natasha L. Bell<sup>1,2</sup>, Robin C. Anderson<sup>3</sup>, Todd R. Callaway<sup>3</sup>, Marcia O. Franco<sup>4</sup>, and Tryon A. Wickersham<sup>1</sup>, <sup>1</sup>Texas A&M University, College Station, TX, <sup>2</sup>Texas A&M University-Kingsville, Kingsville, TX, <sup>3</sup>Southern Plains Agricultural Research Center, Agricultural Research Service, USDA, College Station, TX, <sup>4</sup>Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil.

Effects of monensin withdrawal and subspecies of cattle on the utilization of bermudagrass hay (13.7% CP) were evaluated using ruminally cannulated steers (5 *Bos indicus*, BI and 5 *Bos taurus*, BT; 398 kg BW). Subspecies were concurrently subjected to a 2 period, 2 treatment cross-over design. Treatments were withdrawal from either 0 (CON) or 200  $\text{mg} \cdot \text{hd}^{-1} \cdot \text{d}^{-1}$  (MON) monensin (Rumensin 90; Elanco Animal Health, Greenfield, IN) in 0.91 kg DDGS, fed for 42 d. Withdrawal was evaluated for a 28 d period. Rumen fluid was collected 2 h after feeding on d 1, 4, 7, 14, and 21 after the cessation of MON feeding for determination of pH, VFA, ruminal ammonia-N, rate of ammonia production and methane-producing activity. No subspecies  $\times$  treatment or subspecies  $\times$  treatment  $\times$  day interactions were observed ( $P \geq 0.16$ ). Treatment  $\times$  day interactions ( $P \leq 0.01$ ) were observed for acetate:propionate ratio and molar percentages of acetate and propionate. There was a marked decrease in molar % propionate between d 1 and 4 from 19.1 to 18.0%; however, molar % propionate remained greater ( $P \leq 0.09$ ) for MON than CON through d 7. Molar percentage of acetate increased with MON withdrawal from 68.8 to 69.8 between d 0 and 4. Acetate:propionate ratio was lower ( $P \leq 0.01$ ) on d 0 for MON than CON (3.4 vs. 4.0). By d 4, MON had increased to 3.8, and was not different ( $P = 0.14$ ) from CON. By d 14, no differences ( $P \geq 0.88$ ) between MON and CON remained for acetate, propionate, and acetate:propionate ratio. Steers previously fed monensin had similar ( $P \geq 0.12$ ) total VFA concentrations by d 4. Greater ( $P < 0.01$ ) ruminal ammonia-N concentrations (1.88 vs. 1.73 mM) were observed in CON than MON steers. Monensin had no effect ( $P \geq 0.69$ ) on rate of ammonia production or methane-producing activity. A treatment  $\times$  day interaction occurred ( $P \leq 0.05$ ) for pH with MON steers having higher pH than CON on d 1, 7, and 14. *Bos indicus* steers tended to have higher ( $P = 0.08$ ) pH than BT (6.53 vs 6.46); no other subspecies effects were observed. Results indicate that the monensin induced reduction in acetate:propionate ratio persists for at least 7 d post-withdrawal.

**Key Words:** cattle subspecies, ionophore, VFA

**830 Effects of solid feed level and roughage-to-concentrate ratio on ruminal drinking and passage kinetics of milk, concentrates, and roughage in veal calves.** Harma Berends<sup>1</sup>, Joost van den Borne<sup>1</sup>, Norbert Stockhofe<sup>2</sup>, Myrthe Gilbert<sup>1</sup>, Tamme Zandstra<sup>1</sup>,

Wilbert Pellikaan<sup>1</sup>, Kees van Reenen<sup>3</sup>, Eddie Bokkers<sup>4</sup>, and Walter Gerrits<sup>1</sup>, <sup>1</sup>Animal Nutrition Group, Wageningen University, Wageningen, Gelderland, the Netherlands, <sup>2</sup>Central Veterinary Institute, Lelystad, Flevoland, the Netherlands, <sup>3</sup>Livestock Research, Wageningen, Gelderland, the Netherlands, <sup>4</sup>Animal Production Systems Group, Wageningen University, Wageningen, Gelderland, the Netherlands.

Effects of solid feed (SF) level and roughage-to-concentrate (R:C) ratio on ruminal drinking and passage kinetics of milk, concentrate, and roughage in veal calves were studied. Eighty calves (2 wk of age, 45 kg bodyweight) were divided over 16 pens. Pens were randomly assigned to a low (LSF) or a high (HSF) SF level, and to one of 2 R:C ratios; 20:80 or 50:50 on a DM basis. Roughage was composed of 50% corn silage and 50% chopped straw on a DM basis. During the measurement period at 27 wk of age, SF intake was 1.2 kg DM/d for LSF and 3.0 kg DM/d for HSF, and milk (replacer) intake averaged 2.3 kg DM/d for LSF and 1.3 kg DM/d for HSF. To estimate passage kinetics of milk, concentrate, and straw, indigestible markers (respectively CoEDTA, hexatriacontane  $\text{C}_{36}$ , Cr-NDF) were supplied with the feed as a single dose at respectively 4, 24, and 48 h before slaughter. At slaughter, marker recovery was quantified in the rumen, abomasum, small intestine, and large intestine. Rumen Co recovery averaged 20% of the last milk meal. Recoveries of milk remained largely unaffected by SF level and R:C ratio. Ruminal recovery of  $\text{C}_{36}$  and Cr was unaffected by R:C ratio. Rumen fractional passage rate of concentrate was estimated from recovery of  $\text{C}_{36}$  in the rumen and increased ( $P < 0.001$ ) from 3.3%/h for LSF to 4.9%/h for HSF. Rumen fractional passage rate of straw was estimated from Cr recovery in the rumen and increased ( $P < 0.01$ ) from 1.3%/h for LSF to 1.7%/h for HSF. A greater SF level increased ( $P < 0.001$ ) fresh and dry rumen contents. In HSF calves, pH decreased (from 6.9 to 6.0;  $P < 0.01$ ) and VFA concentrations increased ( $P < 0.05$ ) with a lower R:C ratio, indicating increased fermentation. The Cr: $\text{C}_{36}$  ratio was similar in the small and large intestine, indicating that passage of concentrate and straw was mainly determined by rumen and abomasum emptying. In conclusion, SF level rather than R:C ratio influences rumen passage of concentrate and roughage. Our data provide insight in passage kinetics of milk and SF and may contribute to the development of feed evaluation models for veal calves.

**Key Words:** fractional passage rate, rumen retention time, marker recovery

**831 Effect of dietary supplementation with resveratrol on nutrient digestibility, methanogenesis and ruminal microbial flora in sheep.** Tao Ma<sup>1</sup>, Dan-dan Chen<sup>1</sup>, Yan Tu<sup>1</sup>, Nai-feng Zhang<sup>1</sup>, Bing-wen Si<sup>1</sup>, Kai-dong Deng<sup>2</sup>, and Qi-yu Diao<sup>1\*</sup>, <sup>1</sup>Feed Research Institute, Chinese Academy of Agricultural Sciences, Key Laboratory of Feed Biotechnology of the Ministry of Agriculture, Beijing, China, <sup>2</sup>College of Animal Science, Jinling Institute of Technology, Nanjing, Jiangsu, China.

Two experiments were conducted to evaluate the effect of resveratrol on methanogenesis and microbial flora in Dorper  $\times$  thin-tailed Han crossbred ewes. In experiment 1, Eighteen ewes ( $60.0 \pm 1.73$  kg BW) were assigned to 2 dietary treatments, a basal diet and a basal diet supplemented with resveratrol (0.25 g/head·d), to investigate the effect of resveratrol on nutrient digestibility and nitrogen balance. In experiment 2, 6 ewes ( $64.0 \pm 1.85$  kg BW) with ruminal cannulas were assigned to the identical dietary treatments used in experiment 1 according to change over design, to investigate supplementary resveratrol on ruminal fermentation and microbial flora using q-PCR. The digestibility and nitrogen balance data were analyzed using *t*-test procedures in SAS. Data referring to ruminal fermentation parameters (ruminal pH, ammonia,

and volatile fatty acid) and microbial flora measured at each sampling time were analyzed using a repeated measures one-way ANOVA. Significant differences were accepted when  $P < 0.05$ . The results showed that supplementary resveratrol improved the digestibility of organic matter ( $P < 0.001$ ), nitrogen ( $P = 0.007$ ), neutral detergent fiber ( $P < 0.001$ ), and acid detergent fiber ( $P < 0.001$ ). The excretion of fecal N was reduced ( $P = 0.007$ ), whereas that of urinary N increased ( $P = 0.002$ ), which led to an unchanged N retention ( $P = 0.157$ ). Both  $\text{CO}_2$  and  $\text{CH}_4$  output scaled to digestible dry matter intake decreased from 602.5 to 518.7 ( $P = 0.039$ ) and 68.2 to 56.6 ( $P < 0.001$ ), respectively. Ruminal pH ( $P = 0.341$ ), ammonia ( $P = 0.512$ ), and total volatile fatty acid ( $P = 0.249$ ) were unaffected by resveratrol. The molar proportion of propionate increased from 13.1 to 17.5% ( $P < 0.001$ ) while that of butyrate decreased from 11.0 to 9.55% ( $P < 0.001$ ). The ratio of acetate to propionate decreased from 5.44 to 3.96 ( $P < 0.001$ ). Supplementary resveratrol increased ruminal population of *F. succinogenes*, *R. albus*, and *B. fibrisolvens* ( $P < 0.001$ ) while decreased protozoa and methanogens ( $P < 0.001$ ). In conclusion, dietary resveratrol inhibited methanogenesis without adversely affecting ruminal fermentation.

**Key Words:**  $\text{CH}_4$ , resveratrol, sheep

**832 Essential oils from goat weed (*Ageratum conyzoides*) and African basil (*Ocimum gratissimum*) can reduce in vitro enteric methane production.** Musibau A. Bamikole<sup>1,2</sup>, Ibukun M. Ogunade\*<sup>1</sup>, Felipe Amaro<sup>1</sup>, Yun Jiang<sup>1</sup>, Thiago F. Bernardes<sup>1</sup>, Vania R. Vasconcelos<sup>1</sup>, Darren D. Henry<sup>3</sup>, F. O. Ugiagbe<sup>2</sup>, U. J. Ikhatua<sup>2</sup>, Nicolas DiLorenzo<sup>3</sup>, and Adegbola T. Adesogan<sup>1</sup>, <sup>1</sup>University of Florida, Gainesville, FL, <sup>2</sup>University of Benin, Benin City, Nigeria, <sup>3</sup>North Florida Research and Education Center, University of Florida, Marianna, FL.

The potential of essential oils (EO) from leaves of goat weed (EOG) and African basil (EOB) to reduce in vitro enteric methane production and improve rumen fermentation was examined. A corn silage-based TMR (0.5 g; CP 16.6%; NDF 35.9%) was treated with EOG or EOB at rates of 0 (Control), 10 (Low), 20 (Med) and 30 (High)  $\mu\text{L}/50$  mL of rumen fluid - buffer inoculum (ratio 1:2) and with monensin (0.6 mg/50 mL). Each suspension was incubated in a 120-mL gas-tight culture bottle in triplicate at 39°C for 24 h in each of 2 runs. Fermentation parameters, gas and methane production, in vitro DM digestibility (DMD), and fermentation efficiency (DMD  $\text{g kg}^{-1}/\text{gas volume}$ ) were measured. Data for each EO were separately analyzed with the Glimmix procedure of SAS. Compared with respective Controls, monensin and High EOG or EOB reduced ( $P < 0.05$ ) gas volume (66.8, 71.7 and 49.3 vs. 85.0), DMD (526, 520, and 514 vs. 555 g/kg) and methane percentage in total gas (7.95, 9.02 and 7.19 vs. 10.4%) and increased ( $P < 0.05$ ) fermentation efficiency (7.92, 7.39, and 11.58 vs. 6.58). The respective reductions in methane production (mg/g DM digested) were 36.9, 24.1, 57.2%. Ammonia nitrogen concentration, pH of EOB and VFA concentrations of EOG were unaffected ( $P > 0.05$ ) by treatment. However, monensin and High EOG increased ( $P < 0.05$ ) the pH (5.75, 5.75 vs. 5.66, respectively) and monensin and High EOB increased ( $P < 0.05$ ) molar proportions of butyrate and High EOB decreased ( $P < 0.05$ ) that of propionate. Low rates of EOB and EOG increased ( $P < 0.05$ )  $\text{CH}_4$  production (mg/g DM digested) and Med rates decreased ( $P < 0.05$ ) DMD. Low EOG increased ( $P < 0.05$ ) gas volume and methane production (mg/g DM digested), and decreased ( $P < 0.05$ ) fermentation efficiency. A high dose of essential oils from goat weed and African

basil leaves improved fermentation efficiency and reduced DMD and methane production in a manner that is comparable with monensin.

**Key Words:** essential oil, medicinal plant, in vitro fermentation

**833 Effects of supplemental energy and protein source on performance of steers grazing irrigated corn residue.** Benjamin T. Tibbitts\*<sup>1</sup>, Cody A. Welchons<sup>1</sup>, Robert G. Bondurant<sup>1</sup>, Fred H. Hilscher<sup>1</sup>, James C. MacDonald<sup>1</sup>, and Rick N. Funston<sup>2</sup>, <sup>1</sup>University of Nebraska-Lincoln, Lincoln, NE, <sup>2</sup>University of Nebraska West Central Research and Extension Center, North Platte, NE.

Seventy-five crossbred steer calves ( $235 \pm 1.1$  kg) grazing irrigated corn residue were blocked by BW and randomly assigned to 5 treatment groups ( $n = 15$ ) to evaluate the effects of protein and energy supplementation on steer performance. Steers were supplemented daily at 1100–1200 via a Calan gate system for 86 d. Treatment supplements consisted of 60% soy-pass + 40% soybean meal (SP), dried distillers grains (DDG), 89% Corn/6% Molasses/5% Urea (C + RDP), corn only (CRN), and control (NS) fed at 1.59, 1.36, 1.82, 1.7, and 0.0 kg DM/d respectively. Supplements were fed at different DM amounts to provide equal TDN intake. Estimated TDN values by supplement were 87% (SP), 104% (DDG), 87% (C + RDP), and 83% (CRN). Ending BW ( $P < 0.05$ ) differed among treatments and was 291, 286, 254, 245, and 229 (SEM 4.9) kg for SP, DDG, C + RDP, CRN, and NS respectively. Average daily gain among treatments was 0.67, 0.6, 0.24, 0.14, and - 0.09 kg (SEM 0.06) for SP, DDG, C + RDP, CRN, and NS respectively and was significantly different ( $P < 0.05$ ) among all treatments. Treatment groups supplemented with SP and DDG achieved average daily acceptable gains above 0.5 kg, while C + RDP, CRN, and NS treatment groups achieved average daily gains lower than 0.5 kg. The SP treatment provided a combination of RDP and RUP which resulted in greatest ADG among treatments when supplement TDN was similar.

**Key Words:** beef steers, corn residue grazing, supplementation

**834 Variability in predicted weaning weight of nursing calves using four models.** Phillip A. Lancaster\*<sup>1</sup> and Luis O. Tedeschi<sup>2</sup>, <sup>1</sup>Range Cattle Research and Education Center, University of Florida, Ona, FL, <sup>2</sup>Department of Animal Science, Texas A&M University, College Station, TX.

