

## Ruminant Nutrition: Dairy II

**T380 Dietary grain source and oil supplement: Milk fat synthesis and milk fatty acid profile of Holstein cows.** Shahryar Kargar<sup>\*1</sup>, Gholam Reza Ghorbani<sup>2</sup>, Veerle Fievez<sup>3</sup>, and David J. Schingoethe<sup>4</sup>, <sup>1</sup>Shiraz University, Shiraz, Iran, <sup>2</sup>Isfahan University of Technology, Isfahan, Iran, <sup>3</sup>Ghent University, Melle, Belgium, <sup>4</sup>South Dakota State University, Brookings.

Effects of grain type and dietary oil supplement on milk fat depression, milk fatty acid (FA) profile, and lactational performance of dairy cows were evaluated using 8 multiparous Holstein cows in a duplicated 4 × 4 Latin square design with a 2 × 2 factorial arrangement of treatments. Experimental diets contained either ground barley or ground corn supplemented with either fish oil or soybean oil at 2% of dietary dry matter (DM). Experimental diets contained 28.5 and 31.2% of cereal grain in corn- and barley-based diets, respectively, as the sole source of grain. The forage component of the experimental diet was a mixture of corn silage (19.0% of DM) and alfalfa hay (21.0% of DM). Treatment periods were 25 d, with the final 7 d used for sample and data collection. Data were composited within period and subjected to MIXED MODEL procedure of SAS (SAS Institute, 2003) to account for effects of square, period within square, cow within square, treatments (grain type and oil supplement), and the interaction between grain type and oil supplement. Total FA intake was greater in corn-based diets and also in soybean oil supplemented diets ( $P \leq 0.05$ ). Fish oil decreased intake of all 18-carbon FA but increased intakes of other FA including C16:0, C20:5, and C22:6 ( $P \leq 0.05$ ). No significant differences existed in the DM intake and yield of milk or milk components between barley- and corn-based diets ( $P > 0.05$ ). Fish oil negatively affected feed intake and yields of milk and milk components as compared with soybean oil ( $P \leq 0.05$ ). Although milk fat yield was not affected ( $P > 0.05$ ), the barley-based diets increased the concentration and yield of medium-chain FA but decreased the concentration of long-chain FA as compared with corn-based diets ( $P \leq 0.05$ ). Corn-based diets increased concentration and yield of both *trans*-11 C18:1 and *cis*-9, *trans*-11 C18:2 which was a reflection of greater intake of *cis*-9, *cis*-12 C18:2 (25.1 g/d) as substrate for rumen biohydrogenation ( $P \leq 0.05$ ). Severity of MFD was greater for fish oil- vs. soybean oil which evidenced by the increased concentration and yield of biohydrogenation intermediates associated with MFD (especially *trans*-10 C18:1) in milk fat ( $P \leq 0.05$ ). However, fish oil increased concentration and yield of both *trans*-11 C18:1 and *cis*-9, *trans*-11 C18:2 as compared with soybean oil ( $P \leq 0.05$ ). Results indicated that there was no interaction between the type of grain and oil supplement on induction of MFD and milk fat yield. Although milk fat yield was not affected, milk fat composition was differently modified in barley- vs. corn-based diets. Observed lower milk fat yield for fish oil supplemented diets was coincided with the increased concentration and yield of biohydrogenation intermediates associated with MFD in milk fat.

**Key Words:** grain and oil, milk fat depression, dairy cow

**T381 Effect of biotin and pantothenic acid supplementation on performance and concentration of avidin-binding substances (ABS) in lactating dairy cows.** Gonzalo Ferreira, Alston N. Brown\*, and Christy Teets, *Department of Dairy Science, Virginia Polytechnic Institute and State University, Blacksburg, VA.*

We hypothesized that pantothenic acid reduces the absorption of biotin in lactating dairy cows. Twenty-four Holstein cows (8 primiparous and

16 multiparous) were assigned to a 4 × 4 replicated Latin square design with 18-d periods. Cows were 109 ± 30 d in milk and weighed 591 ± 70 kg at the beginning of the experiment. Treatments consisted of a control diet (CON) containing 53% concentrate and 47% forage, CON supplemented with 20 mg/d biotin (BIO), CON supplemented with 475 mg/d pantothenic acid (PAN), and CON supplemented with 20 mg/d biotin + 475 mg/d pantothenic acid (BIOPAN). Data were analyzed with a Proc Mixed model that included the effects of square (fixed, 5 df), treatment (fixed, 3 df), treatment × square interaction (fixed, 15 df), period (random, 3 df), cow within square (random, 18 df), and the residual error (51 df). No differences among treatments were observed for dry matter intake ( $P < 0.46$ ), milk yield ( $P < 0.68$ ), fat concentration ( $P < 0.85$ ) and yield ( $P < 0.88$ ), protein concentration ( $P < 0.28$ ) and yield ( $P < 0.07$ ), lactose concentration ( $P < 0.90$ ) and yield ( $P < 0.25$ ), and milk urea nitrogen ( $P < 0.49$ ). Biotin supplementation increased the concentration of avidin-binding substances (ABS) in milk ( $P < 0.03$ ). This increase was similar for BIOPAN ( $P < 0.01$ ). Relative to CON, pantothenic acid did not reduce the concentration of ABS in milk ( $P < 0.99$ ). In conclusion, under the conditions of this experiment, pantothenic acid did not affect the absorption of biotin in lactating dairy cows.

**Table 1 (Abstr. T381).** Effect of biotin and pantothenic acid supplementation on performance and concentration of avidin-binding substances (ABS) in milk of dairy cows

Item	CON	BIO	PAN	BIOPAN	SEM	$P <$
DMI, kg/d	21.6	21.5	21.2	21.1	0.49	0.46
Milk yield, kg/d	41.9	41.9	42.1	41.1	1.41	0.68
Fat, %	2.83	2.79	2.88	2.90	0.19	0.85
Fat, kg/d	1.18	1.18	1.22	1.18	0.08	0.88
Protein, %	3.01	3.07	3.03	3.04	0.06	0.28
Protein, kg/d	1.26	1.31	1.30	1.25	0.04	0.07
Lactose, %	4.88	4.90	4.89	4.89	0.04	0.90
Lactose, kg/d	2.06	2.09	2.11	2.02	0.07	0.25
MUN, mg/dL	15.39	15.83	15.42	15.73	0.58	0.49
ABS in milk, ng/mL	28.9 <sup>b</sup>	39.6 <sup>a</sup>	28.9 <sup>b</sup>	43.0 <sup>a</sup>	3.36	0.01

**Key Words:** biotin, pantothenic acid, vitamin

**T382 Assessment of in vitro ruminal fermentation characteristics of lactation dairy diets supplemented with slow-release urea using continuous cultures.** F. Mason<sup>1</sup>, K. Neal<sup>2</sup>, S. Y. Yang<sup>2</sup>, J.-S. Eun<sup>\*2</sup>, and M. Spanghero<sup>1</sup>, <sup>1</sup>Department of Agricultural and Environmental Science, University of Udine, Udine, Italy, <sup>2</sup>Department of Animal, Dairy, and Veterinary Sciences, Utah State University, Logan, UT.

The present study investigated the effects of supplementing slow-release urea (SRU; Optigen, Alltech, Nicholasville, KY) in 3 lactation dairy diets with different forage-to-concentrate ratios (F:C) on in vitro ruminal fermentation characteristics. The experiment was performed in a 3 (F:C) × 2 (without vs. with SRU supplementation) factorial design with 4 independent runs of continuous cultures (n = 4). The 3 F:C included high-forage (HF; 64:36), medium-forage (MF; 51:49), and low-forage diet (LF; 34:66) on a DM basis, and alfalfa hay and cone silage were forage sources at equal proportions. Supplementing SRU considerably reduced dietary concentrations of mixture of soybean meal and canola meal (50:50 on a DM basis) in the SRU supplemented diets. Continuous culture apparatus consisted of 700-mL working volume fermentation

vessels to measure major fermentation end products. Each culture was offered a diet of 20 g DM/d in 2 equal portions at 0800 and 2000 h. Decreasing forage proportion reduced ruminal pH ( $P < 0.01$ ), but supplementing SRU did not influence ruminal pH. Manipulating F:C did not affect total VFA concentration; however, under HF supplementing SRU increased total VFA concentration, while SRU supplementation in MF decreased total VFA concentration, leading to an interaction between F:C and SRU ( $P = 0.02$ ). Decreasing forage proportion decreased acetate proportion, but increased propionate proportion, resulting in a tendency to decrease acetate-to-propionate ratio ( $P = 0.09$ ). In contrast, SRU supplementation did not affect individual VFA proportions. Decreasing F:C decreased ammonia-N concentration, whereas supplementation of SRU tended to increase concentration of ammonia-N ( $P = 0.08$ ). While methane production decreased with decreasing F:C ( $P < 0.01$ ), supplementing SRU tended to increase methane production ( $P = 0.07$ ) mainly due to its sizable increase in MF, leading to a tendency of F:C  $\times$  SRU interaction ( $P = 0.06$ ). Overall results in the current study indicate that supplementing SRU would be more beneficial in HF with enhanced ruminal fermentation evidenced by increased total VFA concentration.

**Key Words:** forage-to-concentrate ratio, ruminal fermentation, slow-release urea

**T383 Ruminal fermentation characteristics of lactation dairy diets with different forage-to-concentrate ratios without or with lipid extract algae in continuous cultures.** S. Y. Yang\*<sup>1</sup>, K. Neal<sup>1</sup>, J.-S. Eun<sup>1</sup>, A. J. Young<sup>1</sup>, and R. C. Sims<sup>2</sup>, <sup>1</sup>Department of Animal, Dairy, and Veterinary Sciences, Utah State University, Logan, UT, <sup>2</sup>Department of Biological Engineering, Utah State University, Logan, UT.

The current in vitro experiment was performed to test the effects of supplementing lipid extract algae (LEA) in lactation dairy diets on ruminal fermentation in a 2 (level of forage in diets)  $\times$  2 (without vs. with LEA) factorial design with 4 independent runs of continuous cultures ( $n = 4$ ). Diets with LEA completely replaced mixture of soybean meal and canola meal (50:50 in a DM basis). The data in this experiment were analyzed using the Proc Mixed procedure of SAS using a model that included fixed effects of level of forage, LEA supplementation, and their interaction and a random effect of fermentor within independent run. Feeding LEA decreased culture pH, regardless of level of forage, but the decrease of culture pH was greater under high-forage diet compared with low-forage diet, resulting in an interaction between level of forage and LEA. Under high-forage diet, total VFA concentration increased with feeding LEA, but it was not affected in low-forage diet, leading to a tendency ( $P = 0.08$ ) of level of forage and LEA interaction. Adding LEA decreased ammonia-N concentration both in high- and low-forage diet. Overall results in this experiment indicate that feeding LEA in lactation dairy diets did not interfere with in vitro ruminal fermentation. The decreased ammonia-N concentration due to feeding LEA may have resulted from less degradation of N fraction in LEA compared with mixture of soybean meal and canola meal.

*Contd.*

**Table 1 (Abstr. T383).** In vitro effects of lipid extract algae supplemented in dairy diets

Item	Diet <sup>1</sup>				SEM	<i>P</i> <sup>2</sup>		
	HF		LF			FC	LEA	INT
	-LEA	+LEA	-LEA	+LEA				
Mean culture pH	6.25 <sup>a</sup>	6.04 <sup>b</sup>	6.07	6.00	0.047	<0.01	0.05	0.05
Total VFA, mM	29.1 <sup>b</sup>	34.0 <sup>a</sup>	34.4	32.5	2.77	0.30	0.60	0.08
Individual VFA <sup>3</sup>								
Acetate (A)	46.1	47.3	47.3	47.7	3.92	0.21	0.33	0.53
Propionate (P)	36.5	36.8	40.6	38.7	2.64	0.01	0.45	0.30
A:P	1.26	1.29	1.19	1.24	0.168	0.06	0.54	0.47
Ammonia-N, mg/dL	8.04	5.36	6.54	3.10	0.403	<0.01	<0.01	0.31

<sup>a-b</sup>Means within a row with different superscripts differ ( $P < 0.05$ ).

<sup>1</sup>HF-LEA = high-forage diet (HF; 60% forage:40% concentrate) without lipid extract algae (LEA); HF+LEA = HF with LEA; LF-LEA = low-forage diet (LF; 40% forage:60% concentrate) without LEA; and LF+LEA = LF with LEA.

<sup>2</sup>FC = forage-to-concentrate ratio in the diet; LEA = supplementation of LEA; and INT = interaction between FC and LEA.

<sup>3</sup>Expressed as mol/100 mol.

**Key Words:** continuous culture, lactation dairy diet, lipid extract algae

**T384 In vitro ruminal metabolism of a lactation dairy diet supplemented with virgin coconut oil and pine bark extract in continuous cultures.** S. Y. Yang\*<sup>1</sup>, R. W. S. Ningrat<sup>2</sup>, K. Neal<sup>1</sup>, B. R. Min<sup>3</sup>, and J.-S. Eun<sup>1</sup>, <sup>1</sup>Department of Animal, Dairy, and Veterinary Sciences, Utah State University, Logan, UT, <sup>2</sup>Faculty of Animal Sciences, Andalas University, Padang, Indonesia, <sup>3</sup>Department of Agricultural and Environmental Sciences, Tuskegee University, Tuskegee, AL.

The present study investigated effects of virgin coconut oil (VCO; 7.0% C8:0, 5.4% C10:0, 48.9% C12:0, and 20.2% C14:0) and pine bark extract (PBE; 75% condensed tannins), either separately or in combination as supplements to a lactation dairy diet on in vitro ruminal fermentation profiles. The experiment was performed in a completely randomized design with 4 independent runs of continuous cultures. Four dietary treatments included: 1) control (CONT; TMR without supplement), 2) TMR with 3% VCO (VCOT), 3) TMR with 3% PBE (PBET), and 4) TMR with 3% VCO and 3% PBE (VPT). Each culture of 700-mL working volume fermentation content was offered a diet of 20 g DM/d in 2 equal portions at 0800 and 2000 h. The data in this study were analyzed using the Proc Mixed procedure of SAS using a model that included fixed effect of dietary treatments (CONT, VCOT, PBET, and VPT) and a random effect of fermentor within independent run. Culture pH was maintained at least at 6.13 across dietary treatments, and supplementing VCO and/or PBE did not influence culture pH. Total VFA concentration was similar in response to the supplements. Supplementation of VCO decreased acetate proportion, but did not affect propionate proportion, resulting in a tendency to decrease acetate-to-propionate ratio ( $P = 0.10$ ). In contrast, PBE supplementation increased acetate proportion, while it did not influence propionate proportion, leading to a tendency to increase acetate-to-propionate ratio ( $P = 0.10$ ). Cultures offered VCOT and VPT increased butyrate proportion ( $P = 0.01$ ). Supplementing PBE decreased ( $P < 0.01$ ) ammonia-N concentration both in PBET and VPT, whereas VCO supplementation resulted in no effect on ammonia-N concentration. Cultures offered VCO and PBE supplementation, either separately or in combination, showed no response on methane production, although feeding VCOT numerically decreased methane production by 13.5% compared with CONT. The decrease in ammonia-N concentration when PBE-containing diets (PBET and VPT) were offered is likely attributed

to condensed tannins in PBE; however, their concentration would not be enough to lessen ruminal methanogenesis. In addition, the concentration of VCO used in this study may have not been enough to manipulate ruminal fermentation.

**Key Words:** continuous culture, pine bark extract, virgin coconut oil

**T385 Methane production from dairy cows fed regular or brown midrib corn silage.** Fadi Hassanat\*<sup>1</sup>, Rachel Gervais<sup>2</sup>, and Chaouki Benchaar<sup>1</sup>, <sup>1</sup>*Agriculture and Agri-Food Canada, Dairy and Swine Research and Development Centre, Sherbrooke, QC, Canada*, <sup>2</sup>*Département des Sciences Animales, Université Laval, Québec, QC, Canada*.

This study investigated the effects of feeding dairy cows regular (RCS) or brown midrib corn silage (BMCS) based diets on enteric CH<sub>4</sub> emissions. In a crossover design (35-d periods), 16 lactating multiparous Holstein cows (BW = 691 ± 77 kg; DIM = 119 ± 27; milk yield = 43.5 ± 6.4 kg/d) were fed (ad libitum; 5% orts on as-fed basis) a TMR (65:35 forage:concentrate ratio) containing 59% (dry matter basis) RCS or BMCS. Production of CH<sub>4</sub> was measured (3 consecutive days) using respiration chambers. Rumen fermentation characteristics were assessed over 2 consecutive days, and milk performance and nutrient apparent-total-tract digestibility were determined over 6 consecutive days. Effects of dietary corn silage cultivar were determined using the MIXED Procedure of SAS and significance was declared at  $P \leq 0.05$ . Compared with feeding RCS, feeding BMCS increased ( $P \leq 0.01$ ) dry matter intake (+ 1.6 kg/d), milk yield (+ 3.2 kg/d) without affecting milk fat (3.87% ± 0.11) and milk protein (3.50% ± 0.05) contents. Dietary treatment did not affect ruminal pH, protozoa numbers, volatile fatty acid profile and apparent total-tract digestibility of organic matter, including its fiber fraction. Daily enteric methane emission averaged 476 g/d (±21) and was not affected by dietary treatment. Methane energy losses (proportional to gross energy intake) were 7% lower ( $P = 0.05$ ) in cows fed BMCS (5.16%) compared with cows fed RCS (5.52%). Similarly, CH<sub>4</sub> emitted per kg of milk was lower ( $P = 0.02$ ) in cows fed BMCS (12.7 g/kg) than in cows fed RCS (14.3 g/kg). It is concluded that replacing RCS with BMCS in dairy cow diets has the potential to lower CH<sub>4</sub> production per unit of energy consumed or unit of milk produced. Possible influence of feeding RCS or BMCS-based diets on gaseous emissions from manure needs to be evaluated.

**Key Words:** CH<sub>4</sub> production, dairy cow, brown midrib corn silage

**T386 Determination of in vivo and in situ bioavailability of a rumen-protected lysine product, AjiPro-L.** Makoto Miura\*<sup>1</sup>, Atsushi Haruno<sup>1</sup>, Hiroyuki Sato<sup>1</sup>, Yuki Miyazawa<sup>1</sup>, Eri Ikegami<sup>1</sup>, Takeshi Fujieda<sup>1</sup>, and Izuru Shinzato<sup>2</sup>, <sup>1</sup>*Research Institute for Bioscience Products & Fine Chemicals, Ajinomoto Co. Inc., Kawasaki, Kanagawa, Japan*, <sup>2</sup>*Ajinomoto Heartland Inc., Chicago, IL*.

Past studies indicated that the 2nd generation AjiPro-L (A2G, Ajinomoto Co., Inc.) has higher bioavailability than the 1st generation product (A1G) based on in vivo plasma lysine technique (Whitehouse et al., 2014; Tucker et al., 2014). The objective of this study was to compare the other characteristics between A2G and A1G. Three ruminally fistulated lactating cows fed a diet composed of corn silage, grass silage and concentrates were used to determine in situ rumen stability. A1G and A2G were placed in Nylon bags (1 g per bag, 5 × 7 cm, pore size 53 ± 10 µm) and incubated in the rumen for 6, 12 and 24 h, respectively. After washing and drying bags, lysine (Lys) content in the residual products was analyzed. Three dry cows, fistulated both ruminally and

duodenally and fed a corn silage based diet, were used to determine rumen-bypassed and fecal excreted Lys from each product. A1G or A2G was ruminally administrated along with the highly protected L-arginine (HP-Arg; Robinson et al., 2011) as a control marker. Duodenal digesta was collected every 2 or 3 h for 48 – 54 h and was homogenized to extract free Lys and Arg. Changes of Lys and Arg concentrations in digesta were plotted to calculate the area under the curve (AUC). The proportion of AUC of Lys to Arg was defined as the bypass rate of AjiPro-L. To determine the fecal excretion, feces from each cow were collected for 72 h after ingestion of AjiPro-L. Feces were homogenized with hot water to extract free Lys from the products. Amounts of Lys excreted in feces were calculated by analyzing free Lys concentration in the extract. Statistical differences were tested by an ANOVA. In situ ruminal protection of A2G was lower ( $P < 0.01$ ) than A1G (97 vs. 99% at 6 h, 94 vs 97% at 12 h and 84 vs 90% at 24 h, respectively), but still reasonably high. The bypass rate of Lys from A2G (75 ± 11%) was not different ( $P = 0.45$ ) from A1G (82 ± 14%), but fecal excreted Lys from A2G (33 ± 6%) was significantly less ( $P < 0.01$ ) than A1G (51 ± 1%). Our results indicate that higher bioavailability of A2G than A1G was attributed to improved intestinal digestibility, supporting the results obtained by the in vivo plasma Lys response method.

**Key Words:** rumen-protected lysine, dairy cow, bioavailability

**T387 Handling characteristics of AjiPro-L in the practical use.** Makoto Miura\*<sup>1</sup>, Yuki Miyazawa<sup>1</sup>, Eri Ikegami<sup>1</sup>, Mizuki Tanida<sup>1</sup>, Takeshi Fujieda<sup>1</sup>, and Izuru Shinzato<sup>2</sup>, <sup>1</sup>*Research Institute for Bioscience Products & Fine Chemicals, Ajinomoto Co. Inc., Kawasaki, Kanagawa, Japan*, <sup>2</sup>*Ajinomoto Heartland Inc., Chicago, IL*.

When choosing a rumen-protected lysine product (RPL), not only the product efficacy but also handling stability in the practical use should be paid attention to. The 2nd generation AjiPro-L (A2G; Ajinomoto Co., Inc.) has been proven to possess higher bioavailability than the 1st generation product (A1G) based on in vivo plasma lysine technique (Whitehouse et al., 2014; Tucker et al., 2014). The objective of this study was to compare handling characteristics between A1G and A2G. A1G or A2G was mixed in a concentrate mix, and the mixing homogeneity was examined. The mix was loaded into a feed truck, transported for 160 km, unloaded into a feed bin, and was discharged. Samples were collected at the time of loading into the truck and discharging from the feed bin. The samples were sieved to separate the particles of A1G or A2G to calculate the inclusion rate. Results showed the inclusion rate of A2G remained unchanged between before and after transportation ( $P > 0.05$ ) while that of A1G decreased after transportation (4.0 vs. 3.6%,  $P < 0.05$ ), suggesting superior mixing homogeneity of A2G over A1G probably thanks to a smaller particle size of A2G (1 – 2 mm in diameter) than A1G (6 – 7 mm). Next, TMR mixing stability was evaluated with the same procedure as Ji et al. (2012). Dacron bags containing 1 g of RPL were mixed with TMR for 6 min in a Super Data Ranger. Then, bags were incubated in the rumen of 3 fistulated cows for 6, 12, and 24 h. After incubation, bags were washed and dried. Amounts of Lys in the residual product were analyzed for calculation of in situ protection. In situ protection of A2G mixed with TMR was slightly lower than A1G (97 vs. 99% at 6 h, 93 vs. 97% at 12 h and 85 vs. 91% at 24 h, respectively,  $P < 0.01$ ). The protection of both RPLs was not affected by mechanical mixing ( $P > 0.05$ ). Finally, storage stability of A2G was examined under –22 to 104°F by using the in vitro procedure (Miyazawa et al., 2014). In vitro ruminal protection or intestinal dissolution was not deteriorated at least for 6 mo under the ambient temperature tested.

Results demonstrate that A2G possesses sufficient handling stability for a use under practical conditions.

**Key Words:** rumen-protected lysine, handling, stability

**T388 Effects of corn silage hybrids on metabolic parameters and lactational performance of transition dairy cows.** A. W.

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Developing solutions to lessen metabolic stress experienced by transition dairy cows is very critical to improve lactational performance. The objective of this study was to determine metabolic parameters [concentrations of nonesterified fatty acids (NEFA) and  $\beta$ -hydroxybutyric acid (BHBA)] and lactational performance of dairy cows fed brown midrib corn silage (BMRCs)-based diets during the transition period when compared with conventional corn silage (CCS)-based diets. At 4 wk before parturition, 40 dry multiparous Holstein cows were randomly assigned treatments. The treatment groups consisted of 2 close-up transition diets (CCS-based and BMRCs-based diets) offered to 2 groups of 20 cows each. After calving, 10 cows from each prepartum group were individually fed either a CCS-based lactation diet or a BMRCs-based lactation diet. Four dietary treatments tested postpartum included: 1) CC = CCS-based close-up diet + CCS-based lactation diet; 2) CB = CCS-based close-up diet + BMRCs-based lactation diet; 3) BB = BMRCs-based close-up diet + BMRCs-based lactation diet; and 4) BC = BMRCs-based close-up diet + CCS-based lactation diet. Statistical analysis during the pre- and postpartum period parameters were performed by a split-plot-in-time design with repeated measures procedure using the Proc Mixed procedure of SAS with a model that included effects of treatment and week and their interaction and a random effect of cow within treatment. During the dry period, there was a tendency ( $P = 0.09$ ) for a diet  $\times$  test week interaction for DMI; initially, CCS-based diet had greater DMI, but as parturition neared, BMRCs-based diet increased DMI, resulting in greater DMI than CCS-based diet at parturition. Postpartum concentrations of BHBA and NEFA were lower for BB and BC diets, with CC and CB diets having the greatest concentrations of both BHBA and NEFA ( $P < 0.05$ ). In contrast, cows fed BB and BC diets increased milk yield compared with those fed CC and CB diets. There was a tendency for BB to have greater milk yield than CB on wk 1 (32.7 vs. 27.9 kg/d,  $P = 0.09$ ) and greater milk yield on wk 4 (45.1 vs. 38.5 kg/d). Overall data in the present study indicate that cows fed BMRCs-based diet in the dry period but CCS-based diet in lactation had positive carry-over effects on metabolic parameters and lactational performance possibly due to a consistent pattern of feed intake during dry period.

**Key Words:** brown-midrib corn silage, dairy cow, transition period

**T389 Effects of thermal processed clay in nonmedicated**

**rations in Japanese dairy farms.** Fang Chi\*<sup>1</sup>, Fumiaki Atarashi<sup>2</sup>, Kenji Wada<sup>2</sup>, Hiroshi Endo<sup>2</sup>, San Ching<sup>1</sup>, and LeAnn Johnston<sup>1</sup>, <sup>1</sup>Amlan International, Chicago, IL, <sup>2</sup>Okitama Food Animal Clinic, Federation of Agricultural Mutual Aid Association, Yamagata Prefecture, Japan.

Calibrin-Z (CZ), a thermally processed calcium montmorillonite, has been shown to enhance dairy productivity due to its barrier function in the digestive tract. Two classic dairy farms from different locations in Yamagata Prefecture, Japan, were used in this on-farm feeding trial. The objective was to investigate the effects of CZ feeding on lactating cow performance under a common dairy feeding regimen in Japan. Concentrate feed was purchased from local feed companies, corn and

grass silage was produced on-farm, and CZ was added to the Total Mixed Ration (TMR) with 20 g/cow/d for 3 mo. The TMR was formulated to meet Japanese nutrients requirements standard, and collected for mycotoxin analyses. Milk yield and quality and fecal firmness were recorded daily. Blood was drawn before and 30 d after CZ was fed, and albumin, BUN, GGT, hematocrit, and white blood cell counts were determined. Disease incidence and general health were recorded. Analysis of variance was used to determine the effects of CZ. Mycotoxin concentrations, such as aflatoxin, DON, fumonisin, and zearalenone were detectable in the TMR, but were below Japanese regulations. Manure firmness was numerically increased on one farm, but no changes were found on the other farm over the 3 mo feeding period. There were no differences in general blood chemistry; however, CD4+ and CD14+ were higher ( $P < 0.05$ ) after CZ was fed for 30 d. In addition, CD3+ and CD335+ were numerically higher ( $P > 0.05$ ) when CZ was included in the ration. Interestingly, a 47% (Farm A) or 40% (Farm B) reduction of disease incidences was observed as compared with the same period of the prior year. Among them, mastitis decreased 60% on Farm A, and enteritis decreased 100% on Farm B. Milk production was increased ( $P < 0.05$ ) on both farms (Farm A, 25.0 vs. 26.4 kg; Farm B, 27.0 vs. 28.4 kg/cow/d) after CZ was fed; while milk fat, protein, lactose, bacteria count, urea nitrogen, and somatic cell count remained unchanged. In conclusion, addition of Calibrin-Z to Japanese non-medicated rations may enhance general health in lactating cows; consequently, increased milk production and a reduction of disease occurrence were observed on the studied farms.

