0839 Pastoral systems in the developing world: Trends, needs, and future scenarios.

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Developing-country rangelands are vast and diverse. They are home to millions who are often poor, politically marginalized, and dependent on livestock for survival. Here we summarize experiences from six case-study sites across sub-Saharan Africa, central Asia, and Latin America generally covering the past 25 yr. We examine issues pertaining to population, natural resource management, climate, land use, livestock marketing, social conflict, and pastoral livelihoods. The six study sites differ with respect to human and livestock population dynamics and the resulting pressures on natural resources. Landscape degradation, however, has been commonly observed. Climate change is also having diverse systemic effects often related to increasing aridity. As rangelands become more economically developed pastoral livelihoods may diversify, food security can improve, commercial livestock production expands, but wealth stratification widens. Some significant upgrades in rural infrastructure and public service delivery have occurred; telecommunications are markedly improved due to widespread adoption of mobile phones. Pressures from grazing, farming, mining, and other land uses—combined with drought—can ignite local conflicts over resources, although the intensity and scope of conflict markedly varies across case-study sites. Pastoralists and their herds have become more sedentary overall due to a wide variety of factors, and this can undermine traditional risk-management tactics based on mobility. Remote rangelands still offer safe havens for insurgents, warlords, and criminals, especially in countries where policing remains weak; the resulting civil strife can undermine commerce and public safety. There has been tremendous growth in knowledge concerning developing-country rangelands since 1990, but this has not often translated into improved environmental stewardship or an enhanced well-being for rangeland dwellers. Some examples of demonstrable impact are described, and these typically have involved longer-term investments in capacity building for pastoralists, local professionals, and other stakeholders. Research is shifting from ecologically centered to more human-centered issues; traditional academic approaches are often being augmented with participatory, community-based engagement. Building human or social capital in ways that are integrated with improved natural-resource stewardship offers the greatest returns on research investment. Our future research and outreach priorities include work that fortifies pastoral governance, enhances livelihoods for a diverse array of rangeland residents, and improves land and livestock management in a comprehensive social-ecological systems approach.

Key Words: Bolivian Altiplano, Borana Ethiopia, Kuchi Afghanistan, northern Mexican rangelands, Mongolia, Peruvian Altiplano, Sahelian Zone

LACTATION BIOLOGY

0840 Duration of lactation in first-parity sows: Does it affect piglet growth in second parity?

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It was recently shown that a teat which is not used in first lactation will have reduced development and milk yield in second lactation. This leads to the question of the minimum duration of suckling required in first parity for milk yield not to be hindered in second parity. The goal of the present study was to determine the impacts of a 2 d, 7 d or 21 d suckling period in first lactation on piglet growth, milk composition, and endocrine status of sows in second lactation. Pregnant Yorkshire gilts were divided into 3 groups according to lactation length: 1) 2 d (2D, $n = 20$), 2) 7 d (7D, $n = 20$); and 3) 21 d (21D, $n = 21$). After weaning, sows were bred and kept for a second parity. In both lactations, litters were standardized to 12 piglets with 12 functional teats and surplus teats were sealed with tape. During the second lactation, piglets were weighed on d 2, 7, 14, 21(weaning), 31, and 56 postpartum, and sow feed intake was recorded. Milk samples and jugular blood samples were obtained from sows on d 21 of the second lactation. Concentrations of prolactin, IGF-1, glucose, and urea were measured in blood. The MIXED procedure of SAS using a univariate model (3 levels) was used for statistical analyses and means were compared using Tukey's test. There was a tendency for 21D sows to consume more feed than 2D or 7D sows during the first week of lactation ($P < 0.10$). There was no treatment effect on BW of piglets at any time until d 56 ($P > 0.10$). Concentrations of prolactin, IGF-1, urea and glucose in sows on d 21 of lactation were not affected by treatment ($P > 0.10$). Furthermore, dry matter, fat, protein, and lactose contents in milk were not affected by treatment ($P > 0.10$). Results indicate that increasing the duration of lactation from

2 d to 7 d, or to 21 d in first-parity sows, does not improve growth rate of their piglets in the subsequent lactation. This suggests that suckling of a teat for 2 d during the first lactation is sufficient to ensure optimal mammary development.

Key Words: lactation length, parity, piglet growth

0841 Effects of glucose and amino acids on casein synthesis via JAK2/STAT5 signaling pathway in bovine mammary epithelial cells.

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Studies confirmed that glucose and amino acids (AAs) could be used as signaling factors to regulate milk protein gene transcription and translation through signaling cascade pathways in the mammary gland that affect milk protein synthesis. Some signaling pathways, like Janus kinase-signal transducer and activator of transcription (JAK-STAT), may play a role in the process of milk protein gene transcription. This study employed bovine mammary epithelial cells (BMEC) as a model to investigate the effects of glucose and AAs on β-casein and κ-casein gene transcription, and to determine if effects are mediated through the JAK-STAT signaling pathway. BMEC cells were cultured in specific medium (without D-glucose and AAs), starved overnight, and then subjected to 3 levels of D-Glu (0, 2.5, or 17.5 mmol/L) and 3 levels of AAs (0, 1, or 7.2 mmol/L) in a 3 × 3 factorial arrangement of treatments. After 6 h of culture, the mRNA abundance of *CSN2, CSN3, JAK2,* and *STAT5* genes were measured by qRT-PCR. Statistical analysis of data was performed using SAS 9.2 statistical software package. Differences between experimental groups were considered significant at $P < 0.05$. The results showed that, at the same level of D-Glu or AAs, increasing the concentration of the other to the medium level or higher, substantially increased the mRNA abundance of *CSN2, CSN3, JAK2,* and *STAT5* genes. These results demonstrated that D-Glu or AA supplements could promote transcription of casein genes (β-casein, κ-casein), and that process might be related to JAK-STAT signaling in the bovine mammary epithelial cell. In conclusion, our results provide basic information for the further study of the mechanisms by which glucose or AAs affect casein expression in the bovine mammary epithelial cell.

Key Words: amino acids, glucose, JAK2/STAT5

0842 Repeated mammary tissue collections during lactation have no impact on cow performance.

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Mammary biopsy (MB) collection is a necessary and valuable approach for studies in mammary gland biology, but it is not known if repeated MB impair performance of the lactating cow. Our objective was to examine the effect of multiple MB during lactation on udder health, DMI, and milk yield of dairy cattle. Sixty-four multiparous, mid-lactation Holstein cows were enrolled in a trial and 32 cows were randomly selected for repeated MB. The MB and non-MB cows had similar parity (2.6 ± 0.9, $P = 0.13$) and DIM (96.5 ± 56.3 d, $P = 0.13$) at enrollment. All animals were housed in the same barn and fed the same diet. Cows were milked three times a day and milk yield was recorded at each milking. Milk composition was measured weekly and DMI was recorded daily. Three MB were performed per cow: at enrollment and at 3 and 5 mo post-enrollment. The first and third MB were taken from the left rear quarter whereas the second MB was from the right rear quarter. The experimental procedures were approved by the Institutional Animal Care and Use Committee of the University of Georgia. Before MB, cows were mildly sedated through i.v. injection of xylazine HCL (20 μg/kg of BW). Briefly, the MB was performed using a rotating stainless steel cannula with a retractable blade at the cutting edge connected with a cordless drill. After sanitation with iodine and ethanol, an incision was made through the skin and connective tissue in the middle of the quarter and a core of mammary tissue (0.75 to 1 g) was extracted. The incision was closed using stainless wound clips and sprayed with an antiseptic dressing to avoid infection. After MB, udder health, wound healing of incisions, and appearance of blood in milk were visually examined at each milking. All bloody milk was discarded and blood was cleared from milk 3.86 ± 2.0 d after MB. During the experiment, four MB quarters and 14 non-MB quarters were diagnosed and treated for clinical mastitis. Compared with non-MB cows, MB cows had similar ($P > 0.1$) DMI, milk yield and percentage of fat, lactose, protein, solids-not-fat, and somatic cell score. In conclusion, mid- to late-lactation cows recover rapidly from MB and repeated MB have no impact on DMI, milk yield and composition, and udder health

of lactating dairy cows.

Key Words: lactating cow, lactation performance, mammary biopsy, udder health

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0843 Lack of glucose and amino acids suppresses protein synthesis of bovine mammary epithelial cells by activating AMPK and inhibiting mTORC1 signaling pathways. S. Wang[1,2,3,4], S. Zhao[1,2,5], H. Gao[1,2,3,6], M. Zhang[1,2,3], N. Zheng[1,2,5], Y. Zhang[1,2,5], S. Yan[4], and J. Wang[*2,3,5], *[1]Ministry of Agriculture-Milk Risk Assessment Laboratory, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, [2]State Key Laboratory of Animal Nutrition, Institute of Animal Science, Chinese Academy of Agricultural Sciences, Beijing, China, [3]Ministry of Agriculture-Milk and Dairy Product Inspection Center, Beijing, China, [4]College of Animal Science, Inner Mongolia Agricultural University, Hohhot, China, [5]Ministry of Agriculture-Milk and Dairy Product Inspection Center (Beijing), Beijing, China, [6]College of Animal Science and Technology, Gansu Agricultural University, Lanzhou, China*.