The objective of this study was to assess the variability surrounding predicted weaning weight (WW) of nursing beef calves at 210 d of age using 2 models developed to predict milk yield (MY) and calf forage DMI. These models were developed to predict calf WW based on peak milk (PKMK), calf BW, and forage DE. Equations to predict calf forage DMI were published by Tedeschi et al. (2006; Nutrient Digestion and Utilization in Farm Animals; TED06) and Tedeschi et al. (2009; J. Anim. Sci. 87:3380; TED09). Additionally, we evaluated 2 equations to predict MY: Wood (1967; WOD) and the NRC (2000; NRC). Calf ADG was computed using NRC (2000) equations for energy requirements assuming ME content of 5.29 Mcal/kg for milk. A Monte Carlo simulation with 5,000 iterations assumed normal distribution with mean and SD of  $3 \pm 0.5$  Mcal/kg for forage DE,  $35 \pm 2$  kg for calf birth weight (CBW), and  $550 \pm 50$  kg for final shrunk BW (FSBW), and uniform distribution with minimum and maximum at 3 and 12 kg/d for PKMK. Although predicted WW overlapped for all model combinations, their mean and SD varied considerably:  $147 \pm 72.9$ ,  $219 \pm 95.1$ ,  $262 \pm 91.6$ , and  $278 \pm 82.2$  kg for NRC&TED09, WOD&TED09, NRC&TED06, and WOD&TED06, respectively. Their predicted WW tended to follow



normal distributions, except for NRC&TED09 that was skewed to the right. The percentage of predicted WW within 100 and 300 kg were 75.3, 76, 67, and 62.8%, respectively, and within 200 and 300 kg were 10.1, 36.1, 42.4, and 46.4%, respectively. Forage DE had the greatest Spearman correlation ( $0.74 < r < 0.86$ ) with WW, followed by PKMK ( $38 < r < 44$ ), FSBW ( $0.16 < r < 0.18$ ), and CBW ( $r = 0.01$ ). Forage DE also had the greatest standardized regression coefficient (SRC) for WW: 0.73, 0.84, 0.81, and 0.80 for NRC&TED09, WOD&TED09, NRC&TED06, and WOD&TED06, respectively, and PKMK had the second greatest

SRC: 0.28, 0.39, and 0.43 for WOD&TED09, NRC&TED06, and WOD&TED06, respectively. For NRC&TED09, FSBW had the second greatest SRC (0.18). We concluded that forage DE is the most influential factor that affects calf WW and these predictive models have distinct prediction patterns for calf WW. Future analysis should focus on consolidating predicted MY between these models.

**Key Words:** beef, growth, modeling simulation

## Nonruminant Nutrition: Fiber

**835 Effects of dietary mannan oligosaccharide supplementation on performance and immune response of sows and their offspring.** X. D. Duan<sup>1</sup>, D. W. Chen<sup>1</sup>, P. Zheng<sup>1</sup>, G. Tian<sup>1</sup>, J. P. Wang<sup>1</sup>, X. B. Mao<sup>1</sup>, J. Yu<sup>1</sup>, J. He<sup>1</sup>, B. Li<sup>1</sup>, Z. Q. Huang<sup>1</sup>, Z. G. Ao<sup>2</sup>, and B. Yu\*<sup>1</sup>, <sup>1</sup>*Institute of Animal Nutrition, Sichuan Agricultural University, Chengdu, Sichuan, China*, <sup>2</sup>*Alltech Biological Product (China) Co. Ltd., Beijing, China*.

A study was conducted to determine effects of mannan oligosaccharide (MOS) supplementation of the diets of sows and their offspring on maternal and offspring performance and immune status. Sixty multiparous sows were fed either a control diet (sCON, n = 30) or a diet containing 400 mg/kg MOS (sMOS, n = 30) from d 86 of gestation until the end of lactation. On d 7 of age, offspring were assigned within sow treatments to the piglet basal diet (pCON) or the piglet basal diet + 800 mg/kg MOS (pMOS) for 28 d. Thus, the offspring treatments were sCON-pCON, sCON-pMOS, sMOS-pCON, and sMOS-pMOS, 15 litters per treatment. No dietary effect was observed on the number of total piglets born and born alive and the piglet weight at birth. Serum-specific antibody concentrations of porcine reproductive and respiratory syndrome, classical swine fever, and porcine pseudorabies of sows and immunoglobulins concentrations in colostrum and milk were not affected by sow dietary MOS ( $P > 0.05$ ). However, the weaning to estrus interval of sows was shortened significantly in the sMOS treatment compared with the sCON treatment ( $P = 0.05$ ). The weaning weight ( $P = 0.03$ ) and pre-weaning weight gains ( $P = 0.01$ ) of piglets from the sMOS were greater than those from the sCON. Piglets from sMOS had higher serum concentrations of IgA ( $P < 0.01$ ), IgG ( $P < 0.01$ ), complement 3 (C3) ( $P < 0.01$ ), lysozyme (LYZ) ( $P < 0.01$ ) and complement 4 (C4) ( $P = 0.05$ ) at weaning, as well as C3 ( $P < 0.01$ ) and LYZ ( $P < 0.01$ ) on d 35 of age. MOS supplementation of piglet diet improved both pre- and post-weaning weight gain ( $P < 0.01$ ) and increased serum concentrations of IgA ( $P = 0.01$ ) and IgG ( $P = 0.01$ ) at weaning, as well as C3 ( $P < 0.01$ ), LYZ ( $P < 0.01$ ) and C4 ( $P = 0.01$ ) on d 35 of age. Piglets of the sMOS-pMOS group had higher body weight and better immune status at weaning and on d 35 of age compared with other groups. These results indicate that supplementing sow diets with MOS during late gestation and lactation improved the growth potential and immune responses of nursing piglets. Furthermore, dietary addition of MOS to pre- and post-weaning piglets also has potential growth and immune-promoting effects.

**Key Words:** immune response, mannan oligosaccharide, performance

**836 Dietary chitooligosaccharide supplementation alleviates immune challenge induced by lipopolysaccharide in weaned piglets.** L. Zhang, G. Tian, D. W. Chen, B. Yu\*, Y. Yao, J. Yu, P. Zheng, X. B. Mao, J. He, and Z. Q. Huang, *Animal Nutrition Institute, Sichuan Agricultural University, Chengdu, Sichuan, China*.

A total of 24 weaned piglets ( $8.53 \pm 1.00$  kg) were used to investigate the effects of chitooligosaccharide (COS) supplementation on immunological responses challenged with *E. coli* lipopolysaccharide (LPS). Experimental treatments were arranged in a  $2 \times 2$  factorial design (n = 6/group), with the main effects of COS (0 vs. 300 mg/kg) and LPS challenge (LPS vs. saline). Piglets were raised individually in metabolic cages and fed 0 or 300 mg/kg COS for 18 d. On d 15–17, piglets were challenged with LPS (80  $\mu$ g/kg BW) or saline daily. Blood was obtained at 3 h and 48 h post-injection on d 17. At 48 h post-injection, weight of spleen was recorded, thymus and spleen were sampled. All data were

analyzed by 2-way ANOVA using GLM procedures of SAS, including COS, LPS, and their interaction as the fixed effects. Spleen index was elevated by LPS challenge (+37.88%,  $P < 0.01$ ), without an effect of COS supplementation. Serum TNF- $\alpha$  (+16.99%,  $P < 0.05$ ) concentrations increased and IL-10 (–11.61%,  $P < 0.01$ ) decreased in pigs injected with LPS at 3 h after injection. Serum IL-1  $\beta$ , IL-2 and IL-4 content were increased (+5.58%, +12.69% and +11.77%,  $P < 0.05$ ) at 48 h after injection. Piglets supplemented with COS had lower serum IL-1  $\beta$  and IL-2 at 3 h (–48.36% and –12.14%,  $P < 0.05$ ) post-injection, and lower serum IL-6 at 3 and 48 h post-injection (–10.43% and –13.13%,  $P < 0.05$ ). However, COS supplementation increased serum IL-10 at 3h post-challenge compared with non-supplemented pigs (+8.44%,  $P < 0.01$ ). These alterations in serum inflammatory factor traits in piglets challenged by LPS were accompanied by increased gene expressions of CD14, MyD88 in spleen (+12.63% and +26.77%) and TLR4, NF- $\kappa$ B, TRIF, IRF3 in thymus (+245.81%, +44.44%, +17.13% and +86.55%) that are related to MyD88 dependent and independent signaling pathway ( $P < 0.05$ ), while COS supplementation alleviated most of these changes (–37.23% for CD14, –34.87% for MyD88, –30.43% for NF- $\kappa$ B, –24.30% for TRIF and –31.82% for IRF3,  $P < 0.05$ ). In conclusion, results indicated that COS supplementation attenuated the immune challenge of LPS possibly by inhibiting over-activation of TLR4-MyD88 dependent and independent signaling pathway.

**Key Words:** chitooligosaccharide, lipopolysaccharide, piglet

**837 Effects of feeding fermented wheat with *Lactobacillus reuteri* on nutrient digestibility, growth performance, and intestinal fermentation in weaned pigs.** M. H. A. Le\*<sup>1</sup>, Y. Yang<sup>1</sup>, S. Galle<sup>1</sup>, J. L. Landero<sup>1</sup>, E. Beltrarena<sup>1,2</sup>, M. G. Gänzl<sup>1</sup>, and R. T. Zijlstra<sup>1</sup>, <sup>1</sup>*University of Alberta, Edmonton, AB, Canada*, <sup>2</sup>*Alberta Agriculture and Rural Development, Edmonton, AB, Canada*.

Feeding fermented feed to weaned pigs may improve gut health and thereby reduce diarrhea incidence. Effects of feeding wheat grain fermented for 24 h with *Lactobacillus reuteri* were evaluated in 36 weaned pigs (7.3 kg BW). The fermented wheat contained (DM basis) 14.3% CP, 0.45% chemically available Lys, and 7.8% NDF, whereas the nonfermented wheat contained 17.0% CP, 0.47% chemically available Lys, and 11.7% NDF. Pigs were fed 6 mash wheat-based diets balanced for water content during 2 phases: phase 1 diets for 1 wk (d 0–7) with 20% unfermented or fermented wheat and subsequently phase 2 diets for 2 wk (d 8–21) with 50% unfermented or fermented wheat. The 6 diets were negative control (NC; non-fermented), positive control (PC; nonfermented + organic acid including lactic and glacial acetic acid in the ratio of 4:1), and 4 fermented wheat diets (*L. reuteri* TMW1.656 and *L. reuteri* LTH5794 with or without added sucrose). Diets were formulated to provide 2.5 and 2.4 Mcal NE/kg and 5.3 and 5.0 g SID Lys/Mcal NE for phase 1 and 2 diets, respectively. Data were analyzed using the MIXED procedure with contrast statements to test effects of fermentation with or without sucrose. Feeding fermented wheat reduced ( $P < 0.05$ ) the apparent total-tract digestibility (ATTD) of diet GE (84.4 vs. 85.2%) and CP (81.8 vs. 83.6%) for wk 3 compared with the controls. Weaned pigs fed fermented wheat diets had lower ADFI ( $P < 0.05$ ) than pigs fed NC and PC for wk 1 and for the entire study (271 vs. 300 g of DM/d). Pigs fed fermented wheat diets had lower ADG ( $P < 0.05$ ; 232.8 vs. 260 g/d) and G:F ( $P < 0.10$ ; 0.81 vs. 0.80) vs. pigs fed NC and PC in wk 3, but ADG and G:F did not differ for the entire study. Concentrations of acetic, propionic, branched-chain, and total VFA in feces increased

( $P < 0.05$ ) in pigs fed fermented wheat with added sucrose; however, VFA did not differ in ileal digesta. In conclusion, feeding fermented wheat diets to weaned pigs stimulated hindgut fermentation but did not increase growth performance and ATTD of diet nutrients.

**Key Words:** fermented wheat, performance, weaned pig

**838 Extracted rice bran improves performance and fecal parameters in weaning pigs via prebiotic action.** M. Begum\*, B. Balasubramanian, M. M. Hossain, S. D. Upadhaya, and I. H. Kim, *Department of Animal Resource & Science, Dankook University, Cheonan, Chungnam, South Korea.*

One hundred forty weaning pigs [(Yorkshire  $\times$  Landrace)  $\times$  Duroc] with an average BW of  $5.70 \pm 1.41$  kg (21 d) were used in a 6-wk experiment to evaluate the effect of extracted rice bran (ERB) on growth performance, ATTD of nutrients, diarrhea score, blood profiles, fecal microbial shedding and fecal noxious gas emissions. Weaning pigs were allotted to diets containing 0 or 0.1 g ERB/kg of diet and 2 levels of antibiotic (tiamulin; ANT, 0 and 33 ppm) according to a  $2 \times 2$  factorial arrangement of treatments. There were 7 replicated pens per treatment with 5 pigs per pen. The experiment included 2 phases: 0 to 2 weeks and 2 to 6 weeks. At 2 and 6 wk, 2 pigs from each pen were subjected for the fecal microbiota and blood profiles. All data were statistically

analyzed using the repeated-measure statement of the MIXED procedure of SAS. The model included diet as a fixed effect whereas pig and period were included as random effects. During phase 1 and overall, ERB supplementation increased growth efficiency (G:F) compared with the ERB-free diet in weaning pigs (0.84 vs. 0.80 and 0.69 vs. 0.67;  $P = 0.01$  and  $P = 0.04$ , respectively). Pigs receiving diets supplemented with ANT increased DM and N digestibility than their counterparts during phase 1 (82.89 vs. 81.19% and 82.85 vs. 80.71%;  $P = 0.04$  and 0.01, respectively). Pigs fed ERB had higher N digestibility than pigs fed the non-ERB diet during phase 1 and phase 2 (83.60 vs. 79.96% and 82.48 vs. 79.22%,  $P = 0.01$  and 0.02, respectively). Supplementation of ANT  $\times$  ERB diets had positive effects for DM digestibility ( $P = 0.04$ ). During phase 2, the supplementation of ERB decreased total cholesterol (TC) than pigs fed the diet lacking ERB (97.30 vs. 103.60 mg/dL;  $P = 0.01$ ). During phase 2, the supplementation of ERB increased the *Lactobacillus* and reduced *Salmonella* counts than pigs fed the ERB-free diet (7.72 vs. 7.58  $\log_{10}$  cfu/g and 2.37 vs. 2.49  $\log_{10}$  cfu/g;  $P = 0.04$  and  $P = 0.02$ , respectively). Pigs fed ERB diets reduced ammonia (NH<sub>3</sub>) gas emission compared with non-ERB diets (22.70 vs. 26.46 ppm;  $P = 0.01$ ). In conclusion, results indicated that dietary supplementation of ERB improved performance in weaning pigs.