**Key Words:** dairy milk production, white blood cells, calcium montmorillonite

**T390 Associations of behavior and production in lactating**

**dairy cows.** Carleigh Johnston and Trevor J. DeVries\*, *Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada.*

Identification of the associations of cow behavior with productivity is important for supporting recommendation of strategies that optimize milk quality, efficiency, and yield. Thus, the objective of this study was to collate data from studies of the behavior of lactating dairy cows and identify associations between measures of behavior of dairy cattle and milk production and efficiency. A database containing behavior and production data for 132 dairy cow-week observations (mean of 7 d of consecutive data per cow) was assembled from 5 studies. Cows averaged (mean  $\pm$  SD) 1.8  $\pm$  0.9 lactations, 108.4  $\pm$  42.7 DIM, and 654.6  $\pm$  71.4 kg BW during each observation week. Production data included: DMI (27.0  $\pm$  3.1 kg/d), milk yield (43.0  $\pm$  7.0 kg/d), milk fat content (3.60  $\pm$  0.49%), milk fat yield (1.53  $\pm$  0.30 kg/d), milk protein content (3.05  $\pm$  0.25%), milk protein yield (1.29  $\pm$  0.17 kg/d), and production efficiency (1.60  $\pm$  0.22 kg milk/kg DMI). Behavioral data included: feeding time (230.4  $\pm$  35.5 min/d), feeding rate (0.13  $\pm$  0.03 kg/min), meal frequency (9.0  $\pm$  2.0 meals/d), meal size (3.2  $\pm$  0.9 kg/meal), daily mealtime (279.6  $\pm$  51.7 min/d), rumination time (516.0  $\pm$  90.7 min/d), and lying time (621.5  $\pm$  142.8 min/d). Data were analyzed in multi-variable mixed-effect regression models to identify which behavioral variables, when accounting for other cow-level factors (DIM, parity, BW), were associated with measures of production. DMI was associated with feeding time (+0.02 kg/min;  $P = 0.002$ ) and tended to be associated with rumination time (+0.003 kg/min;  $P = 0.1$ ) and meal frequency (+0.2 kg/meal;  $P = 0.06$ ). Similarly, milk yield was associated with feeding time (+0.03 kg/min;  $P = 0.005$ ) and rumination time (+0.02 kg/min;  $P < 0.001$ ), and tended to be associated with meal frequency (+0.3 kg/meal;  $P = 0.1$ ). Milk fat yield was associated with meal frequency

(+0.02%/meal;  $P = 0.05$ ). Efficiency of production was associated with rumination time (+0.0006/min;  $P < 0.001$ ) and tended to be associated with meal time per day (+0.0005/min;  $P = 0.09$ ). Overall, our results indicate that nutrition, management, and housing factors that improve time spent feeding, in more frequent meals, and time spent ruminating may have a positive impact on milk yield and component production, as well as production efficiency.

**Key Words:** behavior, production, rumination

**T391 Growth performance and sorting behavior of heifers offered diets with forage dilution.** Wayne Coblenz<sup>\*1</sup>, Nancy Esser<sup>2</sup>, Patrick Hoffman<sup>3</sup>, and Matt Akins<sup>3</sup>, <sup>1</sup>US Dairy Forage Research Center, Marshfield, WI, <sup>2</sup>University of Wisconsin, Marshfield, WI, <sup>3</sup>University of Wisconsin, Madison, WI.

Dairy heifers consuming high-quality forage diets are susceptible to excessive weight gains and over-conditioning, which often has been controlled by dilution with straw that is sortable by dairy heifers. Our objectives were: i) to compare the growth performance of dairy heifers offered a high-quality forage diet (CONTROL) with diets containing 1 of 3 diluting agents that included eastern gamagrass haylage (EGG), chopped wheat straw (STRAW), or chopped corn fodder (FODDER); and ii) evaluate sorting behaviors of heifers offered these forage diets. Holstein heifers ( $n = 128$ ) were stratified (32 heifers/block) on the basis of initial BW (heavy,  $560 \pm 27.7$  kg; medium-heavy,  $481 \pm 17.7$  kg; medium-light,  $441 \pm 22.0$  kg; and light,  $399 \pm 14.4$  kg), and then assigned to 1 of 16 identical research pens (4 pens/block; 8 heifers/pen) in a randomized complete block design with the 4 research diets as treatments. Diets were offered in a 118-d feeding trial with heifers crowded to 133% of capacity at the feed bunk. Compared with CONTROL, inclusion of low-energy forages was effective in reducing DMI (11.06 vs. 10.04 kg/d;  $P < 0.01$ ) and energy intake (7.39 vs. 5.95 kg TDN/d;  $P < 0.01$ ). Concentrations of physically effective fiber (pef) particles did not change during the 24-h period following feeding for either the CONTROL ( $P \geq 0.56$ ) or EGG ( $P \geq 0.75$ ) diets; however, this response for pef particles masked the competing (and cancelling) responses for individual large and medium particles comprising pef, which heifers sorted with discrimination and preference, respectively. Sorting against pef particles was detected for STRAW as a linear ( $P < 0.01$ ) function of time from feeding, and much more severely for the FODDER diet, which exhibited linear, quadratic and cubic effects ( $P < 0.01$ ). Sorting of forage particles by heifers could not be related directly to heifer performance. Compared with CONTROL, ADG was reduced by dilution in all cases (1.16 vs. 0.91 kg/d;  $P < 0.01$ ), but ADG for STRAW was approximately 0.2 kg/d less than EGG (0.98 kg/d) or FODDER (0.97 kg/d), despite exhibiting sorting characteristics intermediate between those diets.

**Key Words:** heifer, sorting, dilution

**T392 Response of lactating cows to a blend of essential oils and pepper extract.** Rayana B. Silva<sup>1</sup>, Renata A. N. Pereira<sup>3,2</sup>, Rafael C. Araújo<sup>4</sup>, and Marcos N. Pereira<sup>\*1,2</sup>, <sup>1</sup>Universidade Federal de Lavras, Lavras, MG, Brazil, <sup>2</sup>Better Nature Research Center, Ijaci, MG, Brazil, <sup>3</sup>Empresa de Pesquisa Agropecuária de Minas Gerais, Lavras, MG, Brazil, <sup>4</sup>GRASP Ind. e Com. LTDA, Curitiba, PR, Brazil.

Secondary plant compounds can modify rumen fermentation, cutaneous blood flow, sweat secretion, diet digestibility, and feeding behavior of dairy cows. Twenty-eight, individually fed Holsteins ( $181 \pm 102$  DIM) received a standard diet for 2 wk and 2 dietary treatments for 8 wk, in a

covariate adjusted randomized block design. Treatments were: Control (CTL) or a microencapsulated blend of essential oils and pepper extract (BEO: Carvacrol, cinamaldehyde, eugenol, and capsaicin. Activo Premium. 150 mg/kg DM). BEO decreased DMI (20.1 vs. 19.5 kg/d;  $P = 0.05$ ) and increased milk yield (30.1 vs. 30.8 kg;  $P = 0.04$ ), improving the milk to feed ratio (1.53 vs. 1.63;  $P < 0.01$ ). Milk fat content tended to increase with BEO ( $P = 0.10$ ), but protein, lactose, total solids, MUN, and SCC did not differ ( $P > 0.21$ ). BCS and BW were similar ( $P > 0.39$ ). BEO reduced ruminal acetate (57.8 vs. 51.4% of VFA;  $P = 0.01$ ) and increased propionate (26.1 vs. 31.3% of VFA;  $P = 0.02$ ), decreasing the A/P ratio (2.3 vs. 1.7;  $P = 0.03$ ). Total-tract digestibility of DM, OM, and NDF were similar ( $P > 0.20$ ), but there were trends for increased non-NDF OM digestibility ( $P = 0.07$ ) and digestible OM intake ( $P = 0.06$ ) on BEO. Cows on BEO ingested a greater proportion of the daily feed intake from 6AM to 12PM ( $P = 0.02$ ) and tended to have longer duration of the first meal ( $P = 0.08$ ), whereas CTL cows tended to ingest more from 7PM to 6AM ( $P = 0.06$ ). BEO induced selective sorting in favor of small feed particles ( $P < 0.01$ ) and refusal of large particles ( $P < 0.03$ ) in the morning. Ingestion and rumination times were similar ( $P > 0.19$ ). Viscosity of manure was reduced by BEO ( $P = 0.04$ ). Treatments had no effect on PUN along the day, plasma glucose content, and the urinary secretion of allantoin ( $P > 0.42$ ). BEO increased the concentration of  $O_2$  in jugular blood ( $P < 0.01$ ), but had no effect on pH,  $pCO_2$ ,  $HCO_3^-$ , total  $CO_2$ , and base excess ( $P > 0.46$ ). Sweating rate at d 14 was increased by BEO ( $P = 0.05$ ). No difference was detected in rectal and skin temperatures and in respiratory frequency ( $P > 0.21$ ). The supplementation of dairy cows with BEO affected feeding behavior, reduced the A/P ratio in rumen fluid, and improved feed efficiency.

**Key Words:** capsaicin, essential oil, feeding behavior

**T393 Supplementation of herbage-based diets with corn meal or liquid molasses changes the milk fatty acids profile in grazing dairy cows.** S. F. Reis<sup>1</sup>, A. F. Brito<sup>\*1</sup>, P. Y. Chouinard<sup>3</sup>, K. J. Soder<sup>2</sup>, and S. Ross<sup>1</sup>, <sup>1</sup>University of New Hampshire, Durham, NH, <sup>2</sup>USDA-Agricultural Research Service, University Park, PA, <sup>3</sup>Université Laval, Québec City, Québec, Canada.

Previous studies showed that feeding carbohydrate sources with different NSC profiles (e.g., starch vs. sucrose) and rates of ruminal degradation altered the milk fatty acids (FA) profile in dairy cows. This study evaluated the impact of corn meal (CM) or liquid molasses (MOL) on the milk FA profile of organically certified Jersey cows fed herbage-based diets. Ten multiparous (107 DIM) and 10 primiparous (131 DIM) lactating cows were blocked by parity, DIM, and milk yield and randomly assigned to 1 of 2 dietary treatments: CM or MOL. Diets were formulated to contain (DM basis): 74% herbage, 12% grass-legume mix hay, 12% CM or MOL, and 2% minerals and vitamins premix. The study lasted 110 d (June to September). Cows had access to pasture with mixed botanical composition (41.2% grass and 20% legume) for approximately 16 h daily in a strip grazing management system and were milked twice daily. Milk samples were collected monthly for 4 consecutive milkings, pooled by cow/period, and analyzed for milk FA using GLC. Herbage averaged (DM basis) 18.5% CP and 56.3% NDF, and 20% linoleic acid and 41.5%  $\alpha$ -linolenic acid (% total FA). Milk FA was expressed as % total milk FA. Milk proportions of most saturated and unsaturated FA were affected by month, which are explained by variation in herbage botanical and FA composition, and milk yield. The milk proportion of total saturated FA was highest in cows fed MOL (71.8 vs. 71%). Milk proportions of  $\alpha$ -linolenic acid (0.88 vs. 0.77%), *trans*-10,*cis*-12 CLA (0.090 vs. 0.078%), and PUFA (4.57 vs. 4.43%) were highest in cows fed MOL. Conversely, milk linoleic acid (1.12 vs. 1.72%), *trans*-10:18:1

(0.16 vs. 0.19%), *trans*-11 18:1 (2.62 vs. 2.76%), *cis*-9,*trans*-11 CLA (0.83 vs. 0.87%), and MUFA (22.8 vs. 23.7%) were lowest with MOL. The proportion of n-3 FA was highest (1.19 vs. 1.03%), whereas that of n-6 FA (1.30 vs. 1.35%) was lowest in cows fed MOL. Supplementing herbage-based diets with energy sources containing different NSC profiles resulted in marked changes in milk FA composition.

**Key Words:** milk fatty acids, molasses, herbage

**T394 Health, milk, milk components, milk quality and reproduction evaluated in Holstein cows fed OmniGen-AF from dry-off through 120 days in milk.** Amanda E. Holland<sup>\*1</sup>, Frank E. Rivera<sup>1</sup>, James D. Chapman<sup>1</sup>, and Lane O. Ely<sup>2</sup>, <sup>1</sup>Phibro Animal Health Corporation, Quincy, IL, <sup>2</sup>University of Georgia, Athens, GA.

Three hundred sixty-one multiparous late lactation cows were selected using DC305 and randomly assigned to 1 or 2 diet additives before dry-off; basal diet + placebo (PL, n = 181) or basal diet + OmniGen-AF (OG, n = 180). Diet groups were balanced by parity, DIM, calving date, 305ME, milk and health history. Both additives were fed at 56g/h/d, added directly to the dry and lactation TMR's. Cows were fed the assigned diet additives from dry-off through 120 d in milk (DIM) and health, milk, milk quality and reproduction evaluated. Data were analyzed by PROC GLM with significance tested at  $P < 0.05$  and only cows completing 120 DIM were included. Average parity and days dry for cows fed the PL and OG were 3.7, 59.3 d and 3.6, 58.3 d, respectively. Cows fed OG had fewer retained placenta (-6.6%,  $P = 0.023$ ) and metritis (-5%,  $P = 0.044$ ) cases than PL fed cows. Mastitis cases were similar, however cows fed OG tended to have fewer repeat cases ( $P = 0.107$ ). No differences were detected in the other recorded health metrics. Individual milk and milk components were recorded and were used to calculate week-4 milk (W4), energy and fat corrected milks (ECM, FCM). No differences were detected between PL and OG fed cows for test-day (TD) milk 1-4 (44.6kg vs. 45.5kg), W4 (43.5kg vs. 44.7kg) or peak milk (49.9kg vs. 50.4kg). OG fed cows produced more ECM (+1.5kg,  $P = 0.008$ ) and FCM (+1.7kg,  $P = 0.007$ ) through TD 1-4 than PL cows. OG cows had higher % milk fat (MF) at TD 1 (+0.3) and kg MF at TD 1 (+0.13), 2 (+0.09) and 3 (+0.09) and 0.05 kg more milk protein (MP) at TD 2 than PL cows. Compared with PL, the OG cows made milk with more kg MF (1.5 vs. 1.6,  $P = 0.002$ ), kg MP (1.3 vs. 1.5,  $P = 0.053$ ) and % MF (3.45 vs. 3.56,  $P = 0.032$ ). TD 1-4 SCC's were not different. All reproductive measures were not different, however OG cows tended to have fewer days to first breeding ( $P = 0.12$ ) and greater days carried calf ( $P = 0.088$ ) than PL cows. In conclusion, feeding OG to multiparous cows from dry-off through 120 DIM resulted in differences in peripartum disease incidence, milk and milk components.

**Key Words:** OmniGen-AF, health, components

**T395 Supplementing lactating cow diets with long chain fats has minimal effects on total-tract NDF digestibility: A quantitative review.** Kristina A. Weld<sup>\*</sup> and Louis E. Armentano, University of Wisconsin-Madison, Madison, WI.

Supplementing fat to lactating cows may reduce total-tract NDF digestibility (ttNDFd). The objective was to analyze the effects of different types of fat across studies. The numerical difference between ttNDFd and dry matter intake (DMI) for the supplemented minus control diet were calculated within study and these differences were analyzed in SAS 9.4 using Proc Mixed (Table 1). Studies were weighted based on the inverse of the standard error of ttNDFd squared. The model contained the

fixed effect of fat type and study as a random effect. Fatty acid content of treatment diets never exceeded 10% of DM (mean = 6.2, SD = 1.4). Supplementation of fats containing 12 and 14 carbon chain fatty acids (C12, C14) decreased both DMI and ttNDFd. Oil decreased ttNDFd by 2.1 percentage units, but the DMI decrease was not significant. No other type of fat consistently decreased ttNDFd, though calcium salts (n = 13 of palm oil, n = 6 of unsaturated C18) increased ttNDFd by 2.2 percentage units and decreased DMI by 1.45 kg/day. A regression analysis across all types of fat, using Proc Glim in SAS 9.4, of the difference in ttNDFd versus the difference in DMI within study shows a positive relationship. This suggests that the increase in ttNDFd for calcium salts is not explained through the decrease in DMI. Overall, these results indicate that the addition of a fat supplement, in which the fatty acids are C16 or greater in length, has minimal effects on ttNDFd. Feeding calcium salts in place of free oils would likely increase ttNDFd, but decrease DMI.

**Table 1 (Abstr. T395).** Effects of fat supplemented diets compared with control diets by fat type

Fat type	No. of studies	ΔttNDFd		ΔDMI,		ΔFA		content	
		percentage units	SE	P-value	kg/d	SE	P-value	%DM	SE
C12/C14	6	-11.32	1.67	0.0001	-4.04	0.66	0.0001	3.8	0.72
Oil	11	-2.13	0.97	0.04	-0.59	0.39	0.13	2.9	0.41
Animal vegetable blend	8	-1.15	1.36	0.41	-0.64	0.54	0.18	3.8	0.58
High C16	5	-0.31	1.43	0.83	-0.31	0.57	0.34	2.9	0.72
Tallow	22	-0.27	1.17	0.82	-0.88	0.47	0.07	2.5	0.62
Hydrogenated fat	14	0.42	1.36	0.76	0.42	0.54	0.37	3.4	0.48
Calcium salts	19	2.2	0.83	0.01	-1.48	0.33	0.0001	3.2	0.37

**Key Words:** NDF digestibility, fat, DMI

**T396 Sodium and potassium carbonates added to continuous cultures of ruminal microorganisms had similar effects on reducing biohydrogenation intermediates linked to milk fat depression.** Kaylin Young<sup>1</sup>, Elliot Block<sup>2</sup>, Joseph Harrison<sup>3</sup>, and Thomas Jenkins<sup>\*1</sup>, <sup>1</sup>Clemson University, Clemson, SC, <sup>2</sup>Arm and Hammer Animal Nutrition, Princeton, NJ, <sup>3</sup>Washington State University, Puyallup, WA.

In previous studies, the addition of potassium carbonate to continuous cultures of ruminal microorganisms decreased the production of *trans*-10 18:1 and *trans*-10,*cis*-12 conjugated linoleic acid (CLA), consistent with in vivo reports of higher milk fat percentages in lactating cows fed supplemental potassium carbonate. This experiment compared sodium carbonate to potassium carbonate to determine if they had similar effects on biohydrogenation intermediates. A single basal diet was fed (60 g/d) to 4 dual-flow continuous fermenters for four 10-d periods divided into 7 d for adaptation and 3 d for collection of samples. The diet consisted of alfalfa pellets and concentrate mix (47/53, DM basis), and contained 3.3% (DM basis) soybean oil. At each feeding (0800 and 1600 h), 16 mL were injected into each fermenter containing either only distilled water (CON), 1.4 g carbonate as  $K_2CO_3$ , 1.4 g carbonate as  $Na_2CO_3$ , or 1.4 g carbonate as a 50:50 mixture of  $K_2CO_3$  and  $Na_2CO_3$  (MIX). The experiment was analyzed as a randomized block design with differences among treatments determined by 3 orthogonal contrasts: (1) control vs. carbonate; (2) MIX vs. both single carbonates; and (3)  $K_2CO_3$  vs.  $Na_2CO_3$ . Carbonate sources did not differ ( $P > 0.05$ ) so MIX,  $K_2CO_3$ , and  $Na_2CO_3$  means were averaged to give a single combined carbonate value. CON and carbonate values differed ( $P < 0.05$ ) respectively for

culture pH (6.05 and 6.36), acetate (58.9 vs. 65.5 mol/100 mol), propionate (27.2 vs. 22.3 mol/100 mol), acetate/propionate (2.12 and 3.19), and production of *trans*-10 C18:1 (504.1 and 251.7 mg/d), *trans*-11 C18:1 (253.6 and 355.5 mg/d), and *cis*-9, *trans*-11 CLA (8.4 and 11.7 mg/d). CON and carbonate also differed ( $P < 0.10$ ) in production of *trans*-10, *cis*-12 CLA (19.7 and 12.8 mg/d). The results showed that sodium carbonate was equally effective as potassium carbonate in reducing the production of biohydrogenation intermediates that have been linked to the cause of milk fat depression.

**Key Words:** lipid, biohydrogenation, carbonates

**T397 Effects of starch content and fermentability, and culture pH on biohydrogenation of unsaturated fatty acids and NDF digestibility in batch culture.** Yan Sun\*, Michael S. Allen, and Adam L. Lock, *Michigan State University, East Lansing, MI.*

Effects of starch content (SC) and fermentability (SF), and culture pH on biohydrogenation (BH) of unsaturated fatty acids (FA) and NDF digestibility (NDFD) in batch culture were evaluated. Cultures (4 replicates/treatment/time point) were incubated at pH 5.8 or 6.2 for 0, 12, and 24 h and included alfalfa hay (55 or 70% of DM), and either dry ground corn (DC) or high moisture corn (HMC) as starch sources (30 or 45% of DM) for a SC of 22 or 33% DM. The alfalfa hay was treated with corn oil (2% of DM) to increase the total unsaturated FA content of cultures. Effects of SC, SF, culture pH, and their interactions were determined. BH extent for *cis*-9,*cis*-12 18:2 and NDFD increased over time across all treatments (both  $P < 0.001$ ). For main effects of treatments, 33% SC compared with 22% SC and pH 5.8 compared with pH 6.2 reduced BH extent of *cis*-9,*cis*-12 18:2 (35 vs. 39% and 23 vs. 51%, respectively; both  $P < 0.001$ ) and NDFD (13 vs. 16% and 8 vs. 21%, respectively; both  $P < 0.001$ ). Compared with DC, HMC decreased NDFD (13 vs. 16%;  $P < 0.001$ ). HMC increased BH extent of *cis*-9,*cis*-12 18:2 at pH 5.8, and decreased it at pH 6.2 compared with DC (interaction  $P < 0.001$ ) with no overall effect of SF ( $P = 0.90$ ). Contents of *trans*-10,*cis*-12 18:2 (CLA) and *trans*-10 18:1 increased over time for all treatments (both  $P < 0.001$ ). Overall, 33% SC compared with 22% SC and pH 5.8 compared with pH 6.2 increased content of CLA by 20 and 67% respectively (both  $P < 0.001$ ). Although SF did not affect CLA content overall ( $P = 0.13$ ), it interacted with time (interaction  $P < 0.001$ ); HMC increased CLA at 12 h, and decreased it at 24 h compared with DC. The 33% SC compared with 22% SC, HMC compared with DC, and pH 5.8 compared with pH 6.2, increased content of *trans*-10 18:1 by 17, 28, and 43% respectively (all  $P < 0.001$ ). In conclusion, higher SC and lower culture pH decreased BH of *cis*-9,*cis*-12 18:2 and NDFD, and increased formation of CLA and *trans*-10 18:1. Compared with DC, HMC increased formation of *trans*-10 18:1, decreased NDFD, and interacted with culture pH and time for BH of *cis*-9, *cis*-12 18:2 and the formation of CLA.

**Key Words:** biohydrogenation, starch, conjugated linoleic acid

**T398 The effect of replacing corn silage with sugarcane on milk yield and intake of lactating dairy cows: An analysis using CNCPS v6.5.** Edgar A. Collao-Saenz\*<sup>1</sup>, Andreas Foskolos<sup>2</sup>, Ryan J. Higgs<sup>2</sup>, Vera L. Banys<sup>1</sup>, Marcos N. Pereira<sup>3</sup>, and Michael E. Van Amburgh<sup>2</sup>, <sup>1</sup>Universidade Federal de Goiás, Jatai, GO, Brazil, <sup>2</sup>Cornell University, Ithaca, NY, <sup>3</sup>Universidade Federal de Lavras, Lavras, MG, Brazil.

Sugarcane is an option for feeding dairy cattle in the tropics. Reductions in DMI and milk yield (MY) are frequently reported when sugarcane replaces corn silage in dairy rations, due to the low NDF digestibility.

This meta-analysis was conducted to evaluate DMI and MY response of dairy cows to the replacement of corn silage with sugarcane. A database was compiled from 13 Brazilian publications involving 50 treatments from the last 12 years. The nutrient composition of the diets was calculated using CNCPS v. 6.5 feed library. Treatments were stratified in 3 categories: no replacement (SC0), 25–75% replacement (SCA) and 100% replacement (SCB) and the outcomes and predictions were analyzed with a mixed effects model (random: study; fixed: replacement level). Then, a mixed model regression analysis was used to investigate the potential relationships among reported nutrient composition of the diets (CP, NPN, NDF, NFC, Lignin and CNCPS estimated fractions) and MY and DMI as dependent variables. The best models were selected based on the highest BLUP ( $R^2_{BLUP}$ ) correlation coefficient and the lowest root mean square error (RMSE). Total replacement reduced MY (21.9 vs. 18.6 kg/d) and DMI (19.1 vs. 16 kg/d). DMI was the single most important factor explaining MY ( $R^2_{BLUP} = 0.973$ ; RMSE = 0.865). When CP and RUP (%CP) were included into the DMI model, accuracy and precision were increased ( $R^2_{BLUP} = 0.979$ ; RMSE = 0.75). Total NDF of the diet did not affect MY, but sugarcane NDF content affected it negatively. Sugar (CHOA4;  $R^2_{BLUP} = 0.948$ ; RMSE = 0.66) and sugarcane peNDF ( $R^2_{BLUP} = 0.941$ ; RMSE = 0.68) were the 2 primary factors negatively related to DMI. The replacement of corn silage with sugarcane reduced MY and DMI of dairy cows, diet sugarcane NDF content was the nutrient most related to the response.

**Key Words:** DMI, corn silage, NDF

**T399 Effect of dietary energy source and protein supply on dairy cow performance.** Helio Rezende Lima Neto<sup>1</sup>, Helene Lapierre<sup>2</sup>, and Lorraine Doepel\*<sup>1</sup>, <sup>1</sup>University of Calgary, Calgary, Alberta, Canada, <sup>2</sup>Agriculture and Agri-Food Canada, Sherbrooke, Quebec, Canada.

This study examined the interaction of dietary energy source and metabolizable protein (MP) supply on cow performance. Eight Holstein cows were used in a double 4 × 4 Latin square design with 21-d periods, with dietary energy source and MP supply tested in a factorial arrangement: high fiber, high protein (HFHP); high fiber, low protein (HFLP); high starch, high protein (HSHP); and high starch, low protein (HSLP). Diets were formulated to supply 100% of the NE<sub>L</sub> requirement, and 100% (HP) or 70% (LP) of MP requirement. There were no interactions between energy source and MP supply on the parameters measured. Intakes of NE<sub>L</sub> (Mcal/d) and MP (g/d) were: HFHP: 31.5, 2148; HFLP: 29.5, 1492; HSHP: 37.8, 2699; HSLP: 36.2, 2007. Milk yield (34.4 vs. 29.9 kg/d) and DMI (22.6 vs. 18.4 kg/d) were higher in cows fed HS compared with those fed HF, whereas DMI tended to be lower in cows fed LP vs. HP (19.9 vs. 21.1). Milk protein (3.04 vs. 2.85%) and lactose (4.62 vs. 4.56%) concentrations were higher in cows fed HS than those fed HF, but milk fat concentration was higher with HF compared with HS (4.22 vs. 3.49%). Milk protein (1.04 vs. 0.85 kg/d) and lactose (1.59 vs. 1.36 kg/d) yields were higher in cows fed HS than those fed HF. Milk protein yield tended to be higher with HP than with LP (962 vs. 925 g/d), and milk urea-N was higher with HP than LP (14.5 vs. 7.9 mg/dL). Dietary energy affected milk yield and composition more than protein supply did. Although diets were formulated to be iso-energetic, the large decrease in DMI in cows fed HF mainly drove the effect of HS vs. HF. The smaller effects of protein supply were not affected by energy source.

