

Ruminant Nutrition: Dairy Production I

152 Liver gene expression patterns can explain accumulation of lipid in the liver during the transition period. H. R. Khazanehei,* P. Eck, A. Regassa, D. O. Krause, and J. C. Plaizier, *University of Manitoba, Winnipeg, MB, Canada.*

The effects of 2 dry-period managements of dairy cows on the expression of genes related to lipid metabolism in the liver were compared. Twenty-four multiparous cows were paired based on expected calving date, and randomly assigned to treatment within each pair. Treatments were a 60-d dry period (60 d) with separate far-off and close-up diets and a 40-d dry period (40 d) with only close-up diet. The 60-d dry period was divided into a 39-d far-off and a 21-d close-up period. The far-off diet contained 1.28 Mcal/kg net energy for lactation (NEL), 14.7% of crude protein (CP), and 50% of neutral detergent fiber (NDF) on a DM basis. The close-up diet contained 1.43 Mcal/kg NEL, 14.6% of CP, and 38% of NDF. A common diet was fed to all cows after calving, which contained 1.69 Mcal/kg, 17.6% of CP, and 31% of NDF. Liver biopsies were obtained at wk -3, 1 and 4 relative to parturition. Differential gene expression was assessed by Affymetrix microarray analysis and data were normalized through RMA algorithm and then statistically analyzed using FlexArray 1.6.1. An FDR-adjusted P-value lower than or equal to 0.1 and a fold-change greater than 2 were considered as a cut-off point to indicate significant up- or downregulation of genes. Gene networks were assembled using Ingenuity Pathway Analysis. A parallel study showed that lipid accumulation in the liver was greater at wk 1 than and wk -3 and wk 4, and at wk 1, this accumulation was greater for the 40 d than for the 60 d treatment. There was no difference in gene expression between the treatments across the sampling days. Genes involved in lipid accumulation and triacylglycerol synthesis in the liver were upregulated at wk 1. Also mitochondrial fatty acids transporters aiding β -oxidation were upregulated, and genes involved in hydroxylation of fatty acids aiding β -oxidation were downregulated at wk 1. Gene expression at wk 4 was similar as at wk -3. The changes in lipid deposition during the transition period are reflected in changes in the expression of genes related to lipid metabolism in the liver, but the difference between the 40 d and 60 d treatments is not.

Table 1. Comparison of expression of genes involved in liver lipid metabolism between week 3 before and week 1 after calving

Genes	Fold-change +1/-3	
	40-d treatment	60-d treatment
Beta-oxidation		
PPARGC1	1.03‡	1.03†
CPT1B	2.26‡	2.51‡
SLC22A5	1.27‡	1.27‡
Fatty acid hydroxylation		
CYP2C9	-2.01‡	-1.25‡
CYP2E1	-1.43‡	-
CYP2U1	-1.26‡	-1.11‡
Lipid accumulation		
PLIN2	1.47†	1.39†
ELOVL5	1.08‡	1.15‡
ACSL1	1.21‡	1.17‡
GHR	-1.12‡	-1.08‡
APOA1	2.83‡	2.54‡

† $P < 0.1$, ‡ $P < 0.05$.

Key Words: dry period, gene expression, lipid metabolism

153 Effects of nutrition, ketosis, and inflammation on hepatokine and nuclear receptor expression in liver of periparturient Holstein dairy cows. H. Akbar,* J. M. Khan, D. B. Carlson, J. K. Drackley, and J. J. Loor, *University of Illinois, Urbana.*

