

Monday, July 16, 2012

## POSTER PRESENTATIONS

### Animal Health I

#### **M1 Immunological and metabolic responses of Holstein and Jersey cows according to body condition score change prepartum.**

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Objectives of this experiment were to evaluate the immune and metabolic responses of Holstein (H) and Jersey (J) cows according to body condition score (BCS) change (BCSC) prepartum. Data from 2 experiments were used. Experiment 1 was conducted with Holstein cows (n = 29) and experiment 2 was conducted with Jersey cows (n = 68). Cows received BCS on d -22 ± 7 and 1 ± 1 (calving = d0) and were classified as having lost (L) and as having not lost (NC) BCS prepartum. Blood was sampled on d -7, 0, 7, and 14 for determination of neutrophil phagocytosis (PHAGO) and oxidative burst (OXID) and expression of CD18 and L-selectin and concentrations of NEFA and glucose. Data were analyzed by Chi-squared and ANOVA for repeated measures. Percentages of H (65.5%) and J (51.5%) cows losing BCS prepartum were similar (P = 0.20). The interaction between breed and BCSC was associated with most of the innate immunity parameters evaluated because among J cows BCSC was not associated with innate immunity parameters (P > 0.10). On the other hand, HL cows had reduced PHAGO (57.8 ± 3.6 vs NC = 73.9 ± 4.8%; P < 0.01) and OXID (93.2 ± 0.7 vs 96.0 ± 0.9%; P = 0.02) than HNC cows. The interaction between breed and BCSC tended (P = 0.09) to be associated with intensity of OXID because HL cows had reduced OXID intensity than HNC cows (15857 ± 3052 vs 24770 ± 3782; P = 0.07). There was a tendency for the interaction between breed and BCSC to be associated with percentage of neutrophils positive for CD18 (P = 0.08) because HL cows tended to have reduced percentage of neutrophils expressing CD18 than HNC cows (74.1 ± 2.0 vs NC = 80.2 ± 2.5%; P = 0.06). Concentration of NEFA was associated with BCSC (L = 0.533 ± 0.038, NC = 0.429 ± 0.044 mmol/L; P = 0.03) but was not associated with the interaction between breed and BCSC (P = 0.99). On the other hand, glucose concentration was not associated with BCSC (P = 0.50) or with the interaction between breed and BCSC (P = 0.62). Immunological parameters peripartum were differently affected by BCS loss prepartum in Holstein and Jersey cows.

**Key Words:** peripartum cow, body condition score, immunological parameter

#### **M2 Treatment outcomes for clinical mastitis caused by *E. coli* in a Wisconsin dairy herd.** M. J. Fuenzalida\*<sup>1</sup>, W. Oliveira<sup>1</sup>, J. Gaska<sup>2</sup>, and P. L. Ruegg<sup>1</sup>, <sup>1</sup>*Department of Dairy Science, University of Wisconsin, Madison*, <sup>2</sup>*Gaska Dairy Health Services, Columbus, WI.*

The objective of this study is to describe the effect of intramammary antimicrobial treatment (ceftiofur) on milk yield, and somatic cell (SCC) reduction during a 42-d follow-up period. Clinical mastitis (CM) occurring in a single quarter (n = 133) was diagnosed between May 2011 and January 2012. Average milk yield was 42.3 kg/d, parity was 2.5, logSCC 1.95, and DIM was 162. Explanatory variables considered in the models were DIM, parity, previous cases of CM, previous SCC, and previous milk yield after treatment. Of 133 cases of clinical mastitis caused by *E. coli*, 75 were not treated and 58 received intramammary

ceftiofur. PROG GENMOD was used for analyzing the binary outcomes (yes/no for SCC reduction, yes/no for milk yield reduction) and PROC GLM SAS 9.2 was used to analyze the continuous variable milk yield. Milk yield after occurrence of the case was 42.4 and 41.5 for treated and not treated cows, respectively but was not significantly affected by treatment (P = 0.48). One unit increase in logSCC was associated with 2.7 kg/d reduction in milk production. Occurrence of a previous case of CM was associated with about 4 kg/d less milk production. Treatment was not associated with SCC reduction (OR = 1.84, CI 0.32–10.31) or change in milk yield (OR = 0.78, CI 0.27–2.21). In this farm, treatment of CM caused by *E. coli* did not improve milk production or SCC compared with no treatment. Supported by AFRI Competitive Grant no. 2010–85122–20612

**Key Words:** clinical mastitis, SCC, milk yield

#### **M3 Differential expression of the hepatic and adipose transcriptome in periparturient Friesian cows with endometritis.** H. Akbar\*<sup>1</sup>, J. M. Khan<sup>1</sup>, S. Meier<sup>2</sup>, C. Burke<sup>2</sup>, S. McDougall<sup>3</sup>, M. Mitchell<sup>4,5</sup>, S. L. Rodriguez-Zas<sup>1</sup>, R. E. Everts<sup>1</sup>, H. A. Lewin<sup>1</sup>, J. R. Roche<sup>2</sup>, and J. J. Loo<sup>1</sup>, <sup>1</sup>*University of Illinois, Urbana*, <sup>2</sup>*DairyNZ Limited, Hamilton, New Zealand*, <sup>3</sup>*Cognosco, Animal Health, Morrinsville, New Zealand*, <sup>4</sup>*Liggins Institute, University of Auckland, Auckland, New Zealand*, <sup>5</sup>*University of Queensland Centre for Clinical Research, Brisbane, St. Lucia, Australia.*

Inflammation of the endometrial lining of the uterus (endometritis) often develops in dairy cows soon after calving. Afflicted animals can develop chronic infections of the uterus due to pathogenic bacteria. This disease causes poor reproductive performance, decreased energy balance, lower milk yield, and potentially hepatocellular damage. The objective of the current study was to uncover molecular changes induced by endometritis using expression profiling of liver and adipose transcriptomes. Postparturient liver and adipose tissue samples were harvested at slaughter from healthy (NUI; n = 6) cows or cows with endometritis (UIN; n = 6) at 29 d postpartum. Endometritis was defined as > 6% of uterine fluid nucleated cells being polymorphonuclear (PMN) cells; healthy cows had ≤ 1% PMN. Transcriptome expression was performed via bovine microarray (~13,000 oligonucleotides) and RT-PCR. Functional analysis of differentially expressed genes (DEG; P < 0.05, fold-change cut off ≥ or ≤ 1.5 UIN vs. NUI) was carried out using the dynamic impact approach (DIA) with the KEGG pathway database. A total of 97 DEG from liver (35 downregulated, 62 upregulated) and 144 from adipose (82 downregulated, 62 upregulated) were used for functional analysis. In liver, endometritis induced activation of selenoamino acid metabolism, complement and coagulation cascades, glycosphingolipid biosynthesis, and toll-like receptor signaling pathway. Bioinformatics analysis also revealed an inhibition of fatty acid oxidation, PPAR signaling, SNARE interactions in vesicular transport, and N-glycan biosynthesis. Functional analysis of adipose uncovered as the most induced pathways nicotinamide metabolism, glycosaminoglycan biosynthesis, and basal transcription factors. The most-inhibited pathways in adipose were taurine and hypotaurine metabolism, pantothenate and CoA biosynthesis, and TCA cycle. Quantitative PCR of liver tissue further revealed a marked

increase in superoxide dismutase-2 (SOD2) and glutathione peroxidase (GPX1) expression in UIN, suggesting a greater state of oxidative stress in those animals. Overall, these preliminary results suggest that uterine infection after calving induces molecular alterations in peripheral tissues.