*Contd.*

**Table 1 (Abstr. T399).**

Parameter	Treatments				SEM	P-value	
	HFHP	HFLP	HSHP	HSLP		HF vs. HS	HP vs. LP
DMI, kg/d	19.0	17.8	23.1	22.0	0.85	<0.01	0.06
Yield, kg/d							
Milk	30.4	29.4	34.9	33.9	1.28	<0.01	0.20
Protein	0.87	0.83	1.06	1.02	0.04	<0.01	0.09
Fat	1.31	1.22	1.19	1.16	0.07	0.16	0.34
Lactose	1.38	1.34	1.62	1.57	0.06	<0.01	0.24
Milk composition, %							
Protein	2.87	2.85	3.04	3.04	0.06	<0.01	0.84
Fat	4.22	4.21	3.52	3.46	0.16	<0.01	0.80
Lactose	4.56	4.55	4.62	4.63	0.04	0.04	0.91
MUN, mg/dL	15.7	10.4	13.2	5.4	0.80	<0.01	<0.01

**Key Words:** energy source, metabolizable protein, milk yield

**T400 Effect of dietary energy source and protein supply on mammary amino acid metabolism of dairy cows.** Helio Rezende Lima Neto<sup>1</sup>, Helene Lapierre<sup>2</sup>, and Lorraine Doepel\*<sup>1</sup>, <sup>1</sup>University of Calgary, Calgary, AB, Canada, <sup>2</sup>Agriculture and Agri-Food Canada, Sherbrooke, QC, Canada.

The objective of this study was to determine the interaction of dietary energy source and metabolizable protein (MP) supply on AA arterial concentration and mammary arterial-venous difference (AVdiff). Eight Holstein cows were used in a double 4 × 4 Latin square design with 21-d periods. Energy source and MP supply were tested in a factorial arrangement with the following dietary treatments: high fiber, high protein (HFHP); high fiber, low protein (HFLP); high starch, high protein (HSHP); and high starch, low protein (HSLP). Diets were formulated to supply 100% of the NE<sub>L</sub> requirement, and 100% (HP) or 70% (LP) of MP requirement. Arterial concentrations of Ile, Leu, Phe, and the branched-chain AA (BCAA) were higher with HF compared with HS, regardless of MP supply, whereas concentrations of His, Leu, and BCAA were higher, or tended to be higher, with HP compared with LP. Group 1 AA-N (His, Met, Trp, Phe+Tyr) concentration tended to be higher with HP than LP but Group 2 AA-N (Ile, Leu, Ly, Val) concentration was not different between HP and LP. Metabolizable protein supply did not affect AVdiff except for Ile, Leu and BCAA, in which it was greater for HP than LP ( $P < 0.10$ ). Energy source did not affect AVdiff. The concentration of total essential AA-N (TEAAN) was higher with HF and HP relative to HS and LP, but AVdiff was not affected by treatment. Protein supply had a greater influence on AVdiff than did energy source, whereas energy source and MP supply influenced essential AA concentrations. Dietary energy source and MP affected several production variables; those data are presented in a companion abstract.

Contd.

**Table 1 (Abstr. T400).**

Arterial concentration, $\mu\text{M}$	Treatments				SEM	P-value	
	HFHP	HFLP	HSHP	HSLP		HF vs. HS	HP vs. LP
G1AAN	327.9	287.4	306.9	291.9	19.24	0.56	0.07
G2AAN	540.8	481.5	448.4	432.4	24.89	<0.01	0.12
BCAA	438.5	366.0	349.6	329.0	19.78	<0.01	0.02
TEAAN	881.3	799.5	792.7	748.3	40.19	0.06	0.09
Ile	91.4	92.0	75.8	75.9	4.95	<0.01	0.94
Lys	51.2	57.7	49.3	51.9	3.06	0.16	0.11
Met	22.9	21.9	22.8	21.1	1.07	0.62	0.13
Phe	59.2	51.6	47.2	49.7	3.08	0.02	0.36

**Key Words:** metabolizable protein, energy source, amino acid

**T401 Effect of supplemental level of Optigen on the milk performance and plasma biochemical indices of dairy goat.** Wang Hui<sup>1</sup>, Xue Neil<sup>2</sup>, and Luo Jun\*<sup>1</sup>, <sup>1</sup>Alltech-NWAFU Animal Science Research Alliance, Northwest A&F University, Yangling, Shaanxi, China, <sup>2</sup>Alltech China, Chaoyang District, Beijing, China.

The experiment was conducted to investigate the effects of different levels of Optigen, a new slow-release non-protein product, on milk performance and plasma biochemical indices of Xinong Saanen dairy goat, thus to select the optimum supplemental level in the diet. Forty-eight healthy lactating Xinong Saanen dairy goats with average BW of  $60 \pm 8.98$  kg were assigned to 4 groups with 12 goats in each group. Goats in the 4 groups were fed experimental diets containing 0, 0.25, 0.5, or 0.75% Optigen, respectively. There was a pretrial period of 1 wk followed by an experimental period of 16 wk. The results showed that (1) the dietary Optigen level did not significantly affect average daily feed intake ( $P > 0.05$ ); (2) goats in the group with dietary Optigen level of 0.5% have a significantly high average daily milk yield ( $P < 0.05$ ); (3) the dietary Optigen level had significant influences on milk fat ( $P < 0.05$ ), but there was no significant difference among 3 experimental group containing 0.25, 0.5, and 0.75% Optigen in the diet ( $P > 0.05$ ); milk protein percentage, milk lactose percentage and SNF percentage were all unaffected ( $P > 0.05$ ) by the supplementation of Optigen; (4) the dietary Optigen level had significant influences on plasma urea nitrogen ( $P < 0.05$ ), goats with 0.5% Optigen supplement in the diet demonstrated the lowest plasma urea nitrogen content, and followed by that with 0.75% and 0.25% Optigen supplement in the diet. Plasma triglyceride, total cholesterol and total protein were unaffected ( $P > 0.05$ ) by the supplementation of Optigen. In conclusion, dietary supplementation of Optigen can increase milk fat content and reduce plasma urea nitrogen content. 0.5% supplementation of Optigen in the diet can obtain a higher fat content and milk production.

**Key Words:** Optigen, milk performance, dairy goat

**T402 A sensory additive increases milk and protein responses to concentrate supplementation in grazing dairy cows.** R. Pulido<sup>1</sup>, M. Ruiz<sup>1</sup>, F. Bargo\*<sup>2</sup>, G. Tedó<sup>2</sup>, R. Cussen<sup>3</sup>, J. Acuña<sup>3</sup>, J. R. Roche<sup>4</sup>, and I. R. Ipharraguerre<sup>2</sup>, <sup>1</sup>UACH, Valdivia, Chile, <sup>2</sup>Lueta SA, Barcelona, Spain, <sup>3</sup>BestFed, Osorno, Chile, <sup>4</sup>Down to Earth Advice Ltd., Hamilton, New Zealand.

Forty 5 Holstein dairy cows (62 DIM, 501 kg BW) were assigned to a 3 × 2 incomplete Latin square (LS) design replicated 15 times to evaluate the effect of a sensory additive (ProEfficient, PE) on milk response to concentrate supplementation on pasture. Cows were blocked by parity

in 15 blocks and within blocks randomly assigned to 3 concentrate treatments: 1 kg/d mineral concentrate (MC); 7 kg/d starch-based concentrate (CC); and 7 kg/d of CC with 30 g/d of PE (PEC). Cows grazed a perennial ryegrass pasture during 56 d divided into 2 28-d periods. Targeted herbage allowance and pre grazing herbage mass were 25 kg DM/cow and 2400 kg DM/ha, respectively. Concentrates were fed twice daily at milking. The MC was composed of 60% wheat/barley/oat mix, 10% rapeseed meal, and 30% of a mineral premix, and averaged 10.4% CP, 17.2% NDF, and 64.3% in vitro DM digestibility (IVDMD). The CC and PEC were composed of 65% corn, 13.5% triticale/oat/wheat mix, 10% soybean meal, 5% rapeseed meal, 2% sugar beet molasses, and 4.5% mineral premix, and averaged 14.1% CP; 14.7% NDF, 88.4% IVDMD. Ryegrass pasture averaged 22.3% CP, 41.9% NDF, and 77.7% IVDMD. Data were analyzed using a mixed model that included the fixed effects of treatment, block, period, their 2-way interactions, and the random effect of cow within block. On average, supplementation with the starch-based concentrate increased ( $P < 0.05$ ) milk yield 4.4 kg/d compared with MC (24.3 vs. 28.7 kg/d, SEM 0.284). However, cows supplemented with PEC produced more ( $P < 0.05$ ) milk than CC cows (29.0 vs. 28.4 kg/d, respectively; SEM 0.284) because the milk production response to concentrate supplementation was improved 16% ( $P < 0.05$ ) by the addition of PE into the concentrate (0.79 kg vs. 0.68 kg milk/kg concentrate for PEC and CC, respectively). Supplementation with PEC also increased ( $P < 0.05$ ) milk protein percentage (3.74 vs. 3.43%, SEM 0.11) and yield (1.084 vs. 0.978 kg/d, SEM 0.039) compared with CC. The milk and protein responses triggered by PE might be associated with a reduction in the substitution rate of CC for pasture or changes in the concentration of the orexigenic hormone ghrelin.

**Key Words:** sensory additive, milk response to supplementation, grazing dairy cow

**T403 Effect of intensified milk feeding on immune status and hepatic energy metabolism of calves.** Christine T. Schäff<sup>1</sup>, Tadeusz Stefaniak<sup>2</sup>, Paulina Jawor<sup>2</sup>, and Harald M. Hammon<sup>1</sup>, <sup>1</sup>Leibniz Institute for Farm Animal Biology (FBN), Dummerstorf, Germany, <sup>2</sup>Faculty of Veterinary Medicine, Wrocław University of Environmental and Life Science, Wrocław, Poland.

Ad libitum milk feeding of neonatal calves improves growth, which is reflected by increased plasma concentrations of glucose, insulin, and IGF-I. Thus, we hypothesized that ad libitum milk replacer feeding during the first 5 wk of life affects hepatic energy metabolism and immune status before weaning in neonatal calves. Holstein calves were fed colostrum for 3 d followed by milk replacer (MR) feeding (125 g/l water) until wk 9 either restricted to 6 l/d all the time (RES; n = 14) or ad libitum until d 35 and then stepwise reduced in wk 6 to 6 l/d and fed like RES thereafter (ADLIB; n = 14). Calves had free access to concentrate and hay and were slaughtered at d 60 ± 2. Blood samples were taken on d 1, 2, and 7, then weekly until wk 8 for determination of plasma concentrations of immunoglobulin (Ig) G1, G2, M, haptoglobin, and fibrinogen. Liver samples were taken at slaughter to determine glycogen concentration and mRNA abundance of IGF-I, IGF and insulin receptor, IGF binding proteins 2, 3, and 4, glucose-6-phosphatase, pyruvate carboxylase, cytosolic phosphoenolpyruvate carboxykinase (*PCK1*), and glucose transporter GLUT 2 by quantitative real-time PCR. Data were analyzed by Mixed Model (plasma data) and General Linear Model (mRNA and glycogen data) of SAS. Plasma concentrations of all Ig equally increased ( $P < 0.05$ ) after first colostrum intake in both groups. Plasma concentrations of fibrinogen ( $P < 0.05$ ) increased during 1st wk in both group and tended to be greater ( $P < 0.1$ ) in RES than in ADLIB, whereas plasma haptoglobin concentrations were not affected

by feeding. Hepatic glycogen concentration and mRNA abundance were unaffected by feeding except abundance of *PCK1* was greater ( $P < 0.05$ ) in ADLIB than in RES indicating possible influence of intensified milk feeding on gluconeogenesis. The hepatic somatotrophic axis was not influenced at slaughter by different early feeding. Therefore, intensive milk feeding for 5 wk had no long-lasting effects on hepatic energy metabolism and Ig status in blood plasma, but may partly affect hepatic acute phase protein response in milk-fed calves.

**Key Words:** calf, ad libitum feeding, energy metabolism

**T404 Effect of lactation stage and rate of increase of concentrate allowance on rumen adaptation in dairy cows.** Kasper Dieho<sup>1</sup>, André Bannink<sup>2</sup>, Thomas J. Schonewille<sup>3</sup>, and Jan Dijkstra<sup>1</sup>, <sup>1</sup>Animal Nutrition Group, Wageningen University, Wageningen, the Netherlands, <sup>2</sup>Animal Nutrition, Wageningen UR Livestock Research, Wageningen, the Netherlands, <sup>3</sup>Department of Farm Animal Health, Faculty of Veterinary Medicine, Utrecht University, Utrecht, the Netherlands.

We studied the effects of lactation stage and postpartum rate of increase of concentrate allowance on daily fermentable organic matter (FOM) intake, rumen papilla surface area, and fractional VFA absorption rate ( $k_a$ VFA). The concentrate treatment aimed to create a transient difference in daily FOM intake. Twelve rumen-cannulated, Holstein-Friesian cows had free access to either a dry period ration or a basal lactation ration. Starting at 4 DIM, concentrate allowance increased at a high (1.0 kg DM/d; HIGH, n = 6) or a low rate (0.25 kg DM/d; LOW, n = 6) up to a maximum of 10.9 kg DM/d. On sampling days (-50, -30, -10, 3, 9, 16, 30, 44, 60, and 80 DIM), the rumen contents were evacuated and papilla biopsies taken. Thereafter,  $k_a$ VFA was measured using the empty washed rumen technique with 46 L McDougall buffer (pH 6.0, 39°C), containing 120 mM VFA (60% acetic, 25% propionic, and 15% butyric acid) and a marker (Co-EDTA). From -50 to 3 DIM, FOM intake was 5.7 ± 0.3 kg/d ( $P = 0.29$ ), papilla surface area decreased from 34.4 to 28.0 ± 2.0 mm<sup>2</sup> ( $P = 0.02$ ) and  $k_a$ VFA decreased from 0.51 to 0.33 ± 0.03 /h ( $P < 0.01$ ), with no differences between future treatment groups HIGH and LOW ( $P \geq 0.33$ ). From 3 to 80 DIM, FOM intake, papilla surface area, and  $k_a$ VFA increased to 15.0 ± 0.3 kg/d, 63.4 ± 2.0 mm<sup>2</sup>, and 0.75 ± 0.03 /h respectively (all  $P < 0.01$ ). A treatment by DIM interaction occurred for daily FOM intake ( $P < 0.01$ ), which was greater in group HIGH at 16 DIM (12.3 vs. 10.1 ± 0.3 kg/d,  $P < 0.01$ ), and for papilla surface area ( $P = 0.01$ ), which was greater in group HIGH from 16 DIM (46.0 vs. 33.2 ± 2.0 mm<sup>2</sup>,  $P < 0.01$ ) through to 44 DIM (60.5 vs. 49.7 ± 2.0 mm<sup>2</sup>,  $P = 0.01$ ). However, a treatment by DIM interaction did not occur for  $k_a$ VFA ( $P = 0.28$ ), which was similar for group HIGH and LOW at 16 DIM (0.60 vs. 0.56 ± 0.03 /h,  $P = 0.53$ ) and 44 DIM (0.69 vs. 0.69 ± 0.03 /h,  $P = 0.95$ ). In conclusion, lactation stage but not concentrate treatment affected  $k_a$ VFA. This suggests  $k_a$ VFA increases with, but is not limited by, papilla surface area during the first weeks of lactation, which was affected by the rate of increase of concentrate allowance.

**Key Words:** VFA absorption, rumen papillae

**T405 Breed and stage of lactation affect the content of bioactive fatty acids in milk.** Melissa L. Bainbridge<sup>1</sup>, Laura M. Cersosimo<sup>1</sup>, André-Denis G. Wright<sup>2</sup>, and Jana Kraft<sup>1</sup>, <sup>1</sup>University of Vermont, Burlington, VT, <sup>2</sup>University of Arizona, Tucson, AZ.

Dairy products are a significant contributor to the fat intake of the American diet and contain several bioactive fatty acids (FA) that possess health benefits. The objective of this study was to compare the

content and profile of bioactive FA in milk between Holstein (HO), Jersey (JE), and F<sub>1</sub> HO × JE crossbreds (CB). Twenty-two primiparous cows (n = 7 HO, n = 7 CB, n = 8 JE) were followed across a lactation. All cows were fed a consistent TMR at a 70:30 forage to concentrate ratio. Sample periods (SP) were defined as 3 d in milk (DIM), 93 DIM, 183 DIM, and 273 DIM. Milk sample and data collection occurred on days -2 to 2 relative to the SP. Data were analyzed using a repeated measures general linear mixed model in SAS. HO and CB, had a higher content of n-3 FA in milk fat than JE (0.60 and 0.63 vs. 0.47 g/100g FA; *P* < 0.01) and a lower n-6/n-3 ratio (3.2 and 3.1 vs. 3.8; *P* < 0.01). SP had an effect on the n-6/n-3 ratio of milk fat, with the lowest value observed at 3 DIM (2.6; *P* < 0.01) and the highest at 183 DIM (3.9; *P* < 0.01). The content of conjugated linoleic acids (CLA) in milk fat was affected by breed and SP. HO and CB had a higher proportion of CLA than JE (0.50 and 0.48 vs. 0.37 g/100g FA; *P* < 0.01). Overall, CLA content decreased over lactation (0.54, 0.45, 0.40, and 0.40 g/100g FA for 3, 93, 183 and 273 DIM, respectively; *P* < 0.01). Breed had no effect on content of total branched-chain fatty acids (BCFA) in milk fat, however, breed affected individual BCFA. Content of *iso* 14:0 in milk fat was lower in HO than in JE (0.10 vs. 0.13 g/100g FA; *P* < 0.05) and content of *iso* 17:0 was higher in HO and CB than in JE (0.33 and 0.31 vs. 0.27g/100g FA; *P* < 0.01). Total BCFA were affected by SP, with 93 and 183 DIM (1.51 and 1.59 g/100g FA) having a lower proportion of BCFA than 3 and 273 DIM (1.71 and 1.74 g/100g FA; *P* < 0.01). In conclusion, HO and CB exhibited a higher content of bioactive FA in milk than JE. Across a lactation, the greatest content of bioactive FA in milk occurred postpartum.

**Key Words:** branched-chain fatty acids, n-3 fatty acids, CLA

**T406 Rumen microbial protein outflow, and plasma amino acid levels, in early lactation multiparity Holstein cows on commercial California dairy farms.** Nadia Swanepoel<sup>\*1,2</sup>, Peter H. Robinson<sup>1</sup>, and Lourens J. Erasmus<sup>2</sup>, <sup>1</sup>*Department of Animal Science, University of California, Davis, CA*, <sup>2</sup>*Department of Animal and Wildlife Sciences, University of Pretoria, Pretoria, South Africa.*

Metabolic models are unlikely to accurately predict microbial crude protein outflow (MCP) from the rumen and amino acid (AA) limitations since they are generally based on static values of unknown accuracy. Practical and reliable methods to identify MCP and AA limitations are required to evaluate nutritional limitations in commercial groups of dairy cows in 'real time'. Our objective was to determine normal ranges of plasma AA, and MCP, in early lactation multiparity Holstein dairy cows fed a range of rations as a benchmark to determine high, low and normal levels. Multiparity high cow groups on 19 commercial dairies in California were selected. One load of total mixed ration (TMR) was collected at feeding, and milk yields were obtained from monthly milk tests on each dairy. Spot urine samples were collected from voluntarily urinating cows between 38 and 151 d in milk (DIM) to focus on early lactation. Specific gravity of each urine sample was determined to estimate urine flow. Urine samples from each dairy were analyzed for allantoin, which was used, with estimated urine volume, to calculate MCP. Blood was collected from the tail vein of 12 cows/dairy (65 to 76 DIM) and assayed for AA. Group averages among the dairies were 33.6 to 52.5 kg/d milk yield, 1309 to 2149 g/d MCP, 2.8 to 4.4, 8.4 to 13.9, 5.9 to 9.7, 22.9 to 38.5, 16.9 to 27.5 and 11.3 to 18.6 µg/mL for Met, Lys, His, Val, Leu, and Ile respectively. Correlation analysis was performed using the STEPWISE Procedure of SAS with a backward elimination of parameters. Milk and DIM were poor MCP predictors (*r*<sup>2</sup> < 0.01 and 0.05 respectively) and neutral detergent fiber (NDF; *P* = 0.02, *r*<sup>2</sup> = 0.23) was the only TMR nutrient that was predictive of MCP,

with higher diet NDF levels leading to increased MCP flow. Multiple correlation with all TMR nutrients (% DM) created the best fit model: MCP (g/d) = -5793.1 + (118.8 × NDF) + (150.1 × Fat) + (87.0 × Starch) + (74.0 × nonfiber carbohydrate) [*r*<sup>2</sup> = 0.50]. Results show that plasma AA (from 6 cows) and MCP (from 8 cows) are sufficient to determine whether plasma AA and/or MCP values in a group of cows are low, high, or normal thereby suggesting changes in rations to address the identified issue(s).

**Key Words:** allantoin, spot urine samples

**T407 Evaluating different physical concentrate forms in preweaned calves.** Marta Terré<sup>1</sup>, Maria Devant<sup>\*1</sup>, and Alex Bach<sup>2,1</sup>, <sup>1</sup>*IRTA, Caldes de Montbui, Spain*, <sup>2</sup>*ICREA, Barcelona, Spain.*

In Experiment 1, 37 crossbreed calves were used in a cafeteria study to evaluate calves preferences for different physical form presentation of concentrates. The same concentrate with 4 different physical presentations: meal (ML), pellet (PL), pellet and whole cereal grains (WHL), and pellet and flaked cereal grains (FLK) were simultaneously available from 4 to 45 d age (weaning). A preference diet ratio was calculated: daily concentrate DM intake for each concentrate presentation divided by daily total DM intake divided by the total number of concentrate offered, and an ANOVA with repeated measures was used to analyze the preference ratio. In Experiment 2, 63 Holstein calves were randomly distributed in 2x2 factorial design to evaluate performance of calves fed 2 different protein sources in the milk replacer (MR) and 2 concentrates with different presentation form: calves received either a MR containing dried whey as a protein source, and a pellet concentrate with whole grains of corn and barley or corn and barley flakes; or calves received a MR with dried skim milk as protein ingredient, and a pellet concentrate with whole grains of corn and barley or corn and barley flakes. Animals were weighed weekly, and intakes were recorded daily. Data were analyzed with an ANOVA with repeated measures. In Experiment 1, during the first week of study, calves had a lower (*P* < 0.01) preference for FLK than for WHL; however, thereafter calves showed the greatest (*P* < 0.01) preference for FLK at 2, 4, and 5 week of study. In Experiment 2, there were no differences between milk replacer types, and starter concentrate intake was greater (*P* < 0.05) in whole-grain than in flaked-grain concentrates from the 5 wk of study until weaning. However, no differences in growth or gain to feed ratio were observed. It is concluded that preweaned calves prefer concentrates based on pellets and flaked-grains, and when flaked-grains are compared with whole-grains, lower concentrate intake is observed, but their performance was not impaired.

**Key Words:** calves, physical form, preference

**T408 Fetal programming on dairy cows: Effect of dam's parity and days in milk at conception on first-lactation milk yield in dairy cows.** Ayelen Chiarle<sup>1</sup>, Ramiro Rearte<sup>1</sup>, Santiago Corva<sup>1</sup>, R. Luzbel de la Sota<sup>1,2</sup>, Mauricio Giuliodori<sup>1</sup>, and Alejandro Relling<sup>\*1,3</sup>, <sup>1</sup>*Fac. Cs Veterinarias, UNLP, La Plata, Argentina*, <sup>2</sup>*CCT La Plata, CONICET, Argentina*, <sup>3</sup>*IGEVET, CCT La Plata, CONICET, Argentina.*

Fetal programming is defined as nutritional and/or endocrine changes that happen in the cow during gestation that may change expression of some genes on the adult life of the progeny. These changes may have an impact on the progeny's performance. Therefore, the objective of the current study was to evaluate the effect of dam's parity (1 through 8) and days in milk (DIM) at conception on the daughter's first lactation milk yield accumulated up to 150 d (MY150, kg). Data from

dairy cows (n = 159.886) and their daughters (n = 201.547) from 1131 Argentinean dairy herds from ARPECOL (La Plata, BA, Argentina) were included in the study. Data were analyzed as completely randomized block design with a mixed model (SAS 9.3). The model included the fixed effects of parity (1 through 8) and DIM at conception, divided into quartiles (1st: >77 d, 2nd: 77–115 d; 3rd: 115–190 d; 4th: <190); and the random effects of dam and dairy herd (block). Mean DIM for each quartile were 57, 94, 148 and 296 d. Linear, quadratic and cubic polynomial contrasts were used for mean separation. The daughters' MY150 decreases ( $P$ -value Linear < 0.01, Quadratic < 0.01, Cubic > 0.10) when parity number increases. Mean MY150 were 2915, 2879, 2862, 2853, 2844, 2846, 2837 and 2835 kg for 1st, 2nd, 3rd, 4th, 5th, 6th, 7th and 8th parity, respectively. In addition, daughters' MY150 increases ( $P$ -value Linear < 0.01, Quadratic < 0.05, Cubic > 0.10) when DIM at conception increases. Mean MY150 were 2851, 2858, 2863, and 2863 kg for those conceived on average at 57, 94, 148 and 296 DIM, respectively. In summary, physiological changes related to parity and DIM at conception have an impact on progeny's MY150.

**Key Words:** dairy cattle, fetal programming, progeny milk yield

**T409 Transcriptome profile in cows resistant to milk fat depression.** Adriana Siurana<sup>1</sup>, Sergio Calsamiglia<sup>\*1</sup>, David Gallardo<sup>3</sup>, and Angela Canovas<sup>2</sup>, <sup>1</sup>*Animal Nutrition and Welfare Service, Universitat Autònoma de Barcelona, Bellaterra, Spain*, <sup>2</sup>*Servei Veterinari de Genètica Molecular, Universitat Autònoma de Barcelona, Bellaterra, Spain*, <sup>3</sup>*Departament de Genètica Animal, Centre de Recerca en Agrigenòmica, Universitat Autònoma de Barcelona, Bellaterra, Spain*.

Feeding linseed to dairy cows results in milk fat depression (MFD), but there is a wide range of sensitivity among cows. The objectives of this study were (1) To compare the mRNA expression of transcripts expressed in milk somatic cells in cows resistant or sensitive to MFD; (2) to identify metabolic pathways and transcription factors affected by MFD in resistant cows under different dietary conditions (no fat supplemented or linseed rich-diet). Four cows were selected from a dairy farm after a switch from a control diet to a linseed-rich diet. Among them, 2 cows (R-MFD) were resistant to MFD showing high milk fat content in both control (CTR) (4.06%) and linseed-rich diet (LIN) (4.36%); and 2 cows (S-MFD) were sensitive to MFD decreasing milk fat content after the change into the LIN diet (3.56 to 2.54%). Fresh milk samples were collected from each cow the week before and 2 weeks after the diet change. Transcriptome analysis was performed using RNA-sequencing technology with a HiSeq2000 platform. Differential expression analysis between cows S-MFD vs. R-MFD allowed to detect a large number of genes differentially expressed (DE) in both diets, CTR (n = 1,111) and LIN (n = 1,669). In addition, 726 genes were differentially expressed between CTR and LIN diets in cows R-MFD whereas only 38 genes were identified in cows S-MFD. Results showed an overexpression in genes and pathways related to fatty acid synthesis and lipid metabolism such as TR/RXR and PPAR $\alpha$ /RXR $\alpha$  Activation pathways in cows R-MFD (CTR vs. LIN). Several genes and transcription factors such as *FOXO3*, *MTOR*, *PPARA*, *PPARG*, *SREBF1* and *MYC* were identified acting as key regulators in R-MFD cows with linseed-rich diet. These results suggest that R-MFD cows could be activating a compensatory mechanism to increase the fatty acid synthesis in linseed-rich diets. It would be interesting to combine these results with the study of the structural variation in the whole transcriptome of S-MFD and R-MFD cows to contribute to the better understanding of molecular mechanisms affecting the MFD in cows.