Glucose and amino acids (AA) regulate milk protein synthesis via signaling pathways involving AMP-activated protein kinase (AMPK) and mammalian target of rapamycin (mTOR) in current nutrient requirement models. The objective of this study was to investigate the effects of nutritional stress due to lack of glucose and/or AAs and determine the sensitivity of bovine mammary epithelial cells to each stress by measuring cell proliferation and expression of β-casein and signaling proteins. Bovine mammary epithelial cells were cultured in specific medium (without glucose and AAs) and starved overnight, then three levels of glucose (0, 2.5, or 17.5 mmol/L) and AAs (0, 1.03, or 7.2 mmol/L) in a 3 × 3 factorial design were added to the medium. The proliferation of bovine mammary epithelial cells was detected by the thiazolyl blue (MTT) method, and the expression of β-casein and phosphorylation of signaling proteins were detected by Western blot. The results showed that proliferation of bovine mammary epithelial cells and their expression of β-casein was decreased under stress due to lack of glucose and/or AAs. When the concentration of AAs was 7.20 mmol/L and the glucose levels 17.5 and 2.5 mmol/L, cell proliferation and β-casein expression were downregulated 9.44% and 37.53%. When the concentration of glucose was 17.5 mmol/L and the AA levels 7.2 and 1.03 mmol/L, cell proliferation was downregulated 6.67% and 21.98%. Further experiments validated that the phosphorylation of AMP-activated protein kinase (AMPK, $Thr^{183/172}$) was upregulated under glucose and/or AA stress. When the AA level was 7.2 mmol/L and glucose levels were 17.5 and 2.50 mmol/L, the expression of P-AMPK($Thr^{183/172}$) was upregulated 210%. When the concentration of glucose was 17.5 mmol/L and AAs 7.20 and 1.03 mmol/L, the expression of P-AMPK($Thr^{183/172}$) had no effect. In contrast, the phosphorylation of the mammalian target of rapamycin (mTOR, Ser^{2481}), the regulatory associated protein of mTOR (Raptor, Ser^{792}), the ribosome protein subunit 6 kinase 1(S6K1, Thr^{389}), the 4E binding protein 1 (4EBP1, Thr^{37}), and the content of ATP were downregulated under glucose and/or AAs stress, and we also find that these phosphorylation proteins and ATP content are more sensitive to glucose than AAs. Results from this study suggest that stress due to lack of glucose and/or AAs can influence the energy and protein supplies in mammary epithelial cells, causing downregulation of proliferation, activating the fuel sensor AMPK, and suppressing the mTOR signaling pathway, which may lead to decreased β-casein expression. Bovine mammary epithelial cells are more sensitive to glucose stress than to AAs.

Key Words: amino acids, β-casein, glucose

0844 Genome-wide association analysis and pathways enrichment for lactation persistency in Canadian Holstein cattle. D. N. Do[*1,2], N. Bissonnette[1], P. Lacasse[1], M. Sargolzaei[3,4], F. Miglior[4,5], X. Zhao[2], and É. M. Ibeagha-Awemu[1], *[1]Agriculture and Agri-Food Canada, Sherbrooke Research and Development Centre, Sherbrooke, QC, Canada, [2]Department of Animal Science, McGill University, Montreal, QC, Canada, [3]Semex Alliance, Guelph, ON, Canada, [4]Centre for Genetic Improvement of Livestock, Department of Animal Biosciences, University of Guelph, Guelph, ON, Canada, [5]Canadian Dairy Network, Guelph, ON, Canada*.

Lactation persistency (LP), generally defined as the rate of declining milk yield after milk peak, is an economically important trait for dairy cattle. Enhancing LP through genetic improvement is an interesting avenue for increasing overall milk production because it does not cause negative energy balance and related health issues for cows. We performed a genome-wide association study (GWAS) and pathway enrichment to explore the genetic mechanisms underlying LP. A GWAS based on generalized quasi-likelihood score method was performed on LP data of 866 cows and 44,023 single nucleotide polymorphisms (SNPs) with inclusion of a polygenic effect explained by genomic relationship matrix. A total of 29 and 88 SNPs were significantly associated with LP at the adjusted Bonferroni ($P < 2.15 \times 10^{-5}$) and at false discovery rate of 5%, respectively. Important genomic regions that harbored several significant SNPs for LP were: 68.2–71.2 mega base pairs (Mb) on bovine chromosome (BTA) 16, 14–16 Mb on BTA 3, 107–109 Mb on BTA 5, and 133–137 Mb on BTA 1. The most significantly associated SNP (rs41818282) is located in an intronic region of hemicentin 1 gene. Functional annotation of 599 genes in the flanking regions (0.5 Mb) of significant SNPs showed that natural killer cell-mediated cytotoxicity and TGF-β signaling pathways were important for LP. In addition, 8 among 11 gene

ontologies enriched for LP were involved in the immune process. These results suggest that biological processes involving cell death, cell proliferation, and immune response are important in the regulation of LP. However, it is necessary to further characterize the detected regions and enriched pathways for confirmation and validation. In conclusion, this study detected several genomic regions for fine mapping to identify potential markers for LP improvement.

Key Words: cows, genomics, GWAS, lactation persistency, pathways

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0845 Effect of 17β-estradiol on milk production, hormone secretion, and mammary gland gene expression of dairy cows. J. J. Tong[1], I. M. Thompson[2], and P. Lacasse*[3], [1]*Department of Clinical Veterinary Medicine, College of Veterinary Medicine, Northeast Agricultural University, Harbin, China,* [2]*AAFC-Sherbrooke R&D Centre, Sherbrooke, QC, Canada,* [3]*Agriculture and Agri-Food Canada, Sherbrooke Research and Development Centre, Sherbrooke, QC, Canada*.

Estradiol inhibits milk production in dairy cows. The present study evaluated the impact of estradiol injections on prolactin (PRL) secretion and mammary gland responsiveness to PRL. Eight mid-lactation cows received either 17β-estradiol (2.5 mg; E2; $n = 4$) or soy oil (2.5 mL; CTL; $n = 4$) injections during 7 d (period 1). After a resting period of 3 wk, a second treatment period was performed where the cows were switched from treatments. For each period, blood and milk samples were collected from d −4 to 14 (relative to the first injection). In addition, blood samples were collected during the a.m. milking on d −4, 2, and 7 to determine the milking-induced PRL release. Mammary gland biopsies were harvested on the last injection day of both periods. Milk fat samples were collected on d 1, 4, 7, and 14. The mRNA levels of genes encoding proteins related to mammary activity (α-lactalbumin, β-casein, and acetyl-CoA carboxylase), apoptosis (Bax, Bcl2, and caspase-3), PRL receptors (PRLR, long and short forms), LIF, and suppressors of cytokine (SOCS1, 2, and 3) were measured by real-time RT-PCR using RNA extracted from milk fat and mammary gland. Injections of E2 decreased moderately milk production during the treatment period (36.70 ± 0.45 kg/d and 39.74 ± 0.45 kg/d, respectively; $P = 0.01$). The E2 treatment tended to increase fat ($P = 0.08$), increased lactose ($P = 0.04$), and increased protein ($P = 0.03$) content of milk. Serum PRL concentration was increased by E2 injections during the treatment ($P = 0.04$) and the post-treatment ($P = 0.01$) periods. Accordingly, milk PRL concentration tended to be increased by E2 ($P = 0.09$). Milking-induced PRL release was also increased by E2 injections ($P = 0.03$). Injections of E2 increased plasma concentrations of IGF-1 ($P < 0.01$) and estradiol ($P < 0.01$) during the treatment period. Milk BSA was not affected by treatments, suggesting that mammary tight junctions were not impaired by E2. In mammary tissue, the LIF mRNA level tended to be lower during the E2 treatment ($P = 0.09$). In milk fat, PRLRL mRNA level tended to be decreased ($P = 0.07$), and PRLRS mRNA level was decreased ($P = 0.04$) by E2 injections. No difference in expression was observed for the other genes in mammary tissue and milk fat. These results suggest that a decrease of mammary gland responsiveness to PRL may be involved in the estradiol-induced inhibition of milk production.

Key Words: estradiol, gene expression, prolactin

0846 Estimation of quarter vs. composite colostrum composition via Brix refractometry, specific gravity, and visual color appearance in dairy cows. J. J. Gross*, E. C. Kessler, and R. M. Bruckmaier, *Veterinary Physiology, Vetsuisse Faculty, University of Bern, Bern, Switzerland*.

The control of colostrum quality is essential for successful calf rearing. Instruments for on-farm colostrum quality determination are widely used in dairy practice. However, composite colostrum samples have predominantly been considered so far, thereby not taking potential variation between quarters into account. In cases of low quality of composite colostrum, feeding a better quality colostrum from individual quarters might be beneficial. The objective of the present study was to identify relationships between colostrum color, quality (assessed by a colostrometer and a Brix refractometer), and composition (measured by different laboratory methods) of colostrum at a quarter level, for two types of colostrum sampling. Quarter and composite colostrum samples from 17 primiparous and 11 multiparous Holstein cows were analyzed for total IgG, fat, protein and lactose contents, and color was measured by a spectrophotometer. In the present study, an IgG concentration below 50 g/L, as determined by ELISA, was found in 14.3% of the analyzed quarter samples. Concentration and total mass of IgG in composite colostrum samples were greater in multiparous compared with primiparous cows ($P < 0.05$). Specific gravity (SG) of colostrum from individual and composite samples was lower in primiparous than in multiparous cows ($P < 0.05$). Milk fat content was greater in quarter and composite colostrum samples of primiparous compared with multiparous dairy cows ($P < 0.05$). Neither in primiparous nor in multiparous dairy cows were there clear relationships between IgG content and SG, Brix, and the color space coordinates L^*, a^*, and b^*. Interestingly, results indicate that despite a similar range of the variables investigated, correlations between those variables can differ between the quarter and the composite samples. Correlation coefficients with IgG concentration of the respective samples were greater using a composite compared with an individual quarter sample. This was true for SG and Brix determination, and also for the color space coordinates measured. Due to the variation in milk composition between individual quarters of a cow, correlation coefficients between colostral IgG concentration, SG, Brix-values, and colostrum

color were poorer in quarter compared with composite samples. In conclusion, both accuracy and limitations of on-farm instruments estimating colostrum quality apply for quarter colostrum samples as well as composite samples.