**Key Words:** uterine infection, transition cow, genomics

**M4 A comparison of two antibiotics on growth performance in beef cattle treated for bovine respiratory disease (BRD).** N. O. Minton<sup>\*1</sup>, L. L. Hawkins<sup>2</sup>, and M. S. Kerley<sup>1</sup>, <sup>1</sup>University of Missouri, Columbia, <sup>2</sup>Bayer HealthCare, Animal Health, Shawnee Mission, KS.

Therapy for bovine respiratory disease (BRD) commonly occurs throughout the receiving period. Proper treatment of BRD is critical to cattle performance throughout the feeding phase. We hypothesized differences in growth performance occur among different antibiotics used for treatment of BRD. Naïve crossbred steers (n = 102) were monitored for BRD over a 45-d growth study. Steers were randomly assigned to antibiotic treatments which consisted of either enrofloxacin (ENR; n = 51) (Baytril 100 Bayer HealthCare, Animal Health) or florfenicol + flunixin (F+F; n = 51) (Resflor Gold Merck Animal Health). Within treatment steers were categorized as no treatment (control), single treatment (ST), relapse (RL) or chronic. Steers were randomly assigned to 2 pens with treatment evenly distributed within pen. Two consecutive BW were recorded at treatment initiation, d 28, and d 45. Individual FI using the GrowSafe feed intake system and visual assessment were used to identify cases of BRD. Rectal temperatures of  $\geq 40^{\circ}\text{C}$  were diagnosed with BRD and treated with predetermined antibiotic. Control steers represent non treated steers. Chi-squared distribution determined morbidity and relapse rate were not different among ENR and F+F. ENR had greater percentages of chronics (6.2% vs. 2.1%;  $P < 0.001$ ) with lower mortality rate (2.1% vs. 7.2%;  $P < 0.001$ ) than F+F; respectively. IBW was not different among all categories between both ENR and F+F. ADG, F:G and total BW gain were not different between control steers and ENR ST and RL steers. ENR ST and RL steers tended to be more efficient than either category of F+F steers ( $P = 0.11$ ). ENR ST steers had greater ADG (1.76+0.11 vs. 1.37+0.11;  $P < 0.05$ ), FBW (333.63+6.77 vs. 311.86+6.77;  $P < 0.05$ ) and BW gain (77.5+4.96 vs. 60.30+4.96;  $P < 0.05$ ) than ST F+F steers. DMI as a percent of BW was not different on treatment day as well as 3 d before and after treatment of BRD between ST ENR and F+F steers. We conclude utilizing enrofloxacin for therapy of BRD corresponds to an increase in growth performance and feed efficiency during the receiving period in comparison to florfenicol + flunixin.

**Key Words:** health, feedlot, beef

**M5 Feedback on data entry errors effect on the maintenance of accurate and consistent dairy health records.** S. K. Giebel<sup>\*1</sup>, J. R. Wenz<sup>1</sup>, S. A. Poisson<sup>1</sup>, C. S. Schneider<sup>2</sup>, and D. A. Moore<sup>1</sup>, <sup>1</sup>Department of Veterinary Clinical Sciences, Washington State University, Pullman, <sup>2</sup>College of Agricultural and Life Sciences, University of Idaho, Moscow.

Maintenance of data quality is not a one-time effort; it requires continuous auditing and evaluation to ensure quality. One reason for this is that health data entry is not standardized in dairy management software (DMS), like reproductive records. The purpose of this study was to evaluate the time needed for producers to implement and maintain health data entry protocols. Forty-three dairies enrolled in a demonstration project on consistent and accurate health records, 51% from Eastern Washington (EWA), 26% from Western Washington (WWA),

and 23% from Idaho (ID), with an average herd size of 2,683 cows (range 366–9,880). Producers were provided with error reports weekly until the percentage of errors dropped below 5.1% for 2 consecutive reports, at which point the dairy was considered graduated. All dairies continued to receive monthly error reports until January 2012. Reports provided feedback on data entry in comparison to their customized health event data recording protocols. The first report averaged 20.5% errors with a range of 0.0 to 81.3%. It took an average of 9 reports before a dairy graduated to monthly reporting, with a range of 4 to 21. Of those herds that contributed at least 3 mo to the study post graduation, nearly 50% never relapsed (relapse = error rate  $> 5\%$ ) but if they did, it most commonly occurred in the first month post graduation. The average months to relapse was one (range 0–6). The error rate on the last report averaged 4.7% with a range of 0.0–16.6%. If the herd was using the Protocol function in Dairy Comp 305 or the Rx-Plus function in DHI-Plus before the start of the project, months to relapse was shorter than those that did not ( $P = 0.01$ ). Median time to relapse was longer for large herds ( $> 1950$  cows) compared with smaller herds (4 vs. 2 mo;  $P = 0.06$ ). Median time to relapse was 4 mo for EWA, 2 mo for WWA and 2.5 mo for ID ( $P = 0.04$ ). For those producers less comfortable with computer use, there was a tendency for the rate of relapse to be faster compared with those more comfortable ( $P = 0.08$ ). Dairies can implement and maintain consistent and accurate health records when provided with regular feedback.

**Key Words:** data recording protocols, dairy management software, dairies

**M6 Impact of water and feed deprivation on physiological parameters in steers.** J. A. Daniel<sup>\*1</sup>, P. H. Walz<sup>2</sup>, J. A. Carroll<sup>3</sup>, T. H. Elsasser<sup>4</sup>, and B. K. Whitlock<sup>5</sup>, <sup>1</sup>Berry College, Mount Berry, GA, <sup>2</sup>Auburn University, Auburn, AL, <sup>3</sup>USDA-ARS Livestock Issues Research Unit, Lubbock, TX, <sup>4</sup>USDA-ARS Bovine Functional Genomics Laboratory, Beltsville, MD, <sup>5</sup>University of Tennessee, Knoxville.

A report in rats demonstrated that dehydration as the result of 8 d of water deprivation increased leakage of endotoxin from the intestine (Zurovsky and Barbiro, 2000; Exp. Toxicol. Pathol. 52:37–42). Given the large number of bacteria in the rumen of cattle, a much shorter period of water deprivation may result in increased leakage of endotoxin and subsequent changes in the physiologic parameters. Therefore, the purpose of this study was to determine the effect of water and feed deprivation on physiologic parameters in steers. Holstein steers (n = 4) fitted with indwelling jugular and hepatic vein cannulas received a 72-h period of water and feed deprivation followed by reintroduction of water and feed for a 24-h period (96 h total) or a 96-h period with ad libitum water and hay access (control) in a switch back design with a 3-wk wash-out period between repetitions. Blood samples were collected and rectal temperatures (RT) were recorded every 6 h for a total of 96 h. Body weights were recorded every 12 h. Data were tested for effects of treatment, time, treatment  $\times$  time interaction and replication using procedures for repeated measures with JMP software (version 7; SAS Institute Inc.). Water and feed deprivation increased packed cell volume at 30, 54, 60, and 66 h relative to control steers ( $P < 0.05$ ). Total protein in the serum was increased at 48, 54, 66, 72, and 78 h in water and feed deprived steers compared with control steers ( $P < 0.05$ ). The average body weight of water and feed deprived steers was less than control steers at 36, 48, 60, and 84 h ( $P < 0.05$ ), and percent of body weight loss was greater in water and feed deprived steers at 24, 36, 48, 60, 72, and 84 h than control steers ( $P < 0.05$ ). Rectal temperature of water and feed deprived steers was lower than control steers at 6, 12, and 66 h, and RT of water and feed deprived steers was higher than

control steers at 72 h ( $P < 0.05$ ). These results suggest that water and feed deprivation negatively affect steers before typical indicators of dehydration (packed cell volume and total protein) are altered.