**Key Words:** milk fat depression, fatty acid synthesis, RNA-sequencing

**T410 Apparent synthesis of vitamin B<sub>6</sub> and folates in the rumen of lactating dairy cows fed alfalfa or orchardgrass silages.** D. S. Castagnino<sup>\*1,2</sup>, M. Seck<sup>1,2</sup>, K. L. Kammes<sup>3</sup>, J. A. Voelker Linton<sup>3</sup>, M. S. Allen<sup>3</sup>, R. Gervais<sup>2</sup>, P. Y. Chouinard<sup>2</sup>, and C. L. Girard<sup>1</sup>, <sup>1</sup>*Agriculture and Agri-Food Canada, Sherbrooke, QC, Canada*, <sup>2</sup>*Département des Sciences Animales, Université Laval, Québec, QC, Canada*, <sup>3</sup>*Department of Animal Science, Michigan State University, East Lansing, MI*.

Effects of forage family (legume vs. grass) on apparent ruminal synthesis and post-ruminal supply of vitamin B<sub>6</sub> and folates were evaluated in 2 experiments. Diets containing either alfalfa (AL) or orchardgrass (OG) silages as the sole forage were offered to ruminally and duodenally cannulated lactating Holstein cows in crossover design experiments. Experiment 1 (E1) compared diets containing AL and OG (~27% total NDF) offered to 8 cows in 2 15-d treatment periods. Experiment 2 (E2) compared diets containing AL and OG (~30% total NDF) offered to 13 cows in 2 18-d treatment periods. Intake, duodenal flow and apparent ruminal synthesis (ARS) for each vitamin were analyzed using SAS mixed procedure. The experiments were analyzed separately. Differences were declared significant at  $P < 0.05$ . Vitamin B<sub>6</sub> intakes were greater for AL than OG in E1 (368 vs. 236  $\pm$  18.2 mg/d;  $P < 0.01$ ) and E2 (180 vs. 77  $\pm$  5.1 mg/d;  $P < 0.01$ ). In contrast, folates intakes were lower for AL than OG in E1 (7.6 vs. 9.2  $\pm$  0.51 mg/d;  $P = 0.02$ ) and E2 (8.7 vs. 10.2  $\pm$  0.29 mg/d;  $P < 0.01$ ). In E1, duodenal flows of vitamins were not affected by treatment (vitamin B<sub>6</sub>, 72  $\pm$  6.2 mg/d,  $P = 0.63$ ; folates, 33  $\pm$  4.2 mg/d,  $P = 0.15$ ). In E2, duodenal flows of vitamin B<sub>6</sub> were greater for AL than OG (59 vs. 31  $\pm$  5.2 mg/d;  $P < 0.01$ ) but the duodenal flows of folates were not affected by treatment (39  $\pm$  2.7 mg/d;  $P = 0.78$ ). Alfalfa increased the apparent ruminal degradation of vitamin B<sub>6</sub> compared with OG in E1 (-294 vs. -167  $\pm$  14.5 mg/d;  $P < 0.01$ ) and E2 (-121 vs. -46  $\pm$  4.9 mg/d;  $P < 0.01$ ). The ARS of folates were not affected by treatment in either experiment (E1: 25  $\pm$  3.9 mg/d,  $P = 0.19$ ; E2: 30  $\pm$  2.5 mg/d,  $P = 0.81$ ). Combining data from both experiments, vitamin B<sub>6</sub> intake was correlated positively with its duodenal flow ( $r = 0.68$ ,  $P < 0.01$ ) but negatively with ARS synthesis of the vitamin ( $r = -0.98$ ,  $P < 0.01$ ). Folates intake was correlated positively with its duodenal flow ( $r = 0.68$ ,  $P < 0.01$ ) and its ARS ( $r = 0.59$ ,  $P < 0.01$ ). Forage family affected the ruminal balance of vitamin B<sub>6</sub> but had little or no effect on the post-ruminal supply of vitamin B<sub>6</sub> and folates.

**Key Words:** dairy cow, vitamin B<sub>6</sub>, folates

**T411 Noninvasive indicators to identify lactating dairy cows with the greater risk of subacute rumen acidosis.** Xiaosheng Gao<sup>\*</sup> and Masahito Oba, *University of Alberta, Edmonton, Alberta, Canada*.

The objective of the study was to evaluate if milk urea nitrogen (MUN) and milk fat content could be used as the noninvasive indicator to identify cows with a greater or lower risk of subacute ruminal acidosis (SARA). Our hypothesis was that cows greatly vary in their risk to develop SARA even if they are fed a common diet, and that cows with lower MUN and milk fat content would have a greater risk of SARA while cows with higher MUN and milk fat content would have a lower risk of SARA. In a screening study, 35 late-lactating Holstein cows (DIM = 250  $\pm$  71.1; BW = 601  $\pm$  45.4 kg) were fed a high grain diet containing 35% forage and 65% concentrate mix ad libitum for 21 d. Concentration of MUN ranged from 5.7 to 13.9 mg/dL among the 35 cows, and the average milk fat content was 3.5%. Then, 5 cows with highest MUN concentrations with milk fat higher than 3.5% were selected as animals that presumably have a low risk of SARA (LOW), and 5 cows with lowest MUN concentrations with milk fat less than 3.5% were selected as animals

that presumably have a high risk of SARA (HIGH). Then these 10 animals were ruminally cannulated during the subsequent dry period. As one LOW cow was culled due to fatty liver, 9 animals (DIM = 122 ± 33.2; BW = 615 ± 49.1 kg) were used for the subsequent study in the following lactation. All cows were fed a high grain diet consisting of 35% forage and 65% concentrate mix ad libitum for 21 d. Ruminal pH was measured every 30 s for 72 h. All response variables were evaluated for the group effect using the PROC TTEST procedure of SAS (version 9.2, SAS Institute Inc., Cary, NC). Minimum (5.75 vs. 5.30;  $P = 0.02$ ) and mean ruminal pH (6.35 vs. 6.04;  $P = 0.02$ ) was higher for LOW compared with HIGH animals. In addition, duration of rumen pH below 5.8 was shorter in LOW animals (52.5 vs. 395 min/d;  $P = 0.04$ ). These results suggested that MUN and milk fat content in late-lactating cows fed a high grain diet may be used to identify cows that have a higher or lower risk of SARA.

**Key Words:** subacute ruminal acidosis, milk urea nitrogen, milk fat

**T412 Effects of different dosages of *Saccharomyces cerevisiae* fermentation product on lactation performance of dairy cows under heat stress.** Wen Zhu\*<sup>1</sup>, B. X. Zhang<sup>1</sup>, K. Y. Yao<sup>1</sup>, I. Yoon<sup>2</sup>, Ruby Chung<sup>2</sup>, J. K. Wang<sup>1</sup>, J. A. Ye<sup>1</sup>, and J. X. Liu<sup>1</sup>, <sup>1</sup>*Institute of Dairy Science, College of Animal Science, Zhejiang University, Hangzhou, China*, <sup>2</sup>*Diamond V, Cedar Rapids, IA*.

The objective of this study was to evaluate the effects of different dosages of *Saccharomyces cerevisiae* fermentation product (SCFP; Original XP; Diamond V, Cedar Rapids, IA) on lactation performance of mid-lactation dairy cows under heat stress. Eighty-one multiparous Holstein lactating dairy cows were divided into 27 blocks of 3 cows each based on milk yield (23.6 ± 0.20 kg/d), parity (2.88 ± 0.91) and days in milk (204 ± 46 d). The cows were randomly assigned within blocks to one of 3 dietary treatments: 0 (control), 120, or 240 g/d of SCFP mixed with 240, 120, or 0 g of corn meal, respectively. The experiment lasted for 9 weeks, using the first week as adaption period. All data were analyzed through the PROC MIXED program of SAS with the covariance type AR (1) for repeated measures. Means were separated using the PDIF option in the LSMEANS statement. Linear and quadratic effects of treatment were tested using orthogonal polynomial contrasts. During the experimental period, average daily Temperature-Humidity Index (measured at 08:00, 14:00 and 20:00) was above 72 for 47 out of 56 d (84%). Rectal temperatures tended to decrease linearly ( $P = 0.07$ ) for cows supplemented with SCFP compared with control cows at 14:30, but were not different at 06:30 ( $P > 0.05$ ). Dry matter intake was not affected ( $P > 0.05$ ) by SCFP supplementation. Milk yield increased linearly ( $P = 0.02$ ) with increasing dosage of SCFP. Feed efficiency (milk yield/dry matter intake) was highest ( $P = 0.04$ ) for cows fed 240 g/d SCFP. Net energy balance also increased linearly ( $P < 0.01$ ) with increasing dosage of SCFP. Cows supplemented with SCFP gained ( $P < 0.01$ ) body weight, while control cows lost body weight. Concentrations of milk urea nitrogen decreased linearly ( $P < 0.01$ ) with increasing dosage of SCFP, while no difference was observed among the treatments in conversion of dietary N to milk N. In summary, supplementation of SCFP alleviated the negative effect of heat stress in lactating Holstein dairy cows and allowed cows to maintain higher milk production, feed efficiency and net energy balance. Effects of SCFP were dose-dependent and greater effects were observed from higher doses.

**Key Words:** heat stress, yeast culture, lactation performance

**T413 Effects of *Saccharomyces cerevisiae* fermentation products on lactation performance, rumen fermentation and microbial communities in dairy cows fed a diet containing low quality forages.** Wen Zhu\*<sup>1</sup>, Z. H. Wei<sup>1</sup>, N. N. Xu<sup>1</sup>, Fan Yang<sup>1</sup>, I. Yoon<sup>2</sup>, Ruby Chung<sup>2</sup>, J. K. Wang<sup>1</sup>, J. A. Ye<sup>1</sup>, and J. X. Liu<sup>1</sup>, <sup>1</sup>*Institute of Dairy Science, College of Animal Science, Zhejiang University, Hangzhou, China*, <sup>2</sup>*Diamond V, Cedar Rapids, IA*.

The objective of this study was to evaluate the effects of various dosages of *Saccharomyces cerevisiae* fermentation product (SCFP; Original XP; Diamond V) on lactation performance, rumen microbial communities and rumen fermentation in mid-lactation dairy cows fed a total mixed ration containing low quality forages. Eighty multiparous Holstein dairy cows were blocked based on days in milk (180 ± 45 d), parity (3.24 ± 1.06) and milk yield (26.6 ± 0.79 kg/d), and were randomly assigned to one of 4 treatments. Treatments consisted of 0, 60, 120, or 180 g/d of SCFP per head mixed with 180, 120, 60, or 0 g of corn meal, respectively. The forage in the basal diet was comprised (DM basis) of 15% corn stover, 7% Chinese wild ryegrass, and 17.3% corn silage. The experiment lasted for 10 weeks, with the first 2 weeks for adaption. The data were analyzed using the PROC MIXED program of SAS with the covariance type AR (1) for repeated measures. Means were separated by using the PDIF option in the LSMEANS statement. Orthogonal polynomial contrasts were used for linear and quadratic effects testing. Dry matter intake decreased linearly ( $P = 0.02$ ), while milk yield increased linearly ( $P = 0.05$ ) with the increasing dosage of SCFP, resulting in improved ( $P < 0.01$ ) feed efficiency compared with the control. Contents of milk components were similar ( $P > 0.05$ ) among the treatments. Neither rumen pH ( $P = 0.70$ ) nor ammonia-nitrogen concentration ( $P = 0.82$ ) was affected by SCFP supplementation. Increasing dosage of SCFP linearly increased ( $P < 0.05$ ) concentrations of ruminal total volatile fatty acids, acetate, propionate and butyrate, with no difference ( $P > 0.05$ ) in molar proportion of individual acids. The population of fungi and cellulolytic bacteria increased linearly ( $P < 0.01$ ), but those of lactate-utilizing and lactate-producing bacteria decreased linearly ( $P < 0.01$ ) with increasing dosage of SCFP. The estimated microbial protein yield in the rumen increased linearly ( $P < 0.01$ ) in response to SCFP supplementation. In summary, supplementation of SCFP improved the lactation performance of dairy cows consuming diets containing low quality forages by optimizing rumen fermentation through manipulating rumen populations. Effects of SCFP were dose-dependent and greater effects were observed from higher doses.

**Key Words:** low-quality forage, yeast culture, lactation performance

**T414 Response of dairy cows to monensin on diets differing in starch content and source.** Eugenio F. Barbosa<sup>1</sup>, Julia D. L. Dias<sup>1</sup>, Fabiana F. Cardoso<sup>1</sup>, Túlio H. R. Souza<sup>1</sup>, Lucas C. Resende<sup>1</sup>, Ozana F. Zacaroni<sup>1</sup>, Renata A. N. Pereira<sup>3,2</sup>, and Marcos N. Pereira\*<sup>1,2</sup>, <sup>1</sup>*Universidade Federal de Lavras, Lavras, MG, Brazil*, <sup>2</sup>*Better Nature Research Center, Ijaci, MG, Brazil*, <sup>3</sup>*Empresa de Pesquisa Agropecuária de Minas Gerais, Lavras, MG, Brazil*.

The response of dairy cows to monensin can interact with diet composition. We evaluated the response of lactating cows to monensin supplementation of diets differing in rumen fermentable starch. Twenty-eight Holsteins (157 ± 76 DIM) were individually fed a standard TMR for 3 wks and monensin (MON. 300 mg/d) or control (CTL) for 9 wks, in a covariate adjusted randomized block design. A low-starch diet (LS. 25.8% starch in DM) was offered from d 1 to 35 and a high-starch diet (HS. 30% starch in DM) from d 36 to 63. HS was formulated by replacing whole cottonseed and finely ground mature corn by an isonitrogenous mixture of soybean meal and high moisture corn. Data were collected

daily or after adaptation to LS and HS, and were analyzed as repeated measures with PROC MIXED of SAS. The mean square for cow nested within treatment (MON vs. CTL) was the error term for the treatment effect. Digestible OM intake (DOMI) was increased by HS (15.3 vs. 14.4 kg/d,  $P < 0.01$ ), but was similar for MON (14.6 kg/d) and CTL (15.1 kg/d,  $P = 0.41$ ). Milk yield did not respond to MON (32.3 kg/d,  $P = 0.32$ ). MON reduced DMI and increased Milk/DMI, ECM/DMI, and ECM/DOMI only when HS was fed ( $P < 0.01$  for the interaction of treatment and wk/diet). Plasma glucose content was increased by MON and the response interacted similarly with wk/diet ( $P < 0.05$ ). MON reduced the A/P ratio in rumen fluid in both diets (2.43 vs. 1.81,  $P < 0.01$ ) and HS reduced ruminal pH ( $P < 0.01$ ) and increased protozoa content ( $P < 0.01$ ). MON reduced total-tract NDF digestibility more in LS (52.1 vs. 41.3% of intake) than in HS (57.0 vs. 52.0,  $P < 0.05$  for the interaction). The daily urinary allantoin excretion did not respond to MON ( $P = 0.40$ ), neither PUN at 0, 1.5, and 3 h post feeding ( $P > 0.27$ ). MON induced lower degree of refusal of long feed particles and of preferential intake of short particles in the afternoon than CTL ( $P < 0.05$ ). Ingestion time (min/d and min/kg of DMI) and the number of daily meals were reduced by HS ( $P < 0.05$ ). HS increased pCO<sub>2</sub>, HCO<sub>3</sub><sup>-</sup>, total CO<sub>2</sub>, and base excess of jugular blood ( $P < 0.04$ ) and tended to reduce blood pH ( $P = 0.10$ ). The positive effect of MON on feed efficiency occurred only when ruminally available starch was increased in the diet.

**Key Words:** monensin, starch, feed efficiency

#### T415 Growth performance of dairy heifers fed reduced-fat distillers grains in replacement of forage in limit-fed rations.

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Our objective was to determine the maximum inclusion rate of reduced-fat distillers dried grains (RFDDGS) in dairy heifer rations. A 16-wk randomized complete block design study was conducted using 48 Holstein heifers (199 ± 2 d of age) to evaluate effects of diet on growth performance. Treatments were 30% RFDDGS (30DG), 40% RFDDGS (40DG), and 50% RFDDGS (50DG) with the rest of the diets consisting of grass hay and 1.5% mineral mix. Using Calan gates rations were limit-fed at 2.65, 2.50, and 2.35% of BW (DM basis) for 30DG, 40DG, and 50DG, respectively to have similar intakes of CP and energy among treatments. Frame sizes, BW, and BCS were measured on 2 consecutive days during wk 0, 2, 4, 6, 8, 10, 12, 14, and 16 of the feeding period. Data were analyzed using the MIXED procedures with repeated measures in SAS 9.4. Changes per day were analyzed by regression analysis. There were no interactions of treatment by wk. Heifer DMI linearly decreased ( $P < 0.01$ ) with increased dietary concentrations of RFDDGS (6.49, 6.21, and 5.84 kg/d; SEM = 0.117 for 30DG, 40DG, and 50DG, respectively). Body weights (264.1, 266.2, and 266.4 kg; SEM = 4.98) and ADG (0.89, 0.94, and 0.97 ± 0.083 kg/d) were similar ( $P > 0.05$ ) among treatments. Gain to feed (0.14, 0.16, and 0.18 kg; SEM = 0.0034) linearly increased ( $P < 0.01$ ). Hip height (124.8, 124.7, and 124.8 cm; SEM = 0.36) and hip width (35.6, 35.8, and 35.8 cm; SEM = 0.57) were similar ( $P > 0.05$ ) among treatments. Body length (112.5, 112.9, and 113.1 cm; SEM = 0.18) tended ( $P = 0.06$ ) to linearly increase across treatments. There was a quadratic effect ( $P < 0.05$ ) for withers height (120.9, 121.7, and 121.6 cm; SEM = 0.29), paunch girth (172.5, 173.9, and 172.5 cm; SEM = 1.29), and BCS (3.11, 3.12, and 3.07; SEM = 0.018), and a quadratic tendency ( $P = 0.08$ ) for heart girth (140.9, 140.6, and 140.9 cm; SEM = 0.41). Limit-feeding diets with greater inclusion rates of RFDDGS resulted in improved gain to feed ratios

and maintained frame growth without increasing BCS, demonstrating that replacing forage with RFDDGS does not negatively affect heifer growth performance.

**Key Words:** distillers grains, dairy heifer, growth performance

#### T416 Ruminal degradation and intestinal digestibility of camelina and carinata meal compared with other protein sources.

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Our objective was to determine DM and CP ruminal degradability and intestinal digestibility of camelina meal (CM) and carinata meal (CR) compared with other oilseed meals, soybean meal (SBM) and reduced-fat distillers dried grains (DG). In situ measurements were done using 3 multiparous, ruminally-cannulated Holstein cows (BW 848.6 ± 94.7 kg). Six feeds were evaluated: CM, CR, canola meal (CN), linseed meal (LN), DG, and SBM. Duplicate 5 g samples were weighed into 10 × 20 cm nylon bags and ruminally incubated for 0, 2, 4, 8, 12, 24 and 48 h. Six additional bags of each feed were incubated at 12 h for use in determination of in vitro intestinal digestibility of CP. Residues were incubated with pepsin and pancreatin solutions for 1 h and 24 h, respectively. Rumen degradation constants for DM and CP were estimated using the NLIN procedures in SAS 9.3. Intestinally digestible protein (IDP), intestinally absorbable dietary protein (IADP = ruminally undegradable protein (RUP) × IDP), and total digestible protein (TDP = ruminally degradable protein (RDP) + IADP) were evaluated using MIXED procedures in SAS. Ruminally degradable DM (RDDM) was greatest in CM, CR, and SBM. The CM and CR had the greatest RDP and least RUP of the feeds. The IDP was less in CM and CR compared with SBM and LN, but greater than CN and DG. The CR and CM had less IADP compared with the other feeds. The TDP was similar for CM and CR compared with SBM and LN, but greater than CN and DG. Overall, results indicate that CM and CR are highly degradable in the rumen and are comparable protein sources to SBM and LN for total digestibility.

**Table 1 (Abstr. T416).**

Item	CM	CR	CN	LN	DG	SBM	SEM
CP, % DM	40.9	43.9	45.3	40.4	35.0	53.9	—
RDDM, % of DM	65.0 <sup>a</sup>	63.0 <sup>a</sup>	50.9 <sup>c</sup>	55.8 <sup>b</sup>	50.8 <sup>c</sup>	65.0 <sup>a</sup>	0.64
RDP, % of CP	76.4 <sup>a</sup>	70.5 <sup>b</sup>	52.0 <sup>d</sup>	61.2 <sup>c</sup>	44.1 <sup>e</sup>	58.4 <sup>c</sup>	0.95
RUP, % of CP	23.6 <sup>e</sup>	29.4 <sup>d</sup>	48.0 <sup>b</sup>	38.8 <sup>c</sup>	55.9 <sup>a</sup>	41.6 <sup>c</sup>	0.95
IDP, % of RUP	80.9 <sup>b</sup>	80.9 <sup>b</sup>	70.9 <sup>c</sup>	81.6 <sup>b</sup>	63.2 <sup>c</sup>	90.5 <sup>a</sup>	2.61
IADP, % of CP	19.1 <sup>c</sup>	23.8 <sup>c</sup>	34.0 <sup>ab</sup>	31.7 <sup>b</sup>	35.4 <sup>ab</sup>	37.6 <sup>a</sup>	1.42
TDP, % of CP	95.5 <sup>a</sup>	94.4 <sup>a</sup>	86.0 <sup>b</sup>	92.9 <sup>a</sup>	79.5 <sup>c</sup>	96.0 <sup>a</sup>	1.22

<sup>a-c</sup>Values with unlike superscripts differ by  $P < 0.05$ .

**Key Words:** camelina meal, carinata meal, digestibility

#### T417 The decline in digestive efficiency of US dairy cows during the last 44 years. Sarah B. Potts\*, Melissa Shaughnessy, and Richard A. Erdman, University of Maryland, College Park, MD.

Since 1970, US milk production per cow has more than doubled resulting in large increases in feed intake. It is well established that increasing feed intake reduces diet digestibility. Our objective was to determine if the digestive efficiency of US dairy cows has also changed in the last 44 years. We assembled a data set consisting of diet digestibility measured via total collection or iNDF procedures with lactating dairy cows from studies published in the *Journal of Dairy Science* from 1970

to 2014. The data set contained 121 treatment means from 38 studies conducted at 16 US institutions. Based on regression analysis, milk yield and DMI increased by 20 kg/d and 8 kg/d between 1970 and 2014, respectively. Temporal effects on digestibility (DM, CP, or NDF) were determined using the regression model:  $\text{Digestibility} = \text{YEAR}_{1970} + \text{CP} + \text{NDF}$ ; where  $\text{YEAR}_{1970}$  is the publication year minus 1970 and CP and NDF are diet constituents (% of diet DM) included to account for their effects on digestibility. DM digestibility decreased 0.13 percentage units per year ( $P < 0.01$ ) for a total reduction of 5.7 percentage units since 1970. However, there were no temporal effects on CP and NDF digestibilities ( $P > 0.3$ ). To account for the potential effect of feed intake on digestibility, DMI as a percentage of BW ( $\text{DMI}_{\text{BW}}$ ) was included in the regression model. With  $\text{DMI}_{\text{BW}}$  in the model, DM digestibility decreased 0.08 percentage units per year ( $P = 0.03$ ) for a total reduction of 3.5 percentage units. This suggests that only a portion (39%) of the decline in digestibility was due to level of feed intake. In contrast, CP digestibility was unchanged over time ( $P = 0.94$ ), and NDF digestibility increased 0.14 percentage units per year ( $P = 0.05$ ) when the model included  $\text{DMI}_{\text{BW}}$ . Increased NDF digestibility was likely due to the use of more highly digestible NDF sources in more recent studies. These results suggest that digestive efficiency has decreased by nearly 6 percentage units over the last 44 years. Perhaps genetic selection of cows for high milk production has inadvertently resulted in cows with high feed intake capacity irrespective of effects on digestive efficiency.

**Key Words:** digestibility, digestive efficiency, dairy cow

**T418 Ruminal degradation and intestinal digestibility of microbially treated soybean meal and dried distillers grains compared with the original feeds.** Jennifer L. Casperson<sup>1</sup>, Jill L. Anderson\*<sup>1</sup>, Jason R. Croat<sup>2</sup>, and William R. Gibbons<sup>2</sup>, <sup>1</sup>Dairy Science Department, South Dakota State University, Brookings, SD, <sup>2</sup>Department of Biology and Microbiology, South Dakota State University, Brookings, SD.

Our objective was to determine if microbial (fungal) treatment of soybean meal (SBM) and dried distillers grain with solubles (DG) affected ruminal degradation and intestinal digestibility of DM and CP. In situ measurements were done using 3 mid-lactation, multiparous, ruminally-cannulated Holstein cows (BW 847 ± 96). Four feeds were evaluated: SBM, treated SBM (TSBM), DG, and treated DG (TDG). Duplicate 5 g samples in nylon bags (10 × 20 cm) were incubated in the rumen for 0, 1, 2, 4, 8, 12, 24 and 48 h. Ruminal degradation constants for DM and CP were estimated using the NLIN procedures in SAS 9.3. Six additional bags of each feed were ruminally incubated for 12 h. Residues were used to determine in vitro intestinal digestibility of CP via incubation with pepsin and pancreatin solutions for 1 h and 24 h, respectively. Intestinally digestible protein (IDP), intestinally absorbable dietary protein (IADP = Ruminal undegradable protein (RUP) × IDP), and total digestible protein (TDP = Ruminal degradable protein (RDP) + IADP) were compared using MIXED procedures in SAS. Total CP was more in TSBM and TDG compared with original feeds. Ruminally degradable DM (RDDM) was less in TSBM and DG than in SBM, TDG. The RDP was increased in TDG versus DG and decreased in TSBM versus SBM. Effects were opposite for RUP. The IDP, IADP were less in DG and TDG compared with SBM and TSBM. The TDP was greatest in SBM and least in DG. Results indicate that microbial-treatment increased protein content of SBM and DG. Digestibility of CP was increased for TDG versus DG, but was not increased in TSBM versus SBM.

Contd.

**Table 1 (Abstr. T418).**

Item	SBM	TSBM	DG	TDG	SEM
CP, % DM	49.3	59.0	29.5	42.5	—
RDDM, % of DM	67.3 <sup>a</sup>	54.1 <sup>b</sup>	54.3 <sup>b</sup>	67.1 <sup>a</sup>	1.27
RDP, % of CP	57.8 <sup>b</sup>	47.2 <sup>c</sup>	40.5 <sup>c</sup>	69.6 <sup>a</sup>	1.52
RUP, % of CP	42.2 <sup>b</sup>	52.8 <sup>a</sup>	59.5 <sup>a</sup>	30.4 <sup>c</sup>	1.52
IDP, % of RUP	89.3 <sup>a</sup>	84.2 <sup>b</sup>	61.4 <sup>c</sup>	57.7 <sup>c</sup>	0.98
IADP, % of CP	37.8 <sup>ab</sup>	44.5 <sup>a</sup>	36.5 <sup>b</sup>	17.5 <sup>c</sup>	1.52
TDP, % of CP	95.5 <sup>a</sup>	91.7 <sup>b</sup>	77.0 <sup>d</sup>	87.2 <sup>c</sup>	0.50

<sup>a-d</sup>Values with unlike superscripts differ by  $P < 0.05$ .

**Key Words:** microbially treated, rumen degradability

**T419 Meta-analysis of feeding trials to estimate energy requirements of dairy cows under tropical conditions.** André S. Oliveira\*, Universidade Federal de Mato Grosso, Campus Sinop, Sinop, Mato Grosso, Brazil.