In rodents, fibroblast growth factors 21 (FGF21) has emerged as a metabolic regulator produced by liver in response to different physiological conditions that regulate glucose and lipid metabolism. Its upregulation induces glucose uptake in adipose tissue and skeletal muscle; whereas in liver it increases hepatic fatty acid oxidation, reduces triacylglycerol accumulation, and stimulates ketogenesis. The regulatory mechanism and action of FGF21 in ruminants remain unclear. We sought to investigate the mRNA expression of FGF21, FGF receptors, the hepatokine ANGPTL4, and genes involved in regulation of fatty acid oxidation (CPT1A, PPARA) under different nutritional conditions including early postparturient ketosis (KET), periparturient dietary L-carnitine supplementation (CAR), and preparturient level of dietary energy. We also examined hepatic FGF21 in response to intramammary lipopolysaccharide (LPS) challenge at 7 d postparturient. In CAR liver biopsies were harvested at d 2 and d 10 postparturient in cows assigned to control (n = 5), 100 g CAR/d (C100, n = 5), and 200 g CAR/d (C200, n = 4). In KET, liver biopsies were obtained at d 14 postparturient from control (n = 7) and feed-restricted induced ketotic (n = 7) cows. In the LPS study, cows (n = 6/treatment) were fed during the dry period either moderate-energy (M) or control energy (S) and received 0 (M:N, S:N) or 200 μ g LPS (M:Y, S:Y) at 7 d postparturient. Liver biopsies were harvested at d 7 and d 14 postparturient. Expression of FGF21 was analyzed via RT-PCR. There was an interaction effect ($P < 0.05$) of CAR for FGF21 expression due primarily to lower expression at d 10 with C100 and C200 vs. control. The opposite effect was observed for ANGPTL4 expression (a hepatokine) at d 2 which was greater with C100 and C200 vs. control. Subsequently, a marked decrease by d 10 was observed with C200 at which point ANGPTL4 was lower than C100 and controls. The significant interaction ($P < 0.05$) for PPARA expression was due to a marked increase at d 10 with C100. A similar response to C100 was observed for CPT1A and KLB expression. Ketosis induced the expression of FGF21 ($P < 0.05$), KLB ($P < 0.05$) and FGFR1 ($P = 0.08$); whereas, no effect was observed for the expression of FGFR2 and KL. In the LPS study, expression of FGF21 was induced ($P < 0.05$) with both M:Y and S:Y at d 7 but the response was more marked with S. By 14 d expression decreased markedly with S:N but remained nearly unchanged with other treatments. Preparturient M led to greater ($P = 0.06$) overall ANGPTL4 postparturient. Overall, results indicate that in transition cows both L-carnitine and inflammatory challenge regulate FGF21 expression likely through their effects on fatty acid oxidation and ketogenesis, i.e., L-carnitine enhances and inflammation inhibits both processes. Ketosis could upregulate the expression of FGF21 likely via incoming nonesterified fatty acids and potentially via PPAR α . However, the role of PPAR α on FGF21 activation remains unclear because of the inverse response observed with C100.

Key Words: metabolism, transition cow

154 Effects of a moderate-energy diet during the close-up dry period on immunometabolic indices in periparturient dairy cows. J. S. Osorio*¹, E. Trevisi², P. Ji¹, J. K. Drackley¹, G. Bertoni², and J. J. Loor¹, ¹University of Illinois, Urbana, ²Università Cattolica del Sacro Cuore, Piacenza, Italy.

The periparturient period is characterized by marked changes in hormonal, metabolic, and immune/stress-like conditions all of which may

contribute to regulating dry matter intake (DMI) and the supply of nutrients to mammary gland. Twenty 3 multiparous Holstein cows were fed a control diet (CE, $n = 10$; 1.24 Mcal/kg DM; high-straw) during the entire dry period (ca. 50 d) or were switched to a moderate-energy (ME, $n = 13$; 1.47 Mcal/kg DM) diet during the last 21 d prepartum through calving. All cows were fed a common lactation diet (1.67 Mcal/kg DM postpartum). Blood samples were collected at -21, -10, 7, 14, and 21 DIM for profiling of 21 markers of liver function, metabolism, oxidative stress, and inflammation. Concentration of cholesterol (CHOL) and the negative acute-phase protein albumin (ALB) decreased (time $P < 0.05$) through calving but CHOL increased markedly between 7 and 21 DIM regardless of treatment. Paraoxonase concentration through calving followed a similar pattern (time $P < 0.01$) as CHOL and ALB; however, concentration remained unchanged from 7 to 21 DIM. Glutamic-oxaloacetic transaminase (GOT) and bilirubin (BIL) concentration increased (time $P < 0.01$) from -21 to 7 DIM regardless of treatment after which it decreased. Concentration of gamma-glutamyl transpeptidase (GGT) tended (treatment \times time $P = 0.11$) to differ due to greater concentration with ME after 7 DIM. The concentration of haptoglobin, an inflammation marker, increased (time $P < 0.01$) between -21 and -10 DIM after which it remained stable through 7 DIM and decreased gradually by 21 DIM regardless of treatment. In contrast, concentration of ceruloplasmin, another inflammatory marker, increased (time $P < 0.01$) linearly between -21 and 14 DIM regardless of treatment. The concentration of reactive-oxygen metabolites (ROM; diet $P = 0.08$) and urea (diet $P = 0.07$) was greater overall with CE. For ROM, differences between diets were evident at 7 and 14 DIM. The marked increase in haptoglobin at 7 DIM with CE agrees with that of ROM and ceruloplasmin. Overall, preliminary data provide some evidence that plane of dietary energy during the close-up dry period alters immunometabolism.

