

Graduate Student Competition: ADSA Production Division

Graduate Student Oral Competition, MS

80 Automated detection of estrus using multiple commercial precision dairy farming technologies in synchronized dairy cows.

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Detecting dairy cattle estrus helps determine optimal breeding time. Dairy cattle estrus varies in length and intensity, increasing the need for accurate and continuous estrous detection. The objective of this study was to evaluate precision dairy farming technologies (PDFT) for estrous detection. The estrous detection efficiency of PDFT is predicted to be greater than traditional visual observation of estrus. Estrus was synchronized in 24 lactating Holstein dairy cows using a modified G7G-Ovsynch protocol (last GnRH injection withheld to permit expression of estrus) beginning 45 to 85 DIM. Resumption of ovarian cyclicity at enrollment, presence of a corpus luteum (CL) on the day of the final injection (designated experimental day 0), regression of the CL by d 5, and presence of a new CL on d 11 were verified by transrectal ultrasonography. Cows were observed for estrous behaviors for 30 min, 4X per day, on days 2 to 5. Blood samples were collected on d -2, -1, 0, 1, 2, 5, 7, 9, and 11 to quantify progesterone to verify luteal regression and ovulation. Potential periods of estrus (gold standard) were defined by the temporal pattern of progesterone (>1.0 ng/mL on d -2, -1 and 0, <1.0 ng/mL on d 2, and >1.0 ng/mL on d 9 and 11). Detection of estrus by PDFTs, an estrous behavioral scoring system, and by visual observation of standing estrus were compared to the gold standard (Table 1). Sensitivity and specificity for detection of estrus was similar among all PDFT. Only 56% of cows that ovulated were observed standing by visual estrous detection. All systems tested are capable of detecting estrus at least as effectively as visual observation.

Table 1 (Abstr. 80). Detection of estrus using alerts generated by PDFT¹ and visual observation (n=24)

Detection method	TP	FP	TN	FN	Sensitivity Specificity	
					(%)	(%)
Afimilk AfiAct Pedometer Plus	16	0	6	2	89	100
GEA CowScout (leg version)	15	0	6	3	83	100
ENGs Track A Cow	14	0	6	4	78	100
Agis SensoOr	14	0	6	4	78	100
Estrus behavioral score ²	12	1	5	6	67	83
Standing behavior	10	0	6	8	56	100

¹Sensitivity = TP/(TP + FN) and specificity = TN/(TN + FP); where TP = true positive, TN = true negative, FP = false positive, and FN = false negative.

²Scoring system as defined by Van Eerdenburg et al. (1996).

Key Words: automated estrus detection, precision dairy technology, estrous behavior

81 Effect of nutrition and management practices on de novo fatty acid synthesis in northeastern US dairy herds. Melissa E. Woolpert*^{1,2}, Catarina Melilli³, Kurt W. Cotanch¹, Heather M. Dann¹, Rick J. Grant¹, Larry E. Chase³, and David M. Barbano³, ¹*William*

