

# Graduate Student Competition: ADSA Production Division Graduate Student Paper Competition, MS Division

**250 Neutrophil function in the transition period and reproductive tract health in dairy cows.** J. A. M. Wittrock,\* T. F. Duffield, D. Bienzle, and S. J. LeBlanc, *University of Guelph, Guelph, ON, Canada.*

The study objective was to evaluate the relationship between neutrophil function during the transition period and reproductive tract disease in dairy cows. Phagocytic and killing ability of polymorphonuclear neutrophilic leukocytes (PMNL) are commonly reduced around calving. Cows ( $n = 81$ ) were followed for 8 wk through the transition period. At -2, -1, 1, 2, 3, and 5 wk relative to calving, PMNL were isolated from the blood and phagocytosis and oxidative burst function were evaluated by flow cytometry, and serum glucose, NEFA, BHB, haptoglobin, insulin, and IGF-1 were measured. Endometritis and cervicitis were diagnosed at 5 wk postpartum by cytobrush cytology, with a positive diagnosis based on  $>5\%$  PMNL. There were 27 cows (34%) with endometritis, and 22 (27%) with cervicitis. There was no difference in PMNL phagocytosis between cows with or without endometritis ( $P = 0.2$ ). Cows with endometritis at wk 5 had 21% lower PMNL oxidative burst function at 2 wk after calving than unaffected cows ( $P = 0.01$ ). Oxidative burst function of PMNL in cows with cervicitis was not different from healthy animals ( $P = 0.2$ ). Cows with cervicitis had 28% lower PMNL phagocytosis than healthy cows 2 wk before calving ( $P = 0.03$ ). Compromised phagocytosis and killing ability of PMNL may precede and contribute to cervicitis and endometritis after calving.

**Key Words:** transition cow, neutrophil, reproductive health

**251 Effects of intrauterine infusion with *E. coli* lipopolysaccharide on systemic and local inflammatory and immune response.** J. G. N. Moraes\*<sup>1</sup>, P. R. B. Silva<sup>1</sup>, L. G. D. Mendonça<sup>1</sup>, J. Silva<sup>1</sup>, M. A. Ballou<sup>2</sup>, K. N. Galvão<sup>3</sup>, and R. C. Chebel<sup>1</sup>, <sup>1</sup>*Department of Veterinary Population Medicine, University of Minnesota, St. Paul,* <sup>2</sup>*Department of Animal and Food Sciences, Texas Tech University, Lubbock,* <sup>3</sup>*Department of Large Animal Clinical Sciences, University of Florida, Gainesville.*

Objectives were to evaluate the effects intrauterine infusion with *E. coli* lipopolysaccharide (LPS) on periperal neutrophil function, hematology, intrauterine cell population, and expression of endometrial interleukin-1 (IL-1). Cows ( $31 \pm 3$  d postpartum) diagnosed with endometritis by Metrichick were assigned to receive 0 (NC,  $n = 31$ ), 150 (150LPS,  $n = 33$ ), or 300 (300LPS,  $n = 34$ )  $\mu\text{g}$  of *E. coli* LPS. A sub-sample of cows ( $n = 25/\text{treatment}$ ) had blood sampled on the day of infusion (d0) before treatment and at 2 and 6 h after treatment for determination of neutrophil phagocytosis (PHAGO) and oxidative burst (OXID) and expression of CD18 and L-selectin. All cows had blood sampled at 0, 2, 6, 24, 48 h after infusion for hemogram and were examined for endometritis using Metrichick on d 1, 2, 7, and 28 and for sub-clinical endometritis using cytobrush on d 0 (before infusion), 1, 2, and 7. Data was analyzed by ANOVA for repeated measures or by chi-squared using MIXED and FREQ procedures, respectively. Percentage of neutrophils positive for PHAGO was ( $P = 0.02$ ) greater for 300LPS ( $66.8 \pm 1.5\%$ ) compared with NC ( $61.7 \pm 1.5\%$ ) and 150LPS ( $61.9 \pm 1.5\%$ ), but the intensity of PHAGO was not different ( $P = 0.36$ ). Treatment did not affect percentages of neutrophils positive for OXID ( $P = 0.49$ ) and for expression of CD18 ( $P = 0.76$ ) and L-selectin ( $P = 0.80$ ). Intensity of OXID tended ( $P = 0.08$ ) be affected by treatment. Treatment had no effect on

hematological parameters. Percentage of cows with endometritis 7 (59%;  $P = 0.47$ ) and 28 (16.7%;  $P = 0.92$ ) d after treatment was not affected by treatment. No differences in percentage of polymorphonuclear cells in uterine cytology were observed on d0 ( $P = 0.35$ ) or thereafter ( $P = 0.55$ ). Percentage of cows with  $>10\%$  of PMNC in uterine cytology was not different among treatments on d 0 (48.5%;  $P = 0.27$ ) and d 1 (85.3%;  $P = 0.27$ ), but on d 7 150LPS (51.7%) and 300LPS (50%) cows tended ( $P = 0.08$ ) to be less likely to have  $>10\%$  of PMNC in uterine cytology than NC (75.9%) cows. Intrauterine infusion with 300 $\mu\text{g}$  of LPS resulted in slight increase in percentage of neutrophils positive for PHAGO.