Key Words: colostrometer, immunoglobulin G, refractometer

0847 Effects of increasing residual milk on milk yield and composition. L. L. Hernandez[1], V. J. McKeon[*2], E. L. Endres[2], A. de Bruijn[2], A. Kleinhans[2], and D. J. Reinemann[2], *[1]Department of Dairy Science, University of Wisconsin, Madison, [2]University of Wisconsin, Madison.*

The future profitability of the dairy industry depends, in part, on faster milking time and increased milk yield. Currently, milking machines detach from the udder when milk flow decreases to a specified amount. Due to variability in quarter-level milk yield, conventional practice may result in over-milking of up to three quarters and a variable amount of over-milking of the fourth quarter. The objective of this research was to evaluate the effect of residual milk on milk yield and composition. Twelve multiparous (avg. lactation 2.83 ± 0.99) Holstein cows were milked twice daily with a quarter milking machine for 42 d beginning 5 d post calving. Twice-daily quarter milk weights were recorded. Cows were assigned alternating control and treatment half udders depending on calving order, resulting in 12 control (CON; 0% residual milk) and 12 treatment (RES; 30% residual milk) quarters. Milk removed from RES quarters was recalculated weekly to account for an increase in milk production. Milk yield was affected by treatment ($P < 0.001$; 87.64 ± 5.20 kg for CON vs. 63.25 ± 5.20 kg for RES), quarter ($P < 0.001$; 67.10 ± 4.68 kg for front vs. 83.79 ± 4.68 kg for rear), and the treatment by week interaction ($P < 0.01$). Milk samples were collected weekly starting on d 5 and ending on d 47. Milk samples were analyzed for SCC, SNF, protein, MUN, and lactose. There was a week effect for SNF, butterfat, protein, MUN, and lactose ($P < 0.001$). There was a treatment by week by quarter effect on SNF ($P < 0.05$). Milk lactose concentrations were decreased by treatment ($P < 0.05$; 4.86 ± 0.07 kg for CON vs. 4.66 ± 0.07 kg for RES), differed among quarters ($P < 0.05$; 4.81 ± 0.07 kg for front vs. 4.71 ± 0.07 kg for rear), and there was a treatment by quarter interaction ($P < 0.05$; 4.96 ± 0.08 kg for CON front, 4.76 ± 0.08 kg for CON rear, 4.67 ± 0.08 kg for RES front, and 4.66 ± 0.08 kg for RES rear). Treatment increased SCC ($P < 0.05$; 42.78 ± 14.75 kg for CON vs. 89.60 ± 14.75 kg for RES). A treatment by quarter interaction was present for SCC ($P < 0.05$; 40.75 ± 16.58 kg for CON front, 44.82 ± 16.58 kg for CON rear, 111.26 ± 16.58 kg for RES front, and 67.94 ± 16.58 kg for RES rear). Results suggest that leaving 30% residual milk in the udder suppresses milk yield, increases SCC, and decreases milk lactose content. Future research should explore optimal take-off settings of milking machines at the quarter level.

Key Words: quarter variability, residual milk

0848 Nutrient composition of milk from great apes throughout lactation. M. Garcia[*1], M. Power[2], and K. M. Moyes[1], *[1]Department of Animal and Avian Sciences, University of Maryland, College Park, [2]Smithsonian Conservation Biology Institute, Washington, DC.*

For great ape infants, milk is the primary, if not sole, food providing the only supply of water, organic nutrients, and minerals during the first 12 to 18 mo of life. Moreover, when mother's milk is not available to a great ape infant (e.g., maternal death or infant rejection), zoo nutritionists/veterinarians formulate a milk replacer to maximize the chances of neonate survival. However, very limited information on milk composition from non-human apes is currently available. This study aimed to identify the nutrient composition of milk from gorillas and orangutans throughout lactation. Fifty three milk samples from 4 gorillas and 3 orangutans were collected throughout 48 and 22 mo in milk (MIM), respectively. Samples were grouped into 5 stages of lactation (i.e., 0 < MIM = 6, 6 < MIM = 12, 12 < MIM = 18, 18 < MIM = 36, and 36 < MIM = 48). Data were analyzed as a complete randomized design. The analysis to compare gorilla with orangutan included only 4 stages of lactation. Across MIM, protein was greater in gorilla than in orangutan milk (1.27 vs. 0.85%, respectively; $P < 0.01$). Protein, fat, lactose, and gross energy were affected by the interaction of species by MIM ($P < 0.05$). For gorilla milk, all components changed with MIM, whereby protein content was greatest by 48 MIM (1.91%, $P < 0.01$) and lactose was lower by 6 MIM, reaching a nadir by 48 MIM (4.95%, $P < 0.01$). Moreover, fat and gross energy contents followed a similar pattern to that of DM, with the highest content by 36 MIM ($P \leq 0.01$). For orangutan milk, protein, DM, and fat were unaffected by MIM. However, gross energy and lactose contents were steady during the first 18 MIM (7.51% and 0.36 Kcal/g, for lactose and gross energy, respectively) and decreased (lactose, 6.67%, $P < 0.05$) or tended to decrease (gross energy, 0.39 Kcal/g, $P < 0.10$) by 36 MIM. Major macronutrients in milk were similar between orangutan and gorilla, except for a lower protein content in orangutan milk. There was some change in milk composition over lactation. Mean values for mature milk between 6 and 18 MIM are the best values to consider when formulating milk replacers (protein 1.05%, fat 2.12%, and lactose 7.21%). Coupled with immune parameters, these results provide useful information to assist professionals caring for non-human great apes under captivity.

Key Words: gorilla, milk components, orangutan

0849 Milk fat globules as a source of mammary microRNA. D. Lago-Novais[1,2], K. Pawlowski[1], J. A. A. Pires[*1], L. Mobuchon[1,3], S. Bes[1], P. Martin[3], and C. Leroux[1], *[1]UMR1213 Herbivores, INRA, VetAgroSup, Saint-Genes-Champanelle, France, [2]Universidade Federal da Bahia, CEP, Salvador-BA, Brazil, [3]UMR1313 Gabi, INRA, AgroParisTech, Université Paris-Saclay, Jouy-en-Josas, France*.

Tissue for research on mammary gland (MG) gene expression is obtained via invasive and expensive methods (biopsy or post-mortem) that limit high throughput analyses. Milk fat globules (MFG) have been used to assess the mRNA content of the mammary epithelial cells in the bovine and goat (Brenaut et al., 2012; Canovas et al., 2014) for gene expression studies. MFG is therefore a satisfactory alternative source of mammary mRNA. MicroRNAs (miRNA) are small stable noncoding RNAs involved in multiple aspects of mammary gland physiology. Whereas the use of MFG was reported in humans (Munch et al., 2013), until now MFG as the source of miRNA has not been studied in the bovine. The objective of this study was to assess MFG as a source of miRNA, and whether the latter are representative of MG miRNA expression, by comparing targeted miRNA in MFG and MG sampled from mid-lactation Holstein cows. Total RNAs were extracted from MFG (n = 6) and MG (n = 6) using TRIzol (ThermoFisher, Inc, USA). Nine miRNA (*miR-29a*, *miR-125*, *miR-126*, *miR-141*, *miR-148a*, *miR-204*, *miR-223*, *miR-320*, and *miR-494*) were studied by RT-qPCR. The results are expressed as fold change of MFG data relative to MG data using the 2-ΔΔCt method and *U6* as internal reference. Statistical analyses were performed using a t test (DataAssist™ software) and $P < 0.05$ considered as significant. Among the nine miRNA chosen on the basis of the expression in MG, two were not detected in MFG whereas they were highly abundant in MG (*miR-126* and *miR-204*), and three were significantly more abundant in MG than in MFG (*miR-29a*, *miR-125b*, and *miR-148a*, presenting a fold change value of 23.2, 13.9, and 8.7, respectively). Four miRNA were detected at the same level in both MFG and MG. Our results suggest that there are different mechanisms of miRNA transfer to milk. Nevertheless, it is possible that miRNA not present in MFG are not expressed in epithelial cells, but are present in other MG cell-types, and therefore not transferred to milk. In conclusion, MFG can be used as a non-invasive source of microRNA but do not reflect exactly the MG miRNome. Further research is warranted on the composition of MFG miRNome and modulation of their secretion in milk.

Key Words: bovine, microRNA, milk fat globule

0850 Consumption of endophyte-infected fescue seed during the dry period and lactation affects mammary gland gene expression in dairy cows. R. L. Baldwin[*1], C. Li[1], D. M. Bickhart[1], C. M. Evock-Clover[1], P. Grossi[2], R. K. Choudhary[3], T. H. Elsasser[4], G. Bertoni[5], E. Trevisi[6], G. E. Aiken[7], K. R. McLeod[8], and A. Capuco[1], *[1]Animal Genomics and Improvement Laboratory, USDA-ARS, Beltsville, MD, [2]Università Cattolica del Sacro Cuore, Piacenza, Italy, [3]School of Animal Biotechnology, GADVASU, Ludhiana, Punjab, India, [4]USDA-ARS, Animal Biosciences and Biotechnology Laboratory, Beltsville, MD, [5]Istituto di Zootecnica, Università Cattolica del Sacro Cuore, Piacenza, Italy, [6]Università Cattolica del Sacro Cuore, Piacenza, Italy, [7]USDA-ARS, Lexington, KY, [8]University of Kentucky, Lexington*.

Ergot alkaloids in endophyte-infected grasses inhibit prolactin secretion and reduce milk production when fed to lactating cows. However, we have shown this effect is temporal in that prepartum consumption of infected seed throughout the dry period does not inhibit subsequent milk production and, in fact, prior exposure to bromocriptine (ergot peptide) actually increases production. To identify changes in the transcriptome and pathways mediating the mammary gland's response to ergot alkaloids in the diet, RNA sequencing (RNA-Seq) was performed on mammary tissue obtained from 24 multiparous Holstein cows exposed to 1 of 3 treatments. Starting at 90 ± 4 d prepartum, cows were fed endophyte-free fescue seed (control, C), endophyte-free fescue seed plus 3×/wk subcutaneous injections of bromocriptine (0.1 mg/kg BW, B), or endophyte-infected fescue seed (I), as 10% of the diet on an as-fed basis. Mammary biopsies from 4 or 5 cows/treatment at each of 3 distinct phases were obtained: 7 d before dry off during the initial lactation (L1), mid-dry period (D), and 10 d postpartum (L2). Biopsy samples from each treatment group and phase of lactation were used to generate individual RNA-Seq libraries. Normalized reads of the RNA-Seq data were organized into technical and biological replicates before processing with the RSEM software package. Each lactation phase was processed separately with the "rsem-run-ebseq" pipeline, and genes that differed between any of three treatments were identified from program output. A large proportion of genes considered to be differentially expressed in at least one treatment with a posterior probability of differential expression greater than 90% (n = 866) were found to be similarly expressed in B and I treatments, but differentially expressed from C (n = 575, total for all three phases). When phases were compared, 104 genes that were differentially expressed compared to C were found to be common to the L1 and L2 phases. Consistent with the production findings, networks most affected by treatments in L1 and L2 include lipid metabolism, small molecule biochemistry, and molecular transport, while in D networks relate more to

developmental and cellular functions and maintenance. The strong similarity in pattern of expression in B and I treatments during both late and early lactation suggests, at least in part, the involvement of similar cell signaling pathways or mechanisms of action for both B and I and the importance of prolactin messaging pathways.

