466 Effects of including supplemental fat in low and high starch diets on milk production and energy partitioning. Joshua L. Garver*, Jonas De Souza, Michael J. VandeHaar, and Adam L. Lock, *Michigan State University, East Lansing, MI.*

Effects of low or high starch diets with or without supplemental fat on the yield of milk and milk components and energy partitioning were evaluated. Thirty-two Holstein cows (172 ± 35 DIM) were assigned randomly to treatment sequence in replicated 4 × 4 Latin squares with a 2 × 2 factorial arrangement of treatments. Treatment diets contained 16% (LS) or 32% (HS) starch and 0.0% (LF) or 1.5% (HF) palmitic acid-enriched fat supplement (BergaFat F-100). Dietary starch was altered by varying the proportion of ground corn, cottonseed, and soyhulls, with LS and HS diets containing 17 and 21% forage NDF and 42 and 29% NDF, respectively. Treatment periods were 21 d in length with the final 5 d used for data and sample collection. The statistical model included the random effects of cow and period and the fixed effects of dietary starch level, fat supplementation, and their interaction. Compared with LF, the HF treatment increased DMI in the LS diet (26.8 vs. 25.8. kg/d) and decreased DMI in the HS diet (25.9 vs. 26.5 kg/d; interaction $P < 0.01$). The HF treatment increased milk fat yield in the LS diet (1.57 vs. 1.51 kg/d) but not in the HS diet (1.49 vs. 1.51 kg/d; interaction $P = 0.06$). Compared with LF, the HF treatment also increased milk protein concentration and yield in the LS diet (3.40 vs. 3.35% and 1.31 vs. 1.26 kg/d), but not in the HS diet (interaction $P = 0.10$ and $P < 0.05$, respectively). There was no effect of treatments on milk fat and lactose concentrations or lactose yield. Compared with LF, the HF treatment increased 3.5% FCM (42.2 vs. 41.0 kg/d) in the LS diet but not in the HS diet (40.4 vs. 41.1 kg/d; interaction $P = 0.07$). Similarly, the HF treatment increased ECM (42.3 vs. 41.0 kg/d) in the LS diet but not in the HS diet (40.5 vs. 41.2 kg/d; interaction $P < 0.05$). The milk to feed ratio (ECM/DMI) was not affected by treatments. There was a trend for HS diets to increase change in BCS compared with LS diets ($P = 0.08$). However, fat supplementation did not affect change in BCS. Results demonstrate that under the dietary conditions tested, a palmitic acid-enriched fat supplement fed to mid and late lactation dairy cows maximized yield of milk and milk components when fed in a low starch diet.

Key Words: body condition, milk fat, palmitic acid

467 Effect of breed, energy level of diet, and lactation stage on the evolution of milk lipolysis in dairy cow. Elise Vanbergue*[1,2], Luc Delaby[1], Ségolène Colette[3], Yves Gallard[3], and Catherine Hurtaud[1], *[1]INRA-Agrocampus Ouest UMR1348 Pegase, Saint-Gilles, France, [2]Institut de l'Elevage, Le Rheu, France, [3]INRA, Domaine du Pin-au-Haras, Exmes, France.*