**Key Words:** extracted rice bran, microbial shedding, noxious gas emissions



## Production, Management, and the Environment VI

**839 Associations between management practices and reproductive performance in Canadian dairy herds.** José Denis-Robichaud\*<sup>1</sup>, Ronaldo L. A. Cerri<sup>2</sup>, Andria Jones-Bitton<sup>1</sup>, and Stephen J. LeBlanc<sup>1</sup>, <sup>1</sup>*Department of Population Medicine, University of Guelph, Guelph, Ontario, Canada,* <sup>2</sup>*Faculty of Land and Food Systems, University of British Columbia, Vancouver, British Columbia, Canada.*

The objective of this study was to identify management practices associated with reproductive performance on Canadian dairy farms. A questionnaire was distributed online and by mail to Canadian dairy farmers from March to May 2014 to assess reproduction management. From 833 responses, reproduction management was categorized as mainly visual heat detection ( $n = 415$  herds), timed artificial insemination (TAI;  $n = 175$ ), automated activity monitoring (AAM;  $n = 85$ ) where each represented  $>50\%$  of AI, or “combined” practices ( $n = 131$ ). Pregnancy rate (PR), insemination rate (IR), and conception risk (CR) per 21 d for 2013 were extracted from dairy herd information (DHI) files of 346 farms. Univariable linear regression models adjusted for region were used to identify associations of these reproduction performance measures with farms’ characteristics and management practices. The average ( $\pm$ SD) PR, IR, and CR were  $17 \pm 4.7\%$ ,  $43 \pm 11.3\%$ , and  $40 \pm 8.5\%$ , respectively. Pregnancy rate was lower in tiestall barns ( $15.2\%$  CI =  $14.4\text{--}16.0$ ) than in freestall barns ( $17.9\%$  CI =  $17.2\text{--}18.6$ ), in herds inseminating once per day ( $15.7\%$  CI =  $14.9\text{--}16.6$ ) than in herds inseminating twice per day ( $17.8\%$  CI =  $16.8\text{--}18.7$ ), and in herds with  $<100$  lactating cows ( $16.2\%$  CI =  $15.5\text{--}16.8$ ) than in herds with more than 250 lactating cows ( $20.1\%$  CI =  $17.2\text{--}22.9$ ). Pregnancy rates were not significantly different by main reproduction management practice: visual heat detection ( $15.9\%$  CI =  $15.0\text{--}16.8$ ), TAI ( $16.7\%$  CI =  $15.4\text{--}18.0$ ), AAM ( $17.4\%$  CI =  $16.0\text{--}18.9$ ), or combined practices ( $18.2\%$  CI =  $16.6\text{--}19.9$ ). Insemination rates were lower in herds using visual heat detection ( $40.0\%$  CI =  $37.3\text{--}42.6$ ) than in herds using TAI ( $47.4\%$  CI =  $43.8\text{--}50.9$ ), or combined practices ( $46.7\%$  CI =  $42.6\text{--}50.7$ ), but not significantly different from herds using AAM ( $44.0\%$  CI =  $40.3\text{--}47.7$ ). Conception risks were higher in herds using visual heat detection ( $40.7\%$  CI =  $37.2\text{--}44.1$ ) than in herds using TAI ( $36.8\%$  CI =  $33.0\text{--}40.3$ ), but not significantly different from herds using AAM ( $40.0\%$  CI =  $36.1\text{--}43.9$ ), or combined practices ( $39.8\%$  CI =  $35.8\text{--}43.9$ ). Performance was influenced by factors other than the reproductive management program.

**Key Words:** dairy cow, management, reproduction

**840 Evaluation of Ovsynch and CIDR inserted concurrently with or two days after initiation of Ovsynch protocol to improve reproductive performance in lactating dairy cows with low estrus detection efficiency.** Abid Hussain Shahzad\*<sup>1</sup>, Abdul Sattar<sup>1</sup>, Nasim Ahmad<sup>1</sup>, Ijaz Ahmad<sup>1</sup>, Deniz Nak<sup>2</sup>, and Yavuz Nak<sup>2</sup>, <sup>1</sup>*University of Veterinary and Animal Sciences, Lahore, Lahore, Punjab, Pakistan,* <sup>2</sup>*Uludag University, Bursa, Bursa, Turkey.*

To assess the efficiency of 3 synchronization methods as postpartum reproductive management tools in dairy cattle on a commercial dairy farm. Cyclic Holstein lactating cows ( $n = 167$ ), 55–90 DIM, were randomly allotted to 1 of 3 treatments. First group was (Ovsynch;  $n = 58$ ) given a 2-mL i.m. injection of synthetic GnRH on d 0. After 7 d, PGF<sub>2a</sub> (2 mL i.m.) was injected. A second injection of GnRH was administered 48 h after the PGF<sub>2a</sub> injection and all cows were bred by timed AI 16 h after the second GnRH injection. Cows in second group (Ovsynch+P7:

$n = 55$ ) were introduced a modified Ovsynch TAI treatment similar to Ovsynch but CIDR was inserted for a 7-d period (CIDR was inserted with GnRH and removed on PG Shot). In third group (Ovsynch+P5;  $n = 54$ ), cows were subjected to Ovsynch protocol in addition to CIDR insertion for 5 d (CIDR was inserted after 2 d of GnRH and removed with PG shot). P4 conc. was measured on d 30 and 60. Pregnancy diagnosis was carried out at d 30, 60, 90 post insemination in all 3 groups. Progesterone profile was significantly higher ( $P < 0.05$ ) for Ovsynch + P5 ( $7.75 \pm 0.38$  ng/mL) and Ovsynch + P7 ( $7.58 \pm 0.26$  ng/mL) compared with Ovsynch ( $6.52 \pm 0.32$  ng/mL) on d 30 PTAI but nonsignificant among all 3 synchronization treatments (i.e.,  $6.37 \pm 0.49$  in Ovsynch,  $6.75 \pm 0.36$  in Ovsynch+P5 and  $6.80 \pm 0.41$  in Ovsynch+P7 group). Pregnancy rate in Control group was 39.7, 36.2, and 32.80 on d 30, 60 and 90 post insemination. In second modified Ovsynch group with 7d CIDR (Ovsynch+P7) insertion pregnancy rate was 42.6% on d 30, 37% on d 60 and 90 PAI. In third group pregnancy rate was 45.5, 43.6 and 41.8 on corresponding 30, 60, 90 d PAI. Although pregnancy rate were statistically not significant but, by percentage points, improved pregnancy rate was observed on all 3 time points in Ovsynch+P5 group.

**Key Words:** modified Ovsynch, postpartum lactating cow, progesterone

**841 Reproductive outcomes following presynchronization of dairy heifers with a 14-d CIDR and prostaglandin F<sub>2a</sub>.** Courtney K. Claypool\*<sup>1</sup>, Jennifer A. Spencer<sup>1</sup>, Saulo Menegatti Zoca<sup>3</sup>, Bahman Shafiqi<sup>1</sup>, William J. Price<sup>1</sup>, Amin Ahmadzadeh<sup>1</sup>, Neil R. Rimbey<sup>2</sup>, and Joseph C. Dalton<sup>2</sup>, <sup>1</sup>*University of Idaho, Moscow, ID,* <sup>2</sup>*University of Idaho, Caldwell, ID,* <sup>3</sup>*UNESP, Botucatu, Sao Paulo, Brazil.*

Efficient pregnancy production in heifers provides an earlier return on investment for dairy producers. The research hypotheses were that presynchronization would result in a faster insemination rate following entry to the breeding program, and an increased proportion of pregnant heifers within the first week of entry into the breeding program compared with no presynchronization. Heifers were assigned to 1 of 3 groups: (1) 14-d CIDR, (2) prostaglandin F<sub>2a</sub> (2 $\times$  PG), or control (1 $\times$  PG). The 14-d CIDR group ( $n = 119$ ) received a CIDR on d -30, which was removed on d -16. The 14-d CIDR group received an injection of PG (25 mg i.m.) upon entry to the breeding program (d 0). The 2 $\times$  PG group ( $n = 118$ ) received an initial injection of PG on d -11, and a second injection of PG on d 0. The 1 $\times$  PG group ( $n = 121$ ) received an injection of PG on d 0. Data were analyzed using ANOVA procedures within the generalized linear mixed models in SAS (9.4). All animals received tail paint and were observed for behavioral estrus daily. Pre-synchronization affected ( $P < 0.05$ ) the proportion of heifers observed in estrus and inseminated in the first week: 95.8% (14-d CIDR), 74.5% (2 $\times$  PG), 66.9% (1 $\times$  PG). Days to AI (LSM  $\pm$  SEM) following entry to the breeding program were also affected ( $P < 0.05$ ) by treatment:  $3.6 \pm 0.4$  d (14-d CIDR),  $4.9 \pm 0.4$  d (2 $\times$  PG),  $6.8 \pm 0.5$  d (1 $\times$  PG). Proportion of heifers pregnant in the first week of the breeding program also differed ( $P < 0.05$ ): 68% (14-d CIDR), 43% (2 $\times$  PG), 41% (1 $\times$  PG). Conception rates to AI in the first week were 71.9% (14-d CIDR), 57.9% (2 $\times$  PG), and 61.7% (1 $\times$  PG), and were different ( $P < 0.05$ ) between 14-d CIDR and 2 $\times$  PG heifers. Days on feed from entrance to the breeding program to projected calving date were calculated for all heifers:  $295 \pm 2.6$  d (14-d CIDR),  $302 \pm 2.6$  d (2 $\times$  PG), and  $305 \pm 2.5$  d (1 $\times$  PG), and were different ( $P < 0.05$ ) between 14-d CIDR and 1 $\times$  PG heifers. Pre-synchronization with a 14-d CIDR and PG appears to be an effective strategy to increase the

proportion of heifers identified in estrus, inseminated, and pregnant within the first week upon entry to the breeding pen.

**Key Words:** dairy heifer, presynchronization, CIDR

#### **842 Effects of expression of estrus measured by activity monitors on ovarian dynamics and conception risk in Holstein cows.**

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The objective of this study was to determine intensity of physical activity as a result of estrus expression and its relationship with ovarian dynamics and fertility. Estrus events were recorded at 2 commercial dairies. A collar-mounted accelerometer (Collar; SCR Heatime) and a leg-mounted pedometer (Leg1; Boumatic Heat-seeker-TX) were used in farm 1. A second leg-mounted pedometer (Leg2; AfiMilk Pedometer Plus) was used in farm 2. Only spontaneous estrus (SE) events were registered at farm 1 (collar and Leg1; 1,099 events; 318 cows). Cows with Leg2-tags were induced to ovulate (IO; 1,411 events; 1,040 cows) with an E2/P4-based TAI protocol. Blood samples and ovarian ultrasonography were done at detection of activity increase (SE), or at the moment of AI and 10 d after AI (IO). IO cows' were scanned on d 7 after AI to assess CL presence. Pregnancy diagnosis was performed at  $42 \pm 7$  d. Data were analyzed with ANOVA, logistic regression, proc MIXED. Peak estrus activity (PA) for Collar was  $71.6 \pm 20.7$  index value, and for Leg1 and Leg2,  $334.3 \pm 155.7\%$  and  $286.18 \pm 163.3\%$  relative increase. Pre-ovulatory follicle diameter was  $18.8 \pm 0.3$  mm (SE) and  $13.3 \pm 3.5$  mm (IO). Follicle diameter was not correlated with AAM measurements ( $r = 0.007$ ). Cows with high PA had greater E2 concentration ( $9.5 \pm 0.3$  vs.  $8.2 \pm 0.2$  ng/mL [Collar];  $8.9 \pm 0.2$  vs.  $8.1 \pm 0.2$  ng/mL [Leg1]). P4 concentration at d of AI was not correlated with relative increase (Leg2), but it was higher on d 10 for cows that had more intense estrus ( $3.17 \pm 0.11$  ng/mL vs.  $3.73 \pm 0.21$  ng/mL). Cows with greater PA had greater P/AI than those with lower PA ( $36.5\%$  vs.  $24.6\%$  [Collar],  $33.5\%$  vs.  $21.4\%$  [Leg1] and  $42.9\%$  vs.  $28.9\%$  [Leg2]). Likelihood of ovulation was greater for high vs. low relative increase ( $94.6\%$  vs.  $86.2\%$ ). Greater estrus intensity improved ovulation and P/AI. Data from AAM might be used to survey and predict fertility measures in dairy cows. More studies are needed to determine management and selection tools to improve estrus expression, detection, and fertility.

**Key Words:** pedometer, follicle, pregnancy

#### **843 Vitality and morphology of Boer buck spermatozoa stored fresh for 72 hours.** Olumide A. Ajao\*, Daniel M. Barry, and Kow K. Benyi, University of Venda, Thohoyandou, Limpopo Province, South Africa.

The study was aimed at evaluating the effects Biladyl and Triladyl extenders on the percentage live proportion and morphological status of the Boer buck spermatozoa when stored at 5°C, 12°C and 17°C in programmable refrigerators for 72 h. Four ( $n = 4$ ) healthy Boer bucks aged  $3.12 \pm 0.55$  years were ejaculated using an artificial vagina (AV) once every 4 d for 6 replicates. Semen was extended at ratio 1: 5 v/v (semen to extender). All smears were duplicated. Semen samples were evaluated for percentage live spermatozoa and morphological status after every 12 h in 72 h of storage. Data were analyzed by ANOVA using the GLM procedure of Minitab. Spermatozoa viability and morphological correlations between extender type and temperature were assessed using the principal component analysis (PCA). The

interaction between extender type and storage temperature indicated no significant difference ( $P > 0.05$ ) on the proportion of live spermatozoa. The percentage live spermatozoa in Biladyl stored at 12°C was 76.6% compared with the higher percentage of live spermatozoa in Triladyl (80.0%) which were kept at 12°C. Among the conditions storage tested, temperature 12°C kept the least number of spermatozoa alive after the 72 h of storage with 69% of live spermatozoa. The percentage of live spermatozoa found in Triladyl after 72 h at 17°C was highest (86.3%). Spermatozoa kept in Biladyl at 12°C and in Triladyl at 12°C statistically produced higher morphological abnormality with ( $P < 0.05$ ) effect than the morphological abnormality discovered in Biladyl 17°C and Triladyl 17°C. Temperature 12°C had the highest percentage of morphologically abnormal spermatozoa (23.33%) in Biladyl, followed by the Triladyl extender (22.46%) stored at 12°C, followed by the percentage found in Biladyl stored at 12°C (18.68%) and lastly by the 17.46% of sperm found in semen extended with Triladyl stored at 17°C. With appropriate protocol, Biladyl and Triladyl extenders can keep acceptable percentage of goat spermatozoa alive when stored at 5°C, 12°C and 17°C up to 72 h.

**Key Words:** extender, vitality, morphology

#### **844 Milk composition and fatty acids in mare's milk fat produced in the mountains and highlands of Kyrgyz Republic during milking season.** Aichurok T. Mazhitova\*, Asylbek A. Kulmyrzaev, and Zhyldyzai E. Ozbekova, Kyrgyz-Turkish Manas University, Bishkek, Kyrgyzstan.

In Kyrgyzstan, mare's milk is used for preparation of the fermented dairy beverage koumiss and fresh mare's milk has also been used to treat lung diseases as a traditional method of treatment, but mare milk production is seasonal. Accordingly the effect of the milking season on the chemical composition (with  $P < 0.01$ ) and fatty acid profile ( $P < 0.05$ ) of mares' milk of the Kyrgyz Novokirgizskaya horse breed grazing on the pastures at 1700 and 2200 m above the sea level was investigated. The animals were kept under extensive pasture conditions and received no additional feed supplements. Milk samples were collected monthly after machine milking from May to July 2014 and from May to August 2014 from 25 mares grazing at 1700 m and 2200 m, respectively. Total solids (11.84–11.05%), milk fat (2.23–1.23%) and ash (0.53–0.40%) content of milk produced at 1700 m were decreased to the end of milking season, while these constituents in the milk produced at 2200 m (10.93–11.11% total solids; 1.29–1.60% milk fat) and also protein (2.20–2.29%) and lactose (6.98–6.93%) did not change significantly during the milking season, except for ash content (0.46–0.295). Fatty acid composition of the milk obtained at 2200 m (vegetation is dominated by grasses, forbs, sub-shrubs) was richer in unsaturated fatty acids than that of produced at the pastures at 1700 m above the sea level (vegetation is dominated by shrubs and sub-shrubs). The results of the study have show that the chemical composition and fatty acid profile of the mare milk is influenced by milking season and geographical location of pastures.