Key Words: ergot alkaloids, prolactin, RNA sequencing

0851 Intravenous infusion of 5 hydroxy-L-tryptophan, a serotonin precursor, to transition dairy cows pre-calving affects GH-IGF axis gene expression in the mammary gland and liver post-calving.

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The role of the monoamine serotonin in the regulation of growth hormone (GH) and insulin-like growth factor-1 (IGF1) is controversial. Most studies have focused on serotonin produced in the brain. Given that the majority of total-body serotonin is produced outside of the central nervous system, we set out to determine whether infusing a serotonin precursor, 5 hydroxy-L-tryptophan (5-HTP) intravenously would impact the somatotropic axis in transition dairy cows. Multiparous Holstein cows were intravenously infused once daily for approximately 6 d pre-calving (−6 to −1), with either 1 L of saline ($n = 6$) or 1.0 mg/kg BW of 5-HTP ($n = 6$). Blood was collected before and after infusion, and at 2, 4, and 8 h post-infusion, at calving, and on d 3, 5, 7, and 14 post-calving to measure serotonin and IGF1 concentrations in serum. Mammary gland and liver biopsies were collected on d 1 and 7 post-calving to assess gene expression of the somatotropic axis. Gene expression data was analyzed using the DDCt method, using d 1 saline-infused cows as the control. Blood variables were analyzed with a two-way ANOVA (pre- and post-calving separately). Overall, 5-HTP-infused cows had greater serotonin and lower IGF1 concentrations than controls. Specifically, 5-HTP-infused cows had greater serotonin concentrations pre-calving (all days except d −2), at calving (d 0), and post-calving only on d 1 of lactation ($P < 0.05$). Infusion of 5-HTP decreased IGF1 concentrations pre-calving, particularly on d −6 and d −5, and only during the first and second hour post-infusion ($P < 0.05$). No differences in IGF1 were detected on d −4 to d −1 of infusion pre-calving. Post-calving, IGF1 concentrations markedly decreased, compared with pre-calving levels, but there were no differences between treatments on d 3, 5, or 7. On d 14, IGF-I concentrations increased to pre-calving levels only for the saline-infused cows and remained low in the 5-HTP-infused cows ($P < 0.05$). In the liver, 5-HTP treatment upregulated the mRNA expression of IGF1, IGF1-receptor and GH-receptor and IGF1-binding protein 3 on d 1 post-calving ($P < 0.01$), and no differences in gene expression were observed on d 7. In the mammary gland, 5-HTP treatment upregulated the mRNA expression of IGF1 and IGF1-binding protein 2 on both d 1 and 7 post-calving ($P < 0.05$). Regardless of the relative steadiness of circulating IGF1 concentrations post-calving, these data imply a potential benefit of 5-HTP administration pre-calving to improve the expression of genes related to the somatotropic axis in the liver and mammary tissue at the onset of lactation in dairy cows.

Key Words: lactation, serotonin, somatotropic axis

0852 Effect of cortisol on mammary epithelial cell Bax and Bcl-2 gene expression at lactation peak of goats.

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Cortisol is one of the main hormones that characterize stress, and chronically elevated cortisol can induce apoptosis, as well as expression of Bax (pro-apoptotic), which, in high levels, signals to cellular mitochondria causing cell apoptosis. In contrast, Bcl-2 (anti-apoptotic) acts as a "protector" of cells. Thus, the aim of this study was to analyze the effects of different levels of cortisol on Bax and Bcl-2 expression in mammary epithelial cells. Twenty-four Saanen goats were distributed in two groups: ACTH administration (cortisol group) or placebo (control group), during the lactation peak, i.e., 60 d of lactation. Four goats from each group were submitted to a biopsy of mammary gland after 1 h of treatment administration. Epithelial cells were isolated and four different concentrations of cortisol were added to this culture: 0.0 (control), 10, 100, or 1000 ng/mL. After 5 d, mRNA was extracted and Bax and Bcl-2 gene expression was measured by RT-PCR. Statistical analysis was performed using ANOVA, and significance was declared at $P \leq 0.05$. In the cortisol group, there was no effect of pre-treatment on expression of Bax and Bcl-2 in cell culture. However, in the placebo group, there was a significant difference ($P < 0.05$) in Bax expression, such that at higher levels of cortisol in the culture, Bax expression was lower (2.28 ± 0.58 and 2.01 ± 0.4, respectively, for 100 and 1000 ng/mL), compared to the control (4.38 ± 1.1, 0.0 ng/mL). For Bcl-2 there was no significant difference ($P > 0.05$). In summary, high levels of cortisol in mammary epithelial cells can interfere with Bax gene expression.

Key Words: apoptosis, cortisol, mammary gland, stress

0853 Interactions among serotonin and circadian systems in the mammary gland. A. Suárez-Trujillo[1], J. S. Crodian[2], A. M. Shamay[3], S. J. Mabjeesh[*4], K. Plaut[5], and T. M. Casey[6], *[1]Department of Animal Science, Universidad de Las Palmas de Gran Canaria, Arucas, Las Palmas, Spain, [2]Purdue University, West Lafeyette, [3]Agriculture Research Organization, Volcani Center, Bet Dagan, Israel, [4]Department of Animal Sciences, The Robert H. Smith Faculty of Agriculture, Food and Environment, The Hebrew University, Rehovot, Israel, [5]Department of Animal Sciences, Purdue University, West Lafayette, [6]Department of Animal Sciences, Purdue University, West Lafayette*.

The circadian and serotonin systems are reciprocally regulated in the brain and function to maintain homeostasis and respond to internal and external stimuli. Both systems play regulatory functions throughout the lactation cycle in the mammary gland. We hypothesized that the serotoninergic and circadian systems are reciprocally regulated in the mammary gland to mediate mammary homeostasis and respond to metabolic demands of milk synthesis. The objective of this study was to determine whether there is reciprocal regulation among the systems using approaches that encompassed bioinformatics analysis, mammary cell and tissue culture, and temporal expression analysis of mammary gene expression using sheep milk. Bioinformatics analysis of the 2000-bp upstream region of the *SLC6A4* (serotonin reuptake transporter, SERT) gene revealed the presence of a canonical E-box sequence (CACGTG) that circadian transcription factor CLOCK:BMAL1 binds. qPCR analysis of steady-state *SLC6A4* mRNA in samples isolated from sheep milk fat globules showed that this gene exhibits a circadian rhythm of expression. *SLC6A4* pattern of expression was similar to *PER2,* a core clock gene that is a transcriptional target of CLOCK:BMAL1. Moreover, comparison of *PER2* and *SLC6A4* temporal expression in wild-type HC11 cells also showed similar expression patterns in this mammary epithelial cell line. In HC11 cultures that carry shRNA that targeted *CLOCK, SLC6A4* expression was decreased across all time points ($P < 0.05$), supporting that the mammary clock regulates this serotonergic factor. To study the effect of serotonin (5-HT) on the mammary clock, mammary explants were prepared from lactating mice and divided into two treatments: control (lactogenic culture media–prolactin, glucocorticoids, and insulin) and 5-HT (lactogenic culture media and 200 μM of 5-HT). Samples were collected every 4 h over a 24-h period. One-way ANOVA of temporal variation within treatments found time had a significant ($P < 0.05$) impact on *BMAL1* expression in the control, but not the 5-HT treated samples, indicating that 5-HT attenuates *BMAL1* expression rhythm. *PER2* temporal variation was significant in both treatments ($P < 0.001$), but the differences were due to different time points, indicating that 5-HT shifted the phase of expression rhythm. Together, these data support that circadian and serotonergic systems interact in mammary gland. Further studies are needed to understand the significance of these interactions and how they may affect productivity.

Key Words: circadian clocks, mammary gland, serotonin

0854 Effects of stress on IGF-1 plasma concentrations and on expression of GH and IGF-1 receptors in mammary glands. G. F. Bomfim*, *Faculty of Animal Science and Food Engineering–FZEA/USP, Pirassununga/Sao Paulo, Brazil*.

Growth hormone (GH) and insulin-like growth factor-1 (IGF-1) have positive effects on lactation. The action of these hormones on mammary tissue can be regulated by their respective receptors, GHR and IGF-1R. When females undergo a situation of stress during the lactation period, its influence on the expression of these receptors, as well as on concentrations of IGF-1 in the blood, is unknown. The objective of this study was to evaluate the effects of cortisol on IGF-1 plasma concentrations and on the expression of IGF-1R and GHR at the peak and end of lactation. Thirty Saanen goats were distributed in two groups: ACTH administration to induce stress (cortisol group) or placebo (control group), at peak (60 d) and end (180 d) of lactation. Four goats from each group were submitted to a mammary biopsy after 60 min of treatment administration. Mammary tissue was collected to measure mRNA levels for IGF-1R and GHR via RT-PCR, and IGF-1 plasma concentrations were measured (EIA method) in blood samples of goats collected at the time of biopsy. Statistical analyses were performed using ANOVA. There was no treatment effect ($P > 0.10$) on GHR or IGF-1R gene expression or on IGF-1 plasma concentrations at 60 d and 180 d of lactation. These results suggest that an increase in concentrations of cortisol in the bloodstream does not influence GHR and IGF-1R expression in the mammary gland. In addition, IGF-1 plasma concentrations remained constant in both groups during lactation.