Spontaneous lipolysis is the result of the activity of a native milk enzyme, the lipoprotein lipase. Lipolysis leads to a release of free fatty acids (FFA) that cause rancid flavor in milk products. A trial was carried out to study the effects of breed, energy levels, and lactation stage on milk spontaneous lipolysis in dairy cows. Sixty-four cows were used through a year of lactation. Cows were divided into 4 homogenous groups according to their breed [Holstein (HO) or Normande (NO)] and to their feeding system: the intensive system, with a high energy diet (HED) (in winter, corn silage with 30% concentrate; otherwise, pasture with 4 kg/d of concentrate) and the grass system (GS) (in winter, conserved grass with no concentrate; otherwise, pasture with no concentrate). The period of calving was synchronized between January and March. Individual milk samples were collected every month from both morning and evening milkings. The FFA levels were determined by the difference between the FFA levels after milking and the FFA after 24 h of storage at 4°C. Data were analyzed using SAS mixed procedure. We showed that FFA levels were higher in the evening milks (0.45 vs. 0.25 mEq/100 g of fat, $P < 0.001$) and that evening milks were more susceptible to lipolysis variations. HO cows were more susceptible to lipolysis than NO (0.34 vs. 0.20 mEq/100 g of fat, in morning milks; 0.62 vs. 0.33 mEq/100 g of fat in evenings milks, $P < 0.001$). There was no effect of the feeding system on morning milks lipolysis but GS had a tendency to increase milk lipolysis in evening milks (0.41 vs. 0.55 mEq/100 g of fat, $P = 0.06$). Lipolysis was higher in early and late lactation stage, particularly in the GS group. Conserved grass may also affect the lipolysis rate. The energy status in early and late lactation stage is frequently negative so the cows may be more susceptible to a low energy diet. No correlation was found between lipolysis and milk fat content. Poor correlations ($r < 0.4$, $P < 0.001$) were found between lipolysis and milk production, fat globule size, proportion of fatty acid and protein composition, body condition and weight.

Key Words: spontaneous milk lipolysis, production factors

468 Direct and indirect transfer of omega-3 fatty acids to milk fat in dairy cows. Natalie L. Urrutia*, Jackie Y. Ying, Samantha R. McKinney, Michael H. Green, and Kevin J. Harvatine, *The Pennsylvania State University, University Park, PA.*

Transfer of dietary fatty acids (FA) to milk has been proposed to fit a 2-pool model with a fast pool representing direct transfer by chylomicrons and a slow pool representing indirect transfer through tissue recycling. The objective of this experiment was to quantify direct and indirect transfer of omega-3 (n-3) FA to milk after an abomasal bolus infusion of n-3 FA. Ten ruminally cannulated multiparous Holstein cows (247 ± 113 DIM; mean ± SD) were used in a crossover design with 7 d periods. Cows were milked 4 times daily (6 h intervals) starting 2 d before initiation of the experiment. Treatments were abomasal infusion of 120 g (infused over 1 h) of a free FA mixture enriched in α-linolenic acid (18:3 n-3; EALA) or in very long chain n-3 (>18C n-3; EVLC). The EALA and EVLC treatments provided 80.2 g and 87.6 g of n-3 FA, respectively. Milk was sampled at each milking for determination of milk fat yield and FA profile. The day before bolus infusion was used as a baseline. Total transfer of n-3 FA was analyzed in a model that included random effects of cow nested in sequence, sequence and period and fixed effect of treatment and milk yield (JMP Pro). Time course data was analyzed as repeated measures in SAS and resulting least squares means were fit to a double exponential decay function by nonlinear curve fitting (JMP Pro). Total transfer of n-3 FA to milk differed between treatments ($P < 0.001$) and was 48.2 and 32.7% of the bolus for EALA and EVLC, respectively. Milk n-3 FA concentration and yield peaked at 12 h and returned to baseline at 138 h post infusion in both treatments. Time course of n-3 FA transferred to milk fit a biexponential model ($R^2 = 0.99$). The area (% of total) under the first exponential representing direct transfer was 83.9 and 42.2% and the second exponential representing indirect transfer was 16.1 and 57.8% of the total n-3 FA transferred for EALA and EVLC, respectively. In conclusion, n-3 FA differed greatly in their transfer efficiency mainly due to differences in their direct transfer rates. These differences pre-

sumably occur due to trafficking of very long chain n-3 FA into plasma lipid pools unavailable to the mammary gland.