**Key Words:** steers, water, feed

**M7 Implementation of health data entry protocols effect on time for data management.** S. K. Giebel\*<sup>1</sup>, J. R. Wenz<sup>1</sup>, S. A. Poisson<sup>1</sup>, C. S. Schneider<sup>2</sup>, and D. A. Moore<sup>1</sup>, <sup>1</sup>Department of Veterinary Clinical Sciences, Washington State University, Pullman, <sup>2</sup>College of Agricultural and Life Sciences, University of Idaho, Moscow.

Producers want to be assured that it will not add time to the process of data management when implementing protocols for consistent and accurate dairy health data entry. Research done on data capture and entry into dairy management software (DMS) program is limited and dated. The purpose of this study was to assess the time taken to capture and enter health data before and after implementation and maintenance of data entry protocols. Twenty-three dairies enrolled in the demonstration project were used to evaluate the time taken to capture health data and enter it into their DMS program. Information was collected on the general flow of data from cow to computer during 2 visits, one before implementation of accurate and consistent health records and one 4 mo after maintenance. The difference in time, for data capture and entry, between the first and second visit was categorized as increased, decreased or no change. There were 22 dairies with complete data for the capture of health information. On average, it took dairy personnel 21.8 (range 2.3–65.8) seconds per cow to capture information at the first visit. After data entry protocol maintenance, it took an average of 17.6 (range 1.0–46.6) seconds per cow. Using a sign test, herds were more likely to either take less or the same amount of time for data capture between visits ( $P < 0.001$ ). There were 18 dairies with complete data for entry of health information. On average, it took dairy personnel 4.6 (range 0.6–12.8) seconds per cow to enter data into the computer at the first visit. After protocol maintenance, it took an average of 3.3 (range 0.3–9.7) seconds per cow. Using a sign test, herds were more likely to either take less or the same amount of time for data entry between visits ( $P < 0.01$ ). The only factors that appeared to influence data capture were related to on-farm protocols such as recording all hospital cow numbers in the parlor and the frequency of data capture in the parlor. Implementation of health data entry protocols for more accurate and consistent records did not add time to the process of data management.

**Key Words:** data capture, data entry, dairy management software

**M8 Transcriptome analysis of liver tissue from calves infected with bovine viral diarrhea virus and *Mannheimia haemolytica*.** R. L. Mills\*<sup>1,2</sup>, L. Carlos-Valdez<sup>2</sup>, L. O. Burciaga-Robles<sup>2</sup>, D. Stein<sup>2</sup>, D. L. Step<sup>2</sup>, R. W. Fulton<sup>2</sup>, U. DeSilva<sup>2</sup>, and C. R. Krehbiel<sup>2</sup>, <sup>1</sup>Austin Peay State University, Clarksville, TN, <sup>2</sup>Oklahoma State University, Stillwater.

Bovine respiratory disease (BRD) negatively affects animal growth and performance, leading to less desirable carcass characteristics. To understand the effects of BRD on liver gene expression, 16 crossbred steers (280.8 ± 32.5 kg) were divided into 2 treatment groups: the control group (CON) received an intratracheal dose of isotonic saline; an infected group (INF) was exposed to a PI-BVDV calf for 72 h followed by intratracheal inoculation with *Mannheimia haemolytica* serotype A (MH). Liver biopsies were taken before exposure to both pathogens (PRE), 12 (12H), and 24 h (24H) following bacterial inoculation. Total RNA was extracted and oligonucleotide microarray hybridization performed on

PRE and 24H sampling times ( $n = 6$ ). Significance for the microarray was set at fold change greater than |2| and  $P < 0.001$ . Quantitative RT-PCR was performed to examine gene expression at the indicated time points with significance set at  $P < 0.05$ . Pathway analysis was conducted using Ingenuity Pathway Analysis. Microarray analysis found 44 genes were induced and 45 genes were suppressed following 24h of dual infection. Pathway analysis revealed lipid metabolism, molecular transport, cellular assembly and organization, carbohydrate metabolism, and small molecule biochemistry were the molecular and cellular functions most affected. PCR showed that  $\beta$ -site APP-cleaving enzyme 1 (BACE1) was increased 2.3-fold in INF compared with CON ( $P < 0.05$ ). S100 calcium binding protein A8 (S100A8) transcripts increased 28.9-fold in INF versus CON ( $P < 0.001$ ), increased more than 21-fold in 12H and 24H above PRE ( $P < 0.01$ ), and exhibited an infection × time interaction with INF-12H and INF-24H showing mRNA increases of 44- and 42-fold above CON-PRE, respectively ( $P < 0.01$ ). Fatty acid desaturase 2 (FADS2) was suppressed 73.2-fold in INF relative to CON ( $P = 0.05$ ) and tended to be downregulated 29.5- and 95.1-fold at 12H and 24H versus PRE ( $P = 0.09$ ). Gene expression changes in liver are indicative of hepatic inflammation, which may suggest calves with BRD are susceptible to the development of fatty liver.

**Key Words:** beef cattle, bovine respiratory disease, gene expression

**M9 See abstract #100**

**M10 Changes in biomarkers of the nitrooxidative stress response and prolactin signal transduction elements to *E. coli* infection in the mammary gland.** T. H. Elsasser\*<sup>1</sup>, A. V. Capuco<sup>1</sup>, M. Rinaldi<sup>2</sup>, and S. Kahl<sup>1</sup>, <sup>1</sup>USDA-ARS, Beltsville, MD, <sup>2</sup>Ghent University, Ghent, Belgium.

Key features of the mammary gland (MG) response to *E. coli* infection (INFec) include (a) the cellular generation of reactive oxynitrogen molecules (Roxn) derived from nitric oxide (NO) and superoxide anion ( $O_2^{\cdot-}$ ) and (b) loss of responsiveness to lactogenic hormone signaling. Of significance to the MG is the damage done to mammary epithelial cells (MEC) by Roxn generated by MEC and neutrophils (PMN). The aim of this study was to determine the time course of production and cellular localization of biomarkers of the Roxn pathway and to quantify changes in PRL receptor (PRLr)-janus kinase-2 (JAK-2) signaling. As applied across the 4 quarters of the MG, 5 multiparous lactating Holstein cows received treatments consisting of PBS or 400 cfu *E. coli* administered 12 or 24 h before euthanasia. MG tissues (lobulo-alveolar) were prepared for immunohistochemical (IHC) localization of biomarker antigens. IHC biomarkers for the Roxn pathway elements consisted of 3'-nitrotyrosine (3-NT, cellular protein nitration) as well as inducible (i) and constitutive (c) isoforms of nitric oxide synthase (NOS, NO generation), and xanthine oxidase (XO,  $O_2^{\cdot-}$  generating capacity). Prolactin (PRL) signal transduction elements markers were PRLr, JAK-2, and nitrated JAK-2 (ntJAK, Roxn-inactivated JAK-2). Antigenic markers were quantified by image analysis of digitally captured micrographs. Mean pixel densities of all antigenic determinants were affected by time and/or cell type ( $P < 0.02$ ). Infiltrating PMN were present only at 24 h. The 3-NT increase peaked in MEC (4.6-fold) at 12h and in PMN (3.7-fold) at 24 h. Both iNOS and eNOS increased in MEC at 12 and 24 h but only iNOS was increased in PMN at 24 h. XO increased 118% in MEC at 12 h ( $P < 0.04$ ) but normalized by 24h. PRLr was decreased by 29 ( $P < 0.03$ ) and 54% ( $P < 0.01$ ) in MEC at 12 and 24 h, respectively. JAK-2 was decreased 84% ( $P < 0.02$ ) at 24 h, respectively. ntJAK was increased 22- ( $P < 0.005$ ) and 4-fold ( $P < 0.05$ ) at 12 and 24 h, respectively. The

data indicate that changes in these measured cellular responses to INFec occur in the MG as a function of time and cell type.