**Key Words:** lipopolysaccharide, uterus

**252 Effects of fresh-cow diseases on reproduction in a large commercial dairy herd.** R. P. Tollefsrud\*<sup>1</sup>, R. L. Larson<sup>1</sup>, H. M. Scott<sup>1</sup>, G. A. Hanzlicek<sup>1</sup>, and D. E. Little<sup>2</sup>, <sup>1</sup>*Kansas State University, Manhattan,* <sup>2</sup>*DairyNet Incorporated, Brookings, SD.*

The 2007 NAHMS (National Animal Health Monitoring System) survey indicated that early lactation health issues are major factors influencing reproduction and culling on US dairy herds. The objective of this study was to evaluate fresh-cow health during the first 30 d in milk, and its association with days to pregnancy in the concurrent lactation. Data were collected on cattle that calved over a 2-mo period (July and August 2009) on a dairy farm located in the Upper Midwest region of the US Health and production data were collected daily for each cow from the beginning of lactation until the majority of the study population was confirmed pregnant. Both a competing risk analysis and a semi-parametric Cox proportional hazard regression model were completed, and the association between specific health-related events and days to pregnancy was investigated and compared between the models. These analyses showed metritis and dystocia in the first 30 d of lactation were associated with greater days to pregnancy. No difference was noted between parities, except lactation-5 and greater cows were significantly associated with greater days to pregnancy. These 2 analyses showed conflicting significance of association between retained placenta, ketosis, twinning, lameness, and other non-specific illnesses with days to pregnancy. This study found that a competing risk analysis and a semi-parametric regression model were appropriate methods to analyze time sensitive data such as reproductive efficiency. This study supports the evidence that parity, metritis, retained placenta, ketosis, dystocia, twinning, lameness, and other non-specific illnesses can affect reproductive inefficiencies.

**Key Words:** reproduction, animal health, dairy

**253 Using temperature-sensing reticular boluses to aid in the detection of disease in dairy cows.** A. E. Adams,\* F. J. Olea-Popelka, and I. N. Roman-Muniz, *Colorado State University, Fort Collins.*

The objective of this study was to examine the feasibility of using a temperature-sensing reticular bolus (TSRB) to aid in the early detection of common dairy cow diseases. Approximately 2175 primiparous and multiparous Holstein dairy cows were utilized in this study. Each cow received an orally administered TSRB after parturition. Reticular temperature (RT) measurements were obtained for approximately 350 d, as cows equipped with a TSRB passed the reading panels located at

the exit lanes of the milking parlor. Over the 350 d study period, health data were collected for all cows diagnosed by dairy personnel with the following production disorders: mastitis, metritis, lameness, or pneumonia. A retrospective analysis of all health and RT temperature data was conducted. For analysis of RT data, a cow was determined to have a fever if RT exceeded baseline temperature by 0.8°C during the 4 d before diagnosis, determined by a logarithm in the TempTrack software (DVM Systems LLC, Greeley, CO). The health and RT data were combined and analyzed to examine the relationship between fever and disease. A logistical regression model was used to determine the likelihood of a cow with a fever (as defined by TSRB data) being diagnosed with one of the listed diseases by dairy personnel. A total of 218 health events were included in this data set. Odds ratio analysis indicate that a cow presenting a fever is 6.73 times as likely to be diagnosed with mastitis ( $P < 0.0001$ ) and 7.50 times as likely to be diagnosed with pneumonia ( $P = 0.0183$ ), as one that did not present a RT change. No difference in RT was found between cows diagnosed as being lame or with metritis when compared with the “healthy” controls. These data suggest that RT monitoring coupled with routine observation and the prompt assessment of cows has the potential to aid in the early detection of mastitis and pneumonia in dairy cows.

**Key Words:** reticular temperature, dairy cow, disease

**254 Optimization of methods for the detection of *Mycobacterium avium* ssp. *paratuberculosis* in milk and colostrum of naturally infected dairy cows with Johne’s disease.** L. Bradner<sup>\*1</sup>, S. Robbe-Austerman<sup>2</sup>, D. Beitz<sup>1</sup>, and J. Stabel<sup>3</sup>, <sup>1</sup>*Iowa State University, Ames*, <sup>2</sup>*USDA-APHIS, National Veterinary Services Laboratory, Ames, IA*, <sup>3</sup>*USDA-ARS, National Animal Disease Center, Ames, IA*.