Key Words: omega-3, milk fat

469 2-Hydroxy-4-(methylthio)butanoate (HMTBa) supplementation increases milk fat and decreases synthesis of alternate biohydrogenation intermediates in diets with risk for milk fat depression. Michel Baldin*[1], Yun Ying[1], Geoff I. Zanton[2], Heather A. Tucker[2], Mercedes Vazquez-Anon[2], and Kevin J. Harvatine[1], *[1]Penn State University, University Park, PA, [2]Novus International Inc., St. Charles, MO.*

We recently reported that supplementation of 2-hydroxy-4-(methylthio) butanoate (HMTBa) reduced the shift to the alternate biohydrogenation pathway and maintained higher milk fat yield in high producing cows fed diets lower in fiber and higher in unsaturated fatty acids (FA). The objective of this experiment was to verify the effect of HMTBa (Alimet, Novus International, Inc., St. Charles, MO) on biohydrogenation intermediates and milk fat synthesis. Twenty-two rumen cannulated high-producing Holstein cows [168 ± 66 DIM; 42 ± 7 kg milk/d (Mean ± SD)] were used in a randomized design performed in 2 blocks (1 = 14 cows, 2 = 8 cows). Treatments were control (corn carrier) and HMTBa (0.1% of diet DM). The experiment included a 7-d covariate period followed by 3 phases that fed diets with increasing risk of milk fat depression (MFD). The diet during the covariate and low-risk phase (7 d) was 32% NDF with no additional oil. The diet during the moderate-risk phase (17 d) was 29% NDF with 0.75% soybean oil. Soybean oil was increased to 1.5% for the last 4 d. Milk yield and DMI were measured daily. Milk was sampled every 7 d and analyzed for fat, protein and FA profile. Data were analyzed using PROC Mixed with repeated measures and the effect of treatment was tested at each time point. There was no effect of block or interaction of block and other fixed effects. There was no overall effect of treatment or treatment by time interaction for DMI ($P = 0.4$), milk yield ($P = 0.4$), and milk protein concentration ($P = 0.9$) and yield ($P = 0.6$). There was an effect of treatment, but no treatment by dietary phase interaction on milk fat with HMTBa increasing milk fat percent (3.2 vs. 3.6%, $P < 0.01$) and yield (1342 vs. 1543 g/d, $P = 0.02$). Additionally, HMTBa decreased the concentration (1.29 vs. 0.81 g/100 g of total FA, $P = 0.02$) and yield (14.7 vs. 10.9 g/d, $P = 0.01$) of *trans*-10 18:1 in milk across the entire feeding period. In conclusion, HMTBa prevented the increase in *trans* FA associated with MFD and maintained milk fat yield when cows were fed a diet with moderate risk of diet-induced MFD.

Key Words: 2-hydroxy-4-(methylthio)butanoate (HMTBa), milk fat

470 Meta-analysis of the effect of plant oils rich in 18:2n-6 on milk fatty acid composition in lactating dairy cows. Mina Vazirigohar*, Mehdi Dehghan-Banadaky, Kamran Rezayazdi, and Ardeshir Nejati-Javaremi, *Department of Animal Science, Campus of Agriculture and Natural Resources, University of Tehran, Karaj, Alborz, Iran.*