**Key Words:** mammary gland, infection, biomarker

**M11 Associations among subclinical hypocalcemia, neutrophil function, and incidence of uterine disease in dairy cows of low or high risk of developing metritis.** N. Martinez<sup>\*1</sup>, F. S. Lima<sup>1</sup>, R. S. Bisinotto<sup>1</sup>, L. F. Greco<sup>1</sup>, E. S. Ribeiro<sup>1</sup>, F. Maunsell<sup>2</sup>, K. N. Galvão<sup>2</sup>, C. A. Risco<sup>2</sup>, and J. E. P. Santos<sup>1</sup>, <sup>1</sup>Department of Animal Sciences, University of Florida, Gainesville, <sup>2</sup>Department of Large Animal Clinical Sciences, University of Florida, Gainesville.

Objectives were to establish relationships among subclinical hypocalcemia (SCH) and concentrations of energy metabolites, neutrophil (PMN) function and incidence of uterine diseases in dairy cows considered to be of low (LRM; normal calving) or high-risk (HRM; calving problems) of developing metritis. In this prospective cohort study, HRM cows (n = 55) were matched with LRM (n = 55) based on parity and day of calving. Rectal temperature (RT) and vaginal discharge were monitored in the first 12 DIM. Metritis was defined as fetid, watery vaginal discharge, and puerperal metritis (PuMet) was defined as metritis concurrent with RT  $\geq$  39.5°C. Blood was sampled at 0, 1, 2, 3, 4, 7, and 12 DIM and analyzed for Ca, NEFA and BHBA. Neutrophil function was measured at 0, 1, and 3 DIM. Serum Ca  $\leq$  8.59 mg/dL in at least one day within the first 3 DIM defined SCH based on receiver operator characteristic analysis (AUC = 0.77;  $P < 0.01$ ). Data were analyzed using PROC GLIMMIX of SAS. Cows with SCH had fewer ( $P < 0.01$ ) circulating blood PMN (3.0 vs. 4.5  $\times 10^3$  PMN/ $\mu$ L) in the first 3 DIM, and reduced ( $P < 0.05$ ) proportion of PMN with oxidative burst (32.4 vs. 42.5%) and phagocytosis (61.3 vs. 73.1%) at 3 DIM compared with normocalcemic (NC) cows. The mean RT of cows increased ( $P < 0.01$ ) when metritis was associated with SCH (39.0°C) compared with cows with metritis and NC (38.7°C). Among HRM cows, those with SCH had greater ( $P < 0.05$ ) incidence of metritis and PuMet [77.8% (35/45) and 53.5% (24/45)] compared with NC cows [(20.0% (2/10) and 10.0% (1/10)]. Among LRM cows, those with SCH had greater ( $P < 0.05$ ) incidence of metritis and PuMet [40.7% (11/27) and 29.6% (8/27)] compared with NC cows [(14.3% (4/28) and 0.0% (0/28)]. Metritis did not influence concentrations of NEFA or BHBA; however, cows with SCH had greater ( $P < 0.01$ ) NEFA (704.6 vs. 426.8  $\mu$ M) and BHBA (9.9 vs. 7.7 mg/dL) concentrations compared with NC cows. These findings indicate that cows with SCH have impaired innate immunity and increased incidence of uterine diseases compared with NC cows regardless of calving problems.

**Key Words:** subclinical hypocalcemia, neutrophil function, metritis

**M12 Hepatic and peripheral interferon responses to bovine respiratory disease in feedlot steers.** J. O. Baggerman,\* C. A. Gifford, and C. R. Krehbiel, Oklahoma State University, Stillwater.

Bovine respiratory disease (BRD) is the most economically important disease in the feedlot industry. Several pathogens contribute to BRD, yet the mechanism by which these pathogens evade immune response remains unclear. Conflicting evidence suggests that the interferon (IFN) response is impaired by BRD pathogens while others have shown robust IFN response in BRD-infected animals. We hypothesize that the IFN response is muted in cattle that exhibit sustained BRD infection. Liver biopsies were collected and RNA isolated from control calves (CONT, n = 6); calves treated once for BRD (1TRT, n = 6); treated twice for BRD (2TRT, n = 6); or treated 3 times for BRD (3TRT, n = 6). Expression of Interferon Stimulated Gene-15 (*ISG15*) and Myxovirus resistance

protein 1 (*MX1*) mRNA levels was quantified. Relative to CONT, *MX1* mRNA levels increased ( $P < 0.05$ ) over 7-fold in liver biopsies from 1TRT, but were similar ( $P > 0.10$ ) to CONT for 2TRT and 3TRT. Likewise, *ISG15* mRNA levels were 40-fold greater ( $P < 0.05$ ) than CONT for 1TRT, but 2TRT and 3TRT were similar to CONT. To determine if the IFN response is impaired with sustained BRD infection as suggested by the changes in gene expression in liver biopsies, *MX1* mRNA levels in peripheral blood leukocytes (PBL) was evaluated from chronically infected calves (CHRON, n = 3) and controls (CONT, n = 3). Additionally, peripheral blood mononuclear cells (PBMC) were isolated from CHRON (n = 6) and CONT (n = 6) calves. For each animal, PBMC were isolated and plated into 6 well plates (4 million cells/mL; 4 wells/animal). Two wells served as a control and 2 wells were treated with human interferon- $\alpha$  (Sigma-Aldrich; 2.5 ng/mL) for 8 h before harvest for mRNA extraction. In PBL samples, *MX1* mRNA levels were similar ( $P > 0.10$ ) in CHRON and CONT2 indicating that the type I interferon response was not active. However, in PBMC challenged with IFN, *MX1* mRNA increased approximately 100-fold in both CHRON and CONT2 groups compared with their own non-treated controls. Results demonstrate that the IFN response is reduced in calves treated multiple times for BRD, but the IFN pathway is still responsive to exogenous IFN. Thus, IFN might be a viable treatment alternative in calves that remain sick after 1 to 2 treatments for BRD.

**Key Words:** bovine respiratory disease, interferon, interferon stimulated genes

**M13 Meta-analysis of *Trypanosoma* prevalence in livestock in the Americas.** Z. J. Simoni<sup>1</sup>, H. E. Rodulfo<sup>1</sup>, M. De Donato<sup>\*1,2</sup>, M. I. Takeet<sup>3</sup>, S. O. Peters<sup>2,4</sup>, and I. G. Imumorin<sup>2</sup>, <sup>1</sup>IIBCA, Universidad de Oriente, Cumana, Venezuela, <sup>2</sup>Dept. Animal Science, Cornell University, Ithaca, NY, <sup>3</sup>Dept. Veterinary Microbiology & Parasitology, Federal University of Agriculture, Abeokuta, Nigeria, <sup>4</sup>Dept. Animal Breeding and Genetics, University of Agriculture, Abeokuta, Nigeria.

Cattle tripanosomosis is a hemoparasitic disease caused by species of *Trypanosoma*, transmitted through dipterans of the genus *Tabanus*, in Central and South America. *T. evansi* and *T. vivax* are species that infect horses and cattle, respectively. They have been reported in at least 10 South American countries, including Colombia, Venezuela, Bolivia, Peru, and Brazil. We surveyed data on prevalence and clinical signs reported for *T. vivax* in water buffalo and cattle in these countries. Studies of genetic characterization indicate that the circulating species in South America is very similar to *T. vivax* from West Africa. *T. vivax* has been known to produce economic losses in the livestock industry, causing mortality, poor weight gain, lower fertility, abortion, and low production of milk and colostrums, in addition to costs associated with control (drugs and veterinary fees). *T. evansi* can cause a serious anemic disease in horses, camels, dogs and elephants. In South America, species such as capybara, coati, and wild rats have been found infected by *T. evansi*, without showing clinical alterations, thus, being considered as natural reservoirs. Recent studies have demonstrated that *T. evansi* has an aberrant kADN without maxicircles and the genetic analysis of kADN have shown that strains from equids of Africa, Asia y South America have similar sequences in the minicircles. *T. theileri* has been found in cattle in South and North America, and although this species is considered non-pathogenic, its pathogenicity has been linked to coinfection with other hemoparasites or subjected to severe stress. *T. equiperdum* has been reported in South and Central America in horses, but its form of transmission is by sexual intercourse, associated with important clinical symptoms. The effects of these *Trypanosoma* species on livestock throughout this region especially in isolated rural areas

with attendant consequences on the economy of small to medium size producers are highlighted.