*Mycobacterium avium* ssp. *paratuberculosis* (MAP) is the causative agent of Johne’s Disease (JD), a chronic enteritis that occurs in dairy cattle and other ruminants. A 2007 NAHMS Dairy Study demonstrated that over 68% of dairy herds are infected with JD so the risk of exposure within a herd is high. MAP is primarily shed in the feces but it is also shed into the milk and colostrum of infected cows. Because of this, there exists concern about transfer of the organism from dam to calf and the prevalence of MAP in the milk supply. The amount of MAP shed into milk is not well defined because the complexity of the milk matrix and presence of confounding microorganisms makes it difficult to recover MAP via culture methods. The objective of this study is to optimize the decontamination of whole raw milk for the isolation of viable MAP and compare recovery rates in liquid culture mediums. The efficacy of 2 liquid culture mediums, TREK-ESP and BD Bactec 12B were compared for recovery speed and thresholds, incidence of contamination, and reproducibility of results. Milk collected from a non-infected cow was spiked with known concentrations of MAP ( $10^2$  to  $10^8$  cfu/mL). Two chemical decontaminates were investigated, hexadecylpyridinium chloride (HPC) and N-acetyl-L-cysteine-sodium hydroxide (NALC-NaOH). It was found that NALC-NaOH was the superior chemical for decontamination ( $P < 0.01$ ) and that MAP recovery from milk was highest and contamination was lowest when decontaminated with 1.5% NaOH for a 15-min exposure time. In comparing the 2 liquid culture mediums, Bactec 12B was superior in recovery especially at low spiking levels ( $P < 0.01$ ) with a detection threshold of less than  $10^2$  cfu/ml and superior in speed of recovery of viable MAP. TREK-ESP culture demonstrated an increased incidence in false positive and false-negative results that were not observed in Bactec 12B medium. Optimized methods will be used to assess the frequency and level of MAP shed into milk during a complete lactation period of naturally infected dams.

**Key Words:** Johne’s disease, milk, cattle

**255 Effects of transient silage DM concentration variation on dairy cows.** L. R. McBeth<sup>\*1</sup>, W. P. Weiss<sup>1</sup>, N. R. St-Pierre<sup>1</sup>, and D. E. Shoemaker<sup>2</sup>, <sup>1</sup>*The Ohio State University, Wooster*; <sup>2</sup>*Ohio State University Extension, Wooster*.

Transient changes in silage DM concentration, if unaccounted for, will cause a diet to become unbalanced. We hypothesized that a transient decrease in silage DM% would have a negative effect on DMI and milk yield and composition. Holstein cows (24) at 116 DIM were used in 8 replicated  $3 \times 3$  Latin squares with 21-d periods. The treatments were 1) Control (CON), 2) Unbalanced (UNB), and 3) Balanced (BAL). The CON diet was formulated to have a consistent silage DM% and forage:concentrate (F:C) of 55:45 throughout the period. Forage consisted of 67% alfalfa silage (53%DM) and 33% corn silage (36%DM). The UNB and BAL diets were the same as the CON for most of the period except during 2 3-d bouts when water was added to the silage (simulating a rain event) to cause a 10%-unit decrease in silage DM%. During the bouts, the UNB diet was the same as CON on an as-fed basis, but the F:C changed to 49:51 (DM basis), changing diet DM (67.2 vs 64.0%), NDF (32.0 vs 30.7%), and starch (29.8 vs 31.5%). The BAL treatment corrected for the change in silage DM concentration by an increase in silage inclusion to rebalance the F:C to 55:45. Daily DMI and milk yield were measured and milk composition was measured 3 times during the bouts and 3 times during the normal time. A mixed model including parity, cows within parity, period, bout, and treatment with repeated measures (i.e., bout (wet and dry) within period) was used. Over the 21-d period, treatment did not affect DMI (24.0, 24.1 and 23.8 kg/d for CON, UNB and BAL), milk yield (39.4, 39.9 and 39.6 kg/d), milk fat (3.41, 3.42 and 3.33%), and milk protein (2.78, 2.80 and 2.79%). No differences were observed between wet and dry bouts within each treatment for DMI and milk yield; but for UNB, milk fat (3.48 vs 3.35%) increased and milk protein (2.78% vs 2.82%) decreased during wet bouts. A significant decrease in milk yield between wet bouts occurred within the UNB treatment (40.3 vs 39.4 kg/d). Overall, a large decrease in silage DM% (10%-unit) over short-term bouts (with or without TMR adjustment) had only minor effects on DMI, milk yield and composition. Therefore, with drier silages adjusting for short-term, transient changes in silage DM was not necessary.