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This study investigated the effects of management practices and dietary chemical characteristics on de novo fatty acid (FA) concentrations in bulk tank milk from commercial dairy herds. De novo FA are synthesized primarily from rumen fermentation products acetate and butyrate. It was hypothesized that farms with higher de novo FA concentrations will prioritize management and nutrition that promote rumen health. Herds (n = 44) located in Vermont and northern New York State were selected based on high (HDN; 25.8 ± 1.0; mean ± SD) or low (LDN; 23.6 ± 0.9) de novo FA expressed as a proportion of total FA (relative %) in bulk tank milk. Management practices were assessed during one visit to each farm in March or April, 2014. Total mixed ration samples were collected and analyzed by NIR for chemical composition. Data were analyzed using the MIXED procedure of SAS with de novo group as the fixed effect, farm as the random effect, and breed as the covariate in the model. Milk (26.3 vs. 22.7 kg/d, SE = 1.3, P = 0.06), fat (1.1 vs. 0.9 kg/d, SE = 0.1, P < 0.01), true protein (0.9 vs. 0.7 kg/d, SE < 0.1, P < 0.01), and de novo FA (25.6 vs. 23.7 relative %, SE = 0.2, P < 0.01) were higher for HDN vs. LDN farms, respectively. Days in milk (170 ± 7 d; least squares means ± SE), freestall bunk space (41 ± 3 cm), manure score (2.8 ± 0.0), and freestall feeding frequency (1.5 ± 0.4 times/d) were not different between groups. However, tiestall feeding frequency (4.8 vs. 2.8 times/d, SE = 0.7, P < 0.1), freestall stocking density (1.05 vs. 1.20 cows/stall, SE = 0.06, P = 0.07), and body condition score (3.07 vs. 2.95, SE = 0.04, P < 0.01) were different between HDN vs. LDN herds, respectively. Dietary DM (40.7 ± 2.0), CP (16.0 ± 0.6%), ADF (22.9 ± 1.0%), NDF (37.5 ± 1.3%), starch (21.7 ± 1.5%), ash (8.5 ± 0.4%), and forage to concentrate ratio (1.47 ± 0.14) were not different (P > 0.12) between groups. However, dietary ether extract was lower in HDN herds (3.5 vs. 4.5%, SE = 0.2, P < 0.01). Overcrowded freestalls, reduced feeding frequency, and greater dietary ether extract content compromised de novo FA synthesis thereby decreasing milk fat and protein yields.

Key Words: stocking density, milk protein, milk fat

82 Pregnancy outcomes based on milk pregnancy-associated glycoprotein levels. Erin M. Wynands*, Stephen J. LeBlanc, and David F. Kelton, *University of Guelph, Guelph, Ontario, Canada.*

Timely diagnosis of pregnancy and pregnancy losses is economically important. A commercially available pregnancy-associated glycoprotein (PAG) milk assay (Idexx Laboratories Inc.) is offered through routine Dairy Herd Improvement (DHI) testing for diagnosis of pregnancy. The objective of this observational study was to describe the relationship between PAG at various stages of gestation and the likelihood of successful calving. The hypothesis was that higher PAG levels would be associated with successful calving. Data were collected from CanWest DHI for all cows with a milk PAG assay between January 1 and May 31, 2013. The PAG milk assay result is reported as a relative PAG level (S-N value). Cows that tested pregnant (PAG >0.25 according to the manufacturer's cut-point; 6196 cows in 967 herds) were included in this analysis. A dichotomous outcome of calving between 270 and 290 d after the relevant insemination was determined for each cow with 5139 successful outcomes. There was a significant interaction between days in gestation (DIG) at time of PAG assay and PAG, so the data were

stratified by DIG. Milk PAG levels increased after breeding, decreased at 45–75 DIG, then increased through the remainder of gestation. For cows ≤ 45 DIG ($n = 793$) increasing PAG level was associated with an increased likelihood of calving ($P = 0.01$). For cows > 45 and ≤ 75 DIG ($n = 1653$) relative PAG levels ($P = 0.001$) and linear score ($P = 0.01$) were negatively associated with the outcome, while test-day milk yield was positively associated with the outcome ($P = 0.01$). For cows > 75 DIG ($n = 3750$) relative PAG level ($P = 0.001$), and milk yield ($P = 0.005$) were positively associated with full-term pregnancy; linear score was negatively associated with the outcome ($P = 0.05$). These results indicate that while higher PAG levels are positively associated with a positive calving outcome in general, a decrease in PAG level around 45–75 d in gestation was associated with a successful pregnancy outcome.

Key Words: pregnancy-associated glycoprotein, pregnancy diagnosis

83 Prepartum dietary cation-anion difference (DCAD) and 25-hydroxyvitamin D₃ supplementation modulate β -defensin responses in postpartum dairy cattle.