**Key Words:** trypanosomiasis, livestock, parasite

**M14 Cytokine production of isolated CD4+ T-cells from high and low immune responder dairy cows during the peripartum period.** M. A. Paibomesai\* and B. Mallard, *University of Guelph, Guelph, Ontario, Canada.*

The peripartum period is one of transition and high stress for dairy cows. It is associated with higher incidents of both metabolic and pathogenic disease. Both antibody (AMIR; IL-4 dominated) and cell mediated (CMIR; IFN $\gamma$  dominated) immune responses (IR) play a key role in the maintenance of health in cattle protecting against extracellular and intracellular pathogens. These processes are induced and maintained by CD4+ T-helper cells, which undergo specific shifts in population and function in response to parturition effects. The aim of this study was to determine the effects of parturition on cytokine production of CD4+ T-cells isolated from highAMIR/lowCMIR (hiAMIR) and highCMIR/lowAMIR (hiCMIR) cows. Previously IR phenotyped cows were selected based on hiAMIR (n = 12) and hiCMIR (n = 11) responses to test antigens. Isolated CD4+ T-cells collected at 28 d prepartum, 4 d postpartum, and 21 d postpartum from these groups were stimulated with the mitogen, Concanavalin A (ConA) and incubated for 24hrs. Cell culture supernatants were collected and IL4 and IFN $\gamma$  concentrations were quantified by ELISA and analyzed using ANOVA. There was no difference in IL4 concentration across all time points for hiCMIR cows and an increase of IFN $\gamma$  ( $P = 0.0223$ ) was observed between 4 d postpartum (3366 pg/mL) and 21 d postpartum (7755 pg/mL). There was a decrease of IL4 ( $P = 0.0474$ ) from 28 d prepartum (496 pg/mL) to 21 d postpartum (212 pg/mL) in hiAMIR cows and no difference observed for IFN $\gamma$  concentration in this group. There was a difference between hiAMIR and hiCMIR groups at 21 d postpartum with hiCMIR isolated T-cells producing more IL4 than hiAMIR group ( $P = 0.0161$ ) and a tendency to produce more IFN $\gamma$  ( $P = 0.0591$ ). In conclusion, it is evident that T-helper cells isolated from hiAMIR and hiCMIR show different responses to parturition effects in terms of cytokine production. These differences could be in response to biases in CMIR and AMIR in these 2 groups and was most notable at 21 d from parturition.

**Key Words:** peripartum period, cytokines, T-helper cells

**M15 Space allowance influences Holstein bull calf innate immunity after castration.** L. E. Hulbert<sup>1</sup>, M. S. Calvo\*<sup>1</sup>, M. A. Ballou<sup>2</sup>, K. C. Klasing<sup>1</sup>, and F. M. Mitloehner<sup>1</sup>, <sup>1</sup>*Department of Animal Science, University of California, Davis,* <sup>2</sup>*Animal and Food Sciences, Texas Tech University, Lubbock.*

Objectives were to determine if space allowance in wooden hutches influences the innate immune responses of Holstein bull calves after castration. Calves were randomly assigned at 4 d of age to either conventional (Cnv; 0.46 m<sup>2</sup> space; n = 18), medium (Med; 0.70 m<sup>2</sup>space; n = 17), or large (Lrg; 1.39 m<sup>2</sup> space; n = 18) hutches. Calves were surgically castrated at 24 d. Peripheral whole blood (WB) samples were collected at -1, 1, 5, and 12 d relative to castration. All calves had increased plasma cortisol 1 d after castration ( $P \leq 0.01$ ). Plasma haptoglobin (Hap) increased from pre-castration concentrations 1 d after castration in all calves, but Med-calves had greater Hap 5 d after castration compared with pre-castration ( $P \leq 0.05$ ). Cnv-housed calves had greater Hap concentrations than Med- and Lrg-housed calves, especially at 12 d after castration ( $P \leq 0.05$ ). The day before castration, Cnv-calves had

the greatest WB killing (WBK) of *E. coli*, but 1 d after castration, WBK in Cnv-calves decreased to percentages lower than Med- or Lrg-calves ( $P \leq 0.05$ ). Within 12 d after castration, WBK in Cnv-calves returned to similar percentages as pre-castration. The Lrg-calves had the least Tumor Necrosis Factor (TNF)- $\alpha$  concentrations from WB stimulated with endotoxin 1 d after castration ( $P \leq 0.05$ ), but Lrg-calve TNF- $\alpha$  increased at 5 d after castration to concentrations greater than 1 d before castration ( $P \leq 0.05$ ). Percentage of neutrophils undergoing phagocytosis and oxidative burst to *E. coli* tended ( $P = 0.06$ ) to decrease in Lrg-calves following castration, with the least percentage at 12 d after castration. Lrg-calves also had suppressed intensity of neutrophil oxidative burst compared with the other calves ( $P \leq 0.01$ ). Modifying wooden hutches to the maximum space did not seem to positively influence calf innate immune function during castration, when there is a great risk of infection.

**Key Words:** free-space, inflammation, stress

**M16 Effects of *Bacillus cereus* var. *toyoi* (Toyocerin) on the immune system of calves.** A. Aris\*<sup>1</sup>, A. Serrano<sup>1</sup>, M. Terré<sup>1</sup>, G. Jiménez<sup>3</sup>, M. Castillo<sup>3</sup>, and A. Bach<sup>1,2</sup>, <sup>1</sup>*Department of Ruminant Production, IRTA, Caldes de Montbui, Spain,* <sup>2</sup>*Institució Catalana de Recerca i Estudis Avançats (ICREA), Barcelona, Spain,* <sup>3</sup>*Rubinum SA, Rubí, Spain.*

*Bacillus cereus* var. *toyoi* (Toyocerin) has shown beneficial effects on the immune system of pigs and poultry, but it has not been studied on cattle. The objective of this experiment was to evaluate the effects of Toyocerin on the immune system of calves. Twenty-four Holstein calves (75.2  $\pm$  1.87 kg of BW and 77  $\pm$  0.7 d of age), were randomly distributed according to BW in 2 groups: 12 calves were assigned to a conventional concentrate feed (CTR), and the other 12 calves were fed the same concentrate feed supplemented with 2  $\times$  10<sup>8</sup>cfu *Bacillus cereus* var. *toyoi*/kg concentrate (TOY). Animals were vaccinated at d 0 with IBRaxion vaccine (Merial) against infectious bovine rhinotracheitis (IBR) and re-vaccinated at d 20 of the study. On d 0 and 56, blood samples were taken to assess the antibody titer against IBR vaccine. Animals were killed at 60 to 67 d of the study. Before sacrifice, a blood sample was obtained to evaluate the proliferation capacity of peripheral blood mononuclear cells (PBMC). Immediately after sacrifice, animals were abdomen opened, and the whole gastrointestinal tract was removed. Jejunal fragments were sampled to analyze IgA and cytokines expression by qPCR. Data were analyzed by ANOVA using the treatment as the main effect. There were no differences in PBMC proliferation between the 2 treatments. The TOY calves tended ( $P = 0.07$ ) to present a greater antibody response than CTR calves (92.5  $\pm$  1.50% vs. 82.5  $\pm$  3.50%, respectively); whereas no differences in IgA expression were observed at jejune level. Calves in the TOY group showed a clear increase ( $P < 0.05$ ) in the expression level of Th2 cytokines such as IL-10 (3.12  $\times$  10<sup>-3</sup>  $\pm$  0.8 $\times$ 10<sup>-3</sup>) in comparison to calves in the CTR group (1.37  $\times$  10<sup>-3</sup>  $\pm$  0.3  $\times$  10<sup>-3</sup>), which would explain the observed improvement in humoral response to vaccination. On the other hand, the levels of Th1 cytokines related to cellular response, such as IL-12 and IL-1 $\beta$ , were numerically lower in TOY group than in CTR group. In conclusion, these preliminary results indicate that Toyocerin exerts a positive effect on the modulation of the immune response in calves.

**Key Words:** *Bacillus toyoi*, immune response, probiotic

**M17 Space allowance influences the innate immune responses of Holstein calves during weaning.** L. E. Hulbert<sup>1</sup>, M. S. Calvo<sup>1</sup>, M. A. Ballou<sup>2</sup>, K. C. Klasing<sup>1</sup>, and F. M. Mitloehner<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of California, Davis, <sup>2</sup>Animal and Food Sciences, Texas Tech University, Lubbock.

Objectives were to determine if increased space allowance in wooden hutches influences the innate immune responses of Holstein bull calves during weaning. Calves were randomly assigned at 4 d of age to either conventional (Cnv; 0.46 m<sup>2</sup> space; n = 18), medium (Med; 0.70 m<sup>2</sup> space; n = 17), or large (Lrg; 1.39 m<sup>2</sup> space; n = 18) hutches. Calves were fed 227 g as-fed milk replacer (MR) twice daily (am and pm) until weaning and offered a calf starter ad libitum throughout the experiment. Weaning was initiated at age 53 d by removal of the pm MR feeding and was completed at age 64 d by removal of the am MR. Peripheral whole blood (WB) samples were collected before the am MR at age 53, 57, 64, 67 and 71 d. At age 67 and 71 d, all calves had increased circulating cortisol and neutrophil:lymphocyte and decreased Interferon- $\gamma$  secretion from WB stimulated with phytohemagglutinin, compared with pre-weaning measures ( $P \leq 0.01$ ). Also, at age 67 and 71 d, Med-calves had greater tumor necrosis factor- $\alpha$  secretion from WB stimulated with endotoxin than the other treatments ( $P \leq 0.05$ ). In all calves, plasma urea nitrogen (UN) was increased at age 67 d compared with pre-weaning ( $P \leq 0.01$ ) and was greatest among Cnv-calves ( $P \leq 0.05$ ). Plasma haptoglobin (Hap) increased from pre-weaning in Cnv- and Med-calves ( $P \leq 0.01$ ), but did not change over time among Lrg-calves. In addition, Lrg-calves had the least WB killing of *E. coli* at age 64 d ( $P \leq 0.05$ ) and had less neutrophil phagocytosis and oxidative burst responses to heat-killed *E. coli* at age 53 and 64 d compared with the other calves ( $P \leq 0.05$ ). Weaning was stressful for all calves, especially after the second bottle was removed. However, a moderate increase in space allowance for calves housed in wooden hutches may be beneficial to innate immune responses during weaning, while the largest increase in space allowance may negatively affect innate immunity.

**Key Words:** free-space, wean, inflammation

**M18 Group-housed Holstein bull calves have decreased innate immune responses compared to individually housed calves after surgical castration.** L. E. Hulbert<sup>1</sup>, M. S. Calvo<sup>1</sup>, R. A. Kurzbard<sup>1</sup>, M. A. Ballou<sup>1</sup>, K. C. Klasing<sup>1</sup>, and F. M. Mitloehner<sup>1</sup>, <sup>1</sup>Department of Animal Science, University of California, Davis, <sup>2</sup>Animal and Food Sciences, Texas Tech University, Lubbock.

Objectives were to (1) determine if housing calves in groups of 3 (Group; n = 9 pens) or individually (Indv; n = 18) influences the innate immune responses of Holstein bull calves following surgical castration and (2) determine whether the innate immune responses of grouped calves is influenced by their ADG ranking within a pen (low = 0.80  $\pm$  0.116; mid = 1.27  $\pm$  0.116; or high = 1.68  $\pm$  0.116 kg/d. All calves were surgically castrated at 24 d of age. Peripheral whole blood (WB) samples were collected at -1, 1, 5, and 12 d relative to castration. At 12 d after castration, all calves had greater plasma cortisol than pre-castration measures ( $P < 0.01$ ). In Indv calves, neutrophil:lymphocyte (N:L) did not change after castration ( $P > 0.10$ ), but N:L increased in Group calves 1 d after castration ( $P \leq 0.05$ ). In addition, 12 d after castration, Group calves had greater plasma haptoglobin (Hap) than Indv-calves ( $P < 0.01$ ). In contrast, before castration Indv-calves had greater WB killing (WBK) of *E. coli* than Group calves ( $P \leq 0.05$ ) and the day after castration, WBK decreased in Indv calves to percentages lower than Group-calves ( $P \leq 0.05$ ). Within 12 d after castration the WBK of Indv calves had returned to pre-castration measures ( $P \leq 0.05$ ). At 5 and 12 d after castration, all calves had increased tumor necrosis factor (TNF)- $\alpha$  concentrations in

WB stimulated with endotoxin compared with pre-castration ( $P < 0.01$ ). Over the entire study period, neutrophil oxidative burst response to heat-killed *E. coli* was lower in Group calves compared with Indv calves ( $P \leq 0.05$ ). Mid-ranked Group calves had higher interferon- $\gamma$  secretion from whole blood cultures stimulated with phytohemagglutinin 1 and 12 d after castration than either the high- or low-ranked calves ( $P \leq 0.05$ ). These data suggest that group housing calves in wooden hutches during castration may negatively influence innate immune function. However, group-housing had the least negative effect on immune function in mid-performing calves.

**Key Words:** bovine, grouping, stress

**M19 A transient receptor potential channel 4 (TRPC4) gene to study response to gastrointestinal nematode infection in parasite-resistant goats.** M. M. Corley\* and J. Ward, Virginia State University, Petersburg.

Gut expulsion of a variety of mammalian (human, sheep, and mice) nematodes requires mechanisms that enhance gut contractions and glycoprotein hyper-secretion to allow detachment of the nematode from the gut wall. *Haemonchus contortus*, the blood sucking gastrointestinal nematode (GIN), costs the global livestock industry billions of dollars per year in lost production and anthelmintic drug costs. The transient receptor potential cation channels (TRPC) allows membrane depolarization and entry of calcium into the cell, thereby resulting in smooth muscle contraction. Deletion of TRPC4 in mice greatly reduces intestinal motility and contractility. To date, the relationship between gene expression of TRPC4 and the response to GIN infection has not been assessed in goats or any other species. This study evaluated gene expression of TRPC4 in selected pasture exposed parasite resistant goats. Intestinal tissues were harvested from goats exhibiting susceptibility and resistance to *H. contortus*. Primers were designed from conserved regions of TRPC4 human, bovine, mouse and rat nucleotide sequence alignments. Total RNA was extracted from homogenized goat intestinal tissues and real time RT-PCR (qRT-PCR) was performed to determine TRPC4 gene expression. The qRT-PCR results showed that TRPC4 was upregulated ( $P < 0.05$ ) in goats naturally susceptible to *H. contortus* infection. Breed and gender differences ( $P < 0.05$ ) in TRPC4 expression were observed. Overall, there was a strong positive ( $P < 0.05$ ) correlation between TRPC4 gene expression and FAMACHA eye color chart scores and fecal egg counts, and strong negative correlation ( $P < 0.05$ ) with packed cell volume in goats. These data indicate that TRPC4 may be useful in elucidation of the relationship between gastrointestinal contractility and the response to gastrointestinal nematode infection in goats as well as other species.

**Key Words:** goat, gastrointestinal nematode, TRPC4

**M20 Use of selected blood parameters to identify markers of heat-sensitivity in Angus and Romosinuano heifers.** R. Chaffin,\* B. A. Scharf, J. S. Johnson, J. Bryant, D. Kishore, P. A. Eichen, and D. E. Spiers, University of Missouri, Columbia.

A study was performed to determine blood parameter differences in heat-sensitive Angus (ANG) and heat-tolerant Romosinuano (RO) heifers in response to heat stress. Eighteen month-old ANG (n = 11; 306.7  $\pm$  25.9 Kg BW) and RO (n = 10; 312.9  $\pm$  32.0 kg BW) heifers were maintained in the Brody Environmental Center (University of Missouri-Columbia) at cycling thermoneutral air temperature (TN, 18.5–23.5°C) for 7 d, followed by either continued maintenance at TN or exposure to heat stress (HS, 18.5–38°C) for the remaining 12 d of the study. Heifers received

melengestrol acetate (0.5mg/animal/day) to suspend reproductive cycling. Blood samples were obtained via jugular venipuncture at TN (d 5), early HS (EH, d 10), and late HS (LH, d 17), and analyzed for a variety of parameters. Rectal temperature (Tcore) was measured with a thermistor thermometer 6 times daily. On d 5, average Tcore did not differ between breeds ( $P = 0.89$ ). During EH and LH, Tcore was significantly higher in ANG overall ( $P = 0.001$ ), and higher among HS ANG than other groups ( $\sim 1^\circ\text{C}$ ,  $P = 0.025$ ). Though insignificant, Tcore tended to rise continually between EH and LH among HS ANG ( $\sim 0.4^\circ\text{C}$ ) while falling in HS RO ( $\sim 0.3^\circ\text{C}$ ). RO had lower triglycerides (TAG) than ANG on d 5 ( $P = 0.028$ ). On d 10 and 17, RO still exhibited lower TAG than ANG ( $P = 0.004$ ), with no effect of HS on either breed. Cholesterol (CHO) was not different between ANG and RO on d 5 ( $P = 0.36$ ), but ANG showed higher levels of CHO than RO during treatment ( $P = 0.04$ ), with HS exposure causing a further reduction below TN level ( $P = 0.02$ ). On d 5, RO had higher alkaline phosphatase (ALP) than ANG ( $P = 0.012$ ), as well as on d 10 and 17 ( $P = 0.004$ ). In general, HS lowered ALP on d 17 from the pretreatment level ( $P = 0.05$ ). These results suggest that blood TAG and ALP levels may serve as markers for detecting potential heat-sensitivity before stress, while blood CHO and ALP levels may be useful markers for heat-sensitivity in beef heifers during stress.

**Key Words:** heat stress, Angus, Romosinuano

**M21 Variation in innate immune parameters in Holstein calves is influenced by housing environment and physiological period.** M. D. Sellers,\* D. L. Hanson, A. R. Pepper-Yowell, C. J. Cobb, and M. A. Ballou, *Department of Animal and Food Sciences, Texas Tech University, Lubbock.*

Objective was to determine if housing environment and physiological state influence the proportion of variation in innate immune parameters due to time, between calves, and within calves during the neonatal, weaning, and commingling periods in Holstein calves. Ninety-nine calves (2  $\pm$  1 d old) were randomly assigned to 5 treatments: 1, 2, or 3 calves/pen indoors (IN1,  $n = 21$ ; IN2,  $n = 20$ ; IN3,  $n = 18$ ), and 1 or 3 calves/pen outdoors (OUT1,  $n = 21$ ; OUT3,  $n = 20$ ). Weaning started on d 43 with removal of PM milk, and was completed when starter consumption was 800 g/d. Calves were commingled on d 91 by randomizing all calves into outdoor group hutches ( $n = 5$  calves/hutch). Peripheral blood samples were collected during neonatal (d 3, 10, 21), weaning (d 45, 47, 53), and commingling (d 91, 94, 99) periods and analyzed for neutrophil oxidative burst (OB) capacity when cocultured with an *Escherichia coli*, neutrophil L-selectin expression, whole blood secretion of tumor necrosis factor- $\alpha$  (TNF) when cocultured with lipopolysaccharide, and plasma concentration of haptoglobin. For each treatment within a period, variation was partitioned using Type III sums of squares estimates and reported as eta-squared, or the proportion of variation attributable to a given effect compared with total variation. Sources of variation included: variation due to time (DAY), between-calf (BET) and residual (RES). Values are reported as ranges or means across treatments. During the neonatal period, OUT3 calves had a larger BET proportion than OUT1 calves for neutrophil L-selectin expression (0.44 vs. 0.20) and OB capacity (0.54 vs. 0.29), and plasma haptoglobin (0.25 vs. 0.16). During weaning, OUT1 calves had higher BET proportions than OUT3 calves (0.56 vs. 0.23) for haptoglobin. The BET accounted for more variation across treatments during weaning when compared with the neonatal period for TNF secretion (0.57 vs. 0.40), percent of cells OB positive (0.64 vs. 0.51), and OB capacity (0.66 vs. 0.43). Proportion of variation due to DAY was lower during weaning than during the neonatal period (0.13 vs. 0.39). The BET variation for percentage of neutrophils positive for OB decreased during commingling compared with weaning (0.45

vs. 0.64). Differences in housing environment and physiological period influence proportion of residual variation, variation between calves, and variation due to time.

**Key Words:** calf, housing, immune

**M22 Intravaginal administration of lactic acid bacteria modulated innate immune responses of periparturient dairy cows.** Q. Deng, J. F. Odhiambo, T. Lam, S. M. Dunn, and B. N. Ametaj,\* *Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB, Canada.*

Dairy cows are characterized by a suppressed immunity around parturition, which increases their susceptibility to uterine infections. The goal of this investigation was to evaluate innate immune responses of transition dairy cows administered intravaginally with a mixture of lactic acid bacteria (LAB). In total, 152 pregnant Holstein cows were randomly assigned, based on parity and BCS, into 3 groups 2 wk before the expected day of calving. Cows received intravaginally a mixture of LAB or carrier (sterile skim milk) once per week at -2, -1, and +1 wk relative to the day of calving as following: treatment 1 (TRT1) - 2 consecutive LAB and 1 carrier dose; treatment 2 (TRT2) - 3 consecutive LAB doses; control (CTR) - 3 consecutive carrier doses. LAB were a mixture of *Lactobacillus sakei* FUA3089, *Pediococcus acidilactici* FUA3138, and FUA3140 stored in sterilized skim milk. The LAB were infused intravaginally with a sterile insemination pipette at  $10^8$  to  $10^9$  cfu per dose. Blood samples were collected from the tail vein before each treatment and once per wk up to +8 wk. A subset of serum samples from 32 cows were used to analyze TNF- $\alpha$ , haptoglobin, and lipopolysaccharide-binding protein (LBP) by ELISA. Results demonstrated that both probiotics and parity had an effect on the concentration of TNF- $\alpha$  in serum. The concentrations of TNF- $\alpha$  were 231, 706, and 326 pg/mL in TRT1, TRT2, and CTR, respectively ( $P < 0.05$ ). Primiparous cows had lower concentrations of TNF- $\alpha$  than multiparous cows (189 vs. 654 pg/mL) ( $P < 0.01$ ). The interaction between TRT and parity showed a tendency to affect the concentration of TNF- $\alpha$  ( $P < 0.1$ ). Concentrations of haptoglobin in serum were 407, 262, and 390  $\mu\text{g/mL}$  in TRT1, TRT2, and CTR, respectively ( $P > 0.05$ ). There was an interaction of TRT and parity on concentrations of haptoglobin ( $P < 0.05$ ) in the serum, although no effect ( $P > 0.05$ ) of treatments was obtained. Concentrations of LBP in the serum were 7,878, 7,500, and 8,337 ng/mL in TRT1, TRT2, and CTR, respectively ( $P > 0.05$ ). Serum concentrations of both haptoglobin and LBP changed with wk ( $P < 0.01$ ). Overall, intravaginal application of LAB modulated innate immune responses in transition dairy cows.