**Key Words:** silage variability, precision feeding, DM change

**256 Development of a merit-based genetic selection index for dairy grazing systems.** K. D. Gay,<sup>\*</sup> N. J. Olynk, T. D. Nennich, A. P. Schinckel, and M. M. Schutz, *Purdue University, West Lafayette, IN*.

Pasture-based dairy producers face different costs, revenues and management challenges from those with conventional production systems. These differences have led many graziers to question whether the US Lifetime Net Merit (NM\$) index is appropriate to achieve maximum economic gain when selecting dairy cows and bulls for their breeding programs. A Grazing Merit index (GM\$) parallel to the US NM\$ index was constructed using costs, revenues, and management characteristics representative of grazing systems. Inputs were obtained from surveys, literature, and farm financial record summaries. Derived weights for GM\$ were then multiplied by Predicted Transmitting Abilities of 584 active Artificial Insemination Holstein bulls to compare with NM\$. Spearman rank correlation between GM\$ and NM\$ was 0.93 ( $P < 0.0001$ ). Traits included in GM\$ (and their percentage of weight) include: milk volume (24%), fat yield (16%), protein yield (4%), productive life (7%), somatic cell count (–8%), Feet and Leg Composite (4%), Body Size Composite (–3%), Udder Composite (7%), daughter pregnancy rate (18%), calving ability (3%), and dairy form (6%). Weights in NM\$ were 0, 19, 16, 22, 10, 4, 6, 7, 11, 5, and 0% for the same traits, respectively. Dairy form

was added to GM\$ to offset the decrease in strength that arises from selection to reduce stature by selecting against Body Size Composite. Relative importance of milk volume was greater due in part to a difference in the milk prices used and the increased marginal value of milk yield when production is less in grazing systems. There is a large decrease in the emphasis placed on productive life probably because grazing cattle tend to remain in the herd longer; therefore, the marginal value of increasing length of productive life is decreased. This trend is reflected in the relative weight for productive life in GM\$ (18%) vs NM\$ (22%). It appears that NM\$ may provide guidance for pasture-based dairy producers, however a GM\$ index based upon appropriate costs and revenues, is more beneficial.

**Key Words:** grazing, Holstein, selection index

**257 Effects of stocking rate, supplement strategy and breed in a pasture-based automatic milking system.** C. Nieman,\* S. Utsumi, D. K. Beede, and J. Rowntree, *Michigan State University, East Lansing.*

The influence of 2 grazing and supplementation strategies on the performance of 14 United States Holstein ( $564 \pm 38$  kg BW; mean  $\pm$  SD) and 14 New Zealand Friesian ( $393 \pm 27$  kg BW) cows in a pasture-based automatic milking system (AMS) was investigated. The study was a  $2 \times 2$  factorial of the 2 breeds and 2 stocking rate treatments lasting 56 d. The low stocking rate (1.92 cows/ha) treatment was offered pasture plus concentrate (PC), whereas the high stocking rate (2.81 cows/ha) treatment was offered PC plus a partial totally mixed ration (pTMR; 60% forage). Amount of pTMR fed was adjusted weekly and used to

supplement deficits in pasture allowances and growth rate. Each stocking rate treatment had voluntary access to a single stall AMS where cows were offered 1 kg of concentrate per 6 kg of milk. Voluntary access to pastures was controlled via automated gates. Fresh pasture allocations were opened daily at 0500 h. Targeted pre- and post-grazing pasture biomass was  $2,400$  kg/ha and  $1,600 \pm 200$  kg/ha, respectively. Pre- and post-grazing biomass was recorded daily with a rising plate meter. Milk yield (MY), daily milking frequency (MF), Milk fat and Milk protein were analyzed with a repeated measures mixed model for a completely randomized design ( $P < 0.05$ ). There was a week by treatment interaction for MF, likely influenced by weekly MF increases for pTMR. The MY followed a similar week by treatment interaction trend ( $P = 0.08$ ), likely explained by MY increases for pTMR during the final weeks of the study. No treatment by breed or week by breed interaction was detected, but significant differences in milk production and composition were detected between the 2 breeds (Table 1). Results suggest a higher milk productivity for the high stocking rate and pTMR supplemented system and superior milk production for United States Holstein cows.

**Table 1.** Milk production and composition by breed

Variable	New Zealand Friesian	United States Holstein
Milk yield, kg/d	15.3 <sup>b</sup>	27.9 <sup>a</sup>
Milking frequency/d	2.2 <sup>b</sup>	2.5 <sup>a</sup>
Milk fat %	4.2 <sup>a</sup>	3.6 <sup>b</sup>
Milk protein %	3.3 <sup>a</sup>	3.1 <sup>b</sup>

<sup>a,b</sup>Means within a row with different superscripts differ ( $P < 0.05$ ).

**Key Words:** pasture-based dairy, automatic milking, feeding systems