Key Words: inflammation, nutrition, metabolism

155 Integrating control by gene expression in adipose tissue into a mechanistic, dynamic model of metabolism to investigate the biological basis for variation in genetics of feed conversion efficiency in lactating dairy cattle. S. Shields* and J. McNamara, *Washington State University, Pullman*.

Variation in efficiency of feed is a multiple function of metabolic flux in visceral, muscle, and adipose tissues. These processes are affected by genotype, phenotype, and intake, and are under control of hormonal and neural systems. To help identify the patterns of metabolic flux in the most efficient dairy cattle, an existing mechanistic metabolic model (Molly, UC Davis) was used, which explicitly includes elements of genetics, including metabolic interactions in the viscera and body. Our present objective was to integrate changes in gene transcription in adipose tissue into control elements in the model. Data were collected from 2nd to 4th parity cows ($n = 120$) in the first 4 mo postpartum, from studies that measured nutrient intake, milk component output, changes in adipose tissue lipid, visceral and body protein and lipid, and metabolism rates and gene expression in adipose tissue. Explicit inputs into the model included nutrient intake, initial body fat and protein, milk production, fat, and protein output. Visceral energy use averaged 37% of intake energy (range 33 to 46%) and 68% of maintenance energy (range 63 to 73%). The variation in maintenance energy accounted for 37.6% of the variation in milk energy efficiency (milk energy/absorbed energy). Visceral and lean body energy use were the 2 major contributors ($P < 0.05$) to variation in milk production efficiency. Expression of several genes coding for metabolic enzymes in adipose tissue measured by transcriptomic arrays were related to efficiency of milk production. The amount of gene transcripts that control lipolysis including the ADRB2

and LIPE, accounted for 10 to 15% of variation in efficiency ($P < 0.02$). Transcripts of genes controlling lipogenesis, including SREBP, TSHSP14, LPL, and ACACA accounted for 15 to 18% of the variation in efficiency ($P < 0.05$). These results confirm some key metabolic control points that can be targeted for further research to define the genotypic and phenotypic control of metabolic efficiency in dairy animals.

Key Words: systems biology, transcriptomics, metabolic model

156 Dietary manipulation of crude protein and starch content affects energy balance in early lactation dairy cows. S. J. Whelan*^{1,3}, F. J. Mulligan², V. Gath², B. Flynn³, and K. M. Pierce¹, ¹*School of Agriculture and Food Science, University College Dublin, Belfield, Dublin 4, Ireland*, ²*School of Veterinary Medicine, University College Dublin, Belfield, Dublin 4, Ireland*, ³*University College Dublin Lyons Research Farm, Newcastle, Dublin, Ireland*.

During early lactation, high levels of milk production and low dry matter intake result in a state of negative energy balance (NEB) and a metabolic status that can be detrimental to the health and longevity of the dairy cow. Reducing dietary CP and providing glucogenic nutrients have been shown to alter energy balance (EB) and metabolic status when employed as separate dietary strategies. However, simultaneous employment of these strategies has yet to be reported. This experiment investigated whether or not offering diets low in CP and high in starch (LP-HS) can reduce NEB and improve the metabolic status of the early lactation dairy cow compared with a higher CP low starch diet (HP-LS). Ten Holstein Friesian dairy cows were assigned to each dietary treatment in a randomized block design. Diets were isoenergetic (1.79 Mcal NE/ kg DM) and formulated to contain 119 and 275 g/kg DM of CP and starch respectively (LP-HS); or 146 and 57 g/kg DM of CP and starch respectively (HP-LS). The experiment was conducted from d 0 to 63 of lactation. Energy balance was calculated daily and blood samples harvested weekly for metabolite analyses; data were then analyzed using Proc Mixed of SAS. Intake of DM (17.4 ± 0.5 kg/ d, $P = 0.25$) and energy (31.6 ± 1.02 Mcal NE/ d, $P = 0.22$) were not affected by diet. The HP-LS group had a higher milk yield (31.6 vs. 29.2 ± 0.6 kg/ d, $P < 0.01$) versus the LP-HS group. However, the LP-HS group had a more positive EB (2.2 vs. -0.7 ± 1.02 Mcal NE/ d, $P = 0.02$) when compared with the HP-LS group. Blood glucose (3.2 ± 0.1 mmol/ L, $P = 0.36$) was not affected by diet. However, blood urea N (3.5 vs. 1.8 ± 0.2 mmol/ L, $P < 0.01$) was higher, while β hydroxy butyric acid (0.7 vs. 0.8 ± 0.1 mmol/ L, $P = 0.03$) was lower for the HP-LS group compared with the LP-HS group. These data suggest that limiting milk production through offering a low CP, high starch diet reduces both the degree of and length of NEB in early lactation dairy cows. Such strategies may be useful in reducing production diseases associated with severe NEB on dairy farms.

Key Words: dairy cows, early lactation, energy balance

157 Colostrum yield by multiparous cows is positively correlated with prepartum body fat mobilization. N. Litherland,* W. Weich, D. Lobao, and Z. Sawall, *University of Minnesota, St. Paul*.

One hundred and 20 Holstein and Holstein-cross multiparous dairy cows from 2 studies in 2010 and 2011 were used in a correlation analysis to determine if prepartum nutrition, metabolism, and management factors affect colostrum yield (CY). Cows were fed moderate-energy high-forage diets containing corn silage, wheat straw, alfalfa hay, ground corn, and molasses and protein supplements. Prepartum diets averaged 14.5% CP, 40.5% NDF, 1.5 Mcal NEL/kg DM, 16.8% starch, 6.0% sugar,

2.0% fat. Cows averaged 42.7 ± 8.1 d dry, 13.6 ± 0.9 kg DMI/d, 3.1 ± 1.1 lactations and postpartum $305\text{ME} = 10,516 \pm 2,203$ kg. All cows were from the St Paul Dairy Research Unit and were managed similarly. Colostrum yield was not significantly affected by treatment or by study. Samples were collected and analyzed using the same techniques for both studies. Data were pooled and subjected to Pearson Correlation analysis in SAS. Correlation coefficients and *P*-values were calculated among CY, parity, days dry, calf birth weight, average prepartum DMI, crude protein intake (CPI), neutral detergent fiber intake (NDFI), sugar intake (SI), starch intake (STI), mature equivalent 305 milk yield (305 ME), pre- and postpartum serum NEFA concentration, and pre- and postpartum liver triglyceride concentration. Colostrum yield averaged 7.9 ± 4.2 kg with a minimum yield of 0.91 kg and maximum yield of 21.6 kg. Twenty-six percent of cows produced <5.0 kg colostrum. Colostrum yield tended to be correlated ($r = 0.19$; $P = 0.06$) with calf birth weight. Colostrum yield was positively correlated with serum NEFA 7 d prepartum ($r = 0.40$; $P < 0.05$), serum NEFA 1 d postpartum ($r = 0.53$; $P < 0.05$), serum NEFA 7 d postpartum ($r = 0.29$; $P < 0.05$), serum NEFA 14 d postpartum ($r = 0.19$; $P < 0.05$). Colostrum yield was positively correlated with liver triglyceride concentration 7 d postpartum ($r = 0.28$; $P < 0.05$) and 14 d postpartum ($r = 0.24$; $P < 0.05$). Prepartum nutrient intake; DMI, CPI, NDFI, SI, and STI were not significantly correlated with CY. Colostrum yield was not significantly correlated with 305ME ($r = 0.07$; $P = 0.6$).

Key Words: colostrum yield, NEFA, transition cow

158 A starch-binding agent decreases the in vitro rate of fermentation of wheat. F. R. Dunshea^{*1}, V. M. Russo¹, I. Sawyer², and B. J. Leury¹, ¹Melbourne School of Land and Environment, The University of Melbourne, Parkville, Victoria, Australia, ²Feedworks Pty Ltd., Lancefield, Victoria, Australia.

The rapid rumen fermentation of the starch in wheat can result in sub acute ruminal acidosis (SARA) with resultant inhibition of rumen function and loss of milk production in dairy cattle. The aim of the present study was to investigate the effect of a starch binding agent (Bioprotect, RealisticAgri, Ironbridge, UK) on the rate of in vitro gas production. The active ingredient in Bioprotect is a stable non-volatile organic salt that complexes with the hydroxyl groups of starch at neutral or slightly acidic conditions (pH 6 to 7), as observed in the rumen. These complexes decompose under more acidic (pH 2 to 3) conditions as in the abomasum and duodenum, making the starch available for enzymatic digestion. Wheat and maize were ground and passed through a 1 mm sieve. A subsample of the ground wheat was treated with Bioprotect (8 mL/kg). Samples (1.0 g) of the grain ($n = 6$ for each grain) were added to serum flasks containing buffered rumen fluid obtained from lactating dairy cows. The flasks were purged with carbon dioxide and maintained in a shaking water bath at 39°C. Gas production was monitored every 5 min for 48 h using the ANKOM wireless in vitro gas production system. Gas production was modeled using a Gompertz equation to determine the rate and maximum amount of gas production (R_{max}) and the rate constant (β). The R_{max} was slightly higher for the control wheat than the treated wheat and the maize (134 vs 129 and 130 mL/g for wheat, treated wheat and maize, $P = 0.05$). The β was greater for the control wheat than the treated wheat, which in turn was greater than for maize (0.267 vs 0.207 and 0.173 min^{-1} , $P < 0.001$). These data demonstrate that wheat is fermented faster than maize and that Bioprotect can slow the fermentation of wheat. If more starch can pass through the rumen without being fermented it may reduce the incidence of SARA and allow for greater post-ruminal enzymatic digestion of starch.

Key Words: wheat, fermentation, starch

159 Effects of intrajugular glucose infusion and dietary protein concentration on feed intake, milk yield and metabolic responses of postpartum cows. W. E. Brown^{*} and M. S. Allen, Michigan State University, East Lansing.

Effects of glucose infusion on feed intake, milk production, and metabolic responses of postpartum cows fed a low or high protein diet were evaluated utilizing a randomized complete block design with a 2×2 factorial arrangement of treatments. Twenty-four multiparous Holstein cows were blocked by BCS and 305-ME milk production, and randomly assigned at calving to one of 4 treatments. Treatments were continuous intrajugular infusion of glucose (GLU, 1 kg/d as 4 L/d of 25% w/v dextrose) or isotonic saline (SAL, 4 L of 0.9% sodium chloride), and diets containing 13.9% (LP) or 18.4% (HP) crude protein. Treatments were initiated at the first scheduled feeding following parturition and lasted 12 d. Data for DMI was collected daily, blood was collected every 2 d, milk yield was collected at each milking, and milk samples were obtained on d 4, 8, and 12. Data were analyzed by ANOVA, with repeated measures when applicable. The GLU infusion reduced cumulative DMI for HP (164.0 vs. 196.5 kg) but not LP (194.2 vs. 189.7 kg) compared with SAL (interaction $P = 0.04$) and tended (interaction $P = 0.12$) to reduce daily DMI for HP (13.6 vs. 16.4 kg/d) but not LP (16.4 vs. 15.9 kg/d). The GLU infusion did not affect cumulative milk yield for HP (441.6 vs. 437.8 kg) but increased milk yield for LP (469.1 vs. 395.0 kg) compared with SAL (interaction $P = 0.02$). The HP treatment tended to increase loss of body condition from 0.65 to 0.82 BCS units/12 d ($P = 0.06$) compared with LP. Consistent with this, HP increased plasma concentrations of NEFA (1184 vs. 895 $\mu\text{Eq/L}$, $P = 0.01$) and BHBA (13.0 vs. 9.5 mg/dL, $P = 0.03$), milk fat concentration (5.71 vs. 4.72%, $P < 0.01$) and yield (2.14 vs. 1.78 kg/d, $P < 0.01$) and 3.5% fat-corrected milk (50.9 vs. 45.1 kg, $P = 0.02$) compared with LP. Greater dietary protein concentration increased milk fat concentration and yield by mobilization of lipid reserves resulting in greater FCM yield. Furthermore, dietary protein concentration interacted with glucose supply to affect feed intake, milk yield and metabolic responses of postpartum cows.

Key Words: transition cow, glucose infusion, dietary protein

160 Effects of feeding moderate-energy high-forage diets with reduced DCAD for 21 or 42 days prepartum on mineral homeostasis and postpartum performance by multiparous dairy cows. W. D. Weich^{*1}, E. Block², and N. B. Litherland¹, ¹University of Minnesota Department of Animal Science, St. Paul, ²Church and Dwight Co. Inc., Arm and Hammer Animal Nutrition, Princeton, NJ.

Objectives were to determine 1) if lowering DCAD is beneficial for cows fed moderate-energy high-fiber (MEHF) diets, 2) if feeding a low DCAD MEHF diet for 21 or 42d prepartum affects postpartum performance and mineral homeostasis differentially. Holstein and Holstein-cross cows were randomly assigned to one of 3 treatments ($n = 20$) 42d before expected calving date. Treatments included: 1) CON, DCAD = +12 mEq/ 100 g DM, 2) 21-ND, DCAD = +12/-16 mEq/ 100 g DM, 3) 42-ND, DCAD = -16 mEq/100 g DM, with treatments containing 38.6% corn silage, 22.8% wheat straw, 17.6% protein mix, 12.1% alfalfa, 6.0% molasses supplement and 2.9% ground corn on a DM basis. Prepartum diets were similar in nutrient composition, averaging 17.0% CP, 42.0% NDF and 1.5 Mcal/kg DM. High and low DCAD diets were created using 2 isonitrogenous protein mixes: 1) 97.5% soybean meal and 2) 52.8% Bio-Chlor®, 45.8% soybean meal. CON was fed high DCAD prepartum for 42d. 21-ND received high DCAD for the first 21d of the dry period, then switched to low DCAD until calving. 42-ND received low DCAD for 42d prepartum. After calving cows received a common lactation diet (16.6% CP, 31.6% NDF, 1.7 Mcal/kg DM). Urine pH for 21-ND and 42-ND were lower ($P < 0.01$) compared with CON, demonstrating effectiveness of low DCAD.

Pre- and postpartum data were analyzed separately using Mixed models in SAS with repeated measures. Pre- and postpartum DMI was similar among treatments. Milk yield by 21-ND and 42-ND were higher ($P < 0.05$) than CON on wk 2 and 3 and tended ($P < 0.10$) to be greater on wk 1. Serum ionized Ca was similar among treatments. 21-ND and 42-ND had higher ($P < 0.05$) total serum Ca compared with CON at 72h postpartum, and tended ($P < 0.10$) to be higher at 12h postpartum. Biopsies of liver on d -14, 7 and 14 relative to calving had similar contents of triglyceride and glycogen among treatments. Serum concentration of NEFA increased after calving but were similar among treatments. These data suggest low DCAD in MEHF diets during a 21 or 42d transition period had positive effects on postpartum total Ca and milk yield.

Key Words: blood Ca, negative DCAD, transition cow

161 Comparison of methane prediction for pasture fed dairy cows using a simulation model (Molly) incorporating revised VFA stoichiometry and microbial pools. J. McNamara*¹, P. Buekes², P. Gregorini², M. Hanigan³, and G. Waghorn², ¹Washington State University, Pullman, ²Dairy New Zealand, Hamilton, New Zealand, ³Virginia Tech University, Blacksburg.

This study compared the outcomes of 3 versions of DairyNZ's metabolic model, Molly, with 2 different VFA stoichiometry constructs, to describe ruminal VFA pattern and methane (CH₄) production (g/d). The models outputs were validated using 30 observations from 30 dairy cattle (82 DIM, 17 kg milk/d) fed fresh cut ryegrass (DMI 12.3 kg DM) in respiration chambers. The model versions were DairyNZ Molly4 (similar to Baldwin, 1995); Molly84, which includes updated ruminal fiber digestive parameters and animal hormonal parameters (Hanigan et al., J. Dairy Sci.); and Molly85, a revised version of Molly84 with new digestive and rumen parameters. The original forage diet VFA construct was compared with a new VFA stoichiometry based on a more recent and larger set of data, including lactate and valerate production and, amylytic, cellulolytic bacteria, as well as protozoal pools. Average observed CH₄ production was 266 (SD 30) (g/d). Mean predicted values for CH₄ production were 287 and 258 (g/d) for Molly4 without and with the new VFA construct, respectively. Molly84 predicted 295 and 288 (g CH₄/d) with and without the new VFA construct ($P > 0.5$) respectively. Molly85 predicted the same CH₄ production (276 g CH₄ /d) with or without the new VFA construct. The incorporation of the new VFA construct did not consistently reduce the mean relative prediction error (RPE %) across the versions of Molly evaluated in the present study. The improvements in the Molly versions from 4, 84 to 85 resulted in a decrease in RPE from 8.6, 8.3 to 4.3%, ($P < 0.05$) respectively. The majority of the root mean square prediction error was apportioned to random bias, e.g., 43, 71 and 70% in Molly4, 84 and 85, respectively. The slope bias was 2% in all cases. It is concluded that DairyNZ present version (Molly85) has the capability to predict CH₄ production of pasture fed dairy cows with acceptable accuracy.