Key Words: ACTH, hormones, lactation

0855 Extracellular matrix molecule d*Eco*RIn signaling pathway gene expression in two bovine mammary cell lines. H. L. M. Tucker*, C. L. M. Parsons, and K. M. Daniels, *Virginia Polytechnic Institute and State University, Blacksburg.*

Previously we showed that d*Eco*RIn, an extracellular matrix (ECM) proteoglycan, is primarily located in mammary stroma and localization depends on physiological stage. Because the mammary gland is a heterogeneous tissue, the primary d*Eco*RIn producing cell populations and primary d*Eco*RIn responsive cell populations have not been identified; knowing this will advance our understanding of mammary ECM remodeling during mammary growth and involution. Our

objective was to characterize known d*Eco*RIn pathway genes in two immortalized bovine cell lines. These were bovine mammary epithelial cells (BME) and mammary fibroblasts (MF-T2). BME and MF-T2 were each grown on 6-well plastic dishes with Dulbecco's Modified Eagle Medium (DMEM) plus 10% fetal bovine serum. Initial densities were 5×10^5 cells/well for BME and 1×10^6 cells/well for MF-T2. After 16 h, media was changed to 100% DMEM. Cell lysates were collected after 16 h of DMEM incubation; total RNA and DNA were quantified. Total RNA was converted into cDNA and used in real-time quantitative PCR (qPCR). Genes of interest were: d*Eco*RIn, transforming growth factor β 1, transforming growth factor receptor 1, insulin-like growth factor 1, insulin-like growth factor receptor 1, epidermal growth factor, epidermal growth factor receptor, hepatocyte growth factor, hepatocyte growth factor receptor, and insulin receptor. Before data analyses, genes of interest were normalized to the average of three internal control genes and the final DNA content of each cell culture well. Triplicate cell culture wells were used and qPCR performed in duplicate. Separate immunocytochemistry experiments were conducted with the similar culture conditions to show cellular localization of the d*Eco*RIn core protein in BME and MF-T2 using a rabbit polyclonal d*Eco*RIn primary antibody (1:50; SC22753) and AlexaFluor 594 goat anti-rabbit secondary antibody (1:200; A11012). MF-T2 produced relatively more d*Eco*RIn core protein mRNA (31× more abundant) and protein than BME when both cell types were cultured under basal conditions. All other genes of interest were detected in both cell types, with transforming growth factor β 1 mRNA also being relatively more abundant in MF-T2 (9× more abundant). Because d*Eco*RIn has known growth regulatory properties, resultant findings will be used to design mechanistic cell culture studies. In the future, d*Eco*RIn signaling pathways may be manipulated in vivo in efforts to increase milk production efficiency via increased epithelial cell proliferation or decreased apoptosis during growth and involution.

Key Words: cell culture, ECM, gene, mammary gland

0856 Associations between quarter-level inflammation status across the dry period and health outcomes in the subsequent lactation. S. A. Metzger*[1], L. L. Hernandez[2], and P. L. Ruegg[1], [1]*Department of Dairy Science, University of Wisconsin-Madison, Madison, WI,* [2]*Department of Dairy Science, University of Wisconsin, Madison.*

The objective of this prospective cohort study was to determine associations of inflammation status of individual mammary quarters of dairy cows across the dry period with health status in the first 150 d of lactation. Milk from all mammary quarters ($n = 649$) of all lactating cows at the University of Wisconsin–Madison Emmons Blaine Dairy Cattle Research Center was sampled at dryoff before administration of dry cow antibiotic therapy and twice within the first 2 wk after the subsequent calving. Quarters with a somatic cell count (SCC) < 100,000 cells/mL and no microbiological growth at all three sampling times (low negative, LN; $n = 76$), quarters with SCC ≥ 150,000 cells/mL and no microbiological growth at the first two sample times and a variable third sample (high negative precalving, HNPre; $n = 17$), quarters with a variable first sample and SCC ≥ 150,000 cells/mL and no microbiological growth at the second and third sample times (high negative postcalving, HNPost; $n = 6$), and quarters with SCC ≥ 150,000 and positive microbiological growth at all three samples (high positive, HPos; $n = 3$) were followed until 150 d in milk (DIM). Foremilk samples were collected weekly for SCC analysis and monthly for microbiological analysis. Log-rank tests were performed to analyze survival to clinical mastitis (CM) and growth in monthly culture and SCC was analyzed with repeated measures analysis. The SCC of LN quarters was lowest throughout the first 150 DIM ($P < 0.001$) and LN quarters were least likely to experience a case of CM ($P < 0.001$). Fifty percent of HNPost quarters experienced CM, as compared to 1 of 3 HP quarters, 3 of 17 HNPre quarters, and only 2 of 76 LN quarters. LN quarters were also least likely to have bacterial growth in monthly samples ($P < 0.001$). All HP and HNPost quarters had bacterial growth in one or more monthly milk samples, while 36% of HNPre quarters and 9.7% of LN quarters had bacterial growth in one or more monthly milk samples. The most common bacteria isolated from monthly aseptic milk samples ($n = 383$) were coagulase-negative *Staphylococcus* species ($n = 22$), followed by streptococcus-like species ($n = 12$), *Corynebacterium bovis* ($n = 4$), yeast species ($n = 3$), and *Klebsiella*species ($n = 1$). Overall, LN quarters had better health outcomes during the first 150 DIM.

Key Words: dry period, inflammation, mastitis

0857 Interaction among energy status, dietary protein, and vitamin A in periparturient dairy cows: Effects on milk fatty acid profile and gross milk yield efficiency. Y. Chen*, K. C. Ramsey, C. Y. Tsai, M. A. McGuire, and P. Rezamand, *University of Idaho, Moscow*.

Diet affects the fatty acid (FA) composition of milk. The objective of this study was to determine the interaction of various rates of dietary protein (11 or 13%), vitamin A (0 or 110 IU/kg BW), and monensin (0 or 400 mg/d per head) fed during the dry period (4 wk before expected calving until calving) on milk FA profiles and feed efficiency of early postpartum dairy cows. Multiparous Holstein dairy cows ($n = 80$ total) were blocked by expected calving date and previous milk yield and randomly assigned to one of eight treatments in a 2 × 2 × 2 factorial arrangement of treatments. Milk yield and composition and feed intake were determined daily from calving (d 0) to 21 d postpartum. Results were analyzed using mixed model repeated measures ANOVA. Significance was declared at $P \leq$

0.05 while $0.05 < P \leq 0.1$ was considered a trend. Dietary vitamin A × monensin affected yield of several FA in milk; cows receiving both vitamin A and monensin during the dry period produced more C18:3 n3 as well as C22:6 compared with other treatment groups ($P = 0.004$ and 0.009, respectively). The interaction of dietary vitamin A × monensin also affected yield of C18:3 n6, sum of saturated FA, sum of unsaturated FA, sum of n3, sum of n6 FA, sum of MUFA, sum of PUFA, and sum of preformed FA. Dietary protein × monensin × vitamin A affected yields of C16:0 + C16:1, C20:4, C20:5, and C22:6 ($P < 0.03$ for all). Furthermore, greater dietary protein reduced the yield of C18:3 n3 and sum of *de novo* FA. Dietary protein × monensin × vitamin A tended to affect yields of C18:1 *cis*, C18:2 *cis*, C18:3 n3, C18:3 n6, sum of n3, sum of n6 FA, and n6:n3. Both energy corrected (ECM) and 3.5% fat corrected milk were interactively affected by dietary protein, monensin, and vitamin A ($P < 0.03$). Gross milk yield efficiency (ECM/DMI) was changed over time but no dietary effect was detected. Overall, diet composition altered the profile and yields of milk FA as well as ECM of early lactating Holstein cows.

Key Words: dairy cow, fatty acids, feed efficiency

0858 Effect of intrammamary infusion of chitosan hydrogels on bovine mammary gland involution after drying-off.

0859 Differences in body condition of gilts that are maintained from mating to the end of gestation affect their mammary development.

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The goal of this project was to determine if different body conditions in late gestation that are due to varying body conditions at mating affect mammary development of gilts. Gilts that were fed ad libitum in the growing period were selected based on their backfat thickness (BF) to form three groups at mating, namely, low (LBF; 12–15 mm, $n = 14$), medium (MBF; 17–19 mm, $n = 15$), and high (HBF; 22–26 mm, $n = 15$) BF. During gestation, LBF, MBF, and HBF gilts were fed approximately 1.25, 1.43, and 1.63 of maintenance requirements to maintain their differences in body condition. Daily feed intake was increased by 1 kg in the last 10 d of gestation. All gilts had their BF measured ultrasonically at mating and every 15 to 20 d thereafter. Blood samples were obtained at mating and on d 109 of gestation to measure concentrations of IGF-1, glucose, insulin, estradiol, urea, FFA, and adiponectin. Gilts were slaughtered on d 110 of gestation to collect mammary glands for compositional analyses and measures of mRNA abundance for selected genes. The MIXED procedure of SAS using a univariate model (3 levels) was used for statistical analyses and means were compared using the Tukey test. Mammary extraparenchymal tissue weight was less in LBF and MBF than in HBF gilts (1259.3, 1402.7, and 1951.5 ± 70.4 g, respectively, $P < 0.01$). Weight of parenchymal tissue was not altered by treatment ($P > 0.10$), but its composition was affected. Concentrations of DNA and RNA decreased as BF increased ($P < 0.01$), whereas percent fat and dry matter increased ($P < 0.01$). Mammary expression of *CSN2* (β-casein) in parenchymal tissue was also lower in HBF than LBF gilts ($P < 0.05$). On d 109 of gestation, concentrations of insulin, IGF-1, and adiponectin were greater ($P < 0.05$) in HBF than in LBF or MBF gilts, whereas those of urea were lower ($P < 0.01$). Maintaining differences in body condition from mating to the end of gestation therefore had an impact on mammary development of gilts. Extraparenchymal tissue mass was affected and, more importantly, composition of parenchymal tissue.

Key Words: body condition, gilt, mammary development

0860 Stem cells and cell hierarchy in the bovine mammary gland.

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Elucidating cell hierarchy and lineage commitment in the mammary gland is fundamental for understanding its development and for establishing methodologies aimed at increased production via stem-cell manipulation. Here, we demonstrate the existence of bovine mammary stem cells and describe enrichment and transplantation methodologies and attempts made to manipulate this population. Lin^- bovine mammary epithelial cells (bMECs) from Holstein heifers were sorted according to CD24 and CD49f expression into four populations. The $CD24^{med}CD49f^{pos}$–enriched population maintained high expression of basal markers. In culture, it generated luminal and basal clones and had high floating-sphere formation and growth rate. Upon transplantation into cleared mouse mammary fat pad, it gave rise to multilayered outgrowths with self-renewing properties. This population was positioned at the top of the cell hierarchy and referred to as the stem cell population. A more committed, bipotent basal population generated both luminal and basal clones in vitro but was almost completely restricted to generating unilayered basal outgrowths. Together with the luminally restricted progenitor population, it may serve as a reservoir for the highly differentiated luminal cells. Two markers, E-cadherin and miR-200c, whose expression levels correlate with differentiation, assisted in more comprehensive delineation of the bovine mammary cell hierarchy. Xanthosine administration did not affect the proportion of stem cells in bovine implants

transplanted into the cleared mouse fat pad. However, it had a latent negative effect on cell proliferation and may, therefore, interfere with mammary gland development and also limit tumor growth. To analyze the development of bovine mammary morphology in the mouse mammary stroma, bMECs were transplanted into the cleared mammary fat pad of immunodeficient mice. Multilayered hollow spheres developed within fibrotic areas. They shared morphological and immunohistochemical characteristics with the heifer gland but did not extend via ductal morphology. Nevertheless, a single case of terminal ductal lobuloalveolar unit (TDLU) development was recorded in mice treated with estrogen and progesterone, implying the feasibility of this representative bovine morphology's development. *In vitro,* paracrine inhibition of bovine epithelial mammosphere development by adipocytes was recorded, and it was antagonized by FGF administration. This indicates an active equilibrium between inhibitory and promotive effects exerted by the adipose and fibrotic regions of the stroma, respectively. Together, these findings imply that unique bovine mammary cell properties are integrated within a conserved mammary cell hierarchy paradigm delineated in their mouse and human counterparts.