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Postpartum hypocalcemia has been associated with suppressed neutrophil and lymphocyte function in dairy cattle. It was hypothesized that mitigation of postpartum hypocalcemia would enhance activation of host-defense mechanisms during the postpartum period. The objective of this study was to determine the effects of pre-partum DCAD in combination with vitamin D supplementation on expression of a cluster of β -defensin (DEFB) host-defense genes. Pregnant dry Holstein cows ($n = 51$, multiparous) of 255 d gestation were blocked by parity and randomly assigned to 4 prepartum diets of positive (+130 mEq/kg) or negative (-130 mEq/kg) DCAD and either 3 mg vitamin D₃ or 3 mg of 25-hydroxyvitamin D₃ (25D) per 11 kg of DM/d. Treatment diets were fed for the last 21 d of gestation, and, upon calving, cows were fed the same lactation ration for the first 49 DIM. Peripheral blood neutrophils and monocyte cultures were collected at d 0 and d 3 of calving and stimulated with 75 ng/mL of 25D and a combination of 100ng/mL of lipopolysaccharide (LPS) and 75 ng/mL of 25D. The mRNA transcripts of DEFB genes were quantified by real-time PCR, and data were analyzed with a general linear mixed model to test for fixed effects and interactions of experimental variables. Neutrophil gene expression of DEFB4 ($P \leq 0.05$), DEFB5 ($P \leq 0.05$), DEFB7 ($P = 0.1$), and DEF10 ($P = 0.1$) was greater in cows fed the negative DCAD. Neutrophils had lower expression of DEFB3, DEFB4, DEFB7, DEFB10, and lingual antimicrobial peptide in response to LPS at d 3 of calving compared with d 0, ($P \leq 0.05$). Cows supplemented with vitamin D₃ showed greater DEFB3, DEFB4, DEFB5, DEFB7, and DEF10 gene expression in monocytes ($P \leq 0.05$) than 25D supplemented cows. In conclusion, pre-partum DCAD and 25D affect neutrophil and monocyte β -defensin responses and have implications for improving transition cow immunity.

Key Words: defensin, DCAD, vitamin D

84 Effects of elevated subcutaneous adipose stores on fractionated peripheral blood mononuclear cells and polymorphonucleocytes fatty acid profile and polymorphonucleocytes gene expression in periparturient dairy cow.

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During the periparturient period, elevated circulating nonesterified fatty acids (NEFA) affect not only the circulating fatty acid (FA) composition, but also that of the peripheral blood mononuclear cells (PBMC) and polymorphonucleocytes (PMN). The changes to specific lipid fractions, however, remain unknown. We hypothesize elevated lipid mobilization will alter FA profiles and therefore gene expression of pro-inflammatory mediators. Starting -28 d before calving, treatment cows ($n = 18$) received a dry cow ration with an additional 10 kg of corn / head per day, while the control cows ($n = 16$) received the dry cow ration (no additional corn) with an additional 400 mg of monensin / head per day to minimize lipid mobilization. Postpartum, treatment cows were fasted for 8 h on d +3. Plasma samples were collected on d -28, +3, +12, and +21 relative to parturition to isolate PBMC and PMN for analysis of fatty acid methyl ester (FAME) of cells' NEFA and phospholipids (PL) fractions. Isolated PMN were analyzed using real-time q-PCR for caspase-1 (CASP), interleukin-8 receptor (IL-8R), and L-selectin (SELL). Data were analyzed as repeated measures ANOVA using mixed model procedures in SAS (9.3) with significance declared at $P \leq 0.05$. Several FA concentrations of NEFA and PL fractions in PBMC and PMN changed over time. Eicosanoid precursor C20:4 in the NEFA fraction of PBMC were greater in treatment than control cows (0.51 vs. 1.29 ± 0.27 g/100g FAME; $P = 0.02$). In contrast, C20:4 of PBMC in PL fraction were greater in control animals (10.06 vs. 8.18 ± 0.56 g/100g FAME; $P = 0.02$). Anti-inflammatory C20:5 concentrations in the PL fraction of PMN were greater in the treatment group and multiparous animals in a treatment across parity effect ($P = 0.05$). Although gene expression of CASP, IL-8R and SELL in PMN changed over time ($P < 0.0001$), no treatment effect was detected. In summary, dietary manipulation altered FA profiles within PBMC and PMN fractions, but did not alter gene expression in PMN.

Key Words: lipid mobilization, immune cell, fatty acid

85 Cows at a high risk for subacute rumen acidosis exhibit different feeding behavior.

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Previous studies reported that cows fed the same diet have a large variation in rumen pH; cows at a high-risk of sub-acute rumen acidosis (SARA) have a lower pH for a longer period of time than low-risk cows. The objective of this study was to determine if feeding behavior is different between cows with a high or low risk for SARA. We hypothesized that high-risk cows would eat longer during each meal, contributing to a lower pH. Sixteen ruminally cannulated lactating cows (DIM = 265 ± 36 ; BW = 632 ± 80 kg) were fed a diet consisting of 35% forage and 65% concentrate once daily to increase the risk of SARA. After a 17-d diet adaptation period, ruminal pH was measured every 30 s over a 24-h period. Cows were classified as high or low risk to SARA based on an acidosis index (area of pH < 5.8 /DMI). Feeding behavior of animals was observed for the same 24-h period; each animal was checked every 5 min and each behavior seen was assumed to last for 5 min. The 24-h observation period was split into 3 8-h time periods following feeding, and eating behavior was summarized for each period. All response variables were evaluated for the group effect using the PROC TTEST procedure of SAS. Minimum pH ranged from 5.13 to 6.10 among the 16 cows fed the same diet. The 7 high-risk cows had a higher acidosis index than the 9 low-risk cows (8.03 vs. 0.11, $P = 0.002$). Despite no significant difference in DMI ($P = 0.87$), high-risk cows spent more time eating in the first 8-h period after feeding (186 vs. 153 min, $P = 0.014$) and less in the third 8-h period (19 vs. 43 min, $P = 0.012$) than low-risk cows. In the third period, high-risk cows spent more time ruminating (243 vs. 197 min, $P = 0.012$). Average meal duration was longer for

high-risk cows (92 vs. 59 min, $P = 0.011$). Our results suggest that feeding behavior may be a contributing factor to SARA, where cows with a high-risk for low rumen pH eat for longer periods of time during the first 8-h after feeding. Increasing feeding frequency to encourage distribution of eating throughout the day may be an effective approach to mitigate SARA, which warrants further investigation.

Key Words: subacute rumen acidosis, feeding behavior

86 Factors associated with aerobic plate count, coliform count, and log reduction of bacteria in automated calf feeders. Alyssa M. Dietrich^{*1}, Whitney A. Knauer², Sandra A. Godden², Christina S. Petersson-Wolfe¹, and Robert E. James¹, ¹Virginia Tech, Blacksburg, VA, ²University of Minnesota, St. Paul, MN.

As automated calf feeders become more widely adopted, there is a need for research on sanitation management. The objective was to identify factors associated with bacteria counts and log reduction (LR) as a result of circuit cleaning (CC) in milk/replacer samples collected from automated calf feeders (Förster Technik, Engen, Germany). It was hypothesized that increased cleaning is associated with low bacteria counts and increased LR. Six VA and 4 MN dairy farms (14 feeders) were visited weekly for 7 mo. Samples of milk/replacer were collected aseptically from each feeder, iced, and frozen at -5°C . For a 4-wk period, VA farms were instructed to collect and freeze samples immediately before and after CC. Samples were thawed and plated on Petrifilms to find \log_{10} aerobic plate count (APC) and \log_{10} coliform count (CCP). From a model of state, farm(state), LSM \pm SE APC was 3.84 ± 0.26 and $5.02 \pm 0.21 \log_{10}$ cfu/mL ($P < 0.01$), and CCP was 0.62 ± 0.47 and $2.89 \pm 0.39 \log_{10}$ cfu/mL ($P < 0.01$) for MN and VA. The LSM by farm ranged from 3.30 to $5.51 \log_{10}$ cfu/mL APC and 0.07 to $3.82 \log_{10}$ cfu/mL CCP. The LR of APC (LRAPC) and CCP (LRCCP) after CC was 0.78 ± 0.82 and $0.67 \pm 1.17 \log_{10}$ cfu/mL (mean \pm SD). By regression, each \log_{10} increase in pre-CC APC and CCP increased LRAPC and LRCCP by $0.45 \log_{10}$ cfu/mL ($P < 0.01$). Farms with higher pre-CC APC and CCP cfu/mL performed more CC/wk and used more mL detergent/L water ($P < 0.05$). More mixer cleanings (MCHE)/d lowered pre-CC APC cfu/mL ($P < 0.05$), whereas use of silicon hoses increased pre-CC CCP cfu/mL ($P = 0.04$), and use of bleach decreased pre-CC CCP cfu/mL ($P < 0.01$). By stepwise regression with farm in the model, more days since last MCHE increased LRCCP ($P < 0.01$) within farm. Number of calves, milk L/d, and feeder visit frequency were not associated with LR of bacteria within farm. LR was more dramatic at high pre-CC bacteria, which was reduced by more MCHE/d and use of bleach.

Key Words: bacterial count, automated calf feeders

87 Effects of supplementing slow-release urea in combination with steam-flaked corn or high-moisture corn on ruminal fermentation and lactational performance of dairy cows. B. M. Tye^{*1}, K. Neal¹, S. Y. Yang¹, J.-S. Eun¹, A. J. Young¹, and K. Mjoun², ¹Department of Animal, Dairy, and Veterinary Sciences, Utah State University, Logan, UT, ²Alltech, Brookings, SD.

Associative effects between slow-release urea (SRU) and different types of corn grains [i.e., steam-flaked corn (SFC) vs. high-moisture corn (HMC)] need to be investigated to improve N utilization and lactational performance of dairy cows. This experiment was performed to test a hypothesis that supplementing SRU (Optigen, Alltech, Nicholasville, KY) in HMC-based diet would improve microbial fermentation and nutrient utilization by lactating dairy cows due to a better synchronous supply of dietary N and ruminally fermentable carbohydrates. Eight

multiparous Holstein cows (32 ± 8.2 DIM) were used in a duplicated 4×4 Latin square design, and within each square, cows were randomly assigned to a sequence of 4 diets during each of the 4 21-d periods (14 d of treatment adaptation and 7 d of data collection). The 4 dietary treatments included: SFC without SRU, SFC with SRU, HMC without SRU, and HMC with SRU. Data were analyzed with a model that included the fixed effect of type of corn grain, SRU supplementation, and their interaction with cow and period as the random effects. Supplementing SRU with SFC but not with HMC increased intake of DM and CP, resulting in tendencies for an interaction between corn grain (CG) and SRU supplementation (CG \times SRU; $P = 0.06$). Milk yield did not differ among treatments (39.0 ± 0.37 kg/d). Milk true protein concentration increased with SFC with SRU, whereas it decreased with HMC with SRU ($P = 0.01$). Dietary treatments did not affect ruminal concentration of volatile fatty acids and ammonia-N. Feeding HMC tended to increase feed efficiency for milk production ($P = 0.09$) and N utilization efficiency for milk N ($P = 0.10$), but supplementing SRU did not influence the efficiency parameters. Supplementing SRU with either SFC or HMC resulted in minor effects on ruminal fermentation and lactational performance possibly due to relatively less dietary concentrations of the corn grains (13.6% DM on average) in high-forage diets (60% of the diet DM). However, feeding HMC improved feed and N utilization efficiency for milk production.

Key Words: steam-flaked corn, high-moisture corn, slow-release urea

88 Regulation of microRNA-221 gene expression in ovarian theca cells of cattle: A possible role in follicular development.

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Cystic ovarian follicles result in reproductively inefficient dairy cows. Fibroblast growth factor-9 (FGF9) gene expression in granulosa cells (GC) is downregulated in cystic vs. non-cystic follicles in cattle. FGF9 is thought to act as an intraovarian regulator by stimulating proliferation and reducing steroid production by GC and theca cells (TC). Analysis using Affymetrix GeneChip bovine Genome Arrays ($n = 8$) revealed that microRNA-221 (*MIR221*) in TC was upregulated by FGF9. We selected this gene for further study due to recent evidence indicating *MIR221* is involved in cell proliferation, angiogenesis and cancer. We hypothesized that *MIR221* is regulated during follicle growth and by steroids and hormones that induce cAMP. Three experiments were conducted to evaluate hormonal regulation of *MIR221* mRNA in TC and its possible role in follicular development using sensitive real-time PCR (*U6* as housekeeping gene). TC were isolated from large (>8 mm) follicles ($n = 6$ to 8 per pool) collected from an abattoir, cultured for 48 h in medium with 10% fetal calf serum, and then treated in serum-free medium for an additional 24 h. Data were analyzed via GLM procedure of SAS. Exp. 1 tested effects of cAMP pharmacologic agents (dbcAMP at 0.1 mg/mL; forskolin at 4.1 $\mu\text{g/mL}$) on FGF9-induced *MIR221* mRNA abundance; neither affected ($P > 0.10$) *MIR221* expression whereas FGF9 increased ($P < 0.05$) *MIR221* abundance 1.94-fold. Exp. 2 tested the effect of 300 ng/mL of steroids (estradiol; androstenedione) and phytoestrogens (genestein; α - and β -zearanol) on *MIR221*; none of the treatments influenced ($P > 0.10$) *MIR221* mRNA levels (3.45 ± 0.62 relative abundance). In Exp. 3, follicular fluid (FF) was collected from ovaries obtained at an abattoir, and *MIR221* mRNA abundance measured; *MIR221* mRNA was 3-fold greater ($P < 0.05$) in FF aspirated from small (1 to 5 mm) follicles compared with FF from large follicles. These novel results indicate *MIR221* is increased by FGF9 but not regulated by steroids or cAMP. Greater levels of *MIR221* in FF

of small vs. large follicles suggest a role for FGF9-induced *MIR221* in follicular development.

Key Words: theca cells, cattle, microRNA-221

89 Feeding a rumen-degradable amino acid can enhance milk production. Kayla M. Hultquist* and David P. Casper, *South Dakota State University, Brookings, SD.*

The study objective was to determine if feeding a rumen degradable AA can increase milk production comparable to growth hormone (GH). Eight multiparous late-lactating (255 ± 26.4 d in milk) Holstein dairy cows were blocked by milk yield (34.1 ± 8.25 kg/d) and randomly assigned to 1 of 4 treatments in a replicated 4×4 Latin square design having 21 d periods with 7 d for dietary adaptation and 14 d for data collection. Treatments were control (C), Posilac (rBST), and valine fed at 40 (V40) and 80 g/d (V80). Cows were fed a total mixed ration (TMR) with a distillers grains carrier at 113.4 g/d containing none or added AA. Rumen and blood samples were collected during wk 2 and 3 for volatile fatty acids and endocrine assays. Dry matter intake was similar ($P > 0.10$) (21.3, 22.0, 22.8, and 21.5 kg/d for C, rBST, V40, and V80, respectively) for cows receiving all treatments, but milk yield (22.0, 26.1, 25.2, and 24.9 kg/d), FCM (22.1, 25.4, 24.4, and 24.3 kg/d), and ECM (22.7, 26.1, 25.1, and 24.9 kg/d) were significantly ($P < 0.05$) increased for cows receiving rBST, V40, and V80 compared with C cows. Milk yield improvements were similar ($P > 0.10$) between rBST, V40, and V80. Milk fat percentages (3.51, 3.36, 3.32, and 3.38%) were highest ($P < 0.05$) for cows receiving C compared with cows receiving V40, while cows receiving other treatments were intermediate and similar ($P > 0.10$). Milk protein percentages (3.20, 3.12, 3.15, and 3.13%) were greatest ($P < 0.05$) for cows receiving C compared with cows receiving rBST. Ruminal isobutyric acid (1.01, 1.05, 1.28, and 1.49 molar %) concentrations were increased ($P < 0.05$) for cows receiving V40 and V80 compared with cows receiving C and rBST treatments. Plasma IGF-1 concentrations (60.4, 106.1, 65.9, and 58.3 ng/mL) were greater ($P < 0.05$) for cows receiving rBST compared to cows receiving other treatments. Plasma GH concentrations (1.78, 1.99, 1.55, and 1.45 ng/mL) were greater ($P < 0.05$) for cows receiving rBST compared with V40 and V80 cows, while C cows were intermediate and similar ($P > 0.10$). This study suggests that feeding a rumen degradable AA can increase milk yield comparable to rBST.

Key Words: milk production, dairy cow, growth hormone

90 Investigation of a new anti-slip flooring technology to address slips and falls by dairy cows. Nancy Franco-Gendron*¹, Renée Bergeron¹, Walt Curilla², Sabine Conte³, Trevor J. DeVries⁴, and Elsa Vasseur¹, ¹*Organic Dairy Research Center, University of Guelph, Alfred Campus, Alfred, Ontario, Canada*, ²*AB Silikal Hygienic Floors, Diamond Hard Surfaces Inc., Calgary, Alberta, Canada*, ³*Centre de Recherche et de Développement sur le Bovin Laitier et le Porc, Agriculture and Agri-Food Canada, Lennoxville, Quebec, Canada*, ⁴*Department of Animal and Poultry Science, University of Guelph, Kemptville Campus, Kemptville, Ontario, Canada.*

Freestall dairy facilities are known to have issues with slips and falls by cows caused by the combination of smooth flooring and manure slurry. Eighteen Holstein dairy cows (parity 1 to 6) selected to maximize gait range were evaluated on various flooring types to test the hypothesis that flooring with greater anti-slip properties will improve cattle's ease of movement. Cows were trained and randomly assigned to walk on 6 flooring types covered with thin coat of manure 3x/wk for 6 wks; 2

controls: rubber mat (Animat), traction milled concrete (Agri-Trac), and 4 different coated treatments: Red (Silikal 0.7–1.2 mm quartz with 1 seal coat), Green (Silikal 0.7–1.2 mm, 2 seal coats), Yellow (Silikal 0.4–0.8 mm, 1 seal coat) and Orange (Silikal 0.4–0.8 mm, 2 seal coats). Cows were filmed walking a straight corridor and a corridor with a turn (5 passages/corridor). Cows had 2 accelerometers on their rear legs to measure the average acceleration of a passage. Reflective markers were placed on both right hooves before walking the straight corridor measuring stride length, foot height, swing and stance time via kinematics. Steps taken, passage time and time/steps were recorded for the curved corridor. Stride for cows on the straight corridor was longer for rubber (1.54 ± 0.026 m; mean \pm SE) compared with all other treatments: 1.44 ± 0.041 m for Red (Wilcoxon sign rank test: $S = 54$; $P = 0.003$), 1.38 ± 0.027 m for Green ($S = 68$; $P < 0.0001$), 1.48 ± 0.031 m for Yellow ($S = 43$; $P = 0.02$), and 1.46 ± 0.033 m for Orange ($S = 47$; $P = 0.01$). Stride of cows walking on the straight corridor was shorter on groove concrete (1.41 ± 0.034 m) compared with Yellow ($S = -52$; $P = 0.02$), but no differences were found with the other treatments. The time taken per step for cows on the curve corridor was not different between flooring (Kruskal-Wallis test: $X^2 = 0.9$; $P > 0.1$). In conclusion, longer strides or faster pace reflects cow ease of movement on a particular flooring type. Based on these 2 variables, cow ease of movement was greater on rubber versus coating treatments, but was better on the Yellow treatment compared to concrete. Other aspects of ease of movement like acceleration or gait need to corroborate these first findings.

Key Words: dairy cows, welfare, flooring

91 2,4-Thiazolidinedione improves liver function but does not affect insulin sensitivity and expression of genes in adipose and mammary tissue of lactating dairy goats. Fernanda T. da Rosa*¹, Johan Osorio¹, Francisco Y. Rivera¹, Erminio Trevisi², Charles T. Estill¹, and Massimo Bionaz¹, ¹*Oregon State University, Corvallis, OR*, ²*Universita Cattolica del Sacro Cuore, Piacenza, PC, Italy.*