The objective of this study was to evaluate the effect of dietary plant oils rich in 18:2n-6 (POLA; including safflower, sunflower, corn, soybean and cottonseed oils) on milk fatty acid (FA) profile in lactating dairy cows, using meta-analysis and meta-regression methods. A total of 20 studies were identified with 27 comparisons between treatment and control groups, which met the selection criteria and included in the analysis. Two levels of POLA supplement (less and more than 30 g/kg diet DM) were evaluated in this meta-analysis. Supplementation of cows with 20 g POLA/kg diet DM reduced milk fat 6:0–16:0, enhanced 18:0, *cis* (Δ9–12, and 15) and *trans* 18:1 (Δ6−14), 18:2n-6, *cis*-9,*trans*-11 CLA and *trans*-10,*cis*-12 CLA concentrations. The weighted mean differences for milk fat content of 16:0, *cis*-9 18:1, 18:2n-6 and *cis*-9,*trans*-11 CLA were –5.687 (95% confidence interval (CI) = –7.558 to –3.816), 3.194 (95% CI = 1.139 to 5.249), 0.810 (95% CI = 0.384 to 1.236) and 0.467 g/100 g total FA (95% CI = 0.281 to 0.653), respectively. Inclusion of 20 g/kg DM POLA had no effect on milk 4:0 and 18:3n-3 concentrations. Results of meta-regression showed that dietary 18:2n-6 content explained more than 55% of the variation in 8:0–15:0 results. Dietary FA content, amount of grain in the basal diets and parity were the sources of heterogeneity for milk *cis*-9 18:1. Milk *trans*-11 18:1 increased with increased dietary 18:2n-6 content and forage level of basal diets. Estimated effect size of milk 18:2n-6, *trans*-10,*cis*-12 CLA and 18:0 were greater in safflower > sunflower > corn > soybean and > cottonseed oils. In conclusion, feeding 20 g POLA/kg diet DM lowered milk fat 16:0, increased *cis*-9 18:1, 18:2n-6, *cis*-9,*trans*-18 CLA, without any changes on 4:0 and 18:3n-3.

Key Words: meta-analysis, milk fatty acid, plant oil

471 Prediction of blood nonesterified fatty acid (NEFA) by FTIR analysis of individual cow milk samples. David M. Barbano*[1], Patrick Cree[3], Tom R. Overton[1], Heather M. Dann[2], and Rick J. Grant[2], *[1]Cornell University, Ithaca, NY, [2]William H. Miner Agricultural Institute, Chazy, NY, [3]Delta Instruments, Drachten, the Netherlands.*

Our objective was to develop and validate a Fourier transform mid-IR-based milk analysis method to estimate blood NEFA concentrations in lactating dairy cows. High blood NEFA indicates that a cow is mobilizing body fat and increases the risk of metabolic disorders. Milk and blood samples were collected from 60 lactating Holsteins once per week for the first 3 weeks of lactation. Cows were milked 3 times per day. Within + or – one milking of the time of blood collection, a milk sample was analyzed using a Delta Instruments (model FTA) mid-IR milk analyzer. A Wako NEFA HR test kit was used as an in vitro enzymatic colorimetric method for the quantitation of NEFA in blood serum and these values were used as reference values for development of a partial least squares (PLS) regression model to predict blood NEFA from the mid-IR milk spectra. There are no NEFA in milk, so a model to predict blood NEFA from a milk sample uses differences in the milk spectra from sample to sample that are correlated with changes in blood NEFA. The final PLS model had 9 factors, used wavelengths in the following ranges (3000 to 2800, 1800 to 1700, 1585 to 1000 cm^{-1}) with a standard error of cross validation of 172 μEq/L. Validation milk and blood sample pairs (n = 53) were collected from Holstein cows from a different herd. The mean value for the blood reference test was 713 μEq/L of serum and the mean value for the milk based blood NEFA prediction was 703 μEq/L of serum with a standard deviation of the difference (SDD) of 218 μEq/L for the 53 validation samples. Blood NEFA measured on blood is a snapshot of the NEFA concentration at an instant in time, while blood NEFA predicted from milk analysis represents a time average for the total time between milkings. The FTIR milk analysis to estimate blood NEFA is rapid (about 10 s), done simultaneously with all other milk component measures, and uses no reagents. This approach could be useful for rapid evaluation of risks of ketosis and reproductive disorders.

