

Physiology and Endocrinology I

666 Gram-negative or gram-positive toxin-induced subclinical mastitis affects preovulatory follicle responses in cows. O. Furman¹, G. Leitner², Z. Roth¹, Y. Lavon³, S. Jacoby⁴, and D. Wolfenson*¹, ¹Faculty of Agriculture, Food and Environment, the Hebrew University, Rehovot, Israel, ²The Veterinary Institute, Bet Dagan, Israel, ³Israel Cattle Breeders Association, Caesarea, Israel, ⁴Institute of Animal Science, Agricultural Research Organization, Bet-Dagan, Israel.

Long-term, subclinical mastitis lowers conception rate. The objective of this study was to induce subclinical mastitis by gram-positive (G+) exosecretions (*S. aureus* ex.) or gram-negative (G-) endotoxin (*E. coli* LPS), and examine its effects on preovulatory follicle responses. Throughout the study, cows were synchronized with series of GnRH and PGF_{2α} administered every 8 d. Cows received intramammary injections every 48 h for 20 d with minimal doses of either G+ (10 µg, n = 10) or G- (0.5 µg, n = 6) toxin, or saline (n = 6). Follicular fluids were aspirated before, at the end of (immediate effect), and 16 and 32 d after toxin exposure (carryover effect). Data were analyzed by ANOVA. During G+ or G- exposure, somatic cells count was higher ($P < 0.05$) than in controls, but body temperature ($38.8 \pm 0.1^\circ\text{C}$), plasma cortisol and haptoglobin concentrations did not differ among groups. In the G-group, a 50 to 60% immediate and 16 d-carryover decrease ($P < 0.05$) in plasma estradiol concentration relative to controls (11.0 ± 1.0 pg/ml) was noted; in the G+ group, only a 16 d-carryover decrease of 25% in plasma estradiol was noted ($P < 0.05$). Neither toxin induced an immediate drop in follicular steroids; however, an immediate decrease in the number of medium-size follicles was noted in the G+ group compared with controls (1.3 ± 0.2 vs. 3.0 ± 0.7 follicles; $P < 0.05$). Most G- group cows (5/6) exhibited a 32 d-carryover decrease ($P < 0.05$) of 70% and 62% in estradiol and androstenedione concentrations, respectively, in the follicular fluids. In the G+ group, only half of the cows exhibited a 33% and 60% carryover decrease in follicular estradiol and androstenedione concentrations, respectively ($P < 0.05$); in the other half, steroid levels were not altered. Results indicate that growth dynamics of medium-size follicles is attenuated mainly by *S. aureus* ex. Small, pre- to early antral follicles are susceptible to both *S. aureus* ex. and *E. coli* LPS toxins, and the induced damage is then carried over to the preovulatory follicles; this corresponds well with the long-term chronic nature of subclinical mastitis.

Key Words: mastitis, steroid, follicles

667 Blood constituents in milk due to changed blood-milk barrier integrity during mastitis. O. Wellnitz, M. Lehmann, and R. M. Bruckmaier,* *Veterinary Physiology, Vetsuisse Faculty, University of Bern, Bern, Switzerland.*

Mastitis is accompanied by changes in milk composition. Besides changes of mammary secretions the blood-milk barrier integrity is impaired and molecules can cross the border from blood into milk or vice versa. This study aimed to investigate the transfer of several blood constituents into milk during a mammary immune response due to intramammary endotoxin challenge. Five cows who received a β -hydroxybutyrate (BHB) clamp infusion to increase BHB in blood to a steady concentration (1.5 to 2.0 mmol/L) were challenged intramammarily into one quarter with 200 µg *E. coli* lipopolysaccharide (LPS). They were immunized against bluetongue virus (BTV) 2 years before. Blood and milk were analyzed hourly. Changes were considered significant if $P < 0.05$. Blood concentrations of LDH, IgG, and specific antibodies against BTV did not change. In milk the somatic cell count

increased in LPS challenged quarters within 4h but not in controls. Lactate dehydrogenase (LDH), BHB, lactate, and immunoglobulin (Ig) G was 33 ± 8 U/L, 0.08 ± 0.01 mmol/L, 8 ± 2 mg/L, and 0.1 ± 0.0 mg/L, respectively, and increased within 3 h and continued to increase through 8h to concentrations of 326 ± 146 U/L, 0.56 ± 0.01 mmol/L, 182 ± 26 mg/L, 0.72 ± 0.24 mg/L, respectively, in milk of LPS challenged quarters but not of control quarters. IgG increase in milk was paralleled by an increase of antibodies against BTV. Lactate concentrations in blood and milk of control quarters increased 2h after challenge and were correlated ($r = 0.74$). The blood constituents LDH, BHB, IgG, and lactate increