

## Animal Health: Transition cow health

**354 Characterizing critical thresholds of subclinical ketosis using the in-line milk monitoring system Herd Navigator.** Elizabeth R. Ellis\*, Tom C. Wright, John P. Cant, and Vern R. Osborne, *Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada.*

Sub-clinical ketosis (SCK) refers to a state of elevated  $\beta$ -hydroxybutyrate (BHBA) without visual signs of acute disease. With a prevalence of 26 to 60% (dependent on BHBA level used to define disease), this costly metabolic disorder often goes undetected. Innovative in-line milk testing technology, Herd Navigator (DeLaval), enables automated milk BHBA analysis of individuals in a herd. The objective of this study was to characterize SCK as it affects milk production using Herd Navigator technology. The study comprised 3 hundred and 56 Holstein cows from 5 dairy farms in Ontario, operating herd-specific SCK treatment protocols based on BHBA thresholds ranging from 1.2 to 1.7 mmol/L. Individual cow data were collected from June 2013 to July 2014 for daily milk yield, days to peak BHBA level, days to peak milk, peak milk yield; total predicted milk yield to 60 d and 305 d were generated for all cows. Based on the maximum BHBA level attained within 60DIM, cows were assigned to 1 of 5 groups (G1  $\geq$  1.7 mmol/L, G2  $\geq$  1.5 mmol/L, G3  $\geq$  1.4 mmol/L, G4  $\geq$  1.2 mmol/L, G5  $<$  1.2 mmol/L) for analysis. Data were analyzed by ANOVA using the maximum BHBA value as covariate and farm as a random effect. Results showed that first-lactation cows in G1 had higher milk yield at 60 DIM (165 kg  $\pm$  91,  $P <$  0.05) than first-lactation cows in G5. In second-lactation cows, there were no differences in milk yield between groups. Third-lactation cows in G2 had higher milk yield at 60 DIM than those in G4 (431 kg  $\pm$  153,  $P <$  0.05) G5 (295 kg  $\pm$  125,  $P <$  0.05). Predicted milk yield at 305 DIM of third lactation cows in G1 was lower (-2505 kg  $\pm$  1318,  $P <$  0.05) than in G2. Across farms, elevated ( $\geq$  1.4 mmol/L) milk BHBA values were associated with higher milk production for mature cows at 305DIM, but not for first- and second-lactation cows. Results of this study indicate that farm-level milk BHBA data from Herd Navigator coupled with available on-farm milk production records could be used to refine the effectiveness of SCK treatment protocols and identify different BHBA treatment thresholds by parity to improve milk yield.

**Key Words:** ketosis, milk production, Herd Navigator

**355 Monitoring rumination in transition dairy cows for the early detection of subclinical ketosis.** Emily I. Kaufman\*<sup>1</sup>, Stephen J. LeBlanc<sup>2</sup>, Brian W. McBride<sup>1</sup>, Todd F. Duffield<sup>2</sup>, and Trevor J. DeVries<sup>1</sup>, <sup>1</sup>*Department of Animal and Poultry Science, University of Guelph, Guelph, ON, Canada,* <sup>2</sup>*Department of Population Medicine, University of Guelph, Guelph, ON, Canada.*

The objective of this study was to characterize the relationship between rumination and subclinical ketosis (SCK) in transition dairy cows. A study was conducted on 4 commercial dairy farms in Eastern Ontario, Canada. A total of 339 dairy cows (107 primiparous and 232 multiparous) were monitored for rumination activity and SCK from 14d before calving until 28d after calving. Rumination was recorded daily using an automated monitoring system. A blood sample was taken from the coccygeal vein of each cow for the measurement of  $\beta$ -hydroxybutyrate (BHBA) 1x/wk throughout the 6-wk observation period. Cows with a BHBA concentration  $\geq$  1.2 mmol/L postpartum were considered to have SCK. Cases of retained placenta, metritis, milk fever, or mastitis during the study period were also recorded. Cows were categorized into 1 of

3 groups: healthy (H) cows had no SCK or any other health issue (n = 139); SCK (K) cows with no other health problems during transition (n = 97); or ketotic plus (K+) cows that had SCK and one or more other health problems (n = 53). Data were summarized by wk and analyzed in a repeated measures general linear mixed model. From 2 wk before (-2) calving to 4 wk after calving (+4), there was no difference in daily rumination time (409  $\pm$  9.8 min/d; mean  $\pm$  SE) among H, K, and K+ cows in their first lactation ( $P = 0.5$ ). Multiparous cows in H spent an average of 459 min/d ruminating from wk -2 to wk +4. Multiparous K cows ruminated 25  $\pm$  12.8 min/d less ( $P = 0.05$ ) than H cows, while K+ cows ruminated 44  $\pm$  15.6 min/d less ( $P = 0.005$ ) than H cows. The largest differences in rumination time between H and K+ cows were seen during wk -1, +1 and +2, when K+ cows ruminated 48  $\pm$  17.2 min/d, 73  $\pm$  16.0 min/d, and 65  $\pm$  19.4 min/d less ( $P \leq 0.005$ ) than H cows, respectively. These results suggest that rumination monitoring across the transition period might contribute to identification of SCK and other health issues in multiparous cows.

**Key Words:** transition cow, rumination, subclinical ketosis

**356 Use of a rumination and activity monitoring for the identification of dairy cows with health disorders.** Matias L. Stangaferro\*, Robert Wijma, Cristian E. Quinteros, Miranda M. Medrano, Magdalena Masello, and Julio O. Giordano, *Department of Animal Science, Cornell University, Ithaca, NY.*

Objectives were to evaluate: 1) the ability of a commercial rumination (Rum) and activity (Act) monitoring system (HR Tags, SCR Dairy) to identify cows with health disorders and 2) the interval between the day of diagnosis of disease and day of alert by the HR system (HR). Holstein cows (n = 1,118; 449 nulliparous and 669 multiparous) were fitted with an HR tag from -28 to 80 DIM. Every 12 h after 1 DIM, an individual cow Health Index (HI) was generated based on Rum and Act. Cows with a HI value  $<$  86 points were flagged by HR. Farm personnel examined cows for signs of clinical disease (CDZ) daily. From 1 to 10 DIM, personnel evaluated: appetite, rectal temperature, ketone bodies in urine, rumen fill and movements, vaginal discharge, daily milk weights, and conductivity. Data from 1,099 cows was available. Number of CDZ events included was: displaced abomasum (DA) 41, ketosis (KET) 57, indigestion (IND) 9, metritis (MET) 360, and mastitis (MAST) 74. Sensitivity (Se) of HR to flag cows with CDZ (farm personnel diagnosis as gold standard) and the interval between day of CDZ diagnosis and the day a cow was flagged by HR was evaluated with PROC FREQ and PROC TTEST of SAS, respectively. The Se of HI was: 97.6% (CI 93-100%) for DA, 84.2% (CI 75-94%) for KET, 88.9% (CI 68-100%) for IND, 45.6% (CI 40-51%) for MET and 49.7% (CI 42-57%) for all MAST. For all DA, KET, and IND combined, Se of HR was 89.7% (CI 84-95%). Sensitivity of HR by MAST pathogen was 66.7% (CI 50-83%) for *E. coli* and *Klebsiella*, 52.6% (CI 37-68%) for *Streptococcus* and *Staphylococcus* spp. combined, and 22.2% (CI 0-49%) for *Staph. aureus*. Mean and 95% CI for interval between day of CDZ to day flagged by HR (cows flagged only) was: -3 (-3.7- -2.3;  $P <$  0.01), -1.6 (-2.3- -1.0;  $P <$  0.01), -0.5 (-1.5- 0.5;  $P = 0.28$ ), -0.8 (-1.2- -0.44;  $P <$  0.01), -0.8 (-1.2- 0.3;  $P <$  0.01), for DA, KET, IND, MET, and all MAST, respectively. We conclude that the HR system is most effective to identify cows suffering metabolic and digestive disorders. A relatively lower Se to identify cows with MET and MAST might be explained by less severe systemic illness and type of mastitis-

causing pathogen. The HR system identified cows with DA, KET, MET and MAST earlier than farm personnel.

**Key Words:** rumination, activity, dairy cow

### 357 Development of a ketosis prevalence tool in Holstein dairy cows based on milk component data and cow test-day information.

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Subclinical ketosis affects between 40 and 60% of dairy cows and negatively affects cow productivity and health. Although cow-side ketone testing strategies are available, many lack sufficient accuracy, are labor-intensive, and can be costly. The objective of this study was to validate the use of multiple regression models to predict blood  $\beta$ -hydroxybutyrate (BHBA) from milk components and continuous test-day variables in early lactation cows for determining ketosis prevalence. Blood samples were collected on the same day as milk test from 658 Holstein cows 5 to 20 DIM on 10 dairy farms. Blood serum was analyzed for concentration of BHBA by colorimetric assay (Stanbio, Boerne, TX). Milk samples were analyzed for milk BHBA and acetone concentrations by Fourier transform infrared spectrometry (FOSS Analytical A/S, Eden Prairie, MN), in addition to standard milk analysis variables. Continuous test-day variables were collected from DairyComp305 (Valley Agricultural Software, Tulare, CA) records. Models were built in the REG procedure of SAS 9.4 (SAS Institute Inc., Cary, NC) using stepwise, forward selection by excluding variables with a  $P$ -value  $< 0.15$  and selection criterion of Akaike's information criterion. Data interrogation justified development of separate models for primiparous and multiparous cows, as well as cows 5 to 11 DIM and cows 12 to 20 DIM. Additionally, disease etiology allowed for unique models for the 5 to 11 and 12 to 20 DIM ranges. Significant variables were milk BHBA, acetone, and fat:protein ratio; parity, previous days dry, previous lactation length, and age at first calving; and DIM and milk production on test day. Overall, model accuracy was 88% for multiparous cows 5 to 11 DIM ( $R^2 = 0.57$ ), 83% for multiparous cows 12 to 20 DIM ( $R^2 = 0.67$ ), 96% for primiparous cows 5 to 11 DIM ( $R^2 = 0.74$ ), and 97% for primiparous cows 5 to 20 DIM ( $R^2 = 0.66$ ). These results suggest that modeling blood BHBA based on milk component data and cow test-day information can serve as a valuable diagnostic tool for monitoring herd-level ketosis prevalence.

**Key Words:** ketosis, linear regression, model

### 358 Evaluation of recurrence of frequent diseases and disorders in early postpartum dairy cows.

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Objective was to evaluate if having disease in one lactation (Lac1) would affect the risk of developing disease and calving related problems in the subsequent lactation (Lac2). Holstein cows ( $n = 1351$ ) from 2 herds in North Florida were used to create 2295 lactation pairs from 4590 lactations occurring between 2006 and 2013. Data were collected within 60 DIM. Data were evaluated using the LOGISTIC procedure of SAS. Having dystocia in Lac1 increased the risk of having it in Lac2 (37.0 vs. 23.7%;  $P < 0.01$ ). Dystocia in Lac2 was also increased by induced parturition and twins in Lac2. Having twins in Lac1 increased

the risk of having it in Lac2 (4.6 vs. 2.4%;  $P < 0.02$ ). Twins in Lac2 tended ( $P = 0.09$ ) to be increased in parity  $> 2$  vs. parity 2 in Lac2. Having stillbirth in Lac1 did not affect the risk of having it in Lac2 (5.7 vs. 3.2%;  $P = 0.12$ ). Stillbirth in Lac2 was increased ( $P < 0.05$ ) by dystocia and twins in Lac2. Having milk fever in Lac1 increased the risk of having it in Lac2 (38.9 vs. 2.6%;  $P < 0.01$ ). Milk fever in Lac2 was also increased ( $P < 0.05$ ) by dystocia and parity  $\geq 2$  in Lac2, but was decreased by induced parturition in Lac2. Having retained placenta in Lac1 increased the risk of having it in Lac2 (13.1 vs. 6.2%;  $P = 0.01$ ). Retained placenta in Lac2 was also increased ( $P < 0.05$ ) by twins and induced parturition in Lac2. Having metritis in Lac1 increased the risk of having it in Lac2 (20.4 vs. 10.4%;  $P < 0.02$ ). Metritis in Lac2 was also increased ( $P < 0.05$ ) by dystocia, induced parturition, twins, stillbirth, retained placenta and ketosis in Lac2. Having ketosis in Lac1 increased the risk of having it in Lac2 (41.9 vs. 17.0%;  $P = 0.01$ ). Ketosis in Lac2 was also increased ( $P < 0.05$ ) by induced parturition, metritis, and parity  $> 2$  in Lac2. Having displaced abomasum in Lac1 had a tendency to increase the risk of having it in Lac2 (10.5 vs. 2.1%;  $P = 0.06$ ). Displaced abomasum in Lac2 was increased ( $P < 0.05$ ) by ketosis in Lac2. Having mastitis in Lac1 had a tendency to increase the risk of having it in Lac2 (22.9 vs. 9.3%;  $P = 0.09$ ). Mastitis in Lac2 was increased ( $P < 0.05$ ) by parity  $> 2$ . In conclusion, with the exception of stillbirth, disease in Lac1 affected the incidence of disease in Lac2.

**Key Words:** dairy cow, postpartum diseases, disease recurrence

### 359 Early lactation disease incidence in Holstein cows across multiple US regions.

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The objective was to assess the incidence of transition cow diseases in Holstein cows ( $n = 10,959$  in 16 herds) calving during the warm (WS: May–Aug) and cool (CS: Oct – Jan) seasons. Cows were enrolled at parturition and monitored weekly for multiple diseases until 60 DIM. Disease included retained fetal membranes (RFM), metritis (MET,  $7 \pm 3$  DIM; foul-smell, watery, brownish vaginal discharge), subclinical ketosis (SKT,  $7 \pm 3$  DIM; serum BHBA  $> 1.0$  mmol/L), mastitis (MAS; farm records), left displaced abomasum (LDA), pneumonia (PN; farm records), clinical endometritis (CE  $28 \pm 3$  DIM; from mucopurulent to fetid vaginal discharge) and lameness (LAM  $35 \pm 3$  DIM; score  $> 3$ ). Study locations included the Northeast (NE; 4 herds), Midwest (MW; 6 herds), Southeast (SE; 1 herd), and the Southwest (SW; 5 herds) regions. Disease incidence was estimated by region and season (top part of Table 1). Associations between disease occurrence and region, calving season, and parity ( $1, \geq 2$ ) were tested by logistic regression. The effect of region on disease occurrence was only significant for LAM ( $P < 0.05$ ). Contrarily, season of calving and parity were significantly associated with the risk of transition diseases in all the disorders analyzed (bottom part of Table 1). This project was funded by USDA-NIFA-AFRI (2013–68004–20361).

*Contd.*

**Table 1 (Abstr. 359).** Incidence (%) of transition cow diseases by region and season of calving (top) and odds ratios (OR) for the risk of disease by region, season of calving, and parity (bottom); references are SW, WS, and parity  $\geq 2$

Item	NE		MW		SE		SW					
	WS	CS	WS	CS	WS	CS	WS	CS				
RFM	8.0	5.9	7.4	5.4	15.0	7.6	4.3	2.9				
MET	21.7	23.8	19.5	20.2	19.7	18.5	27.6	24.8				
SKT	41.8	18.7	25.9	15.5	24.9	20.1	31.3	14.6				
MAS	26.1	16.0	6.1	5.5	18.0	21.3	12.0	8.1				
LDA	3.0	5.6	2.9	1.4	6.0	4.0	1.0	1.0				
PNM	1.1	1.5	1.7	1.8	3.8	13.4	7.1	3.5				
CLE	15.4	32.5	25.9	20.4	23.4	42.9	24.3	26.1				
LAM	11.3	2.6	2.1	8.1	1.7	12.1	5.4	2.0				

  

Region	RFM		MET		SKT		MAS		CLE		LAM	
	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI
MW	1.9	0.7-5.0	0.8	0.5-1.4	0.7	0.2-2.1	0.6	0.2-2.0	1.0	0.6-1.7	3.7	1.3-10.5
NE	2.4	0.8-7.1	1.0	0.6-1.8	1.2	0.4-4.3	2.4	0.8-6.9	1.1	0.6-2.0	0.9	0.3-2.5
SE	4.2	0.8-23.0	0.8	0.3-2.0	1.0	0.1-7.6	2.9	0.5-16.3	1.7	0.7-4.5	1.4	0.3-6.3

  

Season	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI
CS	0.6	0.5-0.7	1.1	1.0-1.2	0.4	0.3-0.4	0.7	0.7-0.9	1.4	1.2-1.5	2.5	1.8-3.4

  

Parity	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI
1	0.4	0.4-0.5	1.8	1.6-2.0	0.6	0.5-0.7	1.3	1.1-1.5	1.1	1.0-1.2	0.2	0.1-0.3

**Key Words:** health, transition, Holstein

**360 Association between dry matter intake pre- and postpartum and postpartum diseases in dairy cows.** Johanny Perez Baez<sup>\*2</sup>, Carlos A. Risco<sup>2</sup>, Jorge A. Hernandez<sup>2</sup>, Gabriel C. Gomes<sup>2</sup>, Leandro F. Greco<sup>1</sup>, Sha Tao<sup>1,3</sup>, Izabella Thompson<sup>1,4</sup>, Bruno do Amaral<sup>1,5</sup>, and Charles Staples<sup>1</sup>, Jose Eduardo P. Santos<sup>1</sup>, and Klibs N. Galvão<sup>2</sup>, <sup>1</sup>Department of Animal Sciences, University of Florida, Gainesville, FL, <sup>2</sup>Department of Large Animal Clinical Sciences, University of Florida, Gainesville, FL, <sup>3</sup>Department of Animal and Dairy Science, University of Georgia, Tifton, GA, <sup>4</sup>Agriculture and Agri-Food Canada, Ottawa, Ontario, Canada, <sup>5</sup>Land O'Lakes, Inc., St. Paul, MN.

The objectives of this retrospective observational study were to determine the association between dry matter intake (DMI) pre- (-14 d to -1) and postpartum (1 to 28 d) and postpartum diseases [retained placenta (RP), metritis (MET), mastitis (MAST), ketosis (KET), and displaced abomasum (DA)] within 28 d postpartum; and to estimate the risk of disease postpartum based on DMI prepartum. Data involving 294 cows from 7 studies were collected. The data were analyzed with the MIXED and GLIMMIX procedures of SAS. Random and repeated variables were cow and day relative to calving, respectively. Models were adjusted for parity, BCS, treatment, study, and interaction between disease and other covariates. Variables with  $P \leq 0.05$  were considered significant. Cows that had RP ate less on d -3 ( $RP \times day P = 0.01$ ) and on d 5, 7, 8, 12, and 19 ( $RP \times day P = 0.10$ ). Cows that had MET ate less on d -3 and -2 ( $MET \times day P = 0.03$ ) and ate less postpartum ( $P < 0.01$ ). Cows that had MAST ate less prepartum ( $P = 0.02$ ) and in the first 14 d postpartum ( $MAST \times day P < 0.01$ ). Cows that had KET ate less pre- and postpartum ( $P < 0.01$ ). Intake of cows that had DA did not differ prepartum ( $P = 0.70$ ) but they ate less postpartum ( $P < 0.01$ ). Cows with at least one disease ate less pre- and postpartum ( $P < 0.01$ ). Moreover, for each kg decrease in DMI in the last week prepartum, there was an increase of 28% in the odds of having KET (OR = 1.28; CI = 1.15-1.41;  $P < 0.01$ ), and 24% in the odds of having at least one disease (OR = 1.24;

CI = 1.13-1.34;  $P < 0.01$ ) postpartum. However, DMI was not associated with the odds of having MAST (OR = 1.09; CI = 0.95-1.23;  $P = 0.23$ ), RP (OR = 1.04; CI = 0.86-1.22;  $P = 0.66$ ), MET (OR = 1.08; CI = 0.97-1.19;  $P = 0.14$ ), or DA (OR = 1.07; CI = 0.82-1.32;  $P = 0.6$ ). Collectively, these data suggest that there is an association between DMI pre- and postpartum and postpartum diseases, and that a reduction in DMI prepartum predisposes cows to disease postpartum.

**Key Words:** postpartum disease, dry matter intake, dairy cow

**361 Laboratory validation of a prototype cow-side instrument for the measurement of blood ionized calcium concentrations in dairy cattle.** Rafael C. Neves<sup>\*</sup>, Tracy Stokol, and Jessica A. A. McArt, Department of Population Medicine & Diagnostic Sciences, Cornell University, Ithaca, NY.

There is currently no efficient and inexpensive method for field measurement of blood calcium concentrations. Ionized calcium (iCa) is the homeostatic form of the mineral and is thought to have greater biological relevance over that of total calcium. The objective of this study was to evaluate the linearity and precision of a prototype cow-side instrument (Horiba, Japan) for measuring blood iCa concentrations. Blood (300 mL) was collected from the right jugular vein of a multiparous dairy cow (4 DIM) into lithium heparin tubes immediately before (T0) and 5 min after (T5) intravenous administration of 500 mL of 23% calcium borogluconate. The iCa concentrations were determined using a blood-gas analyzer (ABL-800 FLEX, Radiometer) as a gold-standard. The T0 sample was diluted using 0.9% saline to create a sample with low iCa (reference interval = 1.10 to 1.35  $\mu\text{mol/L}$ ). The diluted T0 sample was then mixed with the T5 sample in different ratios (100/0, 75/25, 50/50, 25/75, 0/100) to obtain 5 levels of iCa concentrations (0.69, 1.0, 1.28, 1.58, and 1.82  $\mu\text{mol/L}$ ). Each mixture was then analyzed in triplicate using 3 different prototypes under one-point (1P) and 2-point (2P) calibration with the means compared with results from the blood-gas

analyzer. Cumulative sum tests for linearity from Passing and Bablok regressions showed no deviation from linearity for the combined results of all 3 prototypes under 1P vs. the gold-standard ( $P = 0.19$ ) and under 2P vs. the gold-standard ( $P = 0.19$ ). Instrument precision (coefficient of variation; CV) was determined by 10 repeat measurements of the diluted T0 sample, T0, and T5 samples under 1P and 2P calibrations. The CV ranged from 1.3 to 5% for the 3 prototypes. Laboratory results indicate good accuracy and precision for a cow-side instrument at the tested iCa concentrations. Investigation of the instrument under field conditions is warranted.

**Key Words:** ionized calcium, cow-side instrument, dairy cattle

**362 Assessment of daily activity patterns in lactating dairy cows diagnosed with metritis.** Santiago Bas\*, Adrian A. Barragan, Juan M. Piñeiro, Gustavo M. Schuenemann, Päivi J. Rajala-Schultz, and Troy A. Brick, *Department of Veterinary Preventive Medicine, College of Veterinary Medicine, The Ohio State University, Columbus, OH.*

Metritis (MET) is a prevalent uterine disease that affects dairy cows, and causes substantial economic losses due to reduced milk yield, delayed pregnancy, cost of treatments, and increased culling and death rates. The objective was to assess changes in daily activity patterns (i.e., number of steps, number of lying bouts, standing time and lying time) of lactating dairy cows diagnosed with MET using activity monitors (IceQube, IceRobotics, Edinburgh, UK). Lactating dairy cows ( $n = 60$ ) from one commercial dairy herd were enrolled. Primiparous (PRIM;  $n = 12$ ) and multiparous (MULT;  $n = 48$ ) cows were housed in the same pen and were milked 4 times daily. Cows diagnosed with MET ( $n = 30$ ) were matched by lactation number and DIM to cows without MET (noMET;  $n = 30$ ). On study d 1, MET was confirmed (using a metrichk device) by the presence of watery, reddish or brownish foul smelling vaginal discharge. In addition, activity monitors were placed on the hind legs of MET and noMET cows and were kept until study d 7. The daily number of steps (n/d), number of lying bouts (n/d), standing time (min/d) and lying time (min/d) were recorded. Cows showing any other signs of disease (e.g., lameness, mastitis) were not included. Data were analyzed using the MIXED procedure of SAS. Preliminary results showed no difference in the number of steps ( $P = 0.30$ ), number of lying bouts ( $P = 0.93$ ) standing time ( $P = 0.89$ ), or lying time ( $P = 0.89$ ) between MET and noMET cows. However, a different pattern of behavioral activity was observed between PRIM and MULT cows regardless of the MET status. PRIM cows had more steps ( $P < 0.0001$ ; PRIM = 1921; MULT = 1728), more lying bouts ( $P < 0.0001$ ; PRIM = 14; MULT = 11), spent more time standing ( $P = 0.01$ ; PRIM = 860; MULT = 779) and less time lying ( $P = 0.01$ ; PRIM = 580; MULT = 660) than MULT cows. Previous studies have reported differences in activity patterns between cows with and without MET. In the present study, PRIM and MULT cows were housed together and were milked 4 times daily; thus, on-farm management may affect daily behavioral activity patterns of lactating dairy cows regardless of the uterine health status postpartum.

**Key Words:** dairy, metritis, activity

**363 The effect of ketosis on milk production in early lactation.** Khaled Gohary\*, Todd Duffield, and Stephen LeBlanc, *Department of Population Medicine, Ontario Veterinary College, University of Guelph, Guelph, Ontario, Canada.*

The objective of this study was to evaluate the effect of ketosis on milk production in early lactation. Data from 1,156 cows enrolled in a previous study from 5 commercial dairy herds in Southern Ontario were analyzed. Blood samples from all cows were obtained 1 week before calving and once a week for the first 2 weeks following calving. Sera were submitted to the laboratory at the University of Guelph to measure blood analytes including  $\beta$ -hydroxybutyrate (BHBA). Results were categorized from 1.0 to 2.0 mmol/L in 0.2 mmol/L increments. Daily milk production for all cows was recorded until 63 d in milk (DIM). Using weekly milk production as an outcome, for each of the selected thresholds, 3 linear mixed models that accounted for repeated measures within cow were fitted to test the effects of ketosis in wk 1 or wk 2 postpartum, or both. The analyses were performed with farm as a random effect or as a fixed effect. Overall, as the concentration of BHBA increased, milk yield was lower in cows with ketosis compared with cows without ketosis. Ketotic cows produced less total milk to 63 DIM when ketosis persisted for the first 2 weeks after calving than if present in wk 1 or 2 (at BHBA  $\geq 1.4$  mmol/L: -97, -37, and -62 kg, for first 2 weeks after calving, wk 1, and wk 2 after calving, relative to cows without ketosis, respectively,  $P < 0.0001$ ). Cows ketotic at wk 2 only produced more milk in wk 1 than cows without ketosis (at BHBA  $\geq 1.4$  mmol/L:  $25.9 \pm 1.3$  kg/d and  $23.9 \pm 1.2$  kg/d, respectively,  $P = 0.004$ ). When farm was included as a fixed effect, there was a significant interaction between farm and ketosis in wk 2 alone for BHBA  $\geq 1.2$  mmol/L, and in wk 1 and 2 for BHBA  $\geq 1.6$  mmol/L. The effect of ketosis on yield varied among farms from no significant difference to 7.3 kg/d ( $P < 0.0001$ ). The degree and duration of ketosis were negatively associated with milk yield in early lactation, but the timing of onset of ketosis affects the association. The threshold at which ketosis reduces milk production in early lactation may vary among farms and this should be explored in a large data set from many herds.

**Key Words:** dairy cattle, ketosis, milk production

**364 Estimating glucose requirements of an activated immune system in lactating Holstein cows.** Sara K. Stoakes\*, Erin A. Nolan, David J. Valko, Mohannad Abuajamieh, Edith J. Mayorga, Jake Seibert, Maria V. Sanz Fernandez, Patrick J. Gorden, and Lance H. Baumgard, *Iowa State University, Ames, IA.*

Activated immune cells are obligate glucose utilizers; thus, study objectives were to estimate the quantity of whole body glucose utilization during an IV endotoxin challenge. Fasting lactating Holstein cows ( $718 \pm 16$  kg;  $169 \pm 7$  DIM) were jugular catheterized and assigned 1 of 3 bolus treatments: control (CON; 5 mL saline;  $n = 6$ ), lipopolysaccharide infused (LPS;  $1.5 \mu\text{g/kg BW}$ ; *E. coli* 055:B5;  $n = 6$ ), and LPS + euglycemic clamp (LPS-Eu;  $1.5 \mu\text{g/kg BW}$ ; 50% dextrose infusion;  $n = 6$ ). After infusion, blood glucose was determined every 10 min and dextrose infusion was adjusted in LPS-Eu cows to maintain euglycemia. Blood samples were obtained 3, 6, 9, and 12 h post-bolus for further analysis. Cows were milked 6 and 12 h post-bolus. Milk yield decreased in LPS and LPS-Eu cows relative to CON (80%,  $P < 0.01$ ). Milk SCC was increased in LPS relative to CON (48%,  $P = 0.02$ ) while LPS-Eu did not differ from either treatment. LPS and LPS-Eu cows were hyperglycemic for 3 h post-bolus, but thereafter glucose content decreased in LPS relative to LPS-Eu and CON cows (30%,  $P < 0.01$ ). Circulating insulin was and tended to be increased in LPS-Eu (80%,  $P = 0.01$ ) and LPS (72%,  $P = 0.06$ ) relative to CON. Plasma NEFA, BHBA, and Ca were decreased ( $P < 0.01$ ) in LPS and LPS-Eu relative to CON (46, 53, and 46%, respectively). Plasma haptoglobin and L-lactate were increased ( $P < 0.01$ ) in LPS and LPS-Eu cows relative to CON (72 and 62%, respectively). Serum amyloid A was increased in LPS (47%,  $P$

= 0.01) and tended to be increased in LPS-Eu (34%,  $P = 0.09$ ) compared with CON cows. White blood cells were decreased in LPS and LPS-Eu relative to CON cows ( $P < 0.01$ ). Total glucose deficit during the 12 h post-bolus was calculated as the decrease in the amount of glucose required to synthesize milk (relative to pre-infusion levels) plus the amount of glucose infused to maintain euglycemia (in LPS-Eu cows only). Glucose deficit for CON, LPS, and LPS-Eu cows was 483, 1259, and 1553 g, respectively. If this model is a proxy of glucose demands during an immune response, our data indicates an intensely activated immune system uses at least 90 g glucose/h and maintaining euglycemia does not rescue the decrease in milk synthesis.

**Key Words:** lipopolysaccharide

**365 DNA methylation patterns in peripheral blood leukocytes as a marker of uterine function.** Caroline Walker\*<sup>1</sup>, Barbara Kuhn-Sherlock<sup>2</sup>, Susanne Meier<sup>2</sup>, John Roche<sup>2</sup>, and Murray Mitchell<sup>3</sup>, <sup>1</sup>*DairyNZ, Auckland, New Zealand*, <sup>2</sup>*DairyNZ, Hamilton, New Zealand*, <sup>3</sup>*University of Queensland, Queensland, Brisbane, Australia*.

The objective of this study was to assess the suitability of using DNA methylation patterns in peripheral blood leukocytes as a marker of uterine function in the dairy cow. Peripheral blood and endometrial tissues were obtained at 29 d postpartum from cows with subclinical endometritis (SCE,  $n = 6$ ) and control cows (CON,  $n = 6$ ). DNA was extracted from peripheral blood leukocytes and endometrial tissues and DNA methylation was measured using Methylated DNA immunoprecipitation (MeDIP) in combination with microarrays. Genome-wide

DNA methylation was assessed using a custom 400K Agilent microarray (GPL16270). The agreement between DNA methylation measured in leukocytes and in uterine tissue was assessed using Pearson correlation between log ratio in leukocytes and log ratio in intercaruncular and caruncular endometrium. Correlations were considered high if either of the 2 correlation coefficients was greater than 70%. All analyses were performed using SAS 9.2. Genes that were highly correlated between leukocytes and endometrial tissue were submitted for pathway enrichment analysis using PANTHER (Protein Analysis THrough Evolutionary Relationships). 9,733 DNA methylation probes had high (>70%) correlation between blood and endometrial tissue. These probes mapped to a total of 3,329 genes. Enrichment analysis identified several pathways of which the top 5 pathways were; gonadotropin releasing hormone receptor pathway (1.85 fold enrichment,  $P = 1.5 \times 10^{-4}$ ), oxytocin receptor mediated signaling pathway (2.7 fold enrichment,  $P = 3.17 \times 10^{-3}$ ), heterotrimeric G-protein signaling pathway-Gq  $\alpha$  and Go  $\alpha$  mediated pathway (1.9 fold enrichment,  $P = 4.2 \times 10^{-3}$ ), PI3 kinase pathway (2.5 fold enrichment,  $P = 4.3 \times 10^{-3}$ ), and Wnt signaling pathway (1.5 fold enrichment,  $P = 5 \times 10^{-3}$ ). DNA methylation in the blood is highly correlated with DNA methylation status in endometrial tissue. In genes that were highly correlated, there was significant enrichment for several biological pathways that regulate the reproductive and immune systems. This study provides support for the efficacy of peripheral blood leukocytes as a marker of uterine function. Further research will establish if any of these markers are associated with improved reproductive function.

**Key Words:** epigenetics, reproduction