Key Words: blood NEFA, FTIR, milk analysis

472 Increased NEFA availability promotes plasma ceramide accumulation in Holstein cows. J. Eduardo Rico[1], Luciano S. Caixeta[2], Yves R. Boisclair[2], and Joseph W. McFadden*[1], *[1]West Virginia University, Morgantown, WV, [2]Cornell University, Ithaca, NY.*

An increase in plasma NEFA can impair insulin sensitivity in dairy cows by unknown mechanisms. In monogastrics, excess saturated fatty acids can upregulate the hepatic synthesis and secretion of ceramide, a sphingolipid that inhibits insulin action in muscle and adipose tissue. Our objective was to determine whether an increase in plasma NEFA can augment ceramide levels in cows. Six nonpregnant, nonlactating Holstein dairy cows (682 kg ± 22), were used in a crossover design with treatments consisting of i.v. infusion (100 mL/h) of either saline (control) or triacylglycerol (TG) emulsion (Intralipid 20%; Frasenius Kabi) for 16 consecutive hours. The feeding level was set at 120% of estimated energy requirement with meals offered every 2 h. Blood was collected at regular intervals and liver was biopsied at 16 h. LC/MS was used to profile 25 ceramides (Cer), monohexosylceramides (GlcCer), and lactosylceramides (LacCer) in plasma. Data were analyzed using a mixed model with repeated measures (fixed effects of treatment and time). Nonparametric correlations were analyzed. TG infusion increased plasma NEFA by 454% at 3 h relative to control ($P < 0.01$) with no further increase at 16 h. Liver TG were elevated 321% in TG vs control by end of TG infusion ($P < 0.01$). Before infusion, C24:0-Cer, C24:0-GlcCer, and C16:0-LacCer represented 52, 47, and 83%, respectively, of plasma Cer, GlcCer and LacCer. Plasma total Cer increased 149 and 235% by 3 and 16 h of TG infusion, relative to control ($P < 0.01$). Plasma C20:0-, C22:0-, C24:0-, and C26:0-Cer levels increased during TG infusion ($P < 0.01$). Although TG infusion did not modify plasma C16:0-Cer level at 16 h, C16:0-Cer abundance (% of total) was lower at 16 h ($P < 0.01$). TG infusion increased plasma C16:0-, C20:0-, C22:0-, and C24:0-GlcCer levels ($P < 0.01$). Plasma LacCer levels were not modified by TG infusion. Cer levels were positively correlated with NEFA (e.g., C24:0-Cer, r = 0.80; $P < 0.01$). Our data indicate that plasma Cer accumulation occurs in dairy cows concomitantly with increases in plasma NEFA and liver TG. Research is needed to determine whether NEFA-induced insulin resistance is mediated by Cer.

Key Words: ceramide, dairy cow, insulin resistance

473 An acute increase in circulating NEFA does not lower total plasma sphingomyelin levels in Holstein cows. J. Eduardo Rico*[1], Luciano S. Caixeta[2], Yves R. Boisclair[2], and Joseph W. McFadden[1], *[1]West Virginia University, Morgantown, WV, [2]Cornell University, Ithaca, NY.*

Insulin resistance in rodents is characterized by inflammation-mediated activation of sphingomyelinase and the transformation of sphingomyelin (SM) into ceramide (Cer), an effect provoked by saturated fatty acids. Our objective was to determine whether an acute increase in circulating NEFA lowers plasma SM levels in cows. Six nonpregnant, nonlactating Holstein dairy cows (682 kg ± 22), were used in a crossover design with treatments consisting of i.v. infusion (100 mL/h) of either saline (control) or triacylglycerol (TG) emulsion (Intralipid 20%; Frasenius Kabi) for 16 consecutive hours. The feeding level was set at 120% of estimated energy requirement with meals offered every 2 h. Blood was collected at regular intervals. LC/MS was used to profile SM in plasma. Data were analyzed using a mixed model with repeated measures (fixed effects of treatment and time). Nonparametric correlations were analyzed. TG infusion increased plasma NEFA by 454% at 3 h relative to control ($P < 0.01$) with no further increase at 16 h. Total plasma Cer concentrations increased in TG-infused cows, relative to control ($P < 0.01$). Before infusion, C16:0- and C18:1-SM represented 30 and 21%, respectively, of plasma SM. Plasma C16:0-SM levels increased 10% by 16 h of TG infusion, relative to control ($P < 0.05$). Plasma C20:1-SM concentrations were lower in TG-infused cows, relative to control ($P < 0.05$). Plasma C16:1-, C18:0-, C18:1-, C22:0-, C24:0-, and C24:1-SM, as well as total plasma SM were not modified with TG infusion. Before infusion, C20:0-dihydro-SM (DHSM) represented 89% of total plasma DHSM. Plasma C20:0- and C18:0-DHSM increased 79 and 14%, respectively, by 16 h of TG infusion, relative to control ($P < 0.01$). Plasma C18:1- and C20:1-SM were negatively correlated with NEFA (e.g., C20:1-DHSM, r = −0.47; $P < 0.01$). In contrast, plasma C18:0- and C20:0-DHSM, as well as total DHSM were positively correlated with NEFA (e.g., C20:0-DHSM, r = 0.77; $P < 0.01$). An acute increase in plasma NEFA does not appear to promote sphingomyelin hydrolysis in dairy cows thus suggesting that plasma Cer accumulations occurred due to enhanced de novo Cer synthesis.