**Key Words:** mare milk, milking season, geographical location

#### **845 Environmental and economic consequences of subclinical ketosis and related diseases in dairy farming.** P. F. Mostert\*, E. A. M. Bokkers, C. E. Van Middelaar, and I. J. M. De Boer, Wageningen University, Animal Production Systems group, Wageningen, the Netherlands.

Subclinical ketosis (SCK) in dairy cattle is a metabolic disease that occurs around the calving period and increases the risk on other diseases. SCK and other diseases result in, e.g., milk losses, reduced pregnancy rate, culling, and therefore have environmental and economic

consequences. This study aimed to estimate the environmental and economic consequences of SCK and related diseases in dairy farming. A dynamic stochastic simulation model at cow level was developed and combined with a life cycle assessment and partial budget analysis. The model was divided into 4 parts. In part one, cows receive a parity (1–5+) and a potential milk production. Cows subsequently have a risk on getting retained placenta or milk fever (part 2), SCK (part 3), and metritis, displaced abomasum, clinical ketosis, lameness or mastitis (part 4). The risk on diseases depends on parity and previous diseases. The model was parameterized using literature. Inputs are the number of dairy cows, prevalence of diseases and culling rate, outputs are the change in global warming potential (GWP) and profit per case of SCK. Outputs were divided in direct (SCK) and indirect (other diseases due to SCK) consequences, that were estimated with the attributional risk of SCK. Cows with (a combination of) diseases had: a reduced daily milk yield, discarded milk if treated, an increased calving interval, and risk of culling. Monte Carlo simulation was performed to find the variation in the output. Preliminary results showed that the costs increased from €33.0 (±31.3) to €55.2 (±58.3) and GWP increased from 1.3 (±1.3) to 1.8 (±2.0) % CO<sub>2</sub>-e/unit milk per parity based on milk losses per case of SCK. Results differ per parity ( $P < 0.001$ ) due to differences in milk yield and risk on diseases. The highest contribution came from SCK (68%). Other diseases particularly had an effect on the variation of the output. Future calculations will be extended by including reproduction and culling, and by performing sensitivity analyses. In conclusion, SCK has an effect on the environmental and economic performance of dairy farming.

**Key Words:** greenhouse gas emissions, costs, health

#### **846 Characterizing the temporal pattern of leaky gut biomarkers in healthy and ketotic cows during the transition period.**

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The dairy cow transition period is associated with increased incidence of metabolic disorders. Ketosis is the most prevalent metabolic disease and approximately 10–25% of cows experience clinical ketosis following calving. We hypothesize that a compromised gastrointestinal barrier (a.k.a. leaky gut) and subsequent lipopolysaccharide (LPS) infiltration may play a key-role in ketosis etiology. Study objectives were to retrospectively characterize biomarkers of leaky gut during the transition period in healthy and clinically diagnosed ketotic (urine acetoacetic acid >15 mg/dL) cows. Blood samples from lactating Holstein cows ( $n = 164$ , parity 2–7; from a commercial herd) were obtained at 21, 14, and 7 d antepartum (±3 d) and 3, 7, 10, and 14 d postpartum (±1 d). Cows were retrospectively categorized into one of 2 treatments: healthy (H;  $n = 24$ ; no diagnosed health disorder) and ketotic (KET;  $n = 22$ ; cows diagnosed only with ketosis). KET cows had reduced milk production compared with H (10%,  $P < 0.01$ ). Antepartum circulating LPS in KET cows was increased (129%,  $P = 0.02$ ) compared with H, and they tended to have increased NEFA (70%;  $P = 0.08$ ) when compared with H. Postpartum KET cows had increased NEFA (49%;  $P < 0.01$ ), BHBA (16%;  $P = 0.01$ ), SAA (75%;  $P = 0.04$ ), haptoglobin (273%;  $P = 0.03$ ), and L-lactate on d 3 (74%;  $P = 0.02$ ) and tended to have higher LPS binding protein (LBP; 4854 vs. 3569 ng/mL;  $P = 0.07$ ), when compared with H. Postpartum circulating insulin was decreased (11%;  $P = 0.02$ ) in KET cows compared with the H. The postpartum LPS area under the curve (a measure of total LPS exposure over time) tended to be decreased in KET cows ( $P = 0.06$ ), which may be explained by treatment differences

in LBP and its role in LPS removal. In summary, blood bioenergetics and gut integrity biomarkers indicate that ketosis may be closely associated with biomarkers of leaky gut during the transition period.

**Key Words:** ketosis, intestine, lipopolysaccharide

#### **847 A randomized clinical trial assessing the use of a single injection of dexamethasone combined with oral propylene glycol therapy for the treatment of hyperketonemia.** Elise H. Tatone<sup>\*1</sup>, Michael B. Capel<sup>2</sup>, Jessica L. Gordon<sup>1</sup>, Stephen J. LeBlanc<sup>1</sup>, and Todd F. Duffield<sup>1</sup>, <sup>1</sup>*Department of Population Medicine, University of Guelph, Guelph, ON, Canada*, <sup>2</sup>*Perry Veterinary Clinic, Perry, NY*.

The treatment of hyperketonemia [HK, blood  $\beta$ -hydroxybutyrate (BHBA)  $\geq 1.2$  mmol/L] with oral propylene glycol has proven benefits. Parenteral administration of glucocorticoids has been suggested as an adjunctive therapy, however it has not been assessed in a randomized field trial setting. The objective of this research was to evaluate the effect on cure rate of a one-time intramuscular injection of dexamethasone as an adjunctive therapy for hyperketonemia. A randomized control trial was conducted from May to August 2014 on 4 dairy farms in New York State. Cows were tested from 3 to 16 d in milk, once weekly, for blood BHBA using the Precision Xtra device. All cows testing positive for HK were randomly assigned to receive a single intramuscular injection of 20 mg dexamethasone (DX) or an equivalent volume of sterile saline placebo (PB). Both groups received 4 d of oral propylene glycol therapy. Cure risk was determined by blood BHBA obtained the week following enrollment. A multivariable multi-level logistic regression model was constructed for the outcome of being ketotic at first follow-up. Herd was controlled as a random effect. A total of 498 cows were enrolled, 248 treated with DX and 250 receiving PB. At first follow-up, 250 individuals (50.2%) remained HK. Of those, 131 (52.4%) were from the DX group and 115 (46.0%) from the PB group ( $P = 0.26$ ). Lactation number and BHBA at enrollment modified the effect of treatment. The odds of recovering were higher in first lactation animals treated with DX compared with PB treated first-lactation animals (OR: 3.59 CI: 1.29–10.03). Cows enrolled with BHBA at 1.2 and 1.4 mmol/L and treated with DX had odds of recovering 2–3 times higher (OR: 3.09, CI: 1.65–5.75, OR: 2.31, CI: 1.39–3.83, respectively) than those treated with the PB. There is no significant association between treatment and cure risk at higher BHBA at enrollment. Adding DX to HK treatment protocols was beneficial for cows in first lactation or had a blood BHBA at enrollment of 1.2 or 1.4 mmol/L.

**Key Words:** ketosis, treatment, dexamethasone

#### **848 Effects of prepartum digital dermatitis on first-lactation performance.** Arturo Gomez<sup>\*1,2</sup>, Nigel Cook<sup>1</sup>, Mike Socha<sup>2</sup>, and Dörte Döpfer<sup>1</sup>, <sup>1</sup>*University of Wisconsin-Madison, Madison, WI*, <sup>2</sup>*Zinpro Corporation, Eden Prairie, MN*.

The objective of this study was to describe and quantify the effect of prepartum digital dermatitis (DD) on first-lactation performance. A cohort of 719 pregnant heifers was monitored for DD for a period of 6 mo before calving. The heifers were classified by the number of DD events diagnosed as Type I, Type II and Type III (no DD, one DD event, and multiple DD events, respectively) during this period. After calving, health during initial 60 d in milk (DIM), reproductive and hoof health outcomes, and milk production were compared among the 3 group types. Multivariable logistic and linear models were adjusted for age, height, and girth circumference at enrollment, as well as type of trace mineral supplementation during the pre-partum period. Overall, cows



experiencing DD during the rearing period showed inferior production and health outcomes compared with healthy heifers during the first lactation. A numerical difference was found on the percentages of assisted calvings, stillbirths, culls before 60 DIM, and diseased cows during the fresh period between Type III and Type I cows. Significantly lower conception risk at first service (OR [95% CI] = 0.55 [0.33, 0.89]) and increased number of days open (mean [95% CI] = 24 d [5.2, 43]) were observed in Type III compared with Type I cows. In relation to hoof health, a significantly increased risk of DD during first lactation was found in Type II and III cows (OR = 5.16 [3.23, 8.29] and 12.5 [7.52, 21.1], respectively), as well as the earlier occurrence of DD post-calving (mean [95% CI] = 59 d [20, 96], and 74 d [37, 109]). Compared with Type I cows, decreased milk production during initial 305 DIM was estimated at 199 and 335 kg for Type II and III cows, respectively. This difference was due to a greater decline in rate of production (less persistence) after peak yield ( $P < 0.01$ ). Given the long-term effects of DD on health, reproduction, and production, priority should be given to efficient DD prevention and control programs during the rearing period of dairy heifers. Intensive intervention programs are expected to increase overall well-being and farm profitability, based on active long-term DD surveillance, mitigation of risk factors, and prompt treatment.

**Key Words:** digital dermatitis, heifer, milk production

**849 Identification of the most likely classical swine fever outbreak scenarios in the swine industry of Indiana.** Shankar Yadav<sup>\*1</sup>, Nicole Olynk Widmar<sup>2</sup>, and Hsin-Yi Weng<sup>1</sup>, <sup>1</sup>*Department of Comparative Pathobiology, Purdue University, West Lafayette, IN,* <sup>2</sup>*Department of Agricultural Economics, Purdue University, West Lafayette, IN.*

The objectives of this study were to develop metrics using empirical data for the identification of the most likely outbreak scenarios of clas-

sical swine fever (CSF) in Indiana and to describe the characteristics of the outbreaks. Three types of CSF outbreak scenarios were considered: single, multiple, and outbreak due to delay in detection. The data sources included Indiana premise identification database, feral hog population, and US census data. The attributes included in the metrics were distribution of swine premises and operation types, import frequencies (domestic and international), import origins, number of imported pigs, proximity to feral hogs, and immigrant population. Different weights were assigned to each of the attributes based on their importance. The metrics were used to identify the top 10 Indiana's counties that were most likely to initiate a CSF outbreak; premises with high risk of outbreak were identified within the top 10 counties. Each of the identified swine premises represented a single outbreak scenario. The swine import data of the identified premises were used for identifying the multiple outbreak scenarios while the export data for identifying the outbreak due to delay in detection scenarios. These identified outbreak scenarios were simulated to derive the outbreak-related measures. In 2012, there were 8589 swine premises in Indiana. A total of 3,145 import shipments from 27 US states and 3 Canadian provinces were received. Similarly, 3,154 export shipments of live pigs were sent to 41 US states. Nineteen single and 15 multiple outbreak scenarios were identified, while no outbreak due to delay in detection was identified. The median number of premises in the multiple outbreak scenarios was 17 (range: 4–32). The estimated median epidemic durations (days) for single and multiple CSF outbreak scenarios in Indiana were 57 and 121, respectively. The identified most likely CSF outbreak scenarios can be used to estimate epidemic duration and magnitude of an outbreak and provide guidance for developing a risk-based surveillance for the CSF in Indiana.

**Key Words:** classical swine fever, epidemic, risk assessment

## Ruminant Nutrition: Dairy rumen metabolism

**850 A novel inhibitor persistently decreased enteric methane emission and increased weight gain of high-producing Holstein cows without negatively affecting milk production.** Alexander N. Hristov\*<sup>1</sup>, Joonpyo Oh<sup>1</sup>, Fabio Giallongo<sup>1</sup>, Michael T. Harper<sup>1</sup>, Holley Weeks<sup>1</sup>, Antonio F. Branco<sup>2</sup>, Peter J. Moate<sup>3</sup>, Matthew H. Deighton<sup>3</sup>, S. Richard O. Williams<sup>3</sup>, Maik Kindermann<sup>4</sup>, and Stephane Duval<sup>5</sup>, <sup>1</sup>*Department of Animal Science, The Pennsylvania State University, University Park, PA*, <sup>2</sup>*Departamento de Zootecnia, Universidade Estadual de Maringá, Maringá, Parana, Brazil*, <sup>3</sup>*Agriculture Research Division, Department of Economic Development Jobs Transport and Resources, Ellinbank, Victoria, Australia*, <sup>4</sup>*DSM Nutritional Products, Animal Nutrition and Health, Basel, Switzerland*, <sup>5</sup>*DSM Nutritional Products France, Research Centre for Animal Nutrition and Health, Saint Louis Cedex, France*.

This study investigated the effect of a CH<sub>4</sub> inhibitor, 3-nitrooxypropanol (3NOP), on enteric CH<sub>4</sub> emissions in lactating Holstein cows. The experiment was a randomized block design with 48 cows (DIM, 77 ± 3.9; lactations, 2.2 ± 0.15), with a 2-wk covariate and a 12-wk data collection periods. Treatments were: control (no additive) and 3NOP applied at 40, 60, and 80 mg/kg feed DM. 3NOP was mixed with the TMR and cows were fed once daily. Methane and H<sub>2</sub> emissions were measured using 2 methods, the GreenFeed system and the modified SF<sub>6</sub> tracer technique, during the covariate period and experimental wk 2, 6, 9, and 12. Compared with the control, 3NOP decreased the average CH<sub>4</sub> emission by 25, 31, and 32%, respectively, when measured using the GreenFeed system (481, 363, 333, and 329 g/cow/d; SEM = 15.9; *P* < 0.001). Similar inhibition of CH<sub>4</sub> emission by 3NOP was observed when determined using the SF<sub>6</sub> technique. Hydrogen emissions from the control cows were negligible throughout the experiment (on average 0.02 g/cow per day), but increased considerably for the 3NOP treatments (0.48, 0.96, and 1.27 g/cow per day, respectively; SEM = 0.116, *P* < 0.001). Methane emissions per kg of DMI or ECM were on average 29 and 31%, respectively, lower for the 3NOP treatments compared with the control. Treatment had no effect on DMI (27.8 ± 0.45 kg/d), milk production (45.5 ± 1.21 kg/d), and feed efficiency (1.65 kg/kg). Milk composition was not affected by 3NOP. Milk protein and lactose yields were increased (*P* ≤ 0.05) by 3NOP. Body weight gain during the experiment was about 168 g/d greater (*P* = 0.05) for the 3NOP-treated cows. Apparent total-tract digestibility of DM, OM, CP, and ADF were quadratically increased (*P* ≤ 0.06) by 3NOP compared with the control. This experiment demonstrated that enteric CH<sub>4</sub> emissions from ruminant animals can be successfully decreased by the use of a CH<sub>4</sub> inhibitor, without negatively affecting animal productivity. If adopted, this mitigation practice could substantially reduce greenhouse gas emissions from the ruminant livestock sector.

**Key Words:** methane, hydrogen, 3-nitrooxypropanol

**851 Effect of frequency of total mixed ration delivery on digestibility and milk production in lactating dairy cows: A meta-analysis and meta-regression.** Hamidreza Mirzaei-Alamouti\*<sup>1</sup>, Mehdi Kazemi-Joujili<sup>1</sup>, Hamid Amanlou<sup>1</sup>, and Mina Vazirigohar<sup>2</sup>, <sup>1</sup>*Department of Animal Science, Faculty of Agriculture, University of Zanjan, Zanjan, Iran*, <sup>2</sup>*Department of Animal Science, Campus of Agriculture and Natural Resources, University of Tehran, Karaj, Alborz, Iran*.