A meta-analysis of feeding trials was conducted to determine the metabolizable energy requirements for the maintenance ( $\text{ME}_m$ ) and efficiency of metabolizable energy intake (MEI) for the milk production ( $k_L$ ) of *Bos taurus* and *Bos taurus* × *Bos indicus* crossbreds dairy cows in the tropics. The database contained 236 means treatments from 60 feeding trial studies. The data set was classified by genotype: *Bos taurus* (37 studies; n = 150 treatments means;  $\text{MEI} = 45.9 \pm 6.6$  Mcal/d;  $\text{BW} = 570 \pm 45$  kg;  $\text{BW}$  variation =  $0.30 \pm 0.37$  kg/d;  $\text{BCS} = 2.7 \pm 0.2$ ; milk production =  $22.3 \pm 5.0$  kg/d; milk fat =  $3.64 \pm 0.37\%$ ; milk protein =  $3.23 \pm 0.25\%$ ; milk lactose =  $4.43 \pm 0.21\%$ ; and milk energy =  $0.70 \pm 0.04$  Mcal/kg) and *Bos taurus* × *Bos indicus* crossbred (23 studies; n = 86 treatments means;  $\text{MEI} = 31.6 \pm 7.26$  Mcal/d;  $\text{BW} = 487 \pm 48$  kg;  $\text{BW}$  variation =  $0.07 \pm 0.52$  kg/d;  $\text{BCS} = 3.2 \pm 0.5$ ; milk production =  $13.7 \pm 2.0$  kg/d; milk fat =  $3.63 \pm 0.55\%$ ; milk protein =  $3.11 \pm 0.28\%$ ; milk lactose =  $4.49 \pm 0.20\%$ ; and milk energy =  $0.70 \pm 0.06$  Mcal/kg). A linear regression of MEI (Mcal/kg  $\text{BW}^{0.75}/\text{d}$ ) with the milk energy output adjusted to a zero energy balance ( $E_{L(0)}$ , Mcal/kg  $\text{BW}^{0.75}/\text{d}$ ) was fitted using a mixed model with the study as the random effect and the genotype as the fixed effect. The  $\text{ME}_m$  was calculated as the intercept and  $k_L$  as the reciprocal of the slope of the regression. The dairy cow genotype affected the  $\text{ME}_m$  ( $P = 0.013$ ) and  $k_L$  ( $P = 0.033$ ). *Bos taurus* MEI (Mcal/kg  $\text{BW}^{0.75}/\text{d}$ ) =  $0.180 (\pm 0.022, P < 0.001) + 1.546 (\pm 0.153, P < 0.001) \times E_{L(0)}$ ,  $R^2 = 0.675$ ,  $\text{MSPE} = 0.0007$ ; and *Bos taurus* × *Bos indicus* MEI (Mcal/kg  $\text{BW}^{0.75}/\text{d}$ ) =  $0.147 (\pm 0.024, P < 0.001) + 1.771 (\pm 0.288, P < 0.001) \times E_{L(0)}$ ,  $R^2 = 0.859$ ,  $\text{MSPE} = 0.0004$ . *Bos taurus* × *Bos indicus* crossbred dairy cows have ( $P < 0.05$ ) an  $\text{ME}_m$  requirement that is 18% lower ( $0.147 \pm 0.024$  vs.  $0.180 \pm 0.022$  Mcal/kg  $\text{BW}^{0.75}/\text{d}$ ), but a  $k_L$  that is 12% lower ( $0.57 \pm 0.09$  vs.  $0.65 \pm 0.06$ ) than that of *Bos taurus* cows. These results support the hypothesis that *Bos taurus* × *Bos indicus* crossbred dairy cows have a lower  $\text{ME}_m$  requirement but lower energetic efficiency for milk production than do *Bos taurus* dairy cows under tropical conditions.

**Key Words:** *Bos indicus*, maintenance, metabolizable energy

**T420 Effect of a ration change from a TMR to a pasture-based ration on rumen bacteria in dairy cows.** Melanie Schären\*<sup>1</sup>, Ulrich Meyer<sup>1</sup>, Sven Dänicke<sup>1</sup>, and Gerhard Breves<sup>2</sup>, <sup>1</sup>Friedrich Loeffler Institut (FLI)-Federal Research Institute for Animal Health - Institute for Animal Nutrition, Braunschweig, Germany, <sup>2</sup>University of Veterinary Medicine Hannover, Foundation - Institute for Physiology, Hannover, Germany.

In spring, dairy cows are often gradually transitioned from a TMR to a pasture-based ration. This study aims to investigate how the populations of the liquid (LAB), particle (PAB) and epithelium (EAB) associated bacteria in the rumen are influenced by this nutritional change. A 10-wk trial (w1–10) was performed in spring 2014, including 10 rumen-fistulated dairy cows of the German Holstein breed (182 ± 24 d in milk, 23.5 ± 3.5 kg milk/day, means ± SD). The cows were divided into a pasture- and a confinement group (PG and CG, n = 5). The CG stayed on a TMR-based ration (35% corn silage, 35% grass silage, 30% concentrate; DM basis), while the PG was gradually transitioned from a TMR- to a pasture-based ration (w1: TMR-only, w2: 3 h/day on pasture, w3 and 4: 12 h/day on pasture, w5–10: pasture-only).

In w1, w5 and w10 of the trial samples of solid and liquid rumen contents were taken and after emptying the rumen, papillae biopsies were taken from the ventral rumen sac. The bacterial DNA was isolated from each sample and a SSCP-analysis was performed. Similarity and cluster analysis was done using GelComparII. Data were analyzed as repeated measures using PROC MIXED of SAS Enterprise Guide 6.1. Cluster analysis displayed a clustering of LAB, PAB and EAB in both ration types and all three points in time. LAB and PAB had a similarity of 82 ± 5% (means ± SD) and differ strongly from the EAB with a similarity of 39 ± 8% EAB compared with LAB, and 37 ± 8% EAB compared with PAB (p < 0.001). When comparing the samples of the different bacteria populations in w5 and w10 to their reference sample in w1, a significant greater decrease in similarity over time in all three bacteria populations was observed for the PG compared with the CG (p < 0.01; illustrated in Table 1). To further identify the different bacterial populations 16S DNA sequencing of all samples is currently being performed.

**Table 1 (Abstr. T420).** Change in bacterial populations of the LAB, PAB, and EAB over time expressed in average similarity (in %) of samples compared with their reference sample in week 1 (means ± SD)

	LAB CG	PAB CG	EAB CG	LAB PG	PAB PG	EAB PG
Week 5	93 ± 6%	97 ± 2%	91 ± 4%	76 ± 10%	86 ± 8%	82 ± 8%
Week 10	91 ± 3%	94 ± 2%	88 ± 4%	61 ± 13%	85 ± 8%	67 ± 8%

**Key Words:** dairy cow, pasture, rumen microbiota

**T421 Morphology change and expression of genes related to tight junctions, cytokines, proliferation and apoptosis in the rumen of lactating dairy cows fed corn stover or rice straw replacing alfalfa as forage source.** Bing Wang<sup>\*1,3</sup>, Mei Liu<sup>1,3</sup>, X. B. Huang<sup>1,3</sup>, J. S. Wu<sup>2</sup>, and J. X. Liu<sup>1,3</sup>, <sup>1</sup>Institute of Dairy Science, College of Animal Sciences, Zhejiang University, Hangzhou, China, <sup>2</sup>Department of Veterinary Science, College of Animal Sciences, Zhejiang University, Hangzhou, China, <sup>3</sup>MOE Key Laboratory of Molecular Animal Nutrition, Zhejiang University, Hangzhou, China.

The objective of this study was to characterize the morphology and expression of the genes related to tight junction protein, cytokines, proliferation, and apoptosis in the gastrointestinal tracts (GIT) of lactating cows. Eighteen multiparous Holstein dairy cows were individually fed, and randomly assigned into 1 of 3 treatments. Isonitrogenous diets with forage-to-concentrate ratio at 45:55 contained similar concentrate and 15% corn silage, with 3 forage sources (DM basis): 23% alfalfa hay and 7% Chinese wild rye hay (AH); 30% corn stover (CS); and 30% rice straw (RS). The portion with length above 19 mm was the largest in RS, followed by AH, least in CS. After 14-wk feeding, all the cows were slaughtered. The digesta, tissue, and relevant epithelium of the GIT were collected immediately after dissection. The morphology of the GIT

tissues was analyzed according to the hematoxylin-eosin stained method and the histomorphometric analysis was performed using Image J Plus software (Media Cybernetics, Bethesda, MD). The qRT-PCR reaction of the related genes in rumen epithelium was performed using the 2 × SYBR Premix Ex Taq (Tli RNaseH Plus) kit (Takara, Otsu, Japan). The variance of the data was analyzed as a completely randomized design using PROC MIXED of SAS. The DM of rumen digesta was heavier in diet RS than in AH (P < 0.01). The thickness of ruminal mucosa was greater in RS than in AH (P < 0.05) or CS (P < 0.05), but the thickness of mucous epithelium was greater in AH (P < 0.05) or RS (P < 0.05) than in CS. The rumen butyrate concentration was greater in diet CS than RS (P < 0.01) or AH (P < 0.01), and had a negative correlation with the mRNA abundance of BCD (P = 0.01). Abundance of mRNA of IGF-1R was upregulated in AH compared with CS (P = 0.047). Abundance of mRNA of claudin-1 (P = 0.09), claudin-4 (P = 0.10), and ZO-1 (P = 0.07) tended to increase in AH compared with CS. The expression of gene related to cytokines showed no difference among the treatments. The expression of IGF-1 tended to increase in AH (P = 0.066) compared with CS. Our finding suggested that feeding of rice straw to dairy cow caused the change in ruminal mucosa mostly related to cell proliferation and apoptosis, which may be not only attributed to the physically effective fiber effects but also the butyrate factor.

**Key Words:** cow, gastrointestinal tract, morphology

**T422 Effect of a calcium oral bolus administered after calving on the metabolic parameters of dairy cows.** J. M. Béguin<sup>\*1</sup>, R. P. Dagorne<sup>1</sup>, and P. Courty<sup>2</sup>, <sup>1</sup>Néolait, Yffiniac, France, <sup>2</sup>ESITPA, Mont-Saint-Aignan, France.

The objective of this study was to evaluate the effect of postpartum administration of a calcium bolus (Dietevit Flash, Néolait, Yffiniac, France), on the metabolic parameters of multiparous dairy cows. Holstein cows (n = 38) from 3 commercial dairy farms in Western France, were randomly assigned to a control group (group A) and a trial group (group B). Cows in group B received at calving 2 boluses and 2 boluses at 12–18 h after calving. The 4 boluses supplied calcium (65 g as calcium formiate and calcium propionate), magnesium (5 g), vitamin E (4,000 IU), vitamin D3 (50,000 IU) and niacin (12 g). Blood samples were taken 12 d before calving, 12 and 28 h after calving and 10 d after calving. Blood serum was then analyzed for calcium, magnesium and vitamin E at the Frank Duncombe's Lab, France. The metabolic profiles of the 2 groups were compared using repeated measures ANOVA (SPSS v18). The incidence of cows with postpartum hypocalcemia (calcium < 80mg/L) in both groups was compared using the Pearson Chi-squared test. Compared with group A, cows in group B had higher calcemia at 12 h postpartum (87.3 vs. 79.1 mg/L, P = 0.02) and 28 h postpartum (86.3 vs. 76.4 mg/L, P = 0.01), lower magnesemia at 28 h postpartum (19.0 vs. 22.1 mg/L, P = 0.03) and higher blood levels of vitamin E at 28 h postpartum (2.2 vs. 1.4 µg/L, P = 0.04). Prevalence of hypocalcaemia was significantly decreased in group B (7.1% vs. 37.5 and 25% vs. 56.2%, P = 0.01) at respectively 12 and 28 h postpartum. In conclusion, the Dietevit Flash bolus significantly improves calcemia in postpartum multiparous dairy cows. The Dietevit Flash helps secure the difficult peripartum phase by decreasing the prevalence of subclinical hypocalcaemia in a significant way during the first 2 d of lactation.

**Key Words:** dairy cow, bolus, subclinical hypocalcemia

**T423 Urea kinetics in dairy cows fed soybean meal (SBM), canola meal (CM), corn high protein dried distillers grains (HPDDG) or wheat dried distillers grains with solubles (WDDGS).** D. R. Ouellet<sup>\*1</sup>, G. Maxin<sup>2</sup>, and H. Lapierre<sup>1</sup>, <sup>1</sup>Dairy and Swine R&D Centre, Agriculture and Agri-Food Canada, Sherbrooke, QC, Canada, <sup>2</sup>INRA UMR 1213 Herbivores, Saint-Genès-Champagne, France.

Inclusion of CM in dairy rations decreases plasma urea concentration (Martineau et al., J. Dairy Sci. 97:1603). The objective of this study was to compare the effects of feeding diets containing SBM, CM, HPDDG or WDDGS as single protein supplement on urea kinetics. Eight rumen-fistulated Holstein cows were used in a replicated 4 × 4 Latin square with 14-d periods. Dietary treatments consisted of inclusion of these protein in diet: SBM (13.7%), CM (20.8%), HPDDG (20.4%) or WDDGS (22.8%). The 4 diets were isonitrogenous (17.2%CP) and isoenergetic (1.56 Mcal NE<sub>L</sub>/kg DM; NRC 2001), with a fixed forage:concentrate ratio of 62:38. From d 10–13, total collection of urine was performed. On d 12, cows were infused in a jugular vein <sup>15</sup>N<sub>2</sub>-urea (0.5 mmol/h) and urine spot samples were collected after 24 h of infusion; enrichment of <sup>15</sup>N<sub>2</sub>-urea was determined to estimate urea kinetics. Differences among treatments were analyzed using adjusted Tukey-Kramer multiple comparisons test. Results are given as LSM ± SEM for SBM, CM, HPDDG and WDDGS, respectively. The N intake and milk N were similar between treatments (659, 660, 657, and 665 ± 23 g/d and 1.13, 1.15, 1.15, and 1.13 ± 0.01 kg/d; Maxin et al., JDS 96:7806). The RDP supply (NRC 2001) averaged 2622, 2672, 2110 and 2959 g/d. Urea production (UER) tended (*P* = 0.08) to be lower for WDDGS vs. SBM (353 vs. 409 ± 18 g urea-N/d) whereas CM (391) and HPDDG (405) were intermediate. Urea gut entry rate (GER) averaged 265, 272, 267 and 210 ± 18 g urea-N/d, with only WDDGS being lower (*P* < 0.05) than the other diets. The proportion of GER/UER was also lower (*P* < 0.05) for WDDGS: 0.65, 0.69, 0.65, and 0.59 ± 0.02. Urinary N excretion was lower (*P* < 0.06) for CM (118 ± 6 g urea-N/d) than HPDDG (139), WDDGS (144) and SBM (144). Cows fed WDDGS had a lower urea production and GER, but urinary excretion was similar to HPDDG and SBM, in disagreement with estimated higher RDP supply. The intermediate urea production for the CM supplement coupled with the numerically higher proportion returned to the gut lead to a lower urinary urea-N excretion which can explain the decreased plasma urea concentration also observed in this study: 8.5, 7.8, 8.8, and 8.9 ± 0.4 mM (Maxin et al., J. Dairy Sci. 96:7806).

**Key Words:** urea kinetics, protein supplement, dairy cow

**T424 Determining bioavailability of Lys in rumen-protected Lys products using the plasma free AA dose-response technique.** Nancy L. Whitehouse<sup>\*1</sup>, Andre B. D. Pereira<sup>1</sup>, Adam C. Crowther<sup>1</sup>, Devan L. Chirgwin<sup>1</sup>, Andre F. Brito<sup>1</sup>, Charles G. Schwab<sup>2</sup>, and Jack E. Garrett<sup>3</sup>, <sup>1</sup>University of New Hampshire, Durham, NH, <sup>2</sup>Schwab Consulting, LLC, Boscobel, WI, <sup>3</sup>QualiTech Inc, Chaska, MN.

Two experiments were conducted using the technique to determine the bioavailability of Lys in rumen-protected (RP)Lys supplements manufactured by QualiTech Inc. Bioavailability estimates are calculated by dividing the slope of the regression line relating changes in plasma Lys concentrations from feeding an RPLys supplement by the slope of the regression line obtained by continuous abomasal infusion of Lys. Experiment 1 examined the efficacy of the company's first generation product. Experiment 2 was to determine the effect of changes in core/matrix composition and amount of encapsulation on Lys availability. Six multiparous ruminally-cannulated Holstein cows were used in Exp. 1 in a replicated 3 × 3 Latin square (193 DIM). Treatments were 1) 0 g/d Lys, 2) 60 g/d infused Lys from Lys-HCl (80% Lys), and 3) 60 g/d fed Lys

from RPLys (56% Lys). Five multiparous ruminally-cannulated cows were used in Exp. 2 in a 5 × 5 Latin square (108 DIM). Treatments were 1) 0 g/d Lys, 2) 60 g/d infused Lys from Lys-HCl (80% Lys), 3) 60 g/d fed Lys from product A (30.4% Lys), 4) 60 g/d fed Lys from product B (47.4% Lys), and 5) 60 g/d fed Lys from product C (37.3% Lys). Before feeding, the products were mixed with 2 kg of the Lys-adequate basal TMR, placed in ziploc bags, and stored for 8 h at 4°C. Fed treatments were offered to cows 30 min before each of 3 daily feedings. Fed treatments not consumed after 15 min were placed in the rumen. Tail vein samples were collected 2, 4, 6 and 8 h after the morning feeding the last 3 d of each period. Deproteinized plasma samples were composited by cow/d and stored (–80°C) until AA analysis. Lysine concentrations (μM) were expressed as a proportion of total AA before regression analysis. Data were analyzed using the PROC MIXED and REG procedures of SAS. Lysine was the only plasma AA that increased linearly in response to treatments (*P* < 0.02). Calculated Lys bioavailability estimates were 27.1% in Exp. 1, and 17.0, 24.5, and 28.5%, respectively, for Products A, B, and C in Exp. 2. The technique was sensitive enough to confirm differences between Product A vs. products B and C in Exp. 2.

**Key Words:** amino acid, lysine, bioavailability

**T425 Blood ketone bodies incidence and concentration from intensively housed early-lactation dairy cows in Brazil.** Rafahel C. Souza<sup>1</sup>, Rogério C. Souza<sup>1</sup>, Breno M. Sousa<sup>2</sup>, and Andre B. D. Pereira<sup>\*3</sup>, <sup>1</sup>Pontifícia Universidade Católica de Minas Gerais, Betim, MG, Brazil, <sup>2</sup>Centro Universitário UniBH, Belo Horizonte, MG, Brazil, <sup>3</sup>University of New Hampshire, Durham, NH.

Ketosis is a metabolic syndrome that can increase farm costs and result in milk production losses during a lactation. This syndrome mostly affects high production cows and causes are usually associated with negative energy balance during early to mid-lactation. The objective of this study was to evaluate the concentration of ketone bodies in the blood of 732 early-lactation [25.2 ± 15.8 d in milk (DIM)] high producing Holstein cows (32.98 kg/d of milk yield) from 10 intensively managed farms in Brazil. A portable electronic diagnosis equipment (Ketovet, TaiDoc technology, Taiwan, China) was used for measurement of blood ketone bodies and results are expressed in mM. Blood was sampled from the tail's artery or vein, and the volume of 1 drop was added to a reagent strip set for β-hydroxybutyrate (BHBA) analysis and 5 s was the average time needed for the equipment to yield results. Animals with values of blood BHBA of less than 1.1 mM were considered normal and free of metabolic syndrome. Animals with values between 1.2 and 3.5 mM were diagnosed with subclinical ketosis, and animals with values above 3.5 were diagnosed with clinical ketosis. Animals were blocked by days in milk (0–15, 16–30, 31–45, >46) and farm was used as a random effect. Data were analyzed using a t student test of least square difference and significance was declared as *P* < 0.05. From the 732 animals evaluated, 448 (47.85%) were considered free from the syndrome, 279 (38.11%) had subclinical ketosis and 5 (0.68%) had clinical levels of BHBA. Similar BHBA levels were found for cows between 0 and 15 DIM (1.07 mM), between 16 and 30 DIM (1.15 mM) and between 31 and 45 DIM (1.06 mM), but were significantly higher for cows with more than 46 DIM (1.34 mM, SEM = 1.34, *P* < 0.01). Results of this experiment suggest that cows with more than 46 DIM had subclinical ketosis and values of BHBA that were higher than for all cows with less than 45 DIM.

**Key Words:** transition period, metabolic syndrome, ketone bodies

**T426 Evaluation of camelina meal as a feedstuff for growing dairy heifers.** Rhea D. Lawrence\* and Jill L. Anderson, *Dairy Science Department, South Dakota State University, Brookings, SD.*

Our objective was to evaluate growth performance of dairy heifers fed camelina meal (CM) compared with linseed meal (LN) or reduced-fat distillers dried grains with solubles (DG). A 12-wk randomized complete block design study was conducted using 33 Holstein and 9 Brown Swiss heifers (144.8 ± 22 d of age; body weight (BW) 171.8 ± 24.3 kg) with 3 treatments. Treatments were 10% of the diet as CM, LN, or DG (DM basis). Concentrate mixes also included corn and soybean meal, at slightly different inclusion rates to make diets isocaloric and isonitrogenous. All diets contained 60% grass hay and 40% concentrate mix. Diets were limit-fed to 2.65% of BW (DM basis) using a Calan gate feeding system. Frame sizes, BW, and body condition scores (BCS) were measured on 2 d during wk 0, 2, 4, 6, 8, 10, and 12. Data were analyzed using MIXED procedures with repeated measures and ADG was found using REGRESSION procedures of SAS 9.4. Heifer DMI was similar between CM and LN, but greater ( $P = 0.03$ ) for DG (4.83, 4.82, and 5.03 kg/d; SEM = 0.17 for CM, LN, and DG, respectively). Body weights (199.5, 210.9, and 205.1 kg; SEM = 2.86) were found to be less ( $P < 0.01$ ) for heifers fed CM and greatest for LN, which could be due to unequal initial BW. Treatments had similar ( $P > 0.05$ ) ADG (0.67, 0.75, 0.71 kg/d; SE = 0.15). Gain to feed was similar for CM and DG, but greater ( $P < 0.05$ ) for LN (0.14, 0.17, 0.15 kg/d; SEM = 0.006). Hip height (116.3, 115.8, and 115.4 cm; SEM = 0.67), withers height (112.3, 112.3, and 112.4 cm; SEM = 0.47), and heart girth (128.6, 128.3, and 128.9 cm; SEM = 0.81) were similar ( $P > 0.05$ ) among treatments. Hip width (31.7, 31.3, and 31.4 cm; SEM = 0.27) and body length (105.2, 102.0, and 103.3 cm; SEM = 0.77) were greater ( $P < 0.05$ ) for CM compared with LN and DG. Body condition score (3.16, 3.10, and 3.17; SEM = 0.02) was greater ( $P < 0.01$ ) for CM and DG compared with LN. Feeding CM maintained frame growth and ADG but decreased gain to feed compared with DG and LN. The decreased feed efficiency may be related to glucosinolates in the CM. Further research should be conducted before implementing CM as a primary protein source for growing heifers.

**Key Words:** camelina meal, dairy heifer

**T427 Evaluation of the incidence of subclinical ketosis for F<sub>1</sub> Gir x Holstein lactating dairy cows supplemented with medium-chain fatty acids.** Rafahel C. Souza<sup>1</sup>, Rogério C. Souza<sup>1</sup>, Vanessa A. Teixeira<sup>1</sup>, Joaquim H. C. M. Souza Junior<sup>1</sup>, Igor C. Leal<sup>1</sup>, Andre B. D. Pereira<sup>\*2</sup>, and Maria I. V. Melo<sup>1</sup>, <sup>1</sup>*Pontifícia Universidade Católica de Minas Gerais, Betim, MG, Brazil*, <sup>2</sup>*University of New Hampshire, Durham, NH.*

The use of medium-chain fatty acids (MCFA) during the periparturient period can alleviate the effects of negative energy balance and improve animal health in early lactation dairy cows. The objective of this study was to evaluate blood  $\beta$ -hydroxybutyrate (BHBA) of 30 early-lactation F<sub>1</sub> Gir x Holstein cows supplemented with MCFA. Diets were formulated according to the NRC (2001) model and contained 50% forage as corn silage and chopped sugar cane. Concentrate was based on ground corn, soybean meal, citrus pulp and minerals in 2 treatments: (1) no addition of MCFA (0FA); and (2) addition of 40 g of MCFA [40FA, 0.186% of dry matter as toplac rumacel (Nutrifarma, Taio, PR, Brazil)]. The experiment was a completely randomized design and cows were supplemented with MCFA during the periparturient period (from 21 d before parturition until 21 d after). A portable electronic diagnosis equipment (Ketovet, TaiDoc technology, Taiwan, China) was used for measurement of BHBA and results are expressed in mM. Blood was

sampled from the tail artery or vein, and the volume of 1 drop was added to a reagent strip set for BHBA analysis. Samples were collected and analyzed at the day of parturition and then again after 42 and 70 d in milk. Animals with values above 1.2 mM were diagnosed with subclinical ketosis. Data were analyzed using the MIXED procedure of SAS and significance was declared at  $P < 0.05$ . At the day of parturition, 73.4% of cows in 0FA (11 out of 15) had subclinical ketosis while only 6.6% (1 out of 15 cows) of cows on 40FA had subclinical ketosis ( $P < 0.05$ ). The proportion of cows with subclinical ketosis was reduced to 40% for 0FA and slightly increased to 13.4% for 40FA after 42 d in milk and this proportion remained for after 70 d in milk, being significantly different between treatments ( $P < 0.05$ ). Results of this study suggests that F<sub>1</sub> Gir x Holstein cows receiving diets based on corn silage and chopped sugar cane supplemented with MCFA may have lower incidence of subclinical ketosis when compared with non-supplemented cows.

**Key Words:** dairy cow, medium-chain fatty acids, subclinical ketosis.

**T428 Milk production and composition of F<sub>1</sub> Gir x Holstein lactating cows supplemented with medium-chain fatty acids during the periparturient period.** Rafahel C. Souza<sup>1</sup>, Rogério C. Souza<sup>1</sup>, Vanessa A. Teixeira<sup>1</sup>, Igor G. Leal<sup>1</sup>, Joaquim H. C. M. Souza Junior<sup>1</sup>, Andre B. D. Pereira<sup>\*2</sup>, and Maria I. V. Melo<sup>1</sup>, <sup>1</sup>*Pontifícia Universidade Católica de Minas Gerais, Betim, MG, Brazil*, <sup>2</sup>*University of New Hampshire, Durham, NH.*

Subclinical ketosis is usually associated with negative energy balance in early lactation. The use of medium-chain fatty acids (MCFA) during the periparturient period can alleviate the negative energy balance and improve animal health. The objective of this study was to evaluate 30 F<sub>1</sub> Gir x Holstein lactating dairy cows for milk production and composition when supplemented with MCFA during the periparturient period. Diets were formulated according to the NRC (2001) model and contained 50% forage as corn silage and chopped sugar cane. Concentrate was based on ground corn, soybean meal, citrus pulp and minerals in 2 treatments: (1) No addition of MCFA (0FA) and (2) Addition of 40 g of MCFA (40FA, 0.186% of DM) as Toplac Rumacel (Nutrifarma, Taio, PR, Brazil). The experiment was a completely randomized design and cows were supplemented with MCFA during the periparturient period (from 21 d before parturition until 21 d after). Analyses of milk yield and composition were done weekly and collections were made on d 7, 14, 21, 28, 35, 42, 49, 56, 63 and 70 after parturition for each cow. Data were analyzed using the MIXED procedure of SAS and means were compared using the Tukey adjustment with significance declared as  $P < 0.05$ . Milk yield was higher for cows on 40FA (26.97 kg/d) when compared with cows on 0FA (25.50 kg/d,  $P < 0.05$ ), but this trend only happened from 35 d after parturition until 70 d. Milk fat and protein yields were not different between treatments and days sampled. Somatic cell count was lower (64,260 cells/mL) for cows on treatment 40FA compared with 0FA (98,800 cells/mL,  $P < 0.05$ ) suggesting that MCFA can improve mammary health. Results of this study suggest that supplementation of MCFA improved milk production and composition of F<sub>1</sub> Gir x Holstein cows when compared with non-supplemented cows.