Previous studies have shown that daily injection of 2,4-thiazolidinedione (TZD) in lactating dairy goats decreases milk somatic cell count but does not affect milk fat yield. The latter could be due to a lipogenic competition between adipose and mammary tissues, owing to an activation of peroxisome proliferator-activated receptor gamma (PPAR γ), a potential TZD target, in the adipose tissue. This study investigated the hypothesis that activation of PPAR γ improves the response to mastitis and increase milk fat synthesis. To test this, 24 Saanen goats received either TZD or saline and intramammary infusion of either bacteria or saline (6 goats/group). A metabolic and inflammation profiling in blood (including insulin), adipocyte size by histology, and expression by RT-qPCR of PPAR γ target genes in adipose tissue obtained through biopsy and in mammary epithelial cells (MEC) isolated from milk were assessed. In MEC, expression of *CCL2* and *IL8* was also measured. Data were analyzed by GLIMMIX of SAS with Mastitis, TZD, and Time and all interactions as main effects. Significance was determined by Tukey-adjusted $P < 0.05$. Mastitis induction decreased myeloperoxidase but TZD treatment increased haptoglobin, cholesterol, paraoxonase, γ GT, and myeloperoxidase in blood compared with saline, indicating a positive effect on liver and increased neutrophil host defense capability. Insulin sensitivity (estimated using QUICKI) was not affected by TZD but overall was lower in mastitic goats. Adipocyte size increased over time but was not affected by treatments. Sub-clinical mastitis increased expression of *CCL2* and decreased expression of *FASN* in MEC but increased expression of *FASN* and *SREBF1* in adipose. No overall effects of TZD on expression of PPAR γ target genes were observed in MEC and adipose tissue. In summary, the data revealed limited effects

of TZD on the response of goats to mastitis. Furthermore, the data does not support TZD being a strong PPAR γ agonist and insulin sensitizer in dairy goats but indicates that it might improve directly or indirectly the liver function and inflammatory response.

Key Words: goats, mastitis, 2-4-thiazolidinedione

92 Inferring the causal effect of number of lambs born on milk yield in dairy sheep using propensity score methods. Vera C. Ferreira*, Bruno D. Valente, David L. Thomas, and Guilherme J. M. Rosa, *University of Wisconsin, Madison, WI.*

Assigning causal interpretation to associations obtained from observational data is challenging as they are prone to confounding. Number of lambs born (prolificacy) in dairy sheep may be considered a potential factor contributing to milk yield (MY). However, inferring this effect using traditional regression or ANOVA techniques can generate spurious results if confounder variables affect both the outcome (MY) and treatment (prolificacy). Propensity score (PS) methods tackle this issue by balancing baseline covariate distributions between treatment levels, allowing unbiased inference of marginal effects. This method belongs to the framework of causal models dealing with potential outcomes. It intends to mimic aspects of randomized trials, in which the difference

among treatment groups is causally meaningful. Under the assumption that ewe prolificacy affects MY, our objective was to estimate the magnitude of such a causal effect using PS based on Matched Samples. Data comprised 4,319 records from 1,534 crossbred dairy ewes. The set of potential confounders was composed by lactation number (1th, 2nd and 3th – 6th) and dairy breed composition (<.5, 0.5-.75 and > 0.75 of East Friesian or Lacaune). For the treatment variable, single lamb birth was assigned to Group 0, while multiple birth (2, 3 or 4 lambs) was assigned to Group 1. MY represented the volume of milk produced for the whole lactation (mean = 268.5 L and SD = 116.4 L). The R package “nonrandom” was used. A total of 1,166 pairs of treated/nontreated individuals with similar PS values were formed. The criterion for similarity was defined by a caliper size equal to 20% of the sd in the PS logit (0.13) and a ratio of treated/untreated = 1. All covariates were deemed balanced after matching (cutoff for standardized bias = 0.2). The estimated causal effect of prolificacy on MY was 20.52 L, se = 3.77 L, 95% CI = 13.13–27.91 L. This means that ewes that gave birth to a single lamb would be expected to have MY increased by 20.52 L if they had given birth to multiple lambs and all other variables were held constant. This implies that any management practice that increases (decreases) prolificacy would affect MY positively (negatively).

Key Words: causal inference, milk yield, prolificacy