Key Words: dairy cow, insulin resistance, sphingomyelin

474 Temporal changes in plasma sphingolipids during the transition from pregnancy to lactation in Holstein cows. J. Eduardo Rico*, Sina Saed Samii, Alice T. Mathews, and Joseph W. McFadden, *West Virginia University, Morgantown, WV.*

Ceramide (Cer) and sphingomyelin (SM) are sphingolipids associated with conserved cellular processes; however, their involvement in the homeorhetic adaptation to lactation in cows is unknown. Our objectives were to characterize temporal responses in plasma sphingolipids during the periparturient period and determine whether these changes were related to adiposity and lipolysis. Multiparous Holstein cows were grouped by BCS at d −28 prepartum: lean (BCS 2.9 ± 0.13; n = 7) or overweight (OVER; BCS 4.0 ± 0.21; n = 7), fed a balanced diet. Blood samples were collected routinely from d −21 to 21. LC/MS was used to profile 37 Cer, monohexosylceramides (GlcCer), lactosylceramides (LacCer), and SM in plasma. Data were analyzed as repeated measures under a mixed model (fixed effects of BCS and day). Nonparametric correlations were analyzed. Plasma NEFA increased around calving and was higher in OVER from d −5 to 21 ($P < 0.05$). OVER lost more BCS and BW ($P < 0.01$). C16:0-Cer concentration decreased by 40% during transition and was higher in OVER prepartum ($P < 0.05$). C24:0-Cer increased 50% and was 33% higher in OVER at d 21 ($P < 0.05$). C22:0-Cer increased 49 and 66% at d 14 and 21, respectively, in OVER ($P < 0.05$). C24:0-GlcCer increased 52% during transition ($P < 0.05$) and tended to be higher in OVER at d 0 ($P < 0.1$). C16:0-GlcCer increased by 145% at d 4 ($P < 0.01$). C22:0-GlcCer increased over time and was higher in OVER at d 14 ($P = 0.05$). C24:1-GlcCer tended to increase by 100% in OVER during transition ($P < 0.1$). C16:0-LacCer increased postpartum and peaked at d 14 ($P < 0.01$). C18:1- and C24:1-LacCer levels increased during transition and were higher in OVER from d 4 to 21 ($P < 0.01$). Abundant C16:0-, C18:1- and C20:1-SM decreased over time and reached nadir around parturition ($P < 0.01$). C18:1- and C20:1-SM were higher in OVER pre- and postpartum ($P < 0.05$). C20:0-dihydro-SM decreased around calving ($P < 0.01$) and was higher in OVER at d −5 ($P < 0.05$). Cer, GlcCer and LacCer were positively correlated to NEFA and negatively to SM during transition ($P < 0.001$). Our results support a model of NEFA-induced ceramide synthesis during peripartum.