The objectives of this study were to evaluate the effect of frequency of feeding dairy cows with total mixed ration (TMR) on digestibility, feed-

ing behavior, and milk production and composition in lactating cows, using meta-analysis and meta-regression methods. A total of 14 studies with 25 comparisons between treatment (higher feeding frequency) and control (lower feeding frequency) groups, met the selection criteria and included in the analysis. Type and amount of feed ingredients did not differ between treatment and control diets. The mean (±SD) forage and grain amount in the experimental diets were 539 ± 89.9 and 274 ± 104.2 g/kg DM, respectively. Increasing frequency of feeding (in average; 1.0× vs. 3.3×) decreased dry matter intake [DMI; weighted mean differences (WMD) = -0.134 kg/d; 95% confidence interval (CI) = -0.505 to 0.236] but had no effect on milk production. The amount of forage in the basal diet and DIM were the sources of heterogeneity for DMI. Thrice-daily feeding compared with once a day increased milk fat yield (WMD = 15.02 g/d; 95% CI = 10.93 to 40.96), but milk fat content, and milk protein content, and yield remained unchanged. Increasing feeding frequency increased NDF digestion (WMD = 5.663%; 95% CI = 0.631 to 10.69). Results of meta-regression showed that more frequent feeding of diets containing high level of grains improved NDF digestibility in lactating cows, but lowered NDF digestion for high forage diets. Increasing the frequency of feeding did not change total feeding and ruminating times. In conclusion, feeding a TMR containing equal amounts of forage and concentrate 3 times a day vs. once a day decreased DMI without any changes on milk production.

**Key Words:** feeding frequency, lactating cow, meta-analysis

**852 Relationship between rumen molar volatile fatty acid proportions and milk odd- and branched-chain fatty acid concentrations in cows fed diets containing sunflower oil.** Mina Vazirigohar\*, Mehdi Dehghan-Banadaky, Kamran Rezayazdi, and Ardeshir Nejati-Javaremi, *Department of Animal Science, Campus of Agriculture and Natural Resources, University of Tehran, Karaj, Alborz, Iran*.

Principal component analysis (PCA) was used to explore the relationship among molar proportions of individual rumen volatile fatty acid (VFA) and milk odd- and branched-chain fatty acid (OBCFA) concentrations. Data were obtained from an experiment with 24 lactating Holstein cows examining the effects of diets containing variable proportions of forage (39, 43.5 or 48% of forage in total diet DM) and concentrate containing sunflower oil (3% in diet DM) on milk fat content and composition. The data were analyzed using the loading plot of PCA which was performed using PASW (version 18.0). Molar proportions of butyrate were included in the initial evaluation, but these data were subsequently excluded because these did not meet the necessary PCA model fit criteria. Loading plot of PCA revealed that rumen molar acetate proportions clustered with milk *iso* fatty acids of <15 carbon atoms, whereas rumen molar propionate proportions clustered with milk straight odd-chain fatty acids (15:0 and 17:0) and their desaturation products in the opposite side of both PCs. Concentrations of 15:0, *cis*-9 15:1 and *cis*-9 17:1 had high loadings on PC1, whereas 17:0 had high loading on PC2 which was located close to molar propionate proportions. In conclusion, rumen molar acetate and propionate proportions were closely associated with milk fat *iso* and straight odd-chain fatty acids concentrations, respectively, in cows fed diets containing sunflower oil.

**Key Words:** milk odd- and branched-chain fatty acid, principal component analysis, rumen volatile fatty acid

**853 The effect of increasing level of by-product inclusion on milk production, milk composition, nutrient digestibility and nitrogen excretion in early lactation grazing dairy cows.** Billy Carey<sup>1</sup>, Tommy M. Boland\*<sup>1</sup>, Stephen J. Whelan<sup>2</sup>, Gaurav Rajauria<sup>1</sup>, and Karina M. Pierce<sup>1</sup>, <sup>1</sup>*School of Agriculture and Food Science, University College Dublin, Dublin, Ireland*, <sup>2</sup>*DairyCo, Agriculture & Horticulture Development Board, Stoneleigh Park, Kenilworth, United Kingdom*.

Irish dairy farming is characterized by a predominantly spring calving, pasture based production system with strategic use of concentrate supplementation when pasture availability is less than herd demand. Cereal grains are used extensively in concentrates as an energy source, but the use of these human edible energy sources is becoming somewhat controversial. By contrast, by-products, which cannot be consumed by humans, such as maize dried distillers' grains (DDG), palm kernel expeller meal (PKE) and soybean hulls (SH) can provide an economical source of nutrients to complement grazed grass. While these products have been extensively studied in US and New Zealand production systems, there is a paucity of information on their use in moderately yielding (30 kg/d) pasture fed cows. The objective of this research was to evaluate the effect of replacing barley with increasing dietary levels of by-products (DDG, PKE and SH) on milk production and composition, pasture and total dry matter intake (DMI), body condition score (BCS) and body weight (BW) and nitrogen (N) excretion in early lactation dairy cows grazing perennial ryegrass-based pasture. Forty-eight Holstein Friesian dairy cows (64 DIM  $\pm$  24) were blocked by pre-experimental milk yield and offered 1 of 4 supplementary concentrates (5.17 kg of DM/d) containing 35, 55, 75 and 95% by-products for T1, T2, T3 and T4 respectively. The concentrates offered contained DDG, PKE and SH in equal portions and the experiment continued for 70 d. Data were tested using Proc Mixed of SAS with trt, week, parity and their interactions included as fixed effects in the model. Treatment had no effect on milk yield (30.8 kg/d;  $P = 0.78$ ) or fat and protein yield (2.1 kg/d;  $P = 0.57$ ). Similarly, BCS (2.75;  $P = 0.76$ ) and BCS change ( $-0.05$ ;  $P = 0.74$ ) were not affected by treatment. There was also no effect of treatment on pasture DMI (15.73 kg of DM/cow/d;  $P = 0.99$ ), total DMI (21.05 kg of DM/cow/d;  $P = 0.99$ ), digestibility of NDF (0.69;  $P = 0.34$ ), ADF (0.59;  $P = 0.36$ ) or N (0.33;  $P = 0.70$ ) or the excretion of N in urine (0.26 kg/d;  $P = 0.99$ ). The results from this research show that cereals can be replaced with by-products (DDG, PKE and SH) in the diet of grazing dairy cows without any adverse effects on production, feed intake, digestibility, BW, BCS or N excretion.

**Key Words:** dairy, byproduct, cereal

**854 Effects of feeding Fermenten or urea on milk production, rumen nitrogen metabolism, and microbial nitrogen content in lactating dairy cattle.** Samuel W. Fessenden\*<sup>1</sup>, Andreas Foskolos<sup>1</sup>, Elliot Block<sup>2</sup>, and Michael E. Van Amburgh<sup>1</sup>, <sup>1</sup>*Department of Animal Science, Cornell University, Ithaca, NY*, <sup>2</sup>*Arm & Hammer Animal Nutrition, Princeton, NJ*.

The objective of this study was to evaluate effects of 2 different sources of rumen available nitrogen on cattle performance, rumen nitrogen dynamics, and rumen microbial metabolism in lactating dairy cattle. Eight ruminally cannulated multiparous Holstein cows averaging  $60 \pm 10$  DIM and  $637 \pm 38$  kg of BW were assigned to one of 2 treatment sequences in a switchback design. Diets contained (DM basis) 44% corn silage, 13% alfalfa haylage, 12% ground corn, and 31% protein premix containing either a control mix of urea and wheat middlings (CON) or Fermenten at 3% diet inclusion rate (EXP). Both diets provided similar level (DM basis) of aNDFom (31%), CP (16.5%), RDP (8.2%) and

metabolizable energy (64 Mcal ME/d). Diets were formulated to provide approximately 115% of rumen NH<sub>3</sub>-N requirement as predicted by the CNCPS. The trial consisted of 3 28 d experimental periods, where each period consisted of 21 d of diet adaptation and 7 d of data and sample collection. Digestion markers were infused continuously during the sampling period and composited omasal samples were used to calculate nutrient flows. All data were analyzed using the Proc Mixed procedure in SAS. Dry matter intake was 25.5 and 25.2 kg/d for CON and EXP, respectively ( $P = 0.73$ ). Energy corrected milk yield was 42.2 and 43.2 kg/d for CON and EXP, respectively ( $P = 0.43$ ), with no treatment differences ( $P > 0.05$ ) in milk fat and protein yield or content. Compared with CON, EXP increased milk urea nitrogen (10.7 vs. 13.5 mg/dL;  $P = 0.01$ ) and rumen NH<sub>3</sub>-N (4.4 vs. 5.3 mg/dL;  $P = 0.03$ ). Rumen bacteria and omasal protozoa N content tended to increase with EXP inclusion ( $P < 0.1$ ), while rumen protozoa and omasal bacteria N content increased ( $P < 0.05$ ) with EXP inclusion. Effects of Fermenten inclusion on rumen microbial metabolism, especially microbial N content, were consistent with previous observations of increased microbial growth and turnover associated with supplementation of rumen available non-NH<sub>3</sub>-N in N efficient diets.

**Key Words:** microbial protein, omasum, Fermenten

**855 Effect of passage rate and pH on microbial diversity and total methanogens in continuous culture.** Benjamin A. Wenner\*, Jill A. Stiverson, Zhongtang Yu, and Jeffrey L. Firkins, *Department of Animal Sciences, The Ohio State University, Columbus, OH*.

The present study was conducted as a 2  $\times$  2 factorial treatment arrangement in a Latin square design using 4 continuous culture fermenters. Treatments were control pH (CpH; ranging 6.3 to 6.9) or low pH (LpH; 5.8 to 6.4) factorialized with solids passage rates ( $k_p$ ) set to be either low (Lk<sub>p</sub>; 2.5%/h) or high (Hk<sub>p</sub>; 5.0%/h); total buffer  $k_p$  was constant at 7.0%/h. Fermenters were fed once daily (40 g DM; a 50:50 concentrate:forage diet). Periods lasted 10 d, with 3 d of sample collection. Effluent samples were collected once every 24 h for 3 d and pooled. Fermenter contents were sampled at 0, 4, 8 and 12 h post-feeding on d 6. We hypothesized that lowering pH would limit methanogen ability to grow in culture, and increasing  $k_p$  would challenge methanogens to increase growth rate for survival. Further, the combination of these treatments would decrease daily methane production per total archaeal 16S copy outflow and decrease methanogen concentration in culture. DNA was extracted using repeated bead beating protocols and amplified for total bacteria and total archaeal PCR-DGGE using respective universal 16S rRNA gene primers with GC clamps. Banding analysis clearly demonstrated clustering of effluent versus fermenter samples for bacteria and archaea. However, there were no clear banding patterns for treatment combinations or for times post-feeding. Total archaeal 16S rRNA gene copies were quantified using 787f and 1059r primers using qPCR. There was no significant ( $P > 0.10$ ) interaction for treatment by time, and no difference in total archaeal 16S rRNA (copies/mL) between treatments: CpH, Lk<sub>p</sub>:  $1.13 \times 10^5$ , CpH, Hk<sub>p</sub>:  $5.85 \times 10^4$ , LpH, Lk<sub>p</sub>:  $6.84 \times 10^4$  and LpH, Hk<sub>p</sub>:  $4.67 \times 10^4$ . Daily methane production per 16S copy was not significantly different ( $P > 0.10$ ) between treatments: CpH, Lk<sub>p</sub>: 1.12, CpH, Hk<sub>p</sub>: 0.11 LpH, Lk<sub>p</sub>: 0.67 and LpH, Hk<sub>p</sub>: 0.43 nmol CH<sub>4</sub>/archaeal 16S copy. Rumen microbes are resilient to small daily disturbances in pH or slight increases in  $k_p$ , but there is a lack of clear relationship between methanogen concentration and methane production. The poor methane/copy relationship of these treatments indicates a need to move methane mitigation research beyond inhibition toward potential influencers on methanogen metabolism and growth.

**Key Words:** fermenter, methanogen, methane



**856 Lactational performance and ruminal morphometrics when mid-lactation dairy cows are fed *Saccharomyces cerevisiae* fermentation products.** Jon P. Pretz\*<sup>1</sup>, Subash Acharya<sup>1</sup>, Ilky Yoon<sup>2</sup>, Mark Scott<sup>2</sup>, and David P. Casper<sup>1</sup>, <sup>1</sup>South Dakota State University, Brookings, SD, <sup>2</sup>Diamond V Mills Inc., Cedar Rapids, IA.

This study evaluated *Saccharomyces cerevisiae* fermentation products (Diamond V original XPC and 2 prototypes) on lactational performance and ruminal fermentation. Eight ruminally cannulated (132 DIM and 34.4 kg milk) Holstein dairy cows (2 primiparous and 6 multiparous) were blocked by milk yield, DIM and parity and randomly assigned in a replicated 4 × 4 Latin square design. Treatments were (1) Control (C): corn silage and haylage based ration; (2) XPC: C ration with 14 g/hd/d Original XPC; (3) Prototype 1 (P1): C ration with 5 g/hd/d P1; and (4) Prototype 2 (P2): C ration with 19 g/hd/d P2. Treatments were mixed with dried distillers grains and then mixed in the TMR at 454 g/hd/d. Periods were 28 d with the first 21 d for adjustment followed by 7 d of data collection. Milk yield (3×/d) was recorded and milk samples (2 d) during wk 4. On d 25 or 27, rumens were evacuated, weighed, markers added (Co and valeric acid), mixed, block the rumen-omasal orifice using a sponge, and rumen contents returned to the rumen. Ruminal samples were collected for 4 h at 20 min intervals to determine ruminal pH, ammonia, and volatile fatty acid concentrations. After 4 h of sample collection, rumen contents were re-evacuated, re-weighed, rumen-omasal sponge removed, and rumen contents returned. One cow died unrelated to study objectives and data were removed. Milk yield (30.7, 32.3, 32.0, 31.3 kg/d for C, XPC, P1, and P2, respectively) and intake of DM [(DMI); 24.5, 23.6, 23.6, and 25.3 kg/d] were similar ( $P > 0.10$ ) between all cows, but feed efficiency (1.26, 1.36, 1.36, and 1.24 kg/kg milk/DMI) and energy-corrected milk/DMI (1.42, 1.54, 1.52, and 1.38 kg/kg was greater ( $P < 0.01$ ) for cows fed XPC and P1 compared with cows fed C and P2. Milk composition was similar ( $P > 0.10$ ) between cows fed all rations. Ruminal pH (6.06, 6.07, 6.02 and 6.13) was greater ( $P < 0.05$ ) for cows fed P2 compared with cows fed other rations. The feeding of a dairy ration with a *Saccharomyces cerevisiae* fermentation product can improve ruminal pH and feed efficiency of mid-lactation cows.

**Key Words:** dairy cattle, volatile fatty acid, *Saccharomyces cerevisiae* fermentation product

**857 Effects of nitrate and docosahexaenoic acid on methane production in lactating dairy cows.** G. Klop\*<sup>1</sup>, B. Hatew<sup>1</sup>, A. Bannink<sup>2</sup>, and J. Dijkstra<sup>1</sup>, <sup>1</sup>Wageningen University, Animal Nutrition Group, Wageningen, the Netherlands, <sup>2</sup>Wageningen UR Livestock Research, Wageningen, the Netherlands.