Key Words: bovine, cell hierarchy, mammary gland, stem cells

0861 Optimal combination of histidine, lysine, methionine, and leucine affect β-casein synthesis via mTOR signaling pathway in bovine mammary epithelial cells.

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Assessing the regulatory effect of the optimal ratio of the essential amino acids (EAAs) on milk protein synthesis is vital to AA requirement models for lactation. The study employed response surface methodology (RSM) to determine the optimal ratio of histidine, lysine, methionine, and leucine on β-casein expression level in vitro and to clarify the effect of four EAAs on β-casein through mechanistic targeting of the rapamycin (mTOR) signaling pathway. A central composite design containing 5 axial points per EAA, and 28 combinations of the four EAAs was performed for our study. The efficiency of RSM and the changes of the mTOR-related signaling proteins were further verified by western blot. The protein band values from the AA-supplemented cells were related to their AA-deprived controls. The β-casein data from the RSM experimental design were analyzed using a regression model by Design–Expert 8.0.6. The other experimental data were analyzed using Tukey's test for post-hoc multiple comparisons of treatment means by SAS. Differences between experimental groups were considered significant at $P < 0.05$. The results showed that β-casein level was significantly affected by all four EAAs ($P < 0.01$, $R^2 = 0.71$). The optimal conditions for β-casein expression were as follows: His:Lys:Met:Leu = 5:6:1:7. Only a significant interaction of Leu and Met was observed as to β-casein expression ($P < 0.01$). Further experiments validated that under the optimal mixture of four EAAs, the expression of β-casein was 98% as high as the positive control (i.e., media with all AAs). The phosphorylation of mTOR, Raptor, GβL, S6K1, Rps6, and eEF2 was increased with supplementation of either single EAAs or the optimal combination of EAAs. Finding the best combination of these four EAAs promoted β-casein expression, and this appeared to be mediated through activation of the mTORC1 signaling pathway.

Key Words: bovine mammary epithelial cells, optimal ratio of EAAs, mTOR, β-casein

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0862 The goat (*Capra hircus*) mammary gland secretory tissue proteome as influenced by weight loss: A study using label-free proteomics.

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The objective of this work was to study the effect of seasonal weight loss (SWL) on the mammary gland secretory tissue proteome in two goat breeds from the Canary Islands. Two lactating dairy goat breeds from the Canary Islands with different levels of tolerance to SWL were used: Majorera (tolerant) and Palmera (susceptible). Within each breed, goats with the same age and stage of lactation were divided into two groups: control (constant weight) and restricted (15% liveweight reduction). Four groups were established: Palmera control (PC, $n = 6$), Majorera control (MC, $n = 4$), Palmera restricted (PR, $n = 4$), and Majorera restricted (MR, $n = 5$).

Animals on the restricted groups were fed on standard wheat straw, and animals in the control groups were fed on alfalfa hay supplemented with maize, soy, and dehydrated beetroot. At Day 22 of the trial, mammary gland biopsy samples were obtained. Samples (30 mg) were ground in liquid nitrogen with mortar and pestle, added to 500 μL of ammonium bicarbonate 50 mM, urea 8M, and thiourea 2M buffer, homogenized, and centrifuged, and the supernatant recovered. Per sample, 15 μg of proteins were trypsin digested (FASP protocol) and desalted. Peptides were loaded onto reverse-phase C18 columns and analyzed on an LTQ-Orbitrap Velos mass spectrometer. Protein identification and label-free quantification were performed using Mascot (Matrixscience) and Progenesis software (Nonlinear Dynamics). A total of 1010 proteins were identified, from which 96 proteins were considered statistically different among groups (fold change > 1.98 and $P < 0.05$). After SWL, there was an increase of proteins related to apoptosis and stress processes in both breeds. Moreover, both breeds showed a decrease in the number of proteins related to protein, carbohydrate, and fat biosynthesis. When both breeds were compared after SWL, the Majorera breed showed higher expression of immune system related proteins compared to the Palmera breed. In contrast, the Palmera breed showed higher expression of proteins related to apoptosis, ketone body formation (fatty liver), and protein metabolic processes compared to the Majorera breed. In conclusion, the two goat breeds have different metabolic reactions to SWL, highlighting differences particularly related to the immune system (higher expression in the tolerant breed) and apoptosis (higher expression in the susceptible breed).

Key Words: goat, label-free proteomics, mammary gland, seasonal weight loss

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0863 Pre-calving and early lactation factors that predict milk casein and fertility in the transition dairy cow. R. M. Rodney*[1,2], J. K. Hall[3], C. T. Westwood[4], P. Celi[5], and I. J. Lean[1,2], *[1]Scibus, Camden, Australia, [2]University of Sydney, Camden, Australia, [3]Halltech Services, Orange, Australia, [4]Kimihia Research Centre, PGG Wrightson Seeds Limited, Lincoln, Canterbury, New Zealand, [5]Faculty of Veterinary and Agricultural Sciences, University of Melbourne, Parkville, Australia*.

Multiparous Holstein cows ($n = 82$) of either high or low genetic merit (GM) (for milk fat + protein yield) were allocated to one of two diets in a 2 × 2 factorial design. Diets differed in the ratio of rumen-undegradable protein (RUP) to rumen-degradable protein (37% RUP vs. 15% RUP) and were fed from 21 d pre-calving to 150 DIM. This study evaluated the effects of these diets and GM on concentrations of milk casein (CN) variants and aimed to identify pre-calving and early lactation variables that predict milk, CN and protein yield and composition, and fertility of dairy cows. It explored the hypothesis that low milk protein content is associated with lower fertility, extending this to also evaluate the contribution of CN contents. Yields (kg/d) for CN variants were 0.49 and 0.45 of α CN, 0.38 and 0.34 of β CN, 0.07 and 0.06 of κ CN, and 0.10 and 0.09 of γ CN for high and low RUP diets, respectively. Increased RUP increased milk, CN and milk protein yields. Increased GM increased milk and γ CN yields and tended to increase milk protein yield. The effects of indicator variables on CN variant yields and concentrations were largely consistent, with higher body weight and α amino nitrogen resulting in higher yields, but lower concentrations. An increase in cholesterol was associated with decreased CN variant concentrations, while disease lowered CN variant yield. A diet high in RUP increased the proportion of first services that resulted in pregnancy from 41 to 58%. Increased pre-calving metabolizable protein (MP) balance decreased the proportion of first services that resulted in pregnancy when evaluated in a model containing CN %, milk protein yield, diet, and GM. This indicates that the positive effects of a diet high in RUP on fertility may be curvilinear as cows with a very positive MP balance before calving were less fertile than those with a lower, but positive, MP balance. Prepartum MP balance was important to production and reproduction outcomes, while surprisingly, metabolizable energy balance was not. Cows producing the lowest quartile of milk protein percentage were 28% less likely to become pregnant during the first 150 DIM. Milk CN % was similarly positively associated with improved pregnancy at first service. This study demonstrates the importance of protein metabolism to reproductive performance of the dairy cow.

Key Words: casein, fertility, protein degradability

0864 Increasing blood 5-hydroxy-L-tryptophan concentration for prevention of periparturient hypocalcemia in dairy cows. L. E. Hernandez-Castellano*[1], S. R. Weaver[2], L. L. Hernandez[2], and R. M. Bruckmaier[1], *[1]Veterinary Physiology, Vetsuisse Faculty, University of Bern, Bern, Switzerland, [2]Department of Dairy Science, University of Wisconsin, Madison*.

Hypocalcemia in dairy cows is caused by the sudden increase in demand for Ca^{2+} by the mammary gland for milk production after partum and simultaneously the limited ability of Ca^{2+} to be mobilized from bone in a timely manner. Serotonin (5-HT) is a key factor which mediates Ca^{2+} mobilization from bones. Therefore, we hypothesized that administration of 5-hydroxy-L-tryptophan (5-HTP), a 5-HT precursor, would increase 5-HT concentration in blood, and in turn induce Ca^{2+} mobilization from bone. In this study, 20 Holstein dairy cows were randomly assigned to two experimental groups. Ten animals received a daily i.v. infusion of 1 L of 0.9% NaCl (control; C). The other 10 animals received 1 L of 0.9% NaCl containing 1 mg of 5-HTP/kg BW daily (5-HTP group). Infusions were performed beginning on Day 10 before estimated

parturition. Infusions were conducted until the day of parturition, resulting in at least 4 d of infusion. Until parturition, blood samples were collected every morning before the infusions, and after parturition daily until Day 7, and on Day 30. Milk yield was recorded during this period. No differences between groups were observed for blood glucose, Mg^{2+}, β-hydroxybutyrate, and non-esterified fatty acid concentrations. Serum 5-HT concentration was increased until Day 5 after partum in the 5-HTP group compared to the C group. Colostrum 5-HT concentrations were higher in the 5-HTP group than in the controls (37.10 ± 3.12 vs. 25.02 ± 2.75 nM; $P < 0.05$), but differences were undetectable in milk 7 d after partum (13.43 ± 1.12 *vs.* 14.63 ± 1.13 nM; $P > 0.05$). Serum total Ca^{2+} concentrations decreased in both groups around parturition ($P < 0.05$), however, 5-HTP group had higher blood Ca^{2+} concentrations than the controls on Day 1 (1.93 ± 0.06 vs. 1.62 ± 0.09 mM) and Day 2 (2.07 ± 0.04 vs. 1.83 ± 0.07 mM), respectively ($P < 0.05$). Additionally, colostrum yield (first milking) was lower in the 5-HTP group compared to the C group (5.63 ± 0.34 *vs.* 8.56 ± 0.47 kg; $P < 0.05$), but no differences in colostrum IgG concentration were detected (68.41 ± 5.20 vs. 60.70 ± 10.27 mg/mL; $P > 0.05$). Milk yield did not differ between groups during the rest of the experiment. In conclusion, 5-HTP infusions increased blood 5-HT concentration. Moreover, 5-HTP can reduce the decline in blood Ca^{2+} concentration around parturition and hence may influence the occurrence of clinical or subclinical hypocalcemia. Finally, 5-HTP reduced colostrum production without affecting IgG concentrations, suggesting the mass of IgG available from colostrum may be sufficient to fulfill the needs of the offspring.