**Key Words:** dairy cow, milk production and composition, medium-chain fatty acids

**T429 Effects of casein, glucose, and acetate infused into the abomasum of feed-restricted cows on milk and milk components yield.** Marina A. C. Danes<sup>\*1</sup>, Michel A. Wattiaux<sup>1</sup>, and Glen A. Broderick<sup>2</sup>, <sup>1</sup>*University of Wisconsin-Madison, Madison, WI*, <sup>2</sup>*Broderick Nutrition & Research LLC.*

Amino acids and glucose have been shown to regulate protein synthesis in the mammary gland through cell signaling. Acetate might also have an effect through AKT pathway, since it is the main energy source for the mammary secretory cell. Six Holstein multiparous cows, averaging 55 kg milk/d, were used in a 6x6 Latin square with 14-d periods. In each period, cows were fed the same diet for 10 d, after which they were feed-restricted for 4 d to 85% of ad libitum intake and abomasally infused for 1 of 6 treatments: acetate (A), glucose (G), each at 5% of ad libitum metabolizable energy intake (Mcal/d), casein (C) at 15% of ad libitum metabolizable protein intake (g/d), A+C (AC), G+C (GC), or a saline solution (S, negative control). Data from d 13 and 14 of each period were analyzed with Proc Mixed of SAS, using data from the d 9 and 10 as covariate. LS means are reported in the table. Casein by itself had no effect on any of the variables, compared with the negative control. This could be a consequence of lack of energy due to feed restriction, since glucose infused with casein numerically increased milk protein yield by 121 g/d. Glucose increased milk yield and nitrogen use efficiency, as indicated by the lowest MUN. Acetate had no effect on any of the variables, rejecting our hypothesis that it would enhance milk protein synthesis by improving energy status of the cell. Instead, acetate was used for fat synthesis as shown by the numerically greater fat yield for the acetate treatments. Interestingly, the infusion of casein along with acetate increased milk fat secretion more than acetate alone, which might be related to the effect of specific AA on fat synthesizing enzymes recently reported in the literature.

**Table 1 (Abstr. T429).**

Item	A	AC	C	G	GC	S	SEM	P-value
Milk, kg/d	37.2 <sup>B</sup>	39.2 <sup>AB</sup>	38.4 <sup>AB</sup>	42.2 <sup>A</sup>	41.3 <sup>AB</sup>	37.1 <sup>B</sup>	1.8	<0.10
Lactose, kg/d	1.67 <sup>AB</sup>	1.79 <sup>AB</sup>	1.71 <sup>AB</sup>	1.89 <sup>A</sup>	1.84 <sup>AB</sup>	1.65 <sup>B</sup>	0.08	<0.10
Fat, kg/d	1.59	1.7	1.43	1.54	1.44	1.44	0.12	NS
Protein, kg/d	1.13 <sup>B</sup>	1.19 <sup>AB</sup>	1.19 <sup>AB</sup>	1.27 <sup>AB</sup>	1.31 <sup>A</sup>	1.15 <sup>AB</sup>	0.07	0.11
MUN, mg/dL	11.4 <sup>bc</sup>	15.1 <sup>a</sup>	12.2 <sup>bc</sup>	10.4 <sup>c</sup>	11.2 <sup>bc</sup>	12.4 <sup>b</sup>	0.8	<0.01

Means followed by different lowercase (uppercase) letters differ at  $P < 0.10$  ( $P < 0.15$ ).

**Key Words:** milk protein synthesis, mammary metabolism

**T430 The effect of pelletized corn stover replacing alfalfa hay on lactation performance, blood parameters and rumen fermentation in mid-lactation dairy cows.** H. Z. Sun<sup>\*1,2</sup>, Z. H. Wei<sup>1,2</sup>, Wen Zhu<sup>1,2</sup>, X. Xie<sup>1,2</sup>, J. K. Wang<sup>1,2</sup>, L. L. Guan<sup>3</sup>, and J. X. Liu<sup>1,2</sup>, <sup>1</sup>*Institute of Dairy Science, College of Animal Sciences, Zhejiang University, Hangzhou, China*, <sup>2</sup>*MoE Key Laboratory of Molecular Animal Nutrition, Zhejiang University, Hangzhou, China*, <sup>3</sup>*Department of Agricultural, Food & Nutritional Science, University of Alberta, Edmonton, Canada*.

This study was conducted to investigate the effect of pelletized corn stover replacing alfalfa hay on lactation performance, blood parameters and rumen fermentation dairy cows. Twenty multiparous Holstein dairy cows were blocked based on days in milk ( $219 \pm 49.5$  d, mean  $\pm$  SD) and milk yield ( $25.4 \pm 4.03$  kg, mean  $\pm$  SD), and were randomly assigned into one of 2 dietary treatments with similar concentrate mixture, but different forage ingredients (as DM basis): 15% of pelletized corn stover pellets (CS) or 12.2% of alfalfa hay (AH). Two diets had similar content of net energy for lactation (1.57 vs. 1.58 Mcal/kg DM), but differed in crude protein content (15.8 vs. 16.7 for CS and AH). After feeding for 3 weeks, the rumen fluid and blood samples were collected. The data were analyzed using MIXED procedure of SAS software. The DM intake of diet did not differ ( $P = 0.72$ ) between 2 diets, with an average of 16.5

kg/d. Milk yield in AH-fed cows was 0.87 kg/d higher than the CS-fed, though the difference was not significant ( $P = 0.53$ ). No difference was observed between 2 diets in milk protein (3.37 vs. 3.38%,  $P = 0.97$ ), milk fat (4.15 vs. 4.25%,  $P = 0.76$ ), lactose (4.78 vs. 4.76%,  $P = 0.83$ ), total solid (13.7 vs. 12.9%,  $P = 0.16$ ), milk urea nitrogen (16.0 vs. 13.9%,  $P = 0.19$ ), and somatic cell counts ( $206$  vs.  $325 \times 10^3$ /mL,  $P = 0.39$ ). The concentration of plasma glucose was significantly lower in the CS-fed cows than in AH-fed animals (1.82 vs. 2.23 mM,  $P = 0.02$ ), whereas the other blood parameters (NEFA, BHBA, Ca and P) were not different ( $P > 0.05$ ). The concentrations of ruminal total and individual volatile fatty acids showed no difference between 2 groups. From the above results, it is indicated that the pelletized corn stover can be used to replace alfalfa hay in the diet for mid-lactation dairy cows without adverse effects on lactation performance and rumen fermentation.

**Key Words:** pelletized corn stover, alfalfa hay, dairy cow

**T431 Supplementation with artificial sweetener improves milk yield and composition and alters nutrient partitioning in lactating dairy cows.** Emma H. Wall<sup>\*1,2</sup> and David M. Bravo<sup>2</sup>, <sup>1</sup>*Pancosma, Geneva, Switzerland*, <sup>2</sup>*InVivo Animal Nutrition & Health, Saint-Nolff, France*.

Supplementation with Sucram (SUC, Pancosma, Geneva, Switzerland) increases intestinal expression of sodium-glucose co-transporter 1, glucose uptake, and mucosal growth in ruminants. This trial aimed to determine the productive implications of this response in dairy cows. Primi- and multiparous lactating Holstein dairy cows were housed together in a freestall pen and were milked using an automated milking system (AMS). For 8 weeks, SUC was blended with a carrier and was dispensed at the AMS for SUC cows ( $n = 91$ ) at a rate of 2.72 kg/d (SUC dose: 2g/cow/d); control cows received no additive ( $n = 89$ ). Individual cow milk production and composition, milking frequency, and BW were recorded daily. Average DMI of the pen was monitored daily and did not change throughout the study. A treatment by stage of lactation interaction was observed: in cows that were greater than 100 DIM, there was no effect of SUC on any variables measured ( $P > 0.20$ ). In contrast in cows less than 100 DIM, SUC enhanced milk production (44.5 vs. 45.6 kg/d;  $P \leq 0.10$ ). Milk fat (1.62 vs. 1.71 kg/d) and protein (1.41 vs. 1.46 kg/d) yields were also improved with SUC ( $P < 0.02$ ); therefore, energy-corrected milk production was greater for SUC cows (46.0 vs. 48.0 kg/d;  $P < 0.01$ ). There was a parity by treatment interaction for BW such that primiparous cows maintained condition regardless of treatment ( $P > 0.50$ ) whereas multiparous cows supplemented with SUC lost BW during the trial ( $P < 0.001$ ). Interestingly, there was also a parity by treatment interaction for number of daily milkings, which was increased by SUC in primiparous animals only ( $P < 0.001$ ). This indicates that the increase in milk yield in those animals may have been due to greater frequency of milk removal whereas in multiparous cows it was due to mobilization of body reserves. These findings reveal that in early to mid-lactation, supplementation with SUC improves milk production performance and optimizes nutrient partitioning.

**Key Words:** automated milking system, feed additive, Sucram

**T432 Effect of fish oil on transportation of fatty acids in plasma lipoproteins of lactating and nonlactating cows.** Einar Vargas-Bello-Pérez<sup>\*1</sup>, Gonzalo Íñiguez-González<sup>1</sup>, Philip C. Garnsworthy<sup>2</sup>, and Juan J. Lóor<sup>3</sup>, <sup>1</sup>*Pontificia Universidad Católica de Chile, Santiago, Chile*, <sup>2</sup>*The University of Nottingham, Loughborough, UK*, <sup>3</sup>*University of Illinois, Urbana, IL*.

The aim of this study was to elucidate the effect of dietary fish oil (FO) and a blend of FO and hydrogenated palm oil (PO) on transportation of fatty acids (FA) in plasma lipoproteins of lactating and nonlactating cows. Three lactating and 3 nonlactating Holstein cows (fitted with rumen cannulas) were used in 2 different 3 × 3 Latin square designs that included 3 periods of 21 d. Dietary treatments for lactating cows consisted of a basal diet (Control; no fat supplement), and fat-supplemented diets containing FO (500 g/d/cow) and FOPO (250 FO + 250 PO g/d/cow hydrogenated palm oil). For nonlactating cows dietary treatments consisted of a basal diet (Control; no fat supplement), and fat-supplemented diets containing FO (170 g/d/cow) and FOPO (85 FO + 85 PO g/d/cow). Compared with control and FOPO, FO increased plasma contents of C16:0, C16:1, C17:1, C18:2n6t, C18:2 c9t11 and C20:5n3. Nonlactating cows had higher plasma contents of C18:1t11, C18:1n9c, C20:3n6, C18:2n6c, C23:0 and C22:6n3. Only C18:2n6c was higher in lactating than nonlactating cows (37 vs. 26 g/100g). Compared with control and FOPO, FO increased HDL contents of C18:1t11, C18:2n6t, C18:2 c9t11 and C20:5n3, and increased LDL contents of C18:1t9, C18:1t11 and C20:5n3. Nonlactating cows had higher LDL contents of C18:1t9 and C18:1t11 (1.98 vs. 0.89 g/100g and 2.7 vs. 1.14 g/100g) than lactating cows. In plasma, nonlactating cows had higher contents of total SFA (46 vs. 43 g/100g), MUFA (16 vs. 11 g/100g) and C18:1 tFA (5 vs. 3 g/100g) and lower content of total PUFA (39 vs. 46 g/100g) than lactating cows. Total content of C18:1 tFA was higher in nonlactating cows than in lactating cows (7 vs. 3 g/100g). Overall, results demonstrate clear differences in plasma transport of FA, which depend on dietary FA source and lactation state.

**Key Words:** fish oil, plasma, lipoprotein

**T433 Development and evaluation of predictive models of intake for crossbred Holstein-Zebu dairy cows.** V. L. Souza<sup>\*1</sup>, T. Z. Albertini<sup>1</sup>, R. Almeida<sup>2</sup>, G. B. Mourão<sup>1</sup>, J. K. Drackley<sup>3</sup>, and D. P. D. Lanna<sup>1</sup>, <sup>1</sup>Esalq/USP, Piracicaba, SP, Brazil, <sup>2</sup>Universidade Federal do Paraná, Curitiba, PR, Brazil, <sup>3</sup>University of Illinois, Urbana, IL.

Equations to predict dry matter intake (DMI) of crossbred Holstein-Zebu dairy cows were developed and compared by using data of 161 treatment means from 38 Brazilian studies (n = 446 cows, milk production average = 16.60 ± 5.70 kg/d). A data set was developed of Holstein × Zebu lactating dairy cows of different degrees of breeding in confinement or pasture systems. The data were evaluated using mixed nonlinear models including study as a random effect. Body weight (BW), 4% fat corrected milk (4% FCM) and weeks of lactation (WOL) were used as independent variables in the model. The proposed model to estimate DMI (kg/d) of crossbred cows was:  $[0.5552 (\pm 0.06636) \times 4\% \text{ FCM} + 0.06332 (\pm 0.009455) \times \text{BW}^{0.75}] \times [1 - e^{(-0.7732 (\pm 0.7019) \times (\text{WOL} - 1.629 (\pm 1.913))}]$ . The accuracy of the model was compared with 4 previously published equations by use of an independent data set. The mean square error of prediction (MSEP), mean bias, concordance correlation coefficient (CCC), and analysis of linear regression were used for evaluating models. The new model showed the lowest values of MSEP and highest CCC and r<sup>2</sup> compared with the other 4 equations (Table 1). The Brazilian model from Freitas et al. (2006), despite being developed in tropical conditions, showed the highest value of the MSEP. The new equations to predict DMI can be used in the formulation of diets for crossbred dairy cows under tropical conditions.

**Table 1 (Abstr. T433).**

Variable	DMI (kg/d)	New model (2015)	NRC (2001)	Fox et al. (2004)	Traxler (1997)	Freitas et al. (2006)
Mean, kg	16.10	16.88	16.11	15.05	16.95	15.85
Mean bias, Y - X, kg	—	-0.78	-0.01	1.05	-0.85	0.25
MSEP, kg × kg	—	1.64	2.45	2.73	2.76	6.98
MSEP Decomposition, %						
Mean bias, %	—	37.001	0.003	40.458	26.338	0.863
Systematic bias, %	—	3.469	0.052	5.327	0.781	63.86
Random errors, %	—	59.531	99.945	54.215	72.882	35.227
CCC	—	0.90	0.82	0.80	0.80	0.75
r <sup>2</sup>	—	0.88	0.69	0.81	0.74	0.69
P-value (a = 0)	—	0.495	0.889	0.229	0.244	<0.001
P-value (b = 1)	—	0.125	0.884	0.049	0.506	<0.001

**Key Words:** crossbred dairy cow, intake, meta-analysis

**T434 Soil contamination in forages: Estimation and geographical distribution.** J. R. Knapp<sup>\*1</sup>, W. P. Weiss<sup>2</sup>, R. T. Ward<sup>3</sup>, and K. R. Perryman<sup>4</sup>, <sup>1</sup>Fox Hollow Consulting LLC, Columbus, OH, <sup>2</sup>Dept. of Animal Sciences, The Ohio State University, Wooster, OH, <sup>3</sup>Cumberland Valley Analytical Services, Hagerstown, MD, <sup>4</sup>Micronutrients Inc., Indianapolis, IN.

Titanium (Ti) concentration in forages is considered by agronomists to be the gold standard in determining soil contamination of herbaceous materials. However, Ti is not measured in routine nutrient analysis of feed ingredients. Soil contamination of forages reduces the concentration of all nutrients, and soil Fe can decrease the absorption and utilization of dietary copper and perhaps other minerals. The objectives of this study were to determine the relationship between Fe and ash concentrations and to estimate soil contamination from available analytes in commonly used forages in US dairy operations. Data from Cumberland Valley Analytical Services covering the period from 2009 to 2014 for corn silage, legume hay, mixed mostly legume (MML) silage, and mixed mostly grass (MMG) silage were used. Soil contamination was estimated using 2 independent measures, residual ash and Fe enrichment. Residual ash (RA) was calculated according to Cary et al. (1986). Fe enrichment was calculated using the measured forage Fe concentrations and soil Fe concentrations obtained from a US Geological Survey database for the matching location. Linear regression analysis was used to evaluate relationships between total ash, RA, and forage Fe concentrations. As expected, RA was highly correlated with total ash with R<sup>2</sup> > 0.90 (P < 0.001). Fe was also highly correlated with RA and total ash with R<sup>2</sup> ranging from 0.47 to 0.63 (P < 0.001). The relationships between Fe and ash concentrations is good across forages, and 1% or greater soil contamination occurs in a significant portion of forages (Table). Level of soil contamination was associated with geographic location. In particular, corn silages grown in the western US had higher (P < 0.001) levels of total ash, RA, and Fe than corn silages from the eastern and midwestern US.

*Contd.*

**Table 1 (Abstr. T434).** Level of soil contamination (% DM), percentage of samples (Prop), total ash (% DM), and Fe concentrations (mg/kg) of commonly used dairy forages, given as mean  $\pm$  SD

Soil contamination		Corn silage	Legume hay	MML silage	MMG silage
<1%	Prop	63.5	18.3	8.3	19.3
	Total Ash	3.28 $\pm$ 0.47	9.20 $\pm$ 1.24	9.21 $\pm$ 1.19	6.08 $\pm$ 1.56
	Fe	133 $\pm$ 83	212 $\pm$ 128	265 $\pm$ 151	219 $\pm$ 170
1 to 4%	Prop	36.0	71.9	78.1	57.6
	Total Ash	5.39 $\pm$ 0.94	10.68 $\pm$ 1.25	10.55 $\pm$ 1.24	8.41 $\pm$ 1.77
	Fe	234 $\pm$ 139	353 $\pm$ 238	423 $\pm$ 261	355 $\pm$ 252
>4%	Prop	0.5	9.8	13.6	23.1
	Total Ash	8.02 $\pm$ 0.60	13.35 $\pm$ 1.35	13.51 $\pm$ 1.56	12.07 $\pm$ 2.03
	Fe	555 $\pm$ 313	872 $\pm$ 553	1155 $\pm$ 576	850 $\pm$ 605

**Key Words:** trace mineral, soil contamination, copper availability

**T435 Comparison of two methods to quantify free amino acids in cow plasma.** Nelson Lobos<sup>\*1</sup>, Glen Broderick<sup>2</sup>, Nancy Whitehouse<sup>5</sup>, Daniel Luchini<sup>3</sup>, Michel Wattiaux<sup>1</sup>, and Peter Crump<sup>4</sup>, <sup>1</sup>Department of Dairy Science, University of Wisconsin-Madison, Madison, WI, <sup>2</sup>Broderick Nutrition & Research LLC, Madison, WI, <sup>3</sup>Adisseo S.A.S., Alpharetta, GA, <sup>4</sup>Department of Computing and Biometry, University of Wisconsin-Madison, Madison, WI, <sup>5</sup>Department of Biological Sciences, University of New Hampshire, Durham, NH.

The objective was to determine how measurements of plasma free AA by gas chromatography (GC) after chloroformate derivatization compared with values obtained by ion-exchange chromatography (HPLC) followed by ninhydrin derivatization. Plasma was obtained from lactating dairy cows (mean DIM = 148) part of a trial were Met was abomasally infused (12 or 24 g Met/d) or fed in rumen-protected form (15 or 30 g Met/d). Blood samples (n = 89) were collected from coccygeal vessels into evacuated tubes. After centrifugation, plasma was split into 2 aliquots and stored frozen at  $-80^{\circ}\text{C}$ . Each set was analyzed either by HPLC in a commercial laboratory, or in-house by GC using a kit (EZ:faast, Phenomenex). This method involved deproteinization with TCA, AA collection using solid phase extraction, derivatization with chloroformate, GC separation in a capillary column and flame ionization detection. Quantitation was based on area under the curve, using the internal standard ratio method (Norvaline). Data were analyzed by Pearson correlation ( $r$ ; Table 1). However Bland-Altman posited that correlation is misleading to compare methods and instead proposed the inspection of difference plots and limits of agreement. Calculation of concordance correlation coefficients provided a quantitative alternative (CCC; Table 1). Overall correlation between methods was 0.95 ( $P < 0.001$ ); nevertheless, correlations varied greatly for individual AA from 0.97 for Met to 0.15 for Glu. For Met, most data were within the limits of agreement established by Bland-Altman approach. Furthermore among all AA, CCC for Met was the closest to 1 (indicating almost total agreement between methods; Table 1). For research focusing on Met, GC offers a cheaper and faster alternative to HPLC.

*Contd.*

**Table 1 (Abstr. T435).** Free AA (except Arg and Trp) in plasma measured by two methods

Amino acid	GC ( $\mu\text{M}$ )	HPLC ( $\mu\text{M}$ )	$r$	CCC
His	50.2	62.8	0.42	0.18
Ile	107.6	101.6	0.96	0.94
Leu	121.2	145.2	0.92	0.36
Lys	72.4	80.8	0.35	0.67
Met	32.8	33.9	0.97	0.99
Phe	40.0	46.1	0.65	0.37
Thr	186.9	117.9	0.96	0.25
Val	348.0	266.1	0.89	0.25
Ala	351.7	267.9	0.88	0.21
Asn	44.0	57.1	0.84	0.26
Asp	9.4	3.9	0.51	0.07
Cys	18.9	21.0	0.35	0.56
Glu	56.1	39.7	0.15	0.08
Gln	110.6	200.6	0.78	0.10
Gly	463.3	348.8	0.92	0.36
Pro	102.2	81.4	0.84	0.25
Ser	127.1	95.3	0.89	0.32
Tyr	30.5	42.1	0.60	0.31

**Key Words:** amino acid, methionine, chromatography

**T436 Fecal starch and starch digestibility: An indirect inter-relationship.** C. E. Owens<sup>1</sup>, R. A. Zinn<sup>2</sup>, and F. N. Owens<sup>\*3</sup>, <sup>1</sup>Duke University, Durham, NC, <sup>2</sup>University of California, El Centro, CA, <sup>3</sup>DuPont Pioneer, Johnston, IA.

Published equations to calculate total-tract starch digestibility (SD) from the concentration of starch in fecal dry matter (FS) have regression slopes that differ by more than 5 fold. Why these slopes differed and methods to improve the accuracy and precision of prediction were explored. Mathematically, SD and FS are correlated. However, 2 additional factors, starch content of dietary DM (DS) and indigestibility of diet DM (IDM =  $100 - \text{DMD}$ , %) alter this relationship. Published literature data (204 diets fed to lactating dairy cows; 191 diets fed to feedlot cattle) that provided or allowed SD, FS, DS, and IDM to be calculated were compiled to examine this relationship. When DS and IDM were ignored, the relationship was imprecise (SD, % =  $99.16 - 1.07 \text{ FS} \%$ ;  $R^2 = 0.81$  for lactating cows; SD, % =  $99.98 - 0.44 \times \text{FS} \%$ ;  $R^2 = 0.88$  for feedlot cattle). Including DS improved precision (SD, % =  $100.13 - 0.341 \times \text{FS} \% / \text{DS} \%$ ,  $R^2 = 0.92$  for lactating cows; SD, % =  $100.29 - 0.257 \times \text{FS} \% / \text{DS} \%$ ,  $R^2 = 0.91$  for feedlot cattle); adding IDM increased precision further (SD, % =  $100 - 1 \times \text{FS} \% / \text{DS} \% \times \text{IDM} \%$ ;  $R^2 = 1.00$ ;  $P = 1/\infty$  for all cattle). Because starch is a component of dietary DM, as SD increases IDM will decrease; hence the relationship of SD to FS is curvilinear, not linear unless the digestibility of non-starch components increases when SD increases. Knowledge of IDM and the dietary and fecal concentrations of any component of the diet including NDF allows apparent digestibility to be computed directly and avoids the need to predict digestibility from in vitro assays. Direct measurement often proves more economical analytically than in vitro procedures and avoids dependence on tabular feedstuff values in publications or computerized diet formulation programs. Combined with intake of DM, direct digestibility measurement can quantify the amount of digested energy available for production by ruminants. As with all analytical procedures, accuracy of digestibility estimates relies on representative sampling and appropriate analyses of diets and feces.

**Key Words:** starch digestibility, fecal starch, equation

**T437 Starch and NDF digestibility by high-producing lactating cows: A field study.** B. Powel-Smith, L. J. Nuzback, W. C. Mahanna, and F. N. Owens\*, *DuPont Pioneer, Johnston, IA.*

In vivo measurements are the gold standard against which in vitro procedures digestibility estimates must be judged. To quantify the relationships of in vitro ruminal disappearance to total-tract digestibility of starch and NDF, samples were obtained from the top string of lactating cows from 32 commercial herds in the Upper Midwest. Silage, TMR, grain, and fecal samples were assayed at a commercial laboratory for some of the following: starch, 7h in vitro starch digestion, NDF digestibility at 24 and 240 h, and lignin at. For calculating in vivo digestibility, lignin and UNDF served as indigestible markers. All cows were fed kernel processed corn silage (25 to 40% DM) plus dry ground or high moisture ear corn or grain; diets averaged 26% starch and 31% NDF. Except for 2 herds, starch digestibility exceeded 95% being related closely to fecal starch concentration ( $R^2 = 0.98$ ). However, in vitro starch disappearance at 7 h was not related to total-tract starch digestibility ( $R^2 = 0.00$ ). Total-tract digestion averaged 55% for NDF or 60% for available NDF (NDF minus UNDF240). In vivo NDF digestibility of the TMR tended to decline as its UNDF content increased ( $R^2 = 0.21$ ), but 24 h in vitro NDF disappearance of the TMR proved to be poorly related to NDF digestibility in vivo ( $R^2 = 0.13$ ). Kernel processing score was not related to starch digestibility, but all silage had fermented more than 6 mo before feeding. Processing score tended to be greater and presence of whole and half kernels in silage was less with shredlage than conventional kernel processing rolls but total-tract starch digestibility was not altered by processing roll type. Dry matter digestibility calculated from UNDF differed by as much as 10% from digestibility calculated from lignin. Total-tract starch digestibility was not correlated with surface area of particles or mean particle size of the dry ground corn fed. To obtain reliable and accurate estimates of energy availability for ruminants, nutritionists should rely on total-tract digestion measurements.

**Key Words:** in vivo digestibility, NDF, starch

**T438 Effects of diets with different energy levels on growth performance and rumen environment of heifer.** Yan Tu\*<sup>1,2</sup>, Xiang Cui<sup>1</sup>, Tao Ma<sup>1,2</sup>, Bing-wen Si<sup>1,2</sup>, Nai-feng Zhang<sup>1,2</sup>, and Qi-yu Diao<sup>1,2</sup>, <sup>1</sup>*Feed Research Institute of Chinese Academy of Agricultural Sciences, Key Laboratory of Feed Biotechnology of the Ministry of Agriculture, Beijing, China,* <sup>2</sup>*Beijing Key Laboratory for Dairy Cow Nutrition, Beijing, China.*

Thirty-two Chinese Holstein heifers (98 d old) were randomly divided into 4 groups with 8 heifers each (8 replications). Each group was subjected to one of the following 4 total mixed rations (TMR) with 2.52 (A), 2.82 (B), 2.99 (C), and 3.21 (D) Mcal of metabolic energy (ME) per kg (dry matter basis) diet for 82 d. The energy gradient was adjusted by changing the composition of concentrate and adding rumen bypass fat. The concentrate to roughage ratio of the TMR was fixed at 59:52:40:48. Growth performance was determined at 98, 120, 150, and 180 d of age, and rumen fermentation parameters were determined at 98, 120, 150, and 180 d of age, and microbial populations in rumen fluid from 181-d-old heifers were determined by real time polymerase chain reaction analysis. During 98 to 180 d of age, the average daily gain (ADG) did not differ among the groups ( $P > 0.05$ ). However, during 151 to 180 d of age, ADG of the heifers of group D (1.11 kg/d) were greater ( $P < 0.05$ ) than those of group A (0.77 kg/d). Feed utilization efficiency was improved in groups C and D ( $P < 0.05$ ) when compared with that in group A. Dietary ME did not influence ruminal pH value and  $\text{NH}_3\text{-N}$  concentration ( $P > 0.05$ ). Increasing level of ME significantly increased

ruminal proportion of propionate ( $P < 0.01$ ) and reduced that of acetate ( $P < 0.05$ ) and the ratio of acetate to propionate ( $P < 0.01$ ). Dietary treatments had no significant effect on the concentration of total volatile fatty acid ( $P > 0.05$ ). *Lachospira multipara* of group C tended to be higher than that of group D ( $P < 0.10$ ), and the numerical quantity of protozoan, *R. flaeafaciens*, *R. albus*, *B. fibrisolvens* and *P. ruminicola* of group C were greater than the other groups ( $P < 0.05$ ). Current study suggested that heifers supplemented with 2.99 Mcal of ME per kg of diet (group C) had better feed utilization efficiency and more abundant microbial flora compared with those fed other levels of ME.