The objective of this experiment was to study the effects of dietary nitrate (NO<sub>3</sub>) and docosahexaenoic acid (DHA; C22:6 n-3) on enteric methane (CH<sub>4</sub>) production in lactating dairy cows. Twenty-eight lactating Holstein dairy cows were grouped into 7 blocks of 4 cows each. Within blocks, cows were randomly assigned to one of 4 treatments: CON (urea as alternative NPN source); NO<sub>3</sub> (21 g/kg DM); DHA (3 g/kg DM and urea as alternative NPN source); or NO<sub>3</sub> + DHA (21 g/kg DM and 3 g/kg DM, respectively). Cows were fed a total mixed ration consisting of 20% grass silage, 50% corn silage and 30% concentrate on a DM basis. Treatments were included in the concentrates. Methane production was measured during a 5-d period in climate respiration chambers after adaptation to the diet for 12 d. Previous to this 17-d period, cows assigned to a treatment including NO<sub>3</sub> were gradually pre-adapted to the treatment dose of NO<sub>3</sub> over a period of 21 d. Preliminary results show a significant ( $P < 0.05$ ) NO<sub>3</sub> × DHA interaction for CH<sub>4</sub> expressed

in g/d, and cows produced on average 368, 264, 369 and 298 g CH<sub>4</sub>/d on treatments CON, NO<sub>3</sub>, DHA and NO<sub>3</sub> + DHA, respectively. This interaction effect is explained by a lower DMI for the NO<sub>3</sub> treatment despite the restricted feeding regimen during the CH<sub>4</sub> measurement period. Per kg DMI, cows receiving NO<sub>3</sub> produced less CH<sub>4</sub> than cows receiving no NO<sub>3</sub> in their diets (17.5 vs. 22.4 g/kg DMI;  $P < 0.01$ ). Feeding DHA did not affect CH<sub>4</sub> production per kg DMI, but did result in a higher ( $P < 0.01$ ) CH<sub>4</sub> production per kg FPCM. Milk production was not affected by treatment, but FPCM was reduced ( $P < 0.05$ ) by DHA because of a reduced milk fat concentration ( $P < 0.01$ ), which averaged 29.6 and 41.1 g/kg for treatments with or without DHA, respectively. Milk protein concentration was reduced ( $P < 0.05$ ) by NO<sub>3</sub> and averaged 30.0 and 31.0 g/kg for treatments with or without NO<sub>3</sub>, respectively. In conclusion, NO<sub>3</sub> but not DHA reduced enteric CH<sub>4</sub> production and there were no interaction effects on CH<sub>4</sub> production expressed in g/kg DMI and g/kg FPCM.

**Key Words:** methane, nitrate, docosahexaenoic acid

**858 Effects of clay (EcoMix) after a grain challenge on rumen health and metabolism of Holstein cows.** Saige A. Sulzberger\*<sup>1</sup>, Carlie C. Kalebich<sup>1</sup>, Sergey Melnichenko<sup>2</sup>, and Felipe C. Cardoso<sup>1</sup>, <sup>1</sup>University of Illinois, Urbana, IL, <sup>2</sup>United Minerals Group, Kyiv, Ukraine.

Oral supplementation of clay has been reported to function as buffer in dairy cows. However, its effects on rumen, blood, and fecal pH have been variable among studies. Our objective was to determine the effects of 3 levels of dietary clay (EcoMix) supplementation after a grain challenge. Ten multiparous rumen-cannulated Holstein cows (BW = 648 kg ± 12kg), 142 (60–502) DIM, were assigned to 1 of 5 treatments in a completely randomized replicated 5 X 5 Latin Square. Periods consisted of an 18-d adaptation period followed by a 3-d (d 19–21) measurement period. Cow average DMI from d 15–17 was used to restrict feed to 75% on d 18 and to give 20% wheat flour (grain challenge) on d 19, via cannula. Treatments were: CON, no EcoMix and no grain challenge; POS, no EcoMix with a grain challenge; 0.5%, 1%, and 2%, EcoMix as percentages of dietary DMI all with a grain challenge. Statistical analysis was performed using the MIXED procedure of SAS. Two contrasts CONT1 (POS vs. CON), CONT2 (POS vs. average of 0.5%, 1%, and 2%) were compared along with the linear and quadratic treatment effects. Rumen, fecal, and blood pH along with blood metabolites were measured at 0, 4, 8, 12, 16, 20, 24, 36, and 48 h relative to grain challenge. Rumen pH ( $P = 0.003$ ) and fecal pH were lower (CONT1,  $P < 0.001$ ) for POS (6.03 ± 0.06; 6.14 ± 0.04) than CON (6.20 ± 0.06; 6.38 ± 0.04), respectively. There was a linear treatment effect for rumen pH ( $P = 0.001$ ) and fecal pH ( $P = 0.05$ ). Fecal pH (6.22 ± 0.04,  $P < 0.0001$ ) was higher for cows that received EcoMix than POS (6.14 ± 0.04, CONT2). There was a difference for negative incremental area under the curve (Rumen pH below 5.6/24h; 0.5% = 7.93 1% = 8.56, 2% = 7.79, SEM = 0.8,  $P = 0.005$ ) when compared with POS (11.0, CONT2). Cows fed EcoMix had higher milk yield, (0.5% = 28.8 kg, 1% = 30.2 kg, 2% = 29.1 kg, SEM = 3.4), 3.5% FCM (0.5% = 29.9 kg, 1% = 34.1 kg, 2% = 33.1 kg, SEM = 3.4,  $P = 0.02$ ) and ECM (0.5% = 29.1 kg, 1% = 32.8 kg, 2% = 31.6 kg, SEM = 3.3  $P = 0.01$ ) than cows in POS (CONT2, 27.72 kg, 28.0 kg, 27.71kg, respectively). In conclusion, cows that received EcoMix had higher rumen pH, milk yield, ECM and FCM than cows in POS.

**Key Words:** buffer, clay, rumen pH

**859 Magnitude of difference in chemical and nutrient profiles, ruminal degradation kinetics, and intestinal digestion of three different types of co-products from bio-oil processing for dairy cattle.** Xinxin Li<sup>\*1,2</sup>, Yonggen Zhang<sup>1</sup>, and Peiqiang Yu<sup>1,2</sup>, <sup>1</sup>College of Animal Science and Technology, Northeast Agricultural University, Harbin, China, <sup>2</sup>Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, Saskatchewan, Canada.

Co-products from bio-oil industry, rapeseed meal, canola meal and soybean meal, are good sources of feed protein for livestock. The objectives of this study were to determine the magnitude of differences among 3 types of co-products in terms of (1) protein nutrient profile, (2) protein subfractions associated rumen utilization, (3) protein rumen degradation kinetics and (4) intestinal absorbed true protein supply to dairy cattle. Rapeseed meal, canola meal and soybean meal were collected from 3 different sources in 2014. Crude protein was partitioned into 5 subfractions according to the Cornell net carbohydrate and protein system (CNCPS 6.5). Three lactating Holstein cows fitted with permanent rumen cannula were used for determining the protein rumen kinetics in the in situ trial. The estimation of intestinal rumen undegraded feed protein digestibility was determined using a modified 3-step in vitro procedure with 12 h pre-rumen incubation in lactating Holstein cows. In this study, statistical analyses were carried out using the PROC MIXED procedure of SAS 9.3. Significances were declared at  $P < 0.05$ . The results showed that CP content in soybean meal was higher than rapeseed meal and canola meal ( $P < 0.05$ ). Rapeseed meal had the highest NPN content, followed by canola meal with NPN content lowest in soybean meal ( $P < 0.05$ ). The contents of SCP, ADICP and NDICP in rapeseed meal and canola meal were higher than soybean meal ( $P < 0.05$ ). Protein fractions of PA<sub>2</sub>, PB<sub>2</sub> and PC were greater in rapeseed meal and canola meal than soybean meal ( $P < 0.05$ ), but no significant difference was found between rapeseed meal and soybean meal. Both soybean meal and rapeseed meal had higher rumen degradable protein (RDP) than canola meal ( $P < 0.05$ ). Soybean meal was greater than rapeseed meal and canola meal in intestinal digestible protein (IDP,  $P < 0.05$ ). Total digestible protein (TDP) was highest in soybean meal, and lowest in canola meal ( $P < 0.05$ ). Overall, the results indicated that soybean is a greater source of protein feed than canola meal and rapeseed meal.

**Key Words:** soybean meal, rapeseed meal, degradation profile

**860 Amino acid profiles of ruminal microbes, ruminal undegradable protein, and gastrointestinal contents in lactating dairy cows when corn stover or rice straw replaces alfalfa hay.** Bing Wang<sup>\*1,2</sup> and J. X. Liu<sup>1,2</sup>, <sup>1</sup>Institute of Dairy Science, College of Animal Sciences, Hangzhou, P. R. China, <sup>2</sup>MoE Key Laboratory of Molecular Animal Nutrition, Zhejiang University, Hangzhou, P.R. China.

The objective of this study was conducted to evaluate the effects of replacing alfalfa hay with rice straw or corn stover on amino acid (AA) profiles of ruminal microbes, rumen undegradable protein (RUP), and gastrointestinal contents in lactating cows. Eighteen Holstein dairy cows were individually fed, and randomly assigned into one of 3 treatments. Isonitrogenous diets contained similar concentrate and 15% corn silage, with 3 forage sources (DM basis): 23% alfalfa hay and 7% Chinese wild rye hay (AH); 30% corn stover (CS); and 30% rice straw (RS). After 14-wk feeding, all the cows were slaughtered to collect the samples of

rumen fluid and gastrointestinal contents. The AA were analyzed with an AA analyzer (Model L-8800, Hitachi, Tokyo, Japan). The variance of the data was analyzed as a completely randomized design using PROC MIXED of SAS. The AA profiles of ruminal microbes were similar among 3 treatments except for the lower value of Asp in diet RS than in CS ( $P < 0.01$ ) or AH ( $P < 0.02$ ) and the greater value of Gly in AH than in RS ( $P = 0.02$ ). Significant differences were found in the AA profiles among ruminal microbes, fluid, and digesta, with greater percentage of essential AA in digesta than in microbes or fluid ( $P < 0.01$ ), and in microbes than in rumen fluid ( $P < 0.01$ ). The free AA concentration in jejunum was the greatest, but the peptide-binding AA concentration was the greatest in duodenum, resulting in lower ratio of peptide-binding AA to free AA. In addition, the free essential AA profiles in jejunum fluids for all the 3 diets were similar with the recommended digestible AA and milk AA, with the ratio of Lys to Met at 3:1. Most of AA of RUP was independent on forage sources except for Phe ( $P = 0.035$ ) and Gly ( $P = 0.05$ ). There was linear correlation between dietary AA profile and AA composition in ruminal fluids ( $P < 0.01$ ). The AA profiles of ruminal microbes and RUP were constant and independent on forage sources. It is indicated that the AA profile of metabolizable protein may be constant independent on dietary effects.

**Key Words:** amino acid profile, dairy cattle, gastrointestinal content

**861 Effect of increasing concentration of dietary fiber in diets rich in plant oil on milk fat concentration, rumen parameters and feeding behavior of mid-lactating cows.** H. R. Mirzaei Alamouti\* and A. Aghaei, Department of Animal Science, University of Zanjan, Zanjan, Iran.

This experiment was conducted to investigate the effects of different levels of dietary NDF in diets rich in plant oil (sunflower and soybean) on feeding behavior, rumen parameters, milk yield and components of mid-lactating Holstein dairy cows. Four primiparous (BW: 525 ± 30 kg; days-in-milk: 103 ± 6) and 4 multiparous (BW: 587 ± 88 kg; DIM: 99 ± 12) cows were used in a 4 × 4 replicated Latin square design with 21-d experimental periods. Cows were received 1 of 4 dietary treatments: (1) 31% fiber and no supplement plant oil, LFNO; (2) 31% fiber with 3% supplement plant oil, LFHO; (3) 35% fiber with 3% plant oil, MFHO; (4) 39% fiber with 3% plant oil, HFHO. Daily dry matter intake (DMI), milk yield and composition, blood metabolites, dry matter digestibility, rumen fluid characteristics and BW variations were determined. There was no significant difference in milk production among diets. Milk fat ( $P < 0.01$ ) and protein ( $P < 0.05$ ) concentrations were significantly affected by treatments. Dry matter intake and DM digestibility were higher ( $P < 0.01$ ) in LFHO diet. The cows fed HFHO had higher NDF digestibility ( $P < 0.01$ ). Total VFA and acetate concentration were greater for HFHO diet and propionate concentration was greater for LFHO diet ( $P < 0.01$ ) than the others. Rumen fluid pH was increased by increasing dietary NDF concentration ( $P < 0.01$ ). Plasma insulin ( $P < 0.01$ ) and cholesterol ( $P < 0.05$ ) concentration were affected by diets. Chewing activity was positively affected by increasing concentration of dietary NDF ( $P < 0.01$ ). This study showed that diets rich in plant oil and low concentration of NDF induce the milk fat depression in mid-lactating cows, and with increasing dietary NDF concentration severity of milk fat depression can be alleviated.

**Key Words:** feeding behavior, oil supplementation, dietary fiber

## Ruminant Nutrition: Feedlot nutrition

**862 Comparison of methods to calculate metabolizable protein requirements of growing beef cattle.** Andrea K. Watson\*<sup>1</sup>, Terry J. Klopfenstein<sup>1</sup>, Vic A. Wilkerson<sup>2</sup>, Galen E. Erickson<sup>1</sup>, and Jim C. MacDonald<sup>1</sup>, <sup>1</sup>University of Nebraska, Lincoln, NE, <sup>2</sup>Purina Animal Nutrition, Forest Grove, OR.

Data from 3 trials were compiled to calculate microbial CP (MCP) production and MP requirements of growing calves on high forage diets. Individually fed steers (n = 335; 256 ± 15.6 kg midpoint BW) were utilized, each trial lasted 84. Diets consisted of 44% sorghum silage, 44% corn cobs, and 12% protein supplement. Source of protein within the supplement varied and included urea, meat and bone meal, soybean meal, feather meal, poultry by-product meal, or corn gluten meal. All trials included a urea only treatment. Dry matter intake of all calves within a trial was held constant, as a percent of BW, established by the urea supplemented group. The base diet was MP deficient, composition of the protein supplement varied with increasing amounts of test protein replacing urea. As protein in the diet increased, ADG plateaued. This methodology was used by Wilkerson et al. (1993) and is the basis of performance models used by the NRC (1996). Gain ranged from 0.19 to 0.56 kg/d, averaging 0.37 kg/d. Three microbial efficiencies were used to calculate MP. Maximum gain was then regressed against calculated MP to determine MP requirement for maintenance and gain. Method 1 (based on a constant 13% microbial efficiency suggested by the NRC, 1996) predicted an MP requirement of 3.4 g/kg BW<sup>0.75</sup> for maintenance and 461 g/kg gain (r<sup>2</sup> = 0.55). Method 2 (based on an equation developed by Patterson et al., 2006) predicted MP requirements of 2.9 g/kg BW<sup>0.75</sup> and 483 g/kg gain (r<sup>2</sup> = 0.56). Method 3 (based on an equation developed by Galyean et al., 2014) predicted MP requirements of 2.6 g/kg BW<sup>0.75</sup> and 449 g/kg gain (r<sup>2</sup> = 0.59). The factorial method of calculating MP maintenance requirements accounts for scurf, endogenous urinary, and metabolic fecal protein losses and averaged 4.2 ± 0.10 g/kg BW<sup>0.75</sup>, for the 3 trials summarized here. Factors affecting MCP production include TDN, RDP, and microbial efficiency. Dietary TDN and RDP have been measured on a variety of feedstuffs; microbial efficiency is not well defined and is a crucial component in calculating MCP production and MP requirements of growing cattle.