Key Words: dairy cow, transition, sphingolipid

475 Elevations in milk yield from palmitic acid feeding are associated with reduced estimated insulin sensitivity and glucose-stimulated NEFA disappearance. Alice T. Mathews*, J. Eduardo Rico, Neil T. Sprenkle, and Joseph W. McFadden, *West Virginia University, Morgantown, WV.*

The ability of saturated fatty acids (SFA) to enhance milk yield in dairy cows may be due to shifts in glucose utilization by reducing insulin sensitivity in adipose and muscle tissues. Our objective was to evaluate the effects of palmitic acid (PA) on milk production and insulin sensitivity in cows. Twenty multiparous mid-lactation Holstein cows were enrolled in a 68-d study consisting of 3 sequential periods: 5-d covariate, 49-d treatment, and 14-d post. All cows received a sorghum silage-based diet pre- and post-treatment, and were randomly assigned to a common diet (control; no fat, n = 10; 138 ± 45 DIM) or PA at 4% of ration DM (98% PA; Palmit 98; Global Agri-Trade; n = 10; 136 ± 44 DIM). Blood and milk were collected at routine intervals. Intravenous glucose challenges (0.3 g/kg BW; GTT) were performed at d −1, 21, and 49 relative to start of treatments. Data were analyzed as repeated measures using a mixed model (fixed effects of treatment and time), and milk yield served as a covariate. Effects of PA are presented as changes relative to control. PA increased milk yield by wk 7 (30.8 vs. 24.6 kg/d, $P < 0.05$). PA increased milk fat yield (+22 and +18%; $P < 0.05$) and 3.5%-FCM (+19 and +18%; $P < 0.05$) by wk 3 and 7, respectively. PA had no effect on milk protein or lactose content, MUN, SCC, or BW, but increased milk protein yield by wk 7 ($P = 0.05$). PA tended to increase estimated energy balance by wk 7 ($P = 0.06$). PA increased feed efficiency (+10%; 3.5%-FCM/DMI; $P < 0.05$). By d 68, 3.5%-FCM remained elevated in PA cows ($P < 0.05$), while milk yield and components were not different. PA increased plasma NEFA by 48, 93, and 60% by d 4, 6, and 8 ($P < 0.05$), respectively, and had no effect thereafter. PA did not modify plasma insulin or glucose, but reduced estimated insulin sensitivity wk 1 (e.g., −35% on d 8; $P < 0.01$). PA did not modify glucose disposal following GTT; however, PA reduced glucose-stimulated NEFA disappearance by wk 7 ($P < 0.05$). Our data suggest that increased milk yield with PA supplementation may be due in part to alterations in insulin sensitivity.

Key Words: insulin sensitivity, milk production, palmitic acid

476 Effects of timing of chromium propionate supplementation on metabolic and production responses of Holstein cows in early lactation. Michael S. Allen* and Richard Longuski, *Michigan State University, East Lansing, MI.*