Key Words: metabolic cow model, methane prediction, VFA

162 Effects of dry period management and time relative to calving on the expression of genes involved in carbohydrate metabolism in the liver. H. R. Khazanehei,* P. Eck, A. Regassa, D. O. Krause, and J. C. Plaizier, University of Manitoba, Winnipeg, MB, Canada.

The effects of a 60-d dry period with far-off and close-up diets and a 40-d dry period during which only a close-up diet was fed on the expression

of genes involved in carbohydrate metabolism were compared. Twenty-four multiparous cows were paired based on expected calving date, and randomly assigned to treatment within each pair. Treatments were a 60-d dry period (60 d) with separate far-off and close-up diets and a 40-d dry period (40 d) with only close-up diet. The 60-d dry period was divided into a 39-d far-off and a 21-d close-up period. The far-off diet contained 1.28 Mcal/kg net energy for lactation (NEL), 14.7% of crude protein (CP), and 50% of neutral detergent fiber (NDF) on a DM basis. The close-up diet contained 1.43 Mcal/kg NEL, 14.6% of CP, and 38% of NDF. A common diet was fed to all cows after calving, which contained 1.69 Mcal/kg, 17.6% of CP, and 31% of NDF. Liver biopsies were obtained at wk -3, 1 and 4 relative to parturition. Differential gene expression was assessed by affymetrix microarray analysis and data was normalized through RMA algorithm and then statistically analyzed by EB (Wright; Simon) method using FlexArray 1.6.1. An FDR-adjusted P-value lower than or equal to 0.1 and a fold-change greater than 2 were considered as a cut-off point to indicate significant up- or downregulation of genes. Gene networks were assembled using Ingenuity Pathway Analysis. The results showed that in both treatments at wk +1 compared with wk -3, relative to parturition, genes that are involved in glycogenesis downregulated and genes that were involved in gluconeogenesis upregulated. Glucose-6-phosphatase, catalytic subunit, expressed only in 40-d treatment and serine dehydratase expressed only in 60-d treatment. In the 40-d treatment, at wk 4 compared with wk 1, pyruvate carboxylase, which is involved in gluconeogenesis, was downregulated and the PPP1R3C transcript, encoding protein phosphatase 1 regulatory subunit 3C, which is involved in glycogenesis, was upregulated. However, there was no change in gene expression between wk 1 and wk 4 in the 60-d treatment. In conclusion, at the first week after parturition gene expression shows that gluconeogenesis was greater than glycogenesis in both treatments. However, after 4 weeks, cows on the 40 d treatment showed signs of enhanced recovery to normal carbohydrate metabolism in the liver compared with cows on the 60 d treatment.

Table 1. Effects of dry period length on liver gene expression in regard to carbohydrate metabolism

Genes	Name	Fold-change wk +1/-3		Fold-change wk +4/+1	
		40-day trt	60-day trt	40-day trt	60-day trt
Gluconeogenesis					
G6PC	Glucose-6-phosphatase	1.04	-	-	-
PC	Pyruvate carboxylase	2.4	1.73	-1.59	-
SDS	Serine dehydratase	-	1.51	-	-
PPARGC1A	Proxisome proliferator-activated receptor gamma	1.03	1.03	-	-
Glycogenesis					
PPP1R3C	Protein phosphatase 1, subunit 3C	-2.03	-1.61	1.25	-
PPP1R3B	Protein phosphatase 1, subunit 3B	-1.70	-1.34	-	-
IGF1	Insulin-like growth factor 1	-1.71	-2.06	-	-

Key Words: dry period, gene expression, carbohydrate metabolism