**Key Words:** beef cattle, metabolizable protein, microbial crude protein

**863 The effect of zilpaterol hydrochloride supplementation on gain efficiency, harvest yields and carcass grading of steers fed a maintenance intake.** Lee-Anne J. Walter\*<sup>1</sup>, N. Andy Cole<sup>2</sup>, Jenny S. Jennings<sup>3</sup>, John P. Hutcheson<sup>4</sup>, Beverly E. Meyer<sup>2</sup>, Angela N. Schmitz<sup>1</sup>, DeMetris D. Reed<sup>1</sup>, and Ty E. Lawrence<sup>1</sup>, <sup>1</sup>West Texas A&M University, Canyon, TX, <sup>2</sup>USDA ARS, Bushland, TX, <sup>3</sup>Texas A&M AgriLife Research and Extension Center, Amarillo, TX, <sup>4</sup>Merck Animal Health, Summit, NJ.

A trial was conducted to examine gain efficiency, harvest yields and grading characteristics of steers fed only to maintenance intake (MI) and supplemented with zilpaterol hydrochloride (ZH). Beef steers (n = 20; 463 ± 14 kg) blocked (n = 5) by weight and source were individually fed and adapted to MI  $\{[(BW \times 0.891)^{0.75} \times 0.077] / \text{diet NE}_m, \text{BW d} - 1 \text{ and } 1\}$  for 21 d before ZH (90 mg/hd/d) or non-ZH treatment for 20 d (455 ± 14 kg, SOT). During the 21d maintenance period, MI was adjusted based on BW (d -1, 1 vs. 4, 5, 9, 10, 14, 15, 19 and 20) after 10h water and feed withdrawal (no feed remained for any animals at start of with-

drawal periods). Intakes were not adjusted during 20 d treatment period but steers were weighed on d1, 2, 11, 12, 17, and 18 of ZH treatment and on d 1, 2, 5, and 6 of withdrawal. Cattle were harvested after a 6 d withdrawal and carcasses graded 24 h post-harvest. Data were analyzed as a mixed model with fixed effect of ZH and random effect of block. Gain efficiency, ADG and end of ZH BW was not different between treatments (P > 0.10). Control cattle lost more BW (P < 0.01; 9 vs. 2 kg, respectively) than ZH fed cattle during the maintenance collection period (d12–16 of ZH) while BW loss of ZH fed cattle was greater (P < 0.01; 9 vs. 4 kg, respectively) during fasting heat production period (FHP; d18–19 of ZH, 4-d total fast). Harvest BW, exsanguinated BW, empty body weight (EBW) and harvest yields (g/kg EBW) were not different between treatments (P > 0.10). Supplementation of ZH did not improve HCW (P = 0.12) but did increase dressed carcass yield (P = 0.02; 62.12 vs. 60.65%, respectively). Feeding ZH improved LM area (P = 0.02; 77.81 vs. 70.90 cm<sup>2</sup>) and tended to lower USDA calculated yield grade (P = 0.06; 1.8 vs. 2.2). Marbling, 12th rib fat depth and calculated empty body fat did not differ (P > 0.10) between treatments. Results from this trial indicate that ZH supplementation alters carcass traits in steers fed reduced energy intakes by improving dressed carcass yield and LM area (P = 0.02) and exhibiting a tendency to improve yield grade (P = 0.06).

**Key Words:** feed efficiency, carcass grading, zilpaterol

**864 The effect of zilpaterol hydrochloride supplementation on apparent nutrient digestibility and carbon-nitrogen retention of steers fed at maintenance intake.** Lee-Anne J. Walter\*<sup>1</sup>, N. Andy Cole<sup>2</sup>, Jenny S. Jennings<sup>3</sup>, John P. Hutcheson<sup>4</sup>, Beverly E. Meyer<sup>2</sup>, Angela N. Schmitz<sup>1</sup>, DeMetris D. Reed<sup>1</sup>, and Ty E. Lawrence<sup>1</sup>, <sup>1</sup>West Texas A&M University, Canyon, TX, <sup>2</sup>USDA ARS, Bushland, TX, <sup>3</sup>Texas A&M AgriLife Research and Extension Center, Amarillo, TX, <sup>4</sup>Merck Animal Health, Summit, NJ.

A trial was conducted to examine apparent nutrient digestibility and carbon (C) -nitrogen (N) retention of cattle supplemented zilpaterol hydrochloride (ZH). Beef steers (n = 20; 463 ± 14 kg) blocked (n = 5) by weight and source were individually fed and adapted to maintenance intake (MI;  $\{[(BW \times 0.891)^{0.75} \times 0.077] / \text{diet NE}_m, \text{BW d} - 1 \text{ and } 1\}$ ) for 21 d before ZH (90 mg/hd/d) or non-ZH treatment for 20 d (455 ± 14 kg, SOT). During the 21d maintenance period, MI was adjusted based on BW (d -1, 1 vs. 4, 5, 9, 10, 14, 15, 19 and 20) after 10h water and feed withdrawal (no feed remained for any animals at start of withdrawal periods). Intakes were not adjusted during 20 d treatment period. Feces and urine was collected at MI (d12–16 of ZH period) with daily feed and fecal samples analyzed for proximate analysis, ADF, NDF, starch and C; urine was analyzed for C and N. Data were analyzed as a mixed model with fixed effect of ZH and random effects of block and chamber. Dry matter (DM) intake, DM digestibility, fecal or urine output did not differ (P > 0.10) between treatments. Intake, fecal excretion and apparent digestibility of NDF, starch or ether extract did not differ (P > 0.10) between treatments. Control cattle excreted more (P < 0.05) N in urine (39.8 vs. 32.4 g/d, respectively) and tended (P < 0.07) to excrete more N daily than ZH treated cattle. Cattle fed ZH tended to have increased absolute N retention (P = 0.07; 22.14 vs. 14.12 g/d) and increased N retention as a percentage of digested N (P = 0.06; 39.73 vs. 25.49%) while apparent N digested did not differ (P > 0.10) between treatments. Total C loss via, urine, fecal, or CH<sub>4</sub> did not differ (P > 0.10) between



treatments but ZH treated cattle lost more C via CO<sub>2</sub> than control cattle ( $P = 0.04$ ; 1036.9 vs. 974.3 g, respectively). Total C digested and retained did not differ between treatments ( $P > 0.10$ ). Results from this trial indicate that ZH treatment does not alter apparent digestibility of nutrients or C retention but does tend to increase N retention ( $P = 0.07$ ) while increasing C loss through CO<sub>2</sub> production ( $P = 0.04$ )

**Key Words:** beef, carbon nitrogen retention, zilpaterol

### 865 The effect of zilpaterol hydrochloride supplementation on energy metabolism of steers at maintenance and fasting intake levels.

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An indirect calorimetry trial examined the energetic efficiency of cattle supplemented zilpaterol hydrochloride (ZH). Beef steers ( $n = 20$ ;  $463 \pm 14$  kg) blocked ( $n = 5$ ) by weight and source were individually fed and adapted to maintenance intake (MI;  $\{[(BW \times 0.891)^{0.75} \times 0.077] / \text{diet NE}_m, \text{ BW d} - 1 \text{ and } 1\}$ ) for 21 d before ZH (90 mg/hd/d) or non-ZH treatment for 20 d ( $455 \pm 14$  kg, SOT). During the 21d maintenance period, MI was adjusted based on BW (d - 1, 1 vs. d 4, 5, 9, 10, 14, 15, 19 and 20) after 10h water and feed withdrawal (no feed remained for any animals at start of withdrawal periods). Intakes were not adjusted during 20 d treatment period. Respiration chambers were used to quantify energetics for maintenance (d12–16 of ZH period) and fasting heat production (FHP; d19–20 of ZH period, total 4d fast). Daily feed and fecal samples were analyzed for gross energy (GE); urine was analyzed for nitrogen and GE was calculated from the energetic value of urea. Data were analyzed as a mixed model with fixed effect of ZH treatment and random effects of block and chamber. No difference in DMI, O<sub>2</sub> consumption or CH<sub>4</sub> production ( $P > 0.10$ ) was detected between treatments but ZH cattle had higher CO<sub>2</sub> production ( $P = 0.04$ ; 2325 vs. 2185 L/steer; 23.6 vs. 22.4 L/kg BW<sup>0.75</sup>). During FHP, O<sub>2</sub> consumption was not affected ( $P > 0.10$ ) by treatment whereas CO<sub>2</sub> production (L/steer) increased with ZH treatment ( $P = 0.04$ ; 1423 vs. 1338 L/steer). Gross energy, fecal energy, digestible energy and methane energy did not differ between treatments ( $P > 0.10$ ), however urinary energy was higher ( $P = 0.05$ ; 0.091 vs. 0.074 Mcal) in control cattle. There was no difference in metabolizable energy between treatments ( $P > 0.10$ ). Heat production tended ( $P = 0.09$ ) to increase for ZH treated cattle (12.44 vs. 11.69 Mcal, respectively) on an absolute basis, but not on a BW<sup>0.75</sup> basis ( $P = 0.12$ ; 0.126 vs. 0.120, respectively). No difference in FHP was detected between treatments ( $P = 0.32$ ). Results from this trial indicate that ZH treatment tends ( $P = 0.09$ ) to modify heat production during maintenance by increasing CO<sub>2</sub> production ( $P = 0.04$ ).

**Key Words:** beef, energy metabolism, zilpaterol

### 866 Effects of chromium propionate in combination with yeast on growth performance and carcass quality of finishing steers.

Cadra L. Van Bibber-Krueger<sup>\*</sup>, Justin E. Axman, John M. Gonzalez, and Jim S. Drouillard, Kansas State University, Manhattan, KS.

A combination of chromium propionate and yeast supplement (CrY; TruMax, Varied Industries Corp., Mason City, IA) was added to diets of crossbred steers ( $n = 504$ ;  $402 \pm 5.8$  kg initial BW) to evaluate the effect on feedlot performance and carcass quality. Steers were selected on the basis of initial plasma glucose concentrations  $\leq 6.0$  mM, strati-

fied by initial BW, blocked into heavy (H; 420 kg BW) and light (L; 384 kg BW) groups, and assigned to treatments of 0 (CON) or 3.3 g/d CrY (provided 0.5 mg Cr/kg diet DM) with 6 pens/diet within each block. Body weights were measured at 21-d intervals. Finished cattle were weighed and transported to a commercial abattoir where severity of liver abscesses and HCW were collected at harvest, and yield and quality grades, LM area, and 12th rib subcutaneous fat thickness were determined after 36 h of refrigeration. There were no CrY  $\times$  time  $\times$  weight block interactions for ADG, DMI, and G:F ( $P = 0.36$ ;  $P = 0.49$ ;  $P = 0.41$ , respectively), and no CrY  $\times$  block interactions for ADG and DMI ( $P = 0.06$ ;  $P = 0.75$ , respectively), but a CrY  $\times$  block interaction ( $P = 0.03$ ) was observed for G:F; CON-L had poorer efficiency compared with other groups. Yield grade and marbling score tended to be less ( $P > 0.07$ ) for CrY compared with CON, but other carcass measurements were unaffected by treatment ( $P > 0.30$ ). Chromium propionate in combination with yeast may improve feed efficiency of lighter cattle, but other effects were minimal.

**Table 1 (Abstr. 866).** Feedlot performance and carcass traits of steers

Item	CON-L	CrY-L	CON-H	CrY-L	SEM
DMI, kg/d	12.57	12.29	13.00	12.83	0.17
ADG, kg/d	1.70	1.76	1.88	1.84	0.03
G:F <sup>1,3</sup>	0.1349	0.1430	0.1445	0.1434	0.0023
HCW, kg	398	400	408	407	3.65
LM area, cm <sup>2</sup>	88.13	90.06	93.10	92.97	0.96
12th rib fat, cm	1.30	1.22	1.27	1.24	0.43
Yield grade <sup>2,3</sup>	2.79	2.53	2.66	2.66	0.07
Marbling score <sup>3,4</sup>	459	453	430	414	6.54

<sup>1</sup>CrY  $\times$  weight block interaction ( $P = 0.03$ ).

<sup>2</sup>CrY  $\times$  weight block interaction ( $P = 0.08$ ).

<sup>3</sup>Effect of CrY ( $P \leq 0.08$ ).

<sup>4</sup>Marbling score determined by camera imaging; Small = 400 to 499.

**Key Words:** beef cattle, chromium propionate, glucose

### 867 The influence of supplemental Zn-amino acid complex and Optaflexx feeding duration on growth performance and carcass characteristics of finishing beef cattle.

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Previous research indicates that finishing steer ADG and G:F increases linearly with increasing dietary Zn-amino acid complex supplementation. The objective of this study was to determine the influence of supplemental Zn-amino acid complex on growth performance and carcass characteristics of finishing steers fed Optaflexx (OPT; ractopamine hydrochloride) for 0, 28, or 42 d before harvest. This study was organized as 2 groups (Grp) of steers fed concurrently, for 91 (Grp 1) or 84 d (Grp 2). A total of 324 steers ( $463 \pm 23.4$  kg) were fed a corn-based finishing diet supplemented with 60 mg Zn/kg diet DM (as ZnSO<sub>4</sub>). Steers were blocked by weight (6 steers per pen) and assigned to receive either 0 (CON) or 60 mg supplemental Zn/kg DM from a Zn amino-acid complex (ZnAA;  $n = 27$  pens per treatment). Receiving ZnAA for 49 (Exp 1) or 42 d (Exp 2) before start of OPT feeding had no effect on growth of steers ( $P \leq 0.30$ ). Forty-two days before harvest, pens were equally assigned within CON or ZnAA treatments to receive OPT at 300 mg·steer<sup>-1</sup>·d<sup>-1</sup> for 0, 28 (28-OPT) or 42 d (42-OPT) before harvest, creating 6 final treatments ( $n = 9$  pens per treatment). All steers within an experiment were harvested on the same day. Pen was the experimental unit, and the SAS model included the fixed effects of

ZnAA, OPT, and block nested within group, and the random effect of pen. Optaflexx supplementation increased carcass-adjusted ADG ( $P = 0.004$ ), final BW ( $P = 0.004$ ), HCW ( $P = 0.004$ ), and ribeye area ( $P = 0.007$ ). There was an effect of ZnAA within 28-OPT and 42-OPT where carcass-adjusted ADG ( $P \leq 0.10$ ), final BW ( $P \leq 0.05$ ), and HCW ( $P \leq 0.05$ ) were greater in ZnAA supplemented vs. CON steers. However, when steers did not receive OPT there was no effect of ZnAA on final BW ( $P = 0.78$ ), ADG ( $P = 0.98$ ), or HCW ( $P = 0.78$ ). In conclusion, there appears to be a synergistic effect of ZnAA on OPT-induced cattle growth, as supplementing 60 mg Zn/kg DM from ZnAA to cattle fed OPT improved overall growth and HCW.