Forty-eight multiparous Holstein cows were used in a randomized block design experiment with 4 treatments and 12 cows per treatment to determine effects of timing of chromium propionate (CrPr) supplementation on metabolic and production responses in early lactation. Treatments were chromium propionate (C) or control (N), supplemented according to 4 schedules: 1 to 65 d PP (CC); 1–22 d PP (CN); 23–65 d PP (NC); no CrPr supplementation (NN). Supplements were top-dressed at 20 g per cow/d to provide 8 mg of chromium per cow/d for C (KemTRACE Chromium, 0.04% chromium, Kemin Animal Nutrition and Health) or no additional chromium for N (feed-grade limestone). Cows were blocked by calving date, body condition score (BCS) and previous milk yield, and randomly assigned to treatment. Cows were offered a fresh diet (FR) from 1 to 22 d PP and a peak diet (PK) from 23 to 68 ± 3 d PP. CrPr increased daily DMI (2.75 kg/d) and milk yield (4.5 kg/d, both $P < 0.01$), tended to decrease milk fat concentration ($P = 0.07$), and did not affect fat yield compared with control when supplemented during FR only. There were no interactions of the main effects of timing of CrPr supplementation; effects of CrPr supplementation were sustained through most of the experiment when supplementation ceased at 22 d PP and starting supplementation at 23 d PP did not benefit production. Although CrPr supplementation during FR increased milk yield, the effect diminished by 68 d PP, even when supplementation continued during PK. Plasma metabolites and hormones indicate possible effects of CrPr on insulin sensitivity with a reduction in plasma concentrations of NEFA ($P = 0.08$) and BHBA ($P = 0.02$), lower milk fat concentration ($P = 0.07$), and greater BCS over time (interaction $P = 0.06$) during FR compared with control. Supplementation of CrPr during FR increased DMI and milk yield but effects on production diminished over time with no effects of CrPr by 10 weeks PP, even when supplementation continued during PK. Initiating supplementation at 23 d PP did not benefit production in this experiment.

Key Words: postpartum, chromium, insulin sensitivity

477 The effects of dietary niacin supplementation on FoxO1 and genes involved in hepatic glucose production in dairy cows during the transition period. Asako Kinoshita*[1], Kathrin Hansen[3], Lena Locher[1], Ulrich Meyer[2], Sven Dänicke[2], Korinna Huber[3], and Jürgen Rehage[1], *[1]Clinic for cattle, University of Veterinary Medicine Hannover, Hannover, Lower Saxony, Germany, [2]Institute of Animal Nutrition, Friedrich-Loeffler-Institute, Braunschweig, Lower Saxony, Germany, [3]Department of Physiology, University of Veterinary Medicine Hannover, Hannover, Lower Saxony, Germany.*

Forkhead box protein O1 (FoxO1) promotes the hepatic glucose production (HGP) by activating the transcription of gluconeogenic enzymes. As a main target of insulin signaling, FoxO1 is phosphorylated by insulin, leading to inhibition of HGP. Dietary niacin supplementation could affect HGP; for example, by modifying the expression of genes or by inhibiting lipolysis. The objective of this study was to investigate the effects of dietary niacin supplements on protein expression of hepatic FoxO1 and mRNA expression of genes involved in HGP in dairy cows during the transition period. Twenty-one pluriparous German Holstein cows were used for 2 × 2 factorial analysis. Cows received diets containing 0 g (C) or 24 g (N) niacin supplementation from −42 d related to calving (d −42) to d 24. Each group was further divided and received diets with 30% (L) or 60% (H) of concentrate on dry matter basis from d-42 to d0. Dietary concentrate proportion was set to 30% at d 0 for all, and then increased up to 50% within 16 d for CL and NL and within 24 d for CH and NH. Liver biopsies were taken at d −42, d 3, and d 21. Protein expression of FoxO1 and phosphorylated FoxO1 at serine 256 (pFoxO1) was measured semiquantitatively by Western blotting. Real-time RT-PCR was performed to measure the mRNA of FoxO1, glucose-6-phosphatase (G6P), pyruvate carboxylase (PC), cytosolic phosphoenolpyruvate carboxykinase (PCK1), propionyl CoA carboxylase (PCCA), glucose transporter 2 (GLUT2). Data were evaluated by mixed model for repeated measures to test the effect of time, niacin and prepartal concentrate proportion. The protein and mRNA expression of FoxO1 and the protein expression of pFoxO1 were affected neither by time nor by diet. In cows fed with niacin, the relative quantities of mRNA of G6P, GLUT2, PCCA were higher at d 21 and that of GLUT2 was lower at d 1 ($P < 0.05$). Dietary niacin supplements altered the gene expression, increasing HGP in the transition period of cows. However, the regulation of HGP by FoxO1 seemed to be of less importance on the levels of mRNA, protein and phosphorylation.

Key Words: FoxO1, hepatic glucose production, cow