**Key Words:** heifer, rumen fermentation parameter, ruminal microorganism

**T439 Effect of tallow and soybean oil addition to calf starters fed to dairy calves from birth to four months of age on calf performance and digestion.** T. Mark Hill\*, H. Gale Bateman II, James M. Aldrich, James D. Quigley, and Rick L. Schlotterbeck, *Nurture Research Center, ProVimi North America, Brookville, OH.*

Energy demands for calves can increase during periods of heat and cold stress. One way to potentially increase energy intake is to increase the energy density of the feed with fat. Trial 1a compared a control starter with no added fat or oil (CON) to starters with 2% tallow (TAL) and 2% soybean oil (SOY). Starters were 20% CP and 45 to 47% starch. Male Holstein calves (n = 48) that were initially 3 to 5 d of age were fed a 27% CP, 17% fat milk replacer at 0.66 kg DM daily and fully weaned by 42 d of a 56-d trial. Trial 1b estimated the digestion of the diets (employed chromic oxide as an indigestible digesta flow marker) using a subset of 5 weaned calves per treatment between d 52 and 56. Trial 2 used 48 Holstein calves initially 59 to 61 d of age fed starters CON and SOY blended with 5% chopped grass hay over a 56-d trial. Trial 3 used 48 Holstein calves initially 59 to 61 d of age fed starters CON and TAL blended with 5% chopped grass hay over a 56-d trial. Treatments were compared in a randomized ANOVA using repeated measures. In Trials 1a and 1b, pre-planned contrasts compared CON vs. TAL and CON vs. SOY ( $P < 0.05$ ) Compared with CON, calves fed SOY had reduced starter intake, ADG, and digestion of DM, OM, and CP vs. CON before 8 weeks of age ( $P < 0.05$ ). Compared with CON, calves fed SOY had reduced ( $P < 0.05$ ) ADG and change in hip width from 2 to 4 mo of age. Compared with CON, calves fed TAL had reduced ( $P < 0.05$ ) ADG and tended ( $P < 0.1$ ) to have reduced change in hip width from 2 to 4 mo of age. Calculated ME intake was not increased in any trial by added fat or oil. Tallow and soybean oil inclusion at 2% of the starter feed was not advantageous for calf growth before 4 mo of age.

**Key Words:** fat, oil, digestion

**T440 Relationships between udder resistance and dietary levels of copper and zinc.** Alfredo J. Escribano<sup>1</sup>, Juan Jose Mallo\*<sup>1</sup>, Luis Miguel Jiménez<sup>2</sup>, and Nuria Roger<sup>2</sup>, <sup>1</sup>*NOREL Animal Nutrition, Madrid, Spain,* <sup>2</sup>*Servet Talavera S.L., Talavera de la Reina, Toledo, Spain.*

Mastitis constitutes the main health issue in dairy cows farms, because it is widespread and leads to high economic losses. Copper and especially zinc have shown to increase cows' resistance to mammary infections. However, the levels of these minerals in rations varies greatly, and then, there is a gap of knowledge with regard to the relationship between the levels of trace minerals in the diets used to feed dairy cows, and the incidence of mammary diseases. To contribute in this sense, we carried out this study, with the objective to evaluate the relationship between

the level of zinc and copper and udder health parameters. For this purpose, 55 farms representative of the Spanish production systems were selected. The statistical procedure followed was based on bivariate correlation analysis and ANOVA. Results showed that negative and significant correlations existed between the level of dietary copper and the following indicators: Somatic Cell Counts ( $R^2 = 0.138$ ,  $P = 0.008$ ) and monthly rate of clinical mastitis ( $R^2 = 0.093$ ,  $P = 0.031$ ). However, no significant correlations were found with the rest of indicators: mastitis prevalence, new infections rate, curation rate in lactation, curation rate in the dry period, and new infections rate at calving. Regarding zinc levels, a significant correlation was found with the curation rate in the dry period ( $R^2 = 0.136$ ,  $P = 0.008$ ). As it can be observed, weak and few correlations were found. Surprisingly and contrary to most of the scientific knowledge, the effect of zinc had a low impact on udder health and resistance. These results could be showing interactions between minerals levels and other dietary compounds. Also, as levels of zinc were above the recommendations, a low response to increasing zinc levels was found. On the contrary, as not all diets were satisfying animals' needs for copper, higher responses to the level of this mineral were found. Further research is needed to have a deeper knowledge about both minerals interactions and the levels of minerals above which host's resistance do not increase significantly.

**Key Words:** dairy, mastitis, mineral

**T441 Use of a fermented ammoniated condensed whey product in lactating dairy cattle.** John P. McNamara<sup>\*1</sup>, Drina Huisman<sup>1</sup>, Heather M. Hastings<sup>1</sup>, and Gerald Poppy<sup>2</sup>, <sup>1</sup>Washington State University, Pullman, WA, <sup>2</sup>Packerland Whey Product Inc., Luxemburg, WI.

An existing fermented ammoniated condensed whey product (LactoWhey) has been in use in dairy rations for more than 30 years; however the last published studies were almost 30 years ago. It was of interest to test the efficacy of using this product in more modern dairy rations typical of the northwestern US dairy region which contain a wide variety of protein sources and by-product feeds. Therefore, a field trial was conducted at 3 commercial dairies and one university dairy. Lactowhey is liquid fermented ammoniated condensed whey containing 44% CP equivalent and 38% lactic acid equivalent from ammonium lactate. Lactowhey was added to the ration to replace other protein sources (primarily SBM and Canola meal) on a CP equivalent basis. Rations were balanced and fed to meet requirements to multiple pens on each commercial dairy (or through individual feeding gates on the university dairy) starting from an average of 66 DIM for 90 d. On the commercial dairies, milk tank weights and composition were recorded as well as DHIA test day milk and composition. A subset of 18 to 20 cows in each pen was followed for individual milk and composition, and rumen fluid and blood was sampled at 0, 3 and 13 wk of treatment (at the university dairy all cows ( $n = 20$  per treatment group) were also sampled at 6 wk of treatment. Milk production ranged from 35.9 to 45.5 kg/d at start of trial and 41.7 to 35.2 kg/d during treatment period, with no effect of treatment. Milk composition averaged 3.80% and 3.21% and 12.3 mg/dl for fat and protein and MUN across the period and farms, with no effect of treatment. There were no differences due to treatment in BW or BCS change, or rumen pH or in blood NEFA, glucose or BUN. The proportion of rumen bacterial DNA as measured by microbiome analysis was primarily *Prevotella* species, with a variety of other species represented. Species proportions varied among the 4 dairies and among individual cows, with a generally consistent pattern within dairies and individual cows. There was no consistent effect of the fermented whey on bacterial proportions. The work confirms the utility of this fermented

whey product to support high milk production in commercial herd when used to replace other protein sources.

**Key Words:** ammoniated whey, protein substitution, lactation

**T442 Evaluating varying dietary protein and energy levels for economical productive performance of Nili-Ravi buffalo heifer calves.** Zeeshan Muhammad Iqbal<sup>\*1</sup>, Muhammad Abdullah<sup>1</sup>, Khalid Javed<sup>1</sup>, Makhdoom Abdul Jabbar<sup>1</sup>, and Juan J. Loo<sup>2</sup>, <sup>1</sup>University of Veterinary and Animal Sciences, Lahore, Pakistan, <sup>2</sup>University of Illinois at Urbana-Champaign, Urbana, IL.

The optimal dairy management programs are based on rearing of heifers at a low cost without compromising growth rate. The aim of this study was to investigate the effect of dietary protein and energy level on growth and metabolic parameters of Nili-Ravi buffalo heifers. The experiment was conducted with 30 female buffalo calves (6–8 mo age) divided randomly into 3 treatment groups ( $n = 10$ /group). The animals in treatment A, B and C were offered 3 different levels of concentrate (CP 17% and 2.6 Mcal ME/kg) at 0.5%, 1% and 1.5% of body weight, respectively. In addition to this, all animals were given chopped green fodder (sorghum and maize) and fresh water ad libitum. All data were collected on individual animals for a total of 8 mo. As designed, the average daily protein and energy ( $585.94 \pm 15.36$  g,  $13.08 \pm 0.34$  MJ/kg) intake was greater in treatment C and lower ( $374.70 \pm 10.09$  g,  $9.56 \pm 0.26$  MJ/kg) in treatment A. The differences in intake did not affect ( $P > 0.05$ ) average daily gain of heifers ( $497.32.69 \pm 17.92$ ,  $503.63 \pm 19.09$  and  $532.77 \pm 20.67$  g/d). Thus, feed efficiency estimates were greater for treatment A ( $0.135 \pm 0.004$ ) as compared with treatments B and C ( $0.113 \pm 0.003$ ,  $0.108 \pm 0.004$ ). Dietary treatments did not affect ( $P > 0.05$ ) blood profile but serum urea concentration was greater in treatment B and C ( $50.08 \pm 2.05$ ,  $51.41 \pm 2.29$ ) compared with treatment A ( $42.34 \pm 1.59$ ). Increasing nutrient intake had no effect ( $P > 0.05$ ) on the digestibility of organic matter, fat, ash, ADF. However, the digestibility of diet DM, crude protein and ADF was greater in treatment C ( $71.14 \pm 0.96$ ,  $65.85 \pm 0.88$ ,  $56.09 \pm 1.02\%$ ) as compared with treatment A ( $66.06 \pm 2.32$ ,  $61.21 \pm 1.72$ ,  $51.43 \pm 2.13\%$ ) and B ( $67.22 \pm 1.09$ ,  $62.01 \pm 1.20$ ,  $52.91 \pm 1.32\%$ ). The cost for production (USD) of per kg gain was higher in treatment C (\$2.42) and lower in lower in treatment A (\$1.34) and B (\$1.96), respectively. As increasing dietary concentrate intake had no effect on growth rate, the estimated efficiency of utilization of nutrients was better and cost per kg gain was lower with the 0.5% concentrate inclusion. Thus, this level of concentrate in the diet at a rate of 0.5% of body weight appears most economical for rearing Nili-Ravi buffalo heifers from 6 to 14 mo of age.

**Key Words:** protein, energy, heifer

**T443 Trace mineral variation in dairy forages: Where are the hot spots?** J. R. Knapp<sup>\*1</sup>, W. P. Weiss<sup>2</sup>, R. T. Ward<sup>3</sup>, and K. R. Perryman<sup>4</sup>, <sup>1</sup>Fox Hollow Consulting LLC, Columbus, OH, <sup>2</sup>Dept. of Animal Sciences, The Ohio State University, Wooster, OH, <sup>3</sup>Cumberland Valley Analytical Services, Hagerstown, MD, <sup>4</sup>Micronutrients Inc., Indianapolis, IN.

The objectives of this study were to quantify the variation in trace mineral (TM) concentrations in forages, evaluate the contribution of US geographical location and harvest season to the TM variation, and identify areas where high variation in TM concentrations would preclude using standard reference concentrations. Trace minerals of interest were Cu, Mn, Zn, and Fe. Data from Cumberland Valley Analytical Services covering the period from 2009 to 2014 with concentrations of major

nutrients as well as mineral concentrations were used. Data were statistically filtered to remove outliers based on macronutrient concentrations and misidentified feeds before analyses of TM variation. As expected, TM concentrations for corn silage, legume hay, mixed mostly legume (MML) silage, and mixed mostly grass (MMG) silage displayed skewed distributions. TM values were log normalized before ANOVA with location, season, and their interaction as independent effects and total ash concentration as a covariate. Variation in TM concentrations due to location and total ash were significant ( $P < 0.0001$ ) and the largest sources. Season was often non-significant ( $P > 0.20$ ), while the interaction between location and season was significant ( $P < 0.05$ ). Whereas forages in some US locations have consistently low TM concentrations, forages in other areas have both high concentrations and high variation. These results support sampling and analysis of forages and formulation of dairy rations for TM based on analytical results rather than reference values. Recommendations for sampling frequency will vary by location as a function of the observed variation in TM concentrations.

**Table 1 (Abstr. T443).** Lowest and highest variation in observed Cu and Zn concentrations among U.S. geographical locations, given as median and 5th (p5) to 95th (p95) percentiles in mg/kg

Item	Lowest variation			Highest variation		
	Median	p5	p95	Median	p5	p95
<b>Cu</b>						
Corn silage	6.0	4.9	7.3	7.4	3.2	16.4
Legume hay	9.6	7.2	11.4	8.2	6.0	34.0
MML silage	7.8	5.7	9.6	10.9	5.7	30.1
MMG silage	8.8	6.1	9.4	7.8	4.7	37.9
<b>Zn</b>						
Corn silage	23.2	21.4	28.5	26.6	15.3	60.7
Legume hay	24.0	21.1	29.5	27.8	20.9	49.2
MML silage	28.3	25.8	35.0	35.7	21.8	69.7
MMG silage	23.8	20.0	27.6	29.1	18.4	97.5

**Key Words:** copper, zinc, mineral variation

**T444 Evaluating carbon and oxygen flux variability and heat production in dairy cows using a portable automated gas quantification system.** Andre B. D. Pereira<sup>\*1</sup>, Andre F. Brito<sup>1</sup>, Santiago A. Utsumi<sup>2</sup>, Brianna J. Isenberg<sup>1</sup>, and Kelly S. O'Connor<sup>1</sup>, <sup>1</sup>University of New Hampshire, Durham, NH, <sup>2</sup>Michigan State University, W. K. Kellogg Biological Station, Hickory Corners, MI.

The objectives of this study were (1) to evaluate the variability in spot short-term measurements of CO<sub>2</sub> ( $Q_{CO_2}$ ) CH<sub>4</sub> ( $Q_{CH_4}$ ) emissions and O<sub>2</sub> consumption ( $Q_{O_2}$ ), and (2) estimate heat production (HP) from dairy cows fed diets with different RDP and NSC profiles. Eight multiparous and 4 primiparous lactating Holstein cows were blocked by days in milk and milk yield into replicated 4 × 4 Latin squares. Cows received corn silage, grass-legume haylage, roasted soybean, and (1) corn meal and urea (negative control), (2) corn meal, soybean meal, and rumen-protected Lys-Met (positive control), (3) field peas, corn meal, and soybean meal, or (4) field peas, corn meal, soybean meal, and rumen-protected Lys-Met. Gas measurements were performed using a portable automated gas quantification system (Greenfeed, C-Lock Inc., Rapid City, SD) fitted with sensors for CO<sub>2</sub>, CH<sub>4</sub> and O<sub>2</sub>. Cows were sampled 3 times daily (about 8 h apart) for 7 d, with sampling points advanced 2 h in the following day to account for diurnal variation of gas flux. Heart rate was monitored for 4 d to calculate  $Q_{O_2}$ /heartbeat (n = 4 cows). Heat production was calculated according to Kaufmann et al. (2011): [(4.96 + 16.07/respiratory quotient) ×  $Q_{CO_2}$ ]. Filtering

of gas sampling by variable head position or low visitation to the gas quantification system (n = 2) resulted in lower number of gas flux points than anticipated (316 valid measurements of 1,008 possible). Each gas sampling lasted, on average, 3.7 min. For all treatments, the within animal CV averaged 25.5% ( $Q_{CH_4}$ ), 15.4% ( $Q_{CO_2}$ ), and 22.3% ( $Q_{O_2}$ ) and the between animal CV averaged 7.0% ( $Q_{CH_4}$ ), 4.3% ( $Q_{CO_2}$ ), and 5.8% ( $Q_{O_2}$ ). There was no difference between treatments for  $Q_{CH_4}$  (mean = 373.5 g/d),  $Q_{CO_2}$  (mean = 6,293 L/d),  $Q_{O_2}$  (mean = 7,307 L/d), HP (mean = 149 MJ/d),  $Q_{O_2}$ /heartbeat (mean = 66.1 L/heartbeat), and HP/heartbeat (mean = 20.7 MJ/heartbeat). Short-term spot measurements of gas flux can be used to estimate HP in dairy cows. However, sampling rate needs to be optimized, when using the automated gas quantification system, to reduce variability.

**Key Words:** carbon emission, oxygen consumption, heat production

**T445 Influence of a grape seed and grape marc meal extract (GSGME) on performance parameters, hepatic expression of pro-inflammatory and ER stress-related genes and antioxidant status in dairy cows.** Denise K. Gessner<sup>1</sup>, Christian Koch<sup>2</sup>, Franz-Josef Romberg<sup>2</sup>, Anne Winkler<sup>3</sup>, Georg Dusel<sup>3</sup>, Eva Herzog<sup>1</sup>, Erika Most<sup>1</sup>, Anne-Kathrin Blässe<sup>\*4</sup>, Bernhard Eckel<sup>4</sup>, and Klaus Eder<sup>1</sup>, <sup>1</sup>Justus Liebig University Giessen, Giessen, Germany, <sup>2</sup>Lehr- und Versuchsanstalt Hofgut Neumühle, Neumühle, Germany, <sup>3</sup>University of Applied Sciences Bingen, Bingen, Germany, <sup>4</sup>Dr. Eckel GmbH, Niederzissen, Germany.

At the onset of lactation, inflammation and stress of the endoplasmic reticulum (ER) occurs in the liver resulting in impaired hepatic function with undesirable effects on the health status of high yielding dairy cows. The present study investigated the hypothesis that feeding a polyphenol-rich grape seed and grape marc extract (GSGME) has the potential to suppress the inflammatory process and ER stress in the liver during early lactation. Twenty-eight German Holstein cows received either a total mixed ration (TMR, control group) or the same TMR supplemented with 1% GSGME (polyphenol content: 43 mg/g; Anta Ox, Dr. Eckel GmbH) from wk 3 ap to wk 9 pp (experimental unit: single cow). Milk, blood, and liver samples were taken. Data were statistically evaluated by the linear mixed-effects model of R. Dry matter intake of the cows did not differ between the 2 groups. However, the GSGME group had a greater milk performance ( $\Delta$  milk yield: + 3.7 kg/d;  $P < 0.05$ ;  $\Delta$  energy corrected milk: + 2.9 kg,  $P < 0.05$ ) from wk 2 to 9 of lactation. Cows of the GSGME group had lower hepatic mRNA concentrations of FGF21, a key marker of ER stress ( $P < 0.05$ ) and a tendency toward lower hepatic levels of triglycerides and cholesterol ( $P < 0.15$ ). A reduction of mRNA concentrations of various other ER stress response-related and pro-inflammatory genes could be observed in the GSGME group. Feeding GSGME had no effect on the antioxidative status ( $\alpha$ -tocopherol,  $\beta$ -carotene, TBARS and total antioxidant capacity in plasma). In conclusion, the study suggests that feeding polyphenol-rich plant extracts during early lactation could be a useful strategy to prevent inflammatory processes and ER stress in the liver and thus improve milk yield and animal health in dairy cows.

**Key Words:** grape seed extract, grape seed meal extract, dairy cow

**T446 Effects of *Saccharomyces cerevisiae* fermentation products on dairy calf: I. Pre-weaning performance and post-weaning stress.** G. M. Alugongo<sup>\*1</sup>, J. X. Xiao<sup>1</sup>, R. Chung<sup>2</sup>, S. Z. Dong<sup>1</sup>, S. L. Li<sup>1</sup>, I. Yoon<sup>2</sup>, and Z. J. Cao<sup>1</sup>, <sup>1</sup>State Key Laboratory of Animal Nutrition, Department of Animal Nutrition and Feed Sciences, China Agricultural University, Beijing, China, <sup>2</sup>Diamond V, Cedar Rapids, IA.

The aim of the study was to evaluate the effects of *Saccharomyces cerevisiae* fermentation products (SCFP) on performance and health of calves during the first 63 d of age. New Zealand Holstein calves (30 male and 30 female) at 2 d of age were blocked by sex and date of birth then randomly assigned within blocks to one of 3 treatments. A texturized grain starter was fed ad libitum containing 0 (Control), 0.5 or 1% SCFP (Original XPC, Diamond V) of DM. The basal diet contained steam-flaked corn (33.1%), wheat bran (7.6%), Soybean meal (14.3%), Extruded soybean (4.2%), canola meal (8.0%), and DDGS (16.0%) and was without antibiotics. In addition, the supplemented calves were fed 1 g/d SCFP (SmartCare, Diamond V) in milk from d 2 to 30. All calves were fed 4 L of colostrum within 1 h of birth and were subsequently fed milk twice daily until weaned at 56 d of age. Performance and health of all weaned female calves were monitored until 63 d of age to determine the effect of pre-weaning treatment of SCFP on weaning stress. Starter intake, fecal score and medical treatment were recorded daily. Body weight measures and blood samples were collected on d 2, 28, 56, and 63. Serum was analyzed for BUN, NEFA, IGF-1, glucose, albumin, total protein and oxidative biomarkers. Data were analyzed by MIXED procedure in SAS with contrast statement to declare Control vs. all SCFP, and 0.5% SCFP vs. 1% SCFP in starter grains. Calf was the experimental unit. Body weight, DMI, blood parameters and oxidative biomarkers did not differ. Supplementation of SCFP lowered ( $P < 0.05$ ) fecal scores, and diarrhea frequency. Number of medical treatments and medication costs were also lower ( $P < 0.05$ ) with SCFP. After weaning, SCFP reduced diarrhea in a dose-dependent manner with greater effect seen with 1% SCFP in starter grain. Overall, SCFP reduced diarrhea and pre-weaning medical costs and helped calves better manage weaning stress. The effects of SCFP were greater with the higher dosage of SCFP in starter grain.

**Table 1 (Abstr. T446).**

Item <sup>1</sup>	Treatment				Contrasts ( <i>P</i> -value)	
	CON	SCFP1	SCFP2	SEM	CON vs. SCFP1 and SCFP2	SCFP1 vs. SCFP2
ADG (kg/d)						
PrW	0.70	0.72	0.73	0.03	0.29	0.84
PoW	1.09	1.12	1.12	0.11	0.78	0.98
Fecal scores						
PrW	1.86 <sup>a</sup>	1.53 <sup>b</sup>	1.52 <sup>b</sup>	0.04	< 0.001	0.62
PoW	2.19 <sup>a</sup>	1.85 <sup>b</sup>	1.71 <sup>b</sup>	0.12	< 0.001	0.21
Diarrhea frequency						
PrW	20.81 <sup>a</sup>	10.80 <sup>b</sup>	9.20 <sup>b</sup>	1.83	<0.001	0.41
PoW	42.86 <sup>a</sup>	21.43 <sup>b</sup>	2.86 <sup>c</sup>	7.62	<0.001	0.03
Medications						
PrW	0.20 <sup>a</sup>	0.15 <sup>b</sup>	0.15 <sup>b</sup>	0.02	< 0.001	0.86
Price (\$)						
PrW	0.56 <sup>a</sup>	0.44 <sup>b</sup>	0.44 <sup>b</sup>	0.05	0.01	0.97

<sup>a,b,c</sup> $P < 0.05$ .

<sup>1</sup>PrW = pre-weaning; PoW = post-weaning; Medications = average no. of medical treatments/calf/day; Price = average cost of medication/calf/day.

**Key Words:** calf, *Saccharomyces cerevisiae* fermentation product (SCFP), health

**T447 Fibrolytic enzyme and corn silage differing in particle size for lactating dairy cows.** Gilson S. Dias Júnior\*<sup>1</sup>, Alan S. Pereira<sup>1</sup>, Fabiana F. Cardoso<sup>1</sup>, Renata A. N. Pereira<sup>3,2</sup>, and Marcos N. Pereira<sup>1,2</sup>, <sup>1</sup>Universidade Federal de Lavras, Lavras, MG, Brazil, <sup>2</sup>Better Nature Research Center, Ijaci, MG, Brazil, <sup>3</sup>Empresa de Pesquisa Agropecuária de Minas Gerais, Lavras. MG, Brazil.

The majority of Brazilian dairy farms use pull-type, one row forage harvesters for ensiling corn. For such machines, a kernel processor (KP) was developed (MU9000462-0 U2 INPI patent). We evaluated the response of cows to corn silage (CS) short particles-no KP (S, 3 mm theoretical cut) compared with long particles-KP (L, 8.5 mm). The supplementation of fibrolytic enzyme (F, Fibrozyme, Alltech) was evaluated in a 2 × 2 factorial. Sixteen individually fed Holsteins (110 ± 62 DIM) received treatments in 28-d periods, 4 × 4 LS. Corn was harvested at half milk line maturity and ensiled in 3 m diameter bag silos for 138 d. Silage S had 44.8% of particles below the 8 mm screen and L had 25.3%. Diets contained 31.6% of DM as the treatment CS, 17.1% of a standard CS, 1.9% Tifton hay, and 31.7% starch. Diet S had 1.9% of DM as NDF above the 19 mm screen, 12.3% from 8 to 19 mm, and 17.8% below 8 mm, and L had 3.0%, 15.6%, and 13.9%, respectively. The TMR was mixed manually twice per day immediately before feeding to avoid particle size reduction. Silage L increased 4% FCM (29.5 vs. 28.2 kg/d,  $P = 0.09$ ) and fat yield ( $P = 0.04$ ), and had no impact on DMI (22.0 kg/d,  $P = 0.50$ ). There were trends for reduced milk fat content and yield in response to F ( $P < 0.15$ ). Silage S reduced ruminal fluid pH and the proportion of acetate in VFA, but increased butyrate and protozoa content ( $P < 0.05$ ). Total-tract OM and NDF digestibility tended to be increased by F only when S reduced nutrient digestibility ( $P = 0.09$  for interaction of CS and F). The daily urinary allantoin secretion was increased only when F supplemented S ( $P = 0.03$  for interaction of CS and F). Fecal viscosity and DM content were also reduced when F was added to S ( $P < 0.02$  for interaction of CS and F). The loss of visible damaged corn kernels in feces was reduced by F ( $P = 0.05$ ), but the excretion of intact kernels was similar ( $P = 0.92$ ). Silage L tended to increase rumination bouts/bolus ( $P = 0.10$ ) and the duration of the first daily meal ( $P = 0.08$ ), and increased chewing activity per day and per unit of DMI ( $P < 0.03$ ). There was a trend for reduced PUN on F ( $P = 0.07$ ). Fibrolytic enzyme was beneficial to digestion only when fiber digestibility was reduced by short particles CS. Long particles CS increased lactation performance and chewing activity.

**Key Words:** fibrolytic enzyme, corn silage, particle size

**T448 Meta-analysis to examine the effect of supplemental sugar on dairy cow performance as influenced by diet nutrient components.** Stephen M. Emanuele\*<sup>1</sup>, Mary Beth de Ondarza<sup>2</sup>, and Charles J. Sniffen<sup>3</sup>, <sup>1</sup>Quality Liquid Feeds, Dodgeville, WI, <sup>2</sup>Paradox Nutrition LLC, West Chazy, NY, <sup>3</sup>Fencrest LLC, Holderness, NH.