**Key Words:** beef cattle, Optaflexx, zinc

### 868 Residual feed intake in ad libitum and limit-fed steers.

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To determine if nutritional restriction alters the fundamental relationships between intake, growth and efficiency, 60 Angus-Hereford steers ( $310 \pm 33$  d of age,  $263 \pm 33$  kg BW) were randomly assigned to 2 intake groups (IG): ad libitum (AL) or limit-fed (LF, 80% of ad libitum) and fed in individual pens. Intakes were monitored weekly, and steers were weighed monthly. Steers were harvested when they reached a minimum of 12.5 mm backfat, determined by ultrasound at each weighing. Carcass composition was determined from specific gravity. Residual feed intake (RFI) was calculated as the residual of the regression of DMI on average metabolic BW and ADG, with intake group included as a fixed effect; slopes were not found to be heterogeneous ( $P > 0.05$ ). The model was:  $DMI$  (kg/d) =  $IG + 0.0737 BW^{0.75} + 3.296 ADG$ , where  $IG = -0.12$  (AL) or  $-1.11$  (LF). RFI groups were defined as Low (RFI  $< -0.5$  SD from mean), Medium ( $-0.5$  SD  $< RFI < 0.5$  SD) or High (RFI  $> 0.5$  SD). Data were analyzed by regression and ANOVA, including IG and RFI groups as main effects; no interactions were significant. (Minitab Inc., State College, PA). Days on feed were greater ( $P < 0.0001$ ) in LF (198 d) than in AL (175 d) steers, however final BW were not different ( $P > 0.10$ ) between AL and LF groups. DM intakes were lower ( $P < 0.0001$ ) in LF than in AL steers, both in absolute and relative terms (8.71 and 6.60 kg/d and 2.37 and 1.93% of BW, respectively). RFI groups also differed ( $P < 0.001$ ) in DMI, both in absolute and relative terms (8.08, 7.91 and 6.97 kg/d and 2.29, 2.16 and 1.99% of BW, for High, Medium and Low RFI groups, respectively). Gain:feed was different among IG (0.0952 and 0.0868 for AL and LF, respectively,  $P = 0.003$ ) and RFI groups (0.0806, 0.0888 and 0.1036 for High, Medium and Low RFI, respectively;  $P < 0.001$ ). Carcass fat contents were not different ( $P > 0.10$ ) among RFI groups, but tended to be lower ( $P = 0.064$ ) in LF than in AL (28.3 and 30.3%, respectively). These results confirm that nutritional restriction limits rate and efficiency of gain and prolongs the finishing period in beef cattle, and tends to produce leaner carcasses. The lack of any difference in the slopes of the equation used to predict intake between ad libitum- vs. limit-fed steers indicate that feed restriction did not alter the fundamental relationships between intake, metabolic weight and rate of gain.

**Key Words:** efficiency, limit-fed, steer

### 869 Behavioral evaluation when using wet corn gluten feed or wet distillers grains plus solubles to adapt cattle to finishing diets.

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Behavioral responses to adaptation diets using wet corn gluten feed branded Sweet Bran (SB-WCGF) or wet distillers grains plus solubles (WDGS) were evaluated. Six ruminally cannulated steers ( $300 \pm 22$  kg) at 11 mo of age were assigned randomly using a CRD experiment to one of 2 adaptation strategies including either SB-WCGF or WDGS. Steers were fed a series of 6 diets: 4 adaptation steps, a finishing, and a finishing blend diet. The first step included 87.5% DM of either SB-WCGF or WDGS, with 0% dry rolled corn (DRC), and was reduced to 35% of the finishing diet over a period of 4 steps (7 d each), and increasing the level of DRC to 52.5%. Diets also included 7.5% alfalfa hay and 5% supplement. Blend diet (50:50) contained WDGS and SB-WCGF (17.5% of each, DM basis). Behavioral status (24 h) was recorded by video cameras strategically located on top of individual pens. Behavior was evaluated every 5 min during d-4 of each period, and it was noted whether the steers were resting or ruminating, as well as standing up or lying down, eating, or drinking. Data were analyzed using the GLIMMIX procedures of SAS, and blend diet was used as a covariate. Steers fed SB-WCGF strategy spent more time ( $P < 0.10$ ) ruminating while lying down than WDGS in steps 1 and 4 (223 vs. 93; 289 vs. 77 min/d, respectively); and tended ( $P \leq 0.15$ ) for similar pattern for total rumination in steps 1, 2, and 4 (259 vs. 105; 323 vs. 129; and 321 vs. 65 min/d, for SB-WCGF and WDGS strategies, respectively). Steers fed SB-WCGF strategy also spent more time ( $P < 0.10$ ) ruminating per percentage unit of NDF in step 4 (11.38 vs. 2.29 min-d/%NDF, respectively), as well as tended ( $P \leq 0.15$ ) to spend more time chewing (ruminating plus eating activities) than WDGS strategy in step 4 (418 vs. 227 min/d). Steers fed WDGS strategy had greater ( $P < 0.10$ ) time resting while standing up in steps 2 and 3 (267 vs. 174; 283 vs. 211 min/d, respectively). Overall, the SB-WCGF adaptation strategy shows a more desirable rumination pattern during adaptation to DRC-based finishing diets in feedlot steers than strategy using WDGS.

**Key Words:** adaptation, behavior, coproduct

### 870 Effects of *Megasphaera elsdenii* on ruminal pH, VFA, and lactate during transition from 60 to 80% concentrate diet. Jake D. Thieszen\*<sup>1</sup>, Cadra L. Van Bibber-Krueger<sup>1</sup>, Justin E. Axman<sup>1</sup>, Celine C. Aperce<sup>2</sup>, James S. Drouillard<sup>1</sup>, and Kevin A. Miller<sup>2</sup>, <sup>1</sup>Kansas State University, Manhattan, KS, <sup>2</sup>MS Biotec, Wamego, KS.

Our objective of this study was to evaluate changes in ruminal pH, concentrations of VFA and lactate in cattle during a transition from 60% to 80% concentrate diet. *Megasphaera elsdenii* culture (ME; Lactipro, MS-Biotec, Wamego, KS) was administered by oral drench to crossbred heifers (n = 240;  $498 \pm 18$  kg initial BW) at rates of 0, 25, 50, 75, or 100 mL immediately before transition from 60 to 80% concentrate diet. Cattle were fed in pens of 8 head each, and ruminal contents of animals in one pen/treatment were obtained via rumenocentesis for estimation of ruminal pH and concentrations of VFA and lactate at 5, 10, 15, 20, 25, and 30 h after dosing and introduction of the 80% concentrate diet. Lactate concentrations were unaffected by treatment and were  $< 1$  mM ( $P > 0.10$ ). Differences in pH and VFA concentrations were not apparent until 20 to 25 h after dosing, and were characterized by lower propionate production and increased A:P ratios for the 75- and 100-mL dosages compared with other treatments. *Megasphaera elsdenii* may help to avoid pH decline frequently associated with transition to high-concentrate diets.

Contd.

**Table 1 (Abstr. 870).** Ruminal pH in heifers receiving an oral drench of *Megasphaera elsdenii* (ME) culture (0, 25, 50, 75, or 100 mL)

Hour	ME, mL					SEM	P-value
	0	25	50	75	100		
5	6.03	5.80	6.08	5.73	5.89	0.101	0.06
10	5.83	5.78	5.83	5.82	5.78	0.101	0.80
15	6.02	6.10	5.95	6.16	5.93	0.095	0.36
20	5.91 <sup>ac</sup>	6.13 <sup>bc</sup>	5.76 <sup>a</sup>	5.93 <sup>abc</sup>	6.20 <sup>b</sup>	0.101	0.01
25	6.23 <sup>a</sup>	6.12 <sup>a</sup>	6.24 <sup>ab</sup>	6.58 <sup>bc</sup>	6.59 <sup>c</sup>	0.101	0.01
30	5.54	5.60	5.72	5.61	5.75	0.095	0.47

<sup>abc</sup>Within a row, means without a common superscript differ ( $P < 0.05$ ).

**Key Words:** *Megasphaera elsdenii*, feedlot, acidosis.

**871 Dry matter intake patterns of feedlot cattle.** R. B. Hicks<sup>1</sup>, R. P. Lake<sup>2</sup>, and F. N. Owens<sup>\*3</sup>, <sup>1</sup>Oklahoma State University, Goodwell, OK, <sup>2</sup>Hitch Consulting Services, Guymon, OK, <sup>3</sup>DuPont Pioneer, Johnston, IA.

Most intake equations for feedlot cattle strive to predict mean intake for the total feeding period. Yet, DMI of feedlot cattle typically plateaus after 2 to 4 wk on feed and declines steadily thereafter. Consequently, dietary requirements change over time. To quantify intake patterns, weekly DMI data were compiled from 2,329 pens (minimum of 50 steers per pen) fed high concentrate diets an average of 158 d in one southern Great Plains feedlot. Intake of DM differed with initial weight ( $P < 0.01$ ), so pens were grouped by initial weight (216 to 443 kg) into 10 sets. Adjusted for death loss and sick pen days, DMI averaged 9.56 kg/d. Within weight groups, DMI later (wk 8 to 17) was correlated ( $R^2 = 0.77$ ) with DMI earlier (wk 4 through 7). Based on DMI and steer weights, ME realized by each pen of steers was calculated (mean ME =  $3.45 \pm 0.18$  Mcal/kg). Based on initial weight and ME intake, mean weight of steers within each pen each week was calculated. Following 3 wk for diet adaptation, Intake of DM as a percentage of mean weight (DMI<sub>pc</sub>) was lower for pens of steers with greater initial weight and decreased linearly over time (DMI<sub>pc</sub> =  $2.954 - 0.04776 \times \text{week on feed} - 0.000859 \times \text{initial weight, kg}$ ); calculated DMI was related to observed DMI ( $R^2 = 0.79$ ; RMSE = 0.61 kg). Pens of cattle with greater initial weights had greater DMI and ADG but lower DMI<sub>pc</sub> and feed-to-gain ratios. Adjusted for initial weight, and DMI<sub>pc</sub>, expressed as a fraction of current body weight, was greater ( $P < 0.05$ ) during October and November than in January through May or during a summer intake slump (July and August). The dietary CP percentage required, assuming a 64% efficiency of retention of CP intake, added to inevitable N losses was greater for lighter cattle, peaked early, and declined steadily (CP =  $17.1 - 0.1407 \times \text{week} - 0.0142 \times \text{initial weight}$ ;  $R^2 = 0.80$ ). In conclusion, later DMI can be predicted from DMI early in a feeding period. Because DMI and performance vary with initial weight and time on feed, data on interim pen weights, DMI, and ADG should permit nutrient requirements to be predicted more precisely than DMI means for the total feeding period.

**Key Words:** intake, feedlot, protein requirement

**872 Effect of backgrounding system on beef calf performance.** Jordan L. Cox<sup>\*1</sup>, Kristin E. Hales<sup>2</sup>, Kristen M. Ulmer<sup>1</sup>, Rick J. Rasby<sup>1</sup>, Steven D. Shackelford<sup>2</sup>, Harvey C. Freetly<sup>2</sup>, and Mary E. Drenowski<sup>1</sup>, <sup>1</sup>University of Nebraska-Lincoln, Lincoln, NE, <sup>2</sup>USDA-ARS, US Meat Animal Research Center, Clay Center, NE.

In the Midwest opportunity to integrate cattle production into cropping systems abounds. Two winter forage sources are corn residues and double cropped cool season annuals planted after corn silage harvest. The objective of this study was to evaluate backgrounding spring born calves using these feed resources. Composite MARC II steers ( $n = 355$ ) were stratified by BW ( $277.6 \pm 0.52$  kg) and genetic line and assigned to 1 of 3 treatments: (1) corn residue grazing with distillers supplementation (CRD), (2) oat-brassica forage grazing (OBF) or (3) drylotting on a grower ration (DGR). Each treatment had 4 replicates. Calves on CRD were supplemented 6 d a week with 2.77 kg DM/hd of a dried distillers grains mix containing 2% limestone (DM basis). The OBF was planted in early September and the forage produced was 28% purple top turnip, 14% daikon radish and 59% oats (DM basis). The initial forage mass of OBF was  $3516 \pm 121$  kg DM/ha and calves were stocked at  $1538 \pm 37$  kg DM/hd. Both CRD and OBF calves were given access to a free choice mineral containing 1.32 mg monensin per g. The grower ration consisted of 25% alfalfa hay, 51% corn silage, 20% wet distillers grains and 4% supplement containing 732 mg monensin/kg (DM basis). All calves consumed the grower ration before initial weights. The CRD and OBF calves were removed from grazing after 64 d when the OBF biomass was  $1445 \pm 104$  kg DM/ha and calves were fed the grower ration for 6 d and then weighed. The free-choice mineral intake of the CRD calves (173 g/hd/d) was greater ( $P < 0.01$ ; SEM  $\pm 9.6$ ) than OBF calves (121 g/hd/d). The backgrounding phase of DGR calves ended after 54 d when they reached 365 kg BW. Intake of DGR calves during backgrounding was  $8.27 \pm 0.031$  kg/hd. During the backgrounding phase the ADG of the DGR calves (1.57 kg/d) was greater ( $P < 0.01$ ; SEM  $\pm 0.034$ ) than both OBF (1.00 kg/d) and CRD (0.75 kg/d) and ADG of OBF was greater ( $P < 0.01$ ) than CRD. Although the calves drylotting and fed a grower ration had a greater rate of gain, the lower cost of gain associated with the grazing systems make these backgrounding methods economically competitive.

**Key Words:** backgrounding calves, brassicas, corn residue

**873 Gene expression of the hypothalamus in steers fed high-concentrate diet upon entering feedlot phase.** Jason E. Griffin<sup>\*1</sup>, Zhongde Wang<sup>2</sup>, Jeffrey A. Clapper<sup>1</sup>, Robbi H. Pritchard<sup>1</sup>, Keith R. Underwood<sup>1</sup>, and Michael G. Gonda<sup>1</sup>, <sup>1</sup>South Dakota State University, Brookings, SD, <sup>2</sup>Utah State University, Logan, UT.

Finishing diets used in beef feedlots are high in energy to allow for rapid growth and fat deposition. Previous studies have shown that the use of high concentrate post-weaning diets changes expression of genes in the hypothalamus involved with regulating feed intake, metabolism, signaling, and neuronal communication. The objective of this study was to investigate whether expression of key growth and developmental genes in the hypothalamus can be altered by feeding a high concentrate diet when calves enter the feedlot. Predominantly Angus steers ( $n = 12$ ) were randomly allocated to 1 of 2 treatment groups: (1) high energy/high concentrate (HE/HC), a diet fed to provide 1.33 mc/kg NE gain; or (2) traditional backgrounding/finishing (B/F), fed to provide 1.10 mc/kg NE gain for the first 84 d on feed (DOF) and 1.43 mc/kg NE gain for the remaining 118 DOF. Growth at d28 ( $P < 0.01$ ) was greater for the HE/HC treatment group; however, no difference between treatments was observed at the beginning ( $P > 0.14$ ) and end ( $P > 0.13$ ) of the feeding trial. At slaughter, hypothalami were dissected and frozen for RNA extraction. RNA was extracted and expression differences between treatments were measured by RNA-seq. Carcass characteristics and composition, and Warner-Bratzler shear force (WBSF) analysis were conducted. Kidney, pelvic, heart fat percentage was higher ( $P <$



0.04) in HE/HC steers; however, no difference in other carcass traits ( $P > 0.10$ ) and WBSF ( $P > 0.22$ ) were detected. One hundred and 30 one genes were expressed only in the B/F steers, while 195 were only expressed in the HE/HC steers. Nine genes were expressed in both treatments; 3 genes had higher expression in the HE/HC steers and 6 genes had higher expression in B/F steers. Gene Ontology terms involved with differentially expressed genes included virus immune response,

cell cycle regulation, signaling pathway regulation, and hydrogen and oxygen transport pathways. Feedlot calves fed a high concentrate diet upon entering the feedlot have altered gene expression in the hypothalamus, with many differentiated genes involved with signaling and metabolic pathways.

**Key Words:** bovine, hypothalamus, expression

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