Key Words: dairy, hypocalcemia, serotonin

0865 Beta-hydroxybutyrate infusion affects glucose metabolism before and after parturition in dairy cows. M. Zarrin[1,2], L. Grossen-Rösti[1], R. M. Bruckmaier[1], and J. J. Gross[*1], *[1]Veterinary Physiology, Vetsuisse Faculty, University of Bern, Bern, Switzerland, [2]Department of Animal Science, Yasouj University, Yasouj, Iran*.

Recent studies in mid- and late-lactating dairy cows showed that β-hydroxybutyrate (BHBA) infusion had considerable impact on glucose metabolism and immune response during intramammary lipopolysaccharide challenge. The objective of the present study was to infuse BHBA during the dry period and after parturition to investigate the effects of elevated plasma BHBA concentrations on metabolism and endocrine changes in transition dairy cows. Eight multiparous Holstein cows in wk −2 (a.p.) and wk +2 (p.p.) relative to calving were infused (from 0800 AM to 1200 AM, 4 h) with a BHBA solution to increase plasma BHBA concentrations to 1.5 to 2.0 mmol/L (HyperB). The same period on the next day (without any infusion) was assigned as the control (Control). Blood samples were taken before the start of infusion as reference samples, and every 30 min during the following 6 h (4 h infusion and 2 h after the stop of infusion) in HyperB and on the control day, and were analyzed for glucose, BHBA, insulin, and glucagon. Plasma BHBA concentrations reached 1.7 ± 0.1 mmol/L (a.p.), and 1.6 ± 0.2 mmol/L (p.p.) in HyperB compared with 0.6 ± 0.1 mmol/L, and 0.6 ± 0.0 mmol/L in Control, respectively. The 4-h average BHBA infusion rate was 12.4 ± 1.0 and 13.3 ± 0.9 μmol/kg BW/min in wk −2 and +2, respectively ($P = 0.13$). BHBA infusion caused a decrease of plasma glucose concentrations, compared with pre-infusion levels, both before and after parturition ($P < 0.05$). The glucose response did not differ between a.p. and p.p. infusion even though basal glucose concentrations were different before and after calving (3.7 ± 0.1 vs. 3.2 ± 0.2 mmol/L, $P < 0.05$). BHBA infusion increased plasma insulin a.p. but not p.p. ($P < 0.05$) despite greater basal insulin concentrations before compared with after parturition (29.0 ± 8.4 vs. 5.8 ± 0.8 μU/mL, $P < 0.05$). Though basal glucagon concentrations were not different between wk −2 and +2 ($P = 0.30$), BHBA infusion decreased plasma glucagon only p.p. ($P < 0.05$). These findings show that effects of hyperketonemia on plasma glucose concentrations are independent of lactational stage, but endocrine adaptation to hyperketonemia differs before and after parturition. It can be assumed that BHBA has a glucose sparing effect and is a metabolic key regulator in early lactating dairy cows.

Key Words: β-hydroxybutyrate, glucagon, transition period

0866 Impact of increasing dietary crude protein content on urinary nitrogen excretion and milk nitrogen secretion of lactating sows. T. F. Pedersen[*1], C. Y. Chang[1], T. S. Bruun[2], and P. K. Theil[1], *[1]Aarhus University, Tjele, Denmark, [2]SEGES Pig Research Centre, Copenhagen, Denmark*.

The objective of the current study was to evaluate the effect of increased dietary crude protein (CP) content on urinary nitrogen (N) excretion and milk N secretion during lactation. In total, 36 sows from first to fifth parity were included in the experiment from parturition until weaning at d 28. Sows were allotted to 6 different treatments, with dietary CP contents of 149, 164, 174, 183, 193, and 208 g/kg DM, while dietary contents of SID lysine, methionine, threonine, and tryptophan were kept constant by including crystalline amino acids (AA). Sows were fed individually according to Danish recommendations, except for the recommended content of dietary CP. On d 2 postpartum, litters were equalized to 14 piglets and weighed on d 2, 10, and 17 to calculate milk production. Sows were fitted with urinary catheters on d 3, 10, and 17, and urine was collected three times during a 6-h period each week to estimate the daily urine production and N excretion. Additionally, milk samples were collected on d 3, 10, and 17 to estimate the daily secretion of N in milk. Fixed effects of week, treatment, and interaction were tested using a mixed

model. Overall, there was no effect of treatment on N content in urine ($P > 0.10$) or the amount of urine ($P > 0.10$). However, excretion of urinary N tended ($P < 0.10$) to be lowest for sows fed 164 g CP/kg DM (20.5 g N) and highest (1.5- to 1.8-fold higher) for sows fed 193 and 208 g CP/kg DM, respectively. The urinary N excretion averaged 28 g/d and did not change as lactation progressed ($P > 0.10$). Milk protein content increased with increasing dietary CP content, from 4.8 to 5.4% ($P < 0.05$). Milk production was comparable among treatments ($P = 0.09$) and ranged from 10.7 to 12.7 kg/d. The milk N secretion increased from 76 g/d on d 3 to 109 g/d on d 17, but it was not affected by dietary treatment ($P > 0.10$). In conclusion, the highest milk protein content was observed at 208 g CP/kg DM, whereas the lowest urinary N excretion was observed at 164 g CP/kg DM.

Key Words: milk protein, nitrogen loss, nutrition.

0867 Intramammary prednisolone affects the permeability of the blood-milk barrier during LPS and LTA induced mastitis in dairy cows.

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Mastitis can induce pathogen dependent changes in the permeability of the blood-milk barrier and therefore the paracellular transfer of blood and milk components. Glucocorticoids are known to increase the integrity of this barrier. The objective of this study was to examine the effect of intramammary prednisolone (PRED) on the blood-milk barrier in cows during mastitis induced by lipopolysaccharide (LPS) from *Escherichia coli* or by lipoteichoic acid (LTA) from *Staphylococcus aureus*. Thirty-one dairy cows, divided into 6 groups, were intramammarily challenged in one quarter with LPS, LTA, LPS and PRED, LTA and PRED, saline (control), or PRED. Quarters had a somatic cell count (SCC) of 60 ± 10 × 10^3 cells/mL before the experiment. The chosen doses of LPS and LTA induced a similar increase of SCC to 1420 ± 360 × 10^3 cells/mL at 4 h after challenge. Milk and blood samples were collected hourly from 0 to 8 h after challenge. Milk was analyzed for SCC, immunoglobulin (Ig) G, serum albumin (SA), and lactate dehydrogenase (LDH). Plasma was tested for the milk protein α-lactalbumin (ALA). Differences between treatments were tested by analysis of variance using a MIXED procedure and were considered significant if $P < 0.05$. The SCC in milk of control quarters and quarters treated only with PRED did not change throughout the experiment. In LTA challenged quarters with additional PRED administration, there was a reduction in SCC to control quarter level, whereas in LPS treated quarters, additional PRED administration had no effect on SCC. LDH activity did not significantly increase in LTA treated quarters, but increased in LPS quarters from 27 ± 7 U/L before challenge to 404 ± 115 U/L at 6 h after challenge, and decreased to control quarter levels with additional PRED administration. For SA and IgG, only LPS quarters showed an elevation from 0.25 ± 0.07 and 0.32 ± 0.04 mg/mL to 1.34 ± 0.57 and 1.16 ± 0.52 mg/mL, respectively, at 4 h after challenge. The PRED treatment reduced both concentrations to control quarter levels. There were no differences in plasma ALA concentrations in PRED-treated cows compared to cows that received only LPS or LTA. In conclusion, the pathogen specific appearance of blood constituents in milk during mastitis demonstrates a differential activation of the blood-milk barrier. These differential effects can be influenced by intramammary administration of glucocorticoids in a pathogen specific manner.

Key Words: blood-milk barrier, endotoxin, glucocorticoid, mastitis

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0868 Regulation of sterol regulatory element binding protein-1 in bovine mammary epithelial cells.

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The objective of this study was to investigate the molecular mechanisms by which nutrients regulate sterol regulatory element binding protein-1 (SREBP1) in bovine mammary epithelial cells (Mac-T). Three models were tested. First, the relationship between SREBP1 and the mechanistic target of rapamycin (mTOR) signaling was tested through mTOR activation/inhibition as well as SREBP1 knockdown by siRNA. Second, the relationship between AMPK and SREBP1 was tested in t10,c12-CLA-treated Mac-T cells. Third, the activation of SREBP1 was tested by glucose supplementation. Results showed that mTOR activation increased SREBP1 protein as well as the lipogenic gene expression by over 50%. While inhibition on mTOR failed to increase SREBP1, siRNA-directed SREBP1 knockdown confirmed that insulin enhanced lipogenic gene transcription through SREBP1. Further examination found that mTOR signaling regulates SREBP1 by preventing its proteosomal degradation. t10,c12-CLA decreased SREBP1 protein and lipogenic gene expression through phosphorylation of AMPK, while inhibition of AMPK phosphorylation partially rescued the SREBP1 reduction. Lastly, low glucose (1 mmol/L) was able to increase mature SREBP1 level by 2.2-fold. Increasing glucose concentration increased SREBP-cleavage activating protein, a key regulator of SREBP1 activation, in a dosage- and time-dependent manner. In conclusion, these results showed that major cellular metabolic regulators play roles in SREBP1 activation and degradation thus regulating lipogenesis in response to the nutrients provided.