A data set was compiled from published research (n = 85) that tested the effect of supplemental dietary sugar. Forage NDF ranged from 17 – 29% DM and calculated total sugar from 2.3 - 11% DM. Mixed model analysis was conducted using JMP statistical software (SAS Inst. Inc., Cary, NC) with nutrient parameters (CNCPS 6.1 with NDS platform, RUM&N Sas, Italy) as main effects. Model fit used treatment (Control vs. 1.5–3% added sugar vs. 3–5% added sugar vs. 5–7% added sugar), DIM (<150 or >150 DIM) within treatment, and Milk Yield (<33 or >33 kg/d) within treatment as independent factors with experiment as the random effect. Responses were analyzed with the following nutrients (%DM) in the model: starch, soluble fiber, forage NDF, ammonia, RDP, and Protein B2. Protein B2 is true protein that can contribute to dietary RUP. Number of cows per treatment was included as a weighting

factor. Nutrients with a positive effect on 3.5% FCM yield were sugar (+1.45 kg) ( $P < 0.04$ ), starch ( $P < 0.01$ ) and soluble fiber ( $P < 0.05$ ). Higher producing cows (>33 kg milk/d) had a greater response (2.14 kg FCM/d) to sugar than cows producing <33 kg of milk ( $P < 0.0001$ ). Nutrients with a positive effect on milk protein yield were sugar (+0.07 kg;  $P < 0.05$ ) and starch ( $P < 0.0001$ ). Cows producing > 33kg milk had a greater milk protein response to sugar (0.09 vs. 0.05 kg/d than cows producing <33 kg milk/d, ( $P < 0.001$ ). Sugar did not affect milk fat yield ( $P = 0.16$ ), although starch and soluble fiber had a positive effect ( $P < 0.05$ ). Sugar alone did not affect feed efficiency but starch ( $P = 0.04$ ), soluble fiber ( $P = 0.01$ ), and Protein B2 ( $P = 0.03$ ), all had a positive effect with a mean increase of 0.06 units with 3–5% supplemental sugar. Nonlinear statistical analysis using JMP Neural Net function determined optimal total calculated sugar to be 6.75% of diet DM. Optimal range for starch when feeding supplemental sugar was 20 – 25% of diet DM. Sugar did not affect milk fat or protein percent. In conclusion, 3 to 5% supplemental sugar (6 to 8% total sugar (%DM)), and starch (%DM) increased FCM (kg/d), and milk protein yield. To optimize response to supplemental sugar, the diet should contain >17% forage NDF, 10–11% RDP and less than 30% starch.

**Key Words:** sugar, meta-analysis, dairy

**T449 Pre- and post weaning performance and health of dairy calves fed milk replacers with plant and milk protein by-product sources balanced for selected amino acids.** Hugh Chester-Jones<sup>\*1</sup>, Dustin Dean<sup>2</sup>, David Ziegler<sup>1</sup>, and Kevin Halpin<sup>2</sup>, <sup>1</sup>University of Minnesota Southern Research and Outreach Center, Waseca, MN, <sup>2</sup>International Ingredients Corporation, St. Louis, MO.

One-hundred three (9 to 12 d old) individually fed Holstein heifer calves (41.8 ± 0.66 kg) were randomly assigned to 1 of 4 treatments to evaluate pre- (d 1–35) and post-weaning (d 36 to 49) calf performance and health when fed milk replacers (MR) with alternative protein sources. The study was conducted between April and August, 2014. All calves were fed a common all-milk non-medicated 22% CP:18% fat as-fed MR before the study was initiated. Calves were fed the non-medicated 22% CP:18% fat MR with either (1) All milk protein; (2) 33% of the protein replaced with dried partially hydrolyzed vegetable protein, partially autolyzed yeast and dairy solids; (3) 33% of the protein replaced with a milk extender made from dairy ingredients, edible lard, hydrolyzed vegetable proteins and yeast; or (4) 25% proteins from co-drying cheese and whey by-products. Milk replacers were formulated to be equivalent in lysine and sulfur amino acids. The MR were fed at 0.284 kg in 1.99 L water (12.5% solids) 2× daily from d 1 to 28 and 1× daily from d 29 to weaning at d 35. Calf starter (CS; 18% CP) and water were fed free choice d 1 to 49. During the first 7d of the study, calves were fed 1 part neomycin to 1 part oxytetracycline added to the MR solution to provide 22 mg/kg BW daily. There were no pre- or post weaning ADG differences ( $P > 0.05$ ). Calves averaged 0.74 kg/d gain for the 49 d study. There were no differences ( $P > 0.05$ ) in CS or total DMI intake which averaged 53.0 and 69.4 kg for the 49-d study, respectively. There were no differences in pre- and post weaning gain/feed. Gain/feed averaged 0.52 kg for the 49 d study. There were no differences in number of scouring d (d ≥ fecal score 3) pre- and post weaning. Days with fecal scores = 4 were higher ( $P < 0.05$ ) for Treatment 2 calves vs. those fed Treatments 3 and 4. Health treatment costs did not differ ( $P > 0.05$ ). Under the conditions of this study, replacing the total milk protein in MR with alternative sources resulted in calf performance and health similar to those fed an all milk protein MR.

**Key Words:** calf performance, milk replacer, alternative protein

**T450 The effect of biochemical fulvic acid (BFA) on heat stress and lactation performance in lactating cows.** Yifan Fan<sup>\*</sup>, Xiaoming Zhang, and Zhijun Cao, State Key Laboratory of Animal Nutrition, Department of Animal Nutrition and Feed Sciences, China Agricultural University, Beijing, China.

There was almost all cows suffered heat stress in hot and humid climate in most regions of China during summer. The objective of this study was to investigate the effect of biochemical fulvic acid (BFA) on heat stress and lactation performance in lactating cows. BFA is an aromatic nitrogen compound (Shenzhoulvyanmuye Co., Hebei Province, China). Its molecular construction determined it can be chelated trace elements. Thirty multiparous Holstein dairy cows were blocked into 2 groups by parity, days in milk, milk yield (MY) and somatic cell counts (SCC). This research was conducted from July through August 2014 (49 d). Two groups were fed same TMR. One was control group (CG) without BFA; the treatment group (TG) was added 25g BFA per cow per day. The quantity of different cows' parities is same in 2 groups (3 preparturient cows, 7 s lactation cows and 5 third lactation cows). The averaged days in milk of TG versus CG were 150 ± 35 d vs. 149 ± 38 d (Mean ± SD). The MY of TG and CG was 38.4 ± 4.8, 37.9 ± 6.6 kg, respectively. The SCC of TG was 4.17 × 10<sup>4</sup> compared with CG (5.06 × 10<sup>4</sup>). Cows were milked 3 times per day at 0430, 1230, and 2030 h. The MY, composition of milk, body temperature, blood samples, and respiratory rate were recorded. Data were statistically analyzed using the two-sample paired *t*-test for means procedure of SAS (version 9.2, SAS Institute Inc., Cary, NC). Although lactose, milk urea nitrogen (MUN), SCC, body temperature, and respiratory rate did not differ between TG and CG, milk yield (37.30 vs. 35.73 kg;  $P < 0.01$ ) and concentration of milk fat (3.54 vs. 3.13%;  $P < 0.05$ ) were significantly higher for TG compared with CG. However, the milk CP (3.29 vs. 3.18%;  $P < 0.01$ ) of CG was higher than TG. In the meanwhile, concentration of serum GSX-px (356 vs 207 μmol/L,  $P < 0.05$ ) was higher for TG compared with CG. These results suggested that adding BFA to lactating dairy cows during summer might increase milk yield, milk fat, relieve heat stress and not change the yield of milk CP.

**Key Words:** fulvic acid, heat stress, milk production

**T451 Pre- and post-weaning performance and health of dairy calves fed all-milk protein milk replacers or partially replacing milk protein with plasma and plant proteins in varying combinations.** Bruce Ziegler<sup>\*1</sup>, David Ziegler<sup>2</sup>, Hugh Chester-Jones<sup>2</sup>, Daniel Shimek<sup>1</sup>, Mary Raeth<sup>3</sup>, and David Cook<sup>4</sup>, <sup>1</sup>Hubbard Feeds, Inc., Mankato, MN, <sup>2</sup>University of Minnesota Southern Research and Outreach Center, Waseca, MN, <sup>3</sup>University of Minnesota Department of Animal Science, St. Paul, MN, <sup>4</sup>Milk Products, Chilton, WI.

One-hundred seven (2–5 d old) individually fed Holstein heifer calves (39.8 ± 0.70kg) were randomly assigned to 1 of 4 treatments to evaluate pre- (d 1–42) and post weaning (d 43–56) calf performance and health when fed milk replacers (MR) with alternative protein sources. The study was conducted between February and April, 2014. Calves were assigned to non-medicated 24% CP:20% fat MR with (1) All milk protein, (2) 25% of total protein from plasma, (3) 25% of total protein from plant peptide proteins, or (4) 12.5% plasma, 12.5% peptide proteins. All calves were fed a non-medicated 24% CP:20% fat MR at 0.34 kg in 2.39 L water (12.5% solids) 2× daily for the first 35 d and 1× daily d 36 to weaning at 42 d. Day 1 to 14, 1:1 neomycin:oxytetracycline was added to the MR solution to provide 22 mg/kg BW/d. Calf starter (CS; 18% CP as-fed) and water were fed free choice from d1. Milk replacer feeding rate was adjusted if ambient temperatures taken 0800 h were –23.3 to –28.3°C (+20% volume) or > –28.9°C (+40% volume). During the study, average

minimum temperatures were - 21.1, - 9.6 and 0.6°C for February (22 d < -17.8°C), March (3 d < -17.8°C) and April respectively. There were no pre- or post weaning ADG differences ( $P > 0.05$ ). Calves averaged 0.73 kg/d gain for the 56-d study. Milk replacer intake was similar for all treatments, averaging 25.1 kg. There were no differences ( $P > 0.05$ ) in CS or total DMI intake which averaged 51.5 and 76.7 kg for the 56-d study, respectively. There were no differences in pre- and post weaning gain/feed. Gain/feed averaged 0.58 kg for the 56 d study. Across treatments, calves doubled their initial BW and gained an average of 11.7 cm in frame growth. There were no differences in number of scouring d pre- and post weaning and health treatment costs. Under the conditions of this study, replacing 25% of the total milk protein in MR with alternative sources resulted in calf performance and health similar to those fed an all milk protein MR.

**Key Words:** calf performance, milk replacer, alternative protein

**T452 Flow of microbial crude protein out of the rumen when dairy cattle are supplemented with 2-hydroxy-4-methylthio-butanoic acid (HMTBa).** C. J. R. Jenkins<sup>1</sup>, S. C. Fernando<sup>1</sup>, C. L. Anderson<sup>3</sup>, N. D. Aluthge<sup>2</sup>, E. Castillo-Lopez<sup>4,1</sup>, H. A. Tucker<sup>5</sup>, G. I. Zanton<sup>5</sup>, D. Hostetler<sup>6</sup>, and P. J. Kononoff<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Nebraska-Lincoln, Lincoln, NE, <sup>2</sup>Food Science and Technology Department, University of Nebraska-Lincoln, Lincoln, NE, <sup>3</sup>School of Biological Sciences, University of Nebraska-Lincoln, Lincoln, NE, <sup>4</sup>Instituto de Investigaciones Oceanológicas, Universidad Autónoma de Baja California, Ensenada, México, <sup>5</sup>Novus International Inc., St. Charles, MO, <sup>6</sup>The School of Veterinary Medicine and Biomedical Sciences, University of Nebraska-Lincoln, Lincoln, NE.

Four multiparous, lactating Holstein cows (average DIM 169.5 ± 20.5 d), fitted with ruminal and duodenal cannulas, were used in a 4 × 4 Latin square with a 2 × 2 factorial arrangement of treatments to investigate the effects of 2-hydroxy-4-methylthio-butanoic acid (HMTBa; Alimet, Novus International, St. Charles, MO) when fed with diets deficient or in excess of metabolizable protein (MP) on milk production and composition, rumen microbial activity and protein flow out of the rumen, and rumen microbial community composition. Cows were housed in individual tiestalls and fed diets designated as “Low MP” or “High MP,” which were top dressed once daily with 250 g of a corn carrier or 250 g of a corn carrier containing 10% Alimet yielding 25 g of Alimet/hd/d. No interactions were observed between HMTBa and level of dietary MP, with the exception of ruminal acetate to propionate ratio ( $P = 0.04$ ). Milk yield was not affected by treatment and averaged 23.8 ± 2.06 kg. There was a tendency ( $P = 0.06$ ) for increased milk protein percent in cows receiving Low MP diets, averaging 3.30 ± 0.09% and 3.21 ± 0.09% for Low MP and High MP, respectively. The DM, OM, NDF, total-tract digestibilities were greater ( $P \leq 0.03$ ) in cows consuming the Low MP diet. Rumen pH was lower ( $P = 0.05$ ) in cows consuming High MP diets as well as in those consuming HMTBa ( $P < 0.01$ ). Rumen ammonia concentrations tended to be greater ( $P = 0.06$ ) in cows consuming HMTBa. Rumen VFA concentrations were greater ( $P = 0.02$ ) in cows consuming HMTBa. Duodenal DM flow, N flow, and bacterial N flow did not differ between treatments ( $P \geq 0.15$ ). The microbial community structure of cows receiving HMTBa was affected at the phylum level, as the relative abundance of *Fibrobacteres* was increased ( $P = 0.04$ ). Several correlations ( $P \leq 0.05$ ) between microorganisms and metadata were observed, including animal, HMTBa supplementation, level of dietary MP, DMI, digestibility, rumen ammonia, microbial N flow, and milk production. Results suggest that HMTBa affects rumen microbial

activity, irrespective of level of MP. Consequently, further investigation is warranted into the mechanism of these effects in the rumen.

**Key Words:** 2-hydroxy-4-methylthio-butanoic acid, microbial protein, rumen

**T453 Differences in microbial community structure associated with metabolizable protein and 2-hydroxy-4-methylthio-butanoic acid (HMTBa) supplementation when using in vitro and in vivo methods.** C. J. R. Jenkins<sup>1</sup>, N. D. Aluthge<sup>2</sup>, C. Anderson<sup>3</sup>, S. C. Fernando<sup>4,1</sup>, H. A. Tucker<sup>4</sup>, G. I. Zanton<sup>4</sup>, D. Hostetler<sup>5</sup>, and P. J. Kononoff<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of Nebraska-Lincoln, Lincoln, NE, <sup>2</sup>Food Science and Technology Department, University of Nebraska-Lincoln, Lincoln, NE, <sup>3</sup>School of Biological Sciences, University of Nebraska-Lincoln, Lincoln, NE, <sup>4</sup>Novus International Inc., St. Charles, MO, <sup>5</sup>The School of Veterinary Medicine and Biomedical Sciences, University of Nebraska-Lincoln, Lincoln, NE.

The objective of the study was to investigate the effects of 2-hydroxy-4-methylthio-butanoic acid (HMTB; Alimet, Novus International, St. Charles, MO) and metabolizable protein, (Low MP or High MP) as observed in vitro and in vivo. Four multiparous, lactating Holstein cows (average DIM 169.5 ± 20.5 d), fitted with a ruminal and duodenal cannula were used in a 4 × 4 Latin square with a 2 × 2 factorial arrangement of treatments. Experimental periods were 28 d in length and cows received a top dress with either 0 or 25 g of HMTBa. On d 23 of each period and 4 h after feeding, 5 kg of whole ruminal contents were collected from all cows and rumen fluid was collected. Inoculum containing rumen fluid from each cow was added to tubes according to treatment and 0.5 g of each dietary treatment placed in a 100 mL fermentation tubes. After 24 h of fermentation, the fermentation was stop by snap freezing and a microbial pellet was isolated. DNA was isolated from the microbial pellet and was used for microbial community analysis. The Operational Taxonomic Unit(s) (OTU) based analysis of OTU in the Core Measureable Microbiome (CMM), revealed that the relative abundance of *Firmicutes* decreased ( $P = 0.01$ ) from the Low MP diet to the High MP diet (41.3 ± 4.9% and 30.2 ± 1.25%, respectively). In contrast, the proportion of *Tenericutes*, and *Spirochaetes* increased ( $P \leq 0.05$ ) from the Low MP to the High MP diet while an interaction between MP and HMTBa was observed ( $P \leq 0.05$ ) in *Bacteroidetes*, *Fibrobacteres*, and *Verrucomicrobia*. The relative abundance of phyla also differed ( $P \leq 0.01$ ) by method (in vitro vs in vivo) in *Firmicutes*, *Bacteroidetes*, *Proteobacteria*, TM7, *Tenericutes*, *Spirochaetes*, SR1, and *Verrucomicrobia*. The proportion of *Actinobacteria* tended to be higher ( $P = 0.02$ ) in vivo. Comparison of methods revealed differences in the average proportion of most phyla suggesting that in vitro fermentation after 24 h was not completely representative of that which was observed the in vivo.

**Key Words:** 2-hydroxy-4-methylthio-butanoic acid, rumen, bioinformatics

**T454 The effect of long-day photoperiod on behavior of lactating dairy cows.** Kira Macmillan\*, Santiago Espinoza, and Masahito Oba, University of Alberta, Edmonton, Alberta, Canada.

The objective of this study was to evaluate effects of photoperiod management on behavioral responses in lactating dairy cows. Thirty lactating cows (days in milk = 115 ± 33, body weight = 617 ± 70 kg) were subjected to a long-day photoperiod (LP; 16 h/d light) or a short day photoperiod (SP; 8 h/d light) in a tiestall barn (n = 15 per treatment). Data and samples were collected before animals were assigned

to treatments and after a 21-dadaptation period. Feeding behavior data were summarized for 4 time periods based on light management: Period 1 (7 p.m. to 3 a.m.; both treatments had no light), Period 2 (3 a.m. to 8 a.m.; only LP treatment had light), Period 3 (8 a.m. to 4 p.m.; both treatments had light), and Period 4 (4 p.m. to 7 p.m.; only LP treatment had light). All response variables were analyzed using a model including fixed effect of treatment, time, and treatment by time interaction with FIT Model Procedure of JMP. Period by treatment interaction was not observed for DMI, milk yield, lying time, or overall feeding behavior. However, in Period 2 (3 a.m. to 8 a.m.), there was a time by treatment interaction for lying time ( $P = 0.015$ ); LP treatment decreased lying time by 5.5 min/h while SP treatment did not. There was also tendency of an interaction for eating time ( $P = 0.064$ ); LP treatment increased eating time by 5.1 min/h while SP treatment did not. In addition, tendency for period by treatment interaction was observed for sorting behavior ( $P = 0.08$ ). The LP cows sorted, to a less extent compared with SP cows, against long particles (sorting index for particles on the screen with 19-mm pores: 91.4 vs. 78.0), and for small particles (sorting index for particles through the screen with 1.18-mm pores: 101.5 vs. 104.2). These results suggest that the provision of supplementary light may reduce sorting and modulate behavior in dairy cows, and that the behavior response to LP may differ depending on time of day in which supplementary light is given.

**Key Words:** long-day photoperiod, feeding behavior, sorting

**T455 Effects of *Saccharomyces cerevisiae* fermentation products on dairy calf: II) rumen fermentation and gastrointestinal development.** Jianxin Xiao\*<sup>1</sup>, Gibson Maswayi Alusingo<sup>1</sup>, Ruby Chung<sup>2</sup>, Dongshuang Zhao<sup>1</sup>, Shengli Li<sup>1</sup>, Ilkyu Yoon<sup>2</sup>, and Zhijun Cao<sup>1</sup>, <sup>1</sup>State Key Laboratory of Animal Nutrition, Department of Animal Nutrition and Feed Sciences, China Agricultural University, Beijing, China, <sup>2</sup>Diamond V, Cedar Rapids, IA.

The aim of the study was to evaluate the effects of *Saccharomyces cerevisiae* fermentation products (SCFP) in calf starter and milk on rumen fermentation and gastrointestinal tract development in the first 56 d of age. Fifteen Holstein bull calves were randomly assigned to one of 3 groups: a texturized calf starter containing 0 (Control), 0.5 or 1% SCFP (Original XPC, Diamond V) of DM from d 4 to 56. In addition, the supplemented calves were fed 1 g/d SCFP (SmartCare, Diamond V) in milk from d 2 to 30. All calves were fed 4 L of colostrum within 1 h of birth and were subsequently fed milk twice daily until weaned at d 56. Rumen fluid was collected by an esophageal tube 4 h after the morning feeding on d 28 and 56 to determine ruminal pH, ammonia N, and volatile fatty acids (VFA). At d 56, all 15 calves were harvested for slaughter weight, and rumen and intestinal morphological parameters. Papilla length (PL) and width (PW) were measured from 5 locations of rumen. Villus height (VH) and width (VW), crypt depth (CD) and villus height to crypt depth ratio (VCR) were measured in duodenum, jejunum and ileum. Data were analyzed by MIXED procedure in SAS with contrast statement to declare Control vs. all SCFP, and 0.5% SCFP vs. 1% SCFP in starter grains. Ruminal pH and ammonia N were not altered by SCFP. However, SCFP supplementation exhibited higher ( $P < 0.05$ ) ruminal butyrate concentration than Control at d 56. Supplementation of SCFP stimulated ( $P < 0.05$ ) PL in the rumen. Although SCFP had limited effect on VH, it reduced ( $P < 0.01$ ) the CD in jejunum, and increased ( $P < 0.05$ ) VCR in all segments of small intestine, especially when supplemented at a higher dosage in starter. In conclusion, SCFP improved gastrointestinal development, possibly

due to increased butyrate production, and greater effect was observed with higher dosage of SCFP.

**Key Words:** calf, *Saccharomyces cerevisiae* fermentation product (SCFP), gastrointestinal morphology

**T456 Effects of exogenous C16:0 and C18 fatty acids (FA) on milk lipid metabolism in bovine mammary epithelial cells.** N. Dan\*<sup>1</sup>, H. Zhang<sup>2</sup>, C. J. Ao<sup>1</sup>, and Khas-Erdene<sup>1</sup>, <sup>1</sup>College of Animal Science, Inner Mongolia Agricultural University, Hohhot, Inner Mongolia, China, <sup>2</sup>College of Animal Science, Inner Mongolia University for the Nationalities, Tongliao, Inner Mongolia, China.

The objective of this study was to examine the regulatory effects of C16:0, C18:0, C18:1c9, C18:2n6, and C18:3n3 on lipid metabolism of bovine mammary epithelial cells. After plating for 48h, bovine mammary epithelial cells cultured from Chinese Holstein cows were bathed in one of the 6 treatments: 100  $\mu$ M of C16:0 + 5  $\mu$ M of C18:0 + 100  $\mu$ M of C18:1c9 + 25  $\mu$ M of C18:2n6 + 1.2  $\mu$ M of C18:3n3 as Control; C18:0 + C18:1c9 + C18:2n6 + C18:3n3 as C16:0 absent treatment (A-C16:0); C16:0 + C18:1c9 + C18:2n6 + C18:3n3 as C18:0 absent treatment (A-C18:0); C16:0 + C18:0 + C18:2n6 + C18:3n3 as C18:1 absent treatment (A-C18:1); C16:0 + C18:0 + C18:1c9 + C18:3n3 as C18:2 absent treatment (A-C18:2); C16:0 + C18:0 + C18:1c9 + C18:2n6 as C18:3 absent treatment (A-C18:3). Key lipogenic genes were analyzed using quantitative PCR, and the FA composition in cells was quantified with gas chromatography. Data were analyzed by the ANOVA procedure of SAS. Cellular triglyceride contents in cells treated with A-C18:0, A-C18:2, A-C18:3 ( $P < 0.01$ ) and A-C18:1 ( $P < 0.05$ ) were decreased compared with the control. Absence of C16:0 induced greater level of LPL ( $P < 0.05$ ). The level of ACSL1 in A-C18:3 was higher ( $P < 0.05$ ) than that of A-C16:0 and A-C18:2. The mRNA abundance of FABP3 was enhanced by A-C18:3 ( $P < 0.05$ ). The expression of SCD was reduced by A-C18:1 ( $P < 0.05$ ). The abundance of CD36 showed reduction in A-C16:0, A-C18:1, and A-C18:2 ( $P < 0.05$ ). Expression of SREBP-1 in A-C18:0, A-C18:1 and A-C18:2 were lower than that of A-C18:3 ( $P < 0.05$ ). No effects of the treatments on ACACA and FASN were observed ( $P > 0.05$ ). Furthermore, C18:2n6 and C18:1c9 compositions in cells were increased by A-C16:0 and A-C18:0 ( $P < 0.05$ ), respectively. Percentage of C16:0 in A-C18:1, and that of C16:0 and C18:0 in A-C18:3 were enhanced ( $P < 0.05$ ), respectively. Cells in A-C18:3 had greater SFA and lower UFA compared with other treatments ( $P < 0.05$ ). In general, extracellular C18 FA availability has a strong effect on cellular triglyceride synthesis; different exogenous long-chain FAs showed no difference on inhibition of de novo FA synthesis, but greatly influence expression of lipogenic genes and milk FA composition.

**Key Words:** milk fat, lipid metabolism, gene expression

**T457 Does ruminal pH affect flow of N fractions in high-producing dairy cows?** Danilo Domingues Millen\*<sup>1</sup>, Charles Schwab<sup>2</sup>, and Sergio Calsamiglia<sup>3</sup>, <sup>1</sup>Sao Paulo State University, Dracena, Sao Paulo, Brazil, <sup>2</sup>Department of Animal and Nutritional Sciences, University of New Hampshire, Durham, NH, <sup>3</sup>Animal Nutrition and Welfare Service, Universidad Autonoma de Barcelona, Bellaterra, Spain.

There is continuous culture (Calsamiglia et al., 2008) and in situ (Devant et al., 2001) evidence that nonammonia nonmicrobial N (NANMN) flow from the rumen is increased under high concentrate-low pH conditions commonly found in high producing dairy cows. The objective of this study was to evaluate the effect of dietary conditions, including rumi-

nal pH, on the ruminal outflow of N fractions in lactating dairy cows. Peer-reviewed published studies ( $n = 45$  papers;  $n = 176$  data points) designed to investigate flow of N fractions to the small intestine in dairy cows were used. Rumen pH, organic matter truly digested in the rumen (OMTDR), efficiency of microbial protein synthesis (EMPS), and intakes of RUP (RUPI), N (NI), OM (OMI), DM (DMI), and concentrate (CONCI) were the independent variables. Microbial N (MN), NANMN, and nonammonia N (NAN) were the dependent variables in the analysis. Study was included as random effect. The relationship among variables was subjected to multivariate regression analysis by MIXED procedure of SAS. The contribution of each independent variable to the changes observed in each dependent variable was assessed by calculating semipartial correlations ( $pR^2$ ). The RUPI ( $P < 0.0001$ ;  $pR^2 = 0.47$ ), OMTDR ( $P < 0.0001$ ;  $pR^2 = 0.10$ ), EMPS ( $P = 0.003$ ;  $pR^2 = 0.02$ ) and CONCI ( $P = 0.05$ ;  $pR^2 = 0.01$ ) explained 60% of the

variation in NANMN. The contribution of rumen pH to the changes in NANMN was close to, but not significant ( $P = 0.13$ ). The average flow of NANMN at  $pH < 5.8$  ( $n = 14$ ) was 325 g N/d compared with 241 g N/d at  $pH > 5.8$  ( $n = 162$ ), with residuals showing some degree of lack of fit at low pH. However, the small number of data at  $pH < 5.8$  limits the power of the test. The EMPS ( $P < 0.0001$ ;  $pR^2 = 0.37$ ), OMTDR ( $P < 0.0001$ ;  $pR^2 = 0.31$ ), and DMI ( $P < 0.0001$ ;  $pR^2 = 0.19$ ) explained 87% of the variation in MN. The RUPI ( $P < 0.0001$ ;  $pR^2 = 0.27$ ), EMPS ( $P < 0.0001$ ;  $pR^2 = 0.21$ ), OMI ( $P < 0.0001$ ;  $pR^2 = 0.09$ ) and NI ( $P = 0.01$ ;  $pR^2 = 0.01$ ) explained 58% of the variation in NAN flow. The limited in vivo data does not allow to confirm the hypothesis that pH affects the flow of NANMN, but suggests that its flow is increased at low pH.

**Key Words:** pH, microbial, nonammonia