**Key Words:** dairy cows, lactic acid bacteria, innate immunity

**M23 Intravaginal administration of a mixture of lactic acid bacteria lowered the incidence of clinical diseases in transition dairy cows.** Q. Deng, J. F. Odhiambo, T. Lam, S. M. Dunn, and B. N. Ametaj,\* *Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB, Canada.*

Infertility is the main cause of culling of dairy cows in Canada causing dairy industry more than \$667 million in losses during 2010. More than 35% of the cows in a herd are affected by various forms of reproductive tract infections. Infertility is highly related to bacterial infections of the uterus immediately after calving and uterine infections are highly associated with other periparturient diseases. The objective of this study was to evaluate whether intravaginal infusion of a mixture of lactic acid bacteria (LAB) would affect the incidence of clinical diseases in transition dairy cows. 152 pregnant Holstein cows were assigned randomly to

3 groups 2 wk before the expected day of calving, based on their parity and BCS. Cows received intravaginal LAB around parturition as follows: treatment 1 (TRT1) - 2 consecutive LAB and 1 carrier (1 mL of sterile skim milk) dose; treatment 2 (TRT2) - 3 consecutive LAB doses; control (CTR) - 3 consecutive carrier doses. LAB used were a mixture of 3 lactic acid bacteria including *Lactobacillus sakei* FUA3089, *Pediococcus acidilactici* FUA3138, and FUA3140 stored in sterilized skim milk and infused at  $10^8$  to  $10^9$  cfu per dose. Cows were checked twice per week after parturition for retained placenta (RP), vaginal purulent discharges, left displaced abomasum (LDA), and lameness. Results indicated an overall incidence rate of vaginal purulent discharges of 25%. Within those cows, 24% were in TRT1, 26% in TRT2, and 50% in CTR ( $P = 0.05$ ). RP had an overall incidence rate of 7.9%. Within the cows with RP, 17% were in TRT1, 33% in TRT2, and 50% in CTR ( $P = 0.07$ ). LDA exhibited an overall incidence rate of 4.0%. Within the cows having LDA, 17% were in TRT1, 50% in TRT2, and 33% in CTR ( $P = 0.07$ ). The overall incidence rate of lameness was 10.5%. Within those lame cows, 31% were in TRT1, 44% in TRT2, and 25% in CTR ( $P = 0.02$ ). It should be pointed out that a major reason for lameness was mechanical injury. In conclusion, intravaginal administration of LAB lowered the incidence of vaginal purulent discharges and RP in periparturient dairy cows.

**Key Words:** dairy cow, lactic acid bacteria, disease incidence

**M24 Acute phase response intensity is related to the metabolic and immunologic statuses of early postpartum dairy cattle.** C. R. Nightingale,\* M. D. Sellers, A. R. Pepper-Yowell, D. L. Hanson, C. J. Cobb, B. S. Obeidat, and M. A. Ballou, *Department of Animal and Food Sciences, Texas Tech University, Lubbock.*

Objective was to describe the relationship between the intensity of the acute phase response and the metabolic and immunologic statuses of early postpartum, multiparous cows. Peripheral blood was collected from 240 Holstein cows, 2–8 DIM and 2nd–8th parity from 8 dairies across 5 d ( $n = 6$  cows/dairy/day). Plasma concentrations of haptoglobin were measured colorimetrically and cows were classified as low (1st quartile), moderate (2nd and 3rd quartiles), or high (4th quartile) responders. Metabolic measurements included plasma  $\beta$ -hydroxybutyrate (BHBA) concentrations. Immunologic measurements included total leukocyte counts and differentials, neutrophil L-selectin expression, neutrophil oxidative burst capacity when cocultured with *Escherichia coli*, as well as the secretion of tumor necrosis factor- $\alpha$  and interferon- $\gamma$  when diluted whole blood was cocultured with lipopolysaccharide and phytohemagglutinin, respectively. All data are reported as Low, Moderate, and High, respectively. Haptoglobin concentrations ranged from 0.42 to 1.04, 1.05–3.50, and 3.51–10.47 adjusted optical density. High cows had greater BHBA concentrations ( $P < 0.05$ ; 509, 548, and  $645 \pm 57.8$

$\mu M$ ), elevated rectal temperatures ( $P < 0.01$ ; 101.3, 101.4, and  $101.7 \pm 0.14^\circ C$ ), and neutropenia ( $P < 0.01$ ;  $3.5, 3.3,$  and  $2.2 \pm 0.34 \times 10^6$  cells/mL). In addition, the innate immune responses of High cows were stimulated as evident by increased secretion of tumor necrosis factor- $\alpha$  ( $P < 0.05$ ; 570, 562, and  $732 \pm 72.5$  pg/mL), expression of L-selectin on neutrophils ( $P < 0.01$ ; 71, 72, and  $119 \pm 10.0$  mean fluorescence intensity), and neutrophil oxidative burst capacity ( $P < 0.05$ ; 117, 123, and  $146 \pm 11.7$  mean fluorescence intensity). In contrast, the secretion of interferon- $\gamma$  was suppressed in both the Moderate and High cows ( $P < 0.01$ ; 707, 420, and  $309 \pm 102.8$  pg/mL). These data indicate that a stronger acute phase response during the early postpartum period is characterized by increased concentrations of ketone bodies, activated innate immune responses, and a suppressed adaptive immune response.

**Key Words:** immune, metabolic, transition cow

**M25 Isolation and analysis of transient receptor potential channel (TRPC) genes in goats: Implications for study of gastrointestinal nematode infection.** M. M. Corley and J. Ward,\* *Virginia State University, Petersburg.*

Transient receptor potential cation channels (TRPC) are involved in several cellular functions. The TRPC channels function as calcium cation channels that cause membrane depolarization and entry of calcium into the cell, thereby resulting in smooth muscle contraction. Specifically, it has been shown that TRPC4 and TRPC6 deletions in mice, impairs intestinal motility and contractility. Goats are plagued with the gastrointestinal nematode *Haemonchus contortus*, that costs the small ruminant industry billions of dollars per annum in livestock loss and drug treatment. The ability of the intestine to expel worms is dependent on many factors, one of which is intestinal contractility. Although the role of TRPC 4 and 6 has been identified as crucial in intestinal contractility in mice, the TRPC 4 and 6 genes have not been isolated in goats, nor evaluated in *H. contortus* infection. Therefore, the objective of this study was to identify and characterize the TRPC4 and TRPC 6 genes of goats. Total RNA was isolated from homogenized intestinal tissue and purified. Reverse transcriptase PCR (RT-PCR) was performed using cross species primers designed from the human, bovine, mouse and rat TRPC4 and 6 gene alignments. The RT-PCR products were visualized via agarose gel electrophoresis. The expected bp RT-PCR products were observed (388 and 350 bp, respectively), indicating successful amplification of the goat TRPC 4 and 6 cDNA. The cDNA PCR products were sequenced. The goat TRPC showed 98, 92, 91, and 90% sequence homology to the bovine, horse, pig, and human TRPC genes, respectively, and 88 and 87% homology to rat and mouse TRPC. Identification and analysis of goat TRPC 4 and 6 genes will help to elucidate the involvement of TRPC genes in the response to *H. contortus* infection in goats.

**Key Words:** goat, gastrointestinal, TRPC genes