Key Words: AMPK, glucose, mTOR, SREBP1, t10,c12-CLA

0869 Efficacy of dual X-ray absorptiometry as a means to measure mammary gland development in dairy heifer calves. A. J. Geiger*, C. L. M. Parsons, and R. M. Akers, *Virginia Tech, Blacksburg.*

A non-invasive means to assess mammary gland growth in dairy heifers is highly desirable from a research and animal welfare standpoint. We evaluated dual X-ray absorptiometry (DXA) as such a tool. Thirty-six Holstein heifer calves were reared on: 1) a control milk replacer (MR) fed at 454 g powder/d (CTL; 20% crude protein [CP], 20% fat), or 2) an enhanced MR fed at 1135 g powder/d (E; 28% CP, 25% fat). The MR was fed for 8 full weeks with weaning (half milk intake) occurring during the last week. Starter feed was offered after week 4 but was balanced between treatments. At weaning, a subset of calves were sacrificed ($n = 6$/diet). Remaining calves received estradiol (E_2) implants and were sacrificed at week 10. The 4 treatments were: 1) CTL, 2) CTL + E_2 (CTL-E2), 3) E, and 4) E + E_2(E-E2). After sacrifice, udders were removed and half-udders were snap-frozen. Frozen mammary glands were scanned and lipid and lean tissue contents were determined with and without the skin intact. Correlations were calculated between mammary gland lipid content via DXA, and biochemical extraction using PROC CORR function in SAS. Correlations were also calculated between mammary gland DXA lean tissue values and mammary parenchyma weight and DNA content. Correlations with skin removed were slightly higher. Overall, lipid content of the mammary gland as determined by DXA was highly correlated with lipid content determined biochemically, regardless of skin presence ($r = 0.92$; $P < 0.01$ for skin intact; $r = 0.94$; $P < 0.01$ for skin removed). With one exception, correlations were similar within treatments ($r = 0.59$, 0.91, 0.94, 0.91, and 0.96, 0.93, 0.97, 0.92 for treatments CTL, CTL-E2, E, and E-E2 with and without skin, respectively; $P < 0.01$ for all except for CTL with skin intact [$P = 0.16$]). Correlations between mammary gland lean tissue content determined by DXA and dissected parenchyma weight ($r = -0.01$; $P = 0.94$) or parenchyma DNA content ($r = 0.22$; $P = 0.21$) were low and nonsignificant. Results indicate that DXA analysis can be used to evaluate fat content of the mammary gland in young heifers but not the mass of mammary parenchymal tissue. It remains to be determined if this technology can be used in intact animals.

Key Words: calf, dual X-ray absorptiometry, mammary gland

0870 Percentages of milk fat, lactose, and protein are affected by diurnal variations in dairy goats. F. Rosa*[1], J. S. Osorio[1], J. Lohakare[1], M. Moridi[2], A. Ferrari[3], E. Trevisi[3], and M. Bionaz[1], *[1]Department of Animal and Rangeland Sciences, Oregon State University, Corvallis, [2]University of Guilan, Rasht, Islamic Republic of Iran, [3]Universita Cattolica del Sacro Cuore, Piacenza, Italy*.

Diurnal variations in milk synthesis in dairy goats are not known but can have important implications for nutrigenomic interventions to improve milk synthesis. The diurnal variation in milk synthesis was evaluated in 12 *Saanen* multiparous goats in early to mid-lactation. Six goats were treated with an intrajugular injection of 2,4-thiazolidinedione (TZD) at 1000 h, and 6 control goats received saline. Goats received an NRC-compliant diet at 0800 h. All goats received an intramammary infusion with *Streptococcus uberis* 10 d before the onset of treatment to induce sub-clinical mastitis in the right half of the udder, the left half being used as control. Goats were milked every 2 h from 0700 h to 1900 h. Besides milk yield, milk samples were collected for components analysis, and jugular blood samples were collected for analysis of NEFA, triacylglycerol (TAG), urea (BUN), and glucose. Data were analyzed using a GLIMMIX procedure of SAS with time, treatment, and treatment × time as fixed effects for blood variables, with the addition of udder halves and relative interactions as fixed effects for milk variables. Goat was used as random effect. Mean separation was done using Tukey's test. The TZD injection did not affect any of the measured milk variables. SCC was not affected by time but was greater in the right vs. left udder half in both treatment groups ($P < 0.05$). Percent milk fat peaked at 0900 h (4.9%) and decreased afterward. The percentages of milk protein and milk urea (MUN) peaked at 1100 h (2.6% and 28.2 mg/dL, respectively) and the percentages of lactose (4.4%) and solid nonfat (SNF; 7.8%) peaked between 1300 h and 1500 h. None of the single blood metabolites were affected by treatment or treatment × time interaction but all were greatly affected by time ($P < 0.05$). Glucose consistently increased until 1300 h, urea reached a peak at 1100 h, NEFA decreased until 1300 h and increased afterward, and TAG consistently decreased throughout the day. The sum of TAG and NEFA, an index of available fatty acids, was affected by treatment ($P < 0.05$), with values being greater in TZD compared with the control. Significant ($P < 0.01$) positive correlations were observed between blood glucose and percent milk lactose, NEFA and SNF, TAG and SCC, and between BUN and milk fat and MUN. Negative correlations ($P < 0.05$) were observed between glucose and percent milk fat, and between TAG and SNF. This data highlights the diurnal variations occurring in milk synthesis in goats, suggesting that synthesis of milk fat is not exclusively driven by the availability of fatty acids.

Key Words: diurnal variations, goat, milk synthesis

0871 Comparative effect of two commercial preparations of bovine somatotropin on milk yield and overall performance in Chilean dairy cows. M. A. Barrios[1], P. Melendez[*2], and M. Duchens[1], [1]*University of Chile, Santiago, Chile,* [2]*University of Missouri, Columbia*.

To compare the effect of two commercial preparations of bSTr available in Chile on milk production and overall performance of dairy cattle, 348 confined Holstein cows from a high-producing farm located in Casablanca, Valparaíso Region (33.31 S, 71.40 W) were used. Cows were randomly assigned from 70–76 DIM to one of two treatment groups. Group 1 (n = 161) was Boostin® (LG LifeSciences, South Korea) and Group 2 (n = 187) was Lactotropin® (Elanco, USA). In both groups, the hormone was administered every 14 d to about 30 d before dry off. Information was obtained from computerized systems and collected until the end of the eighth cycle of treatment (approximately 180 DIM). Milk data was processed through a repeated measures ANOVA. Fertility was analyzed by logistic regression to evaluate the risk of pregnancy at first insemination. Days open were analyzed with Kaplan-Meier survival analysis to compare days at pregnancy. Frequencies of clinical mastitis were analyzed by logistic regression. Monthly SCC linear scores were analyzed using repeated measures ANOVA. Finally, culling rate was assessed by a Kaplan-Meier survival analysis. No significant differences in the interaction of time by treatment were observed for milk yield (P = 0.07), averaging 42.3 L/d for Lactotropin®, and 42.8 L/day for Boostin®. No significant differences on calving-to-conception interval were recorded during the first 180 d of lactation (P = 0.19), showing a median of 101 d in Lactotropin® group and 90 d in cows treated with Boostin®. Conception rate at first insemination was 40% in cows treated with Boostin® and 32.8% in cows treated with Lactotropin® (P = 0.16). Cows treated with Lactotropin® have a higher incidence of clinical mastitis (33.5%) compared to Boostin® cows (21.1%) (P < 0.01). However, no significant differences were observed in SCC between groups. Finally, treatment has no effect on culling rate (P = 0.78). In conclusion, there are no substantial differences between hormone preparations regarding milk production, udder health, and fertility in high-producing Holstein cows.

Key Words: bST, fertility, mastitis, milk yield

LIVESTOCK WATER SYMPOSIUM

0872 Understanding blue and green water for feed production in animal agriculture. J. G. Warren*, *Oklahoma State University, Stillwater*.

Increasing demand for animal protein combined with concern about water scarcity demands thoughtful considerations of the water footprint of animal agriculture. This water footprint can be discussed in the context of green water, which is rainfall that does not become runoff, and blue water, which is surface or groundwater that is consumed as a result of the animal production system. Much of the green and blue water utilized by animal agriculture provides for the production of grain and forages. As such, the type of feed utilized by specific animal production systems can dramatically influence the water footprint of the system. Grazing systems will generally result in larger water footprints than grain-based production systems because the higher quality grain-based systems provide for more gain per unit of water used. However, this reduced water footprint comes with increased environmental impacts such as erosion and offsite nutrient losses from the grain production systems. Furthermore, pasture-based systems overwhelmingly utilize green water, which would likely be consumed at similar rates if the pasture was used for meat production or wildlife habitat. Lastly, regional differences in soil type, rainfall distribution, and atmospheric water demand (evapotranspiration) also influence the water footprint of animal agriculture by impacting crop water use efficiency.

Key Words: life cycle analysis, groundwater, feed production

0873 Mineral balances including TMR, drinking water and assay minerals in the milk. A. R. Castillo*, *UC Cooperative Extension, Merced, CA*.

Drinking water for dairy animals or manure for soil applications can be both a source of mineral nutrients and toxic substances. Commercial dairy production systems (grazing or indoors) are evolving to a larger scale, with more cows per farm and milk production per cow. Including assayed concentrations of minerals in the diet, drinking water and milk could improve the accuracy of calculations of herd or pen mineral balances. The aim of this presentation is to discuss mineral contents in TMR, water, and milk on mineral balances and excretion in lactating dairy cows. A mineral balance study in California on 40 dairies with low total salts (TS) drinking water for lactating dairy animals (0.2 to 1.5 g TS/L) was performed to compare TMR mineral content with the NRC requirement, with and without including minerals in drinking water, and the average NRC values for milk mineral concentrations to assayed minerals in the bulk tank milk. Most TMR minerals were in excess of NRC requirements. When including minerals in drinking water, Mg, Na, Cl, S, and Cu increased TMR median mineral contents by about 5% (ranging from 3.6% for S to 7% for Na). The assayed values of minerals in milk were lower than NRC averages (i.e., Mg, 49%; Na, 58%; Cu, 295%; and Fe, 525%). Estimated excretions of minerals via manure varied substantially across farms. Farms in the 10th percentile had estimated mineral excretions via manure 2 to 3 times less than those in the 90th. For example, daily K median excretion was 321 g/cow, from 240 (10th) to 425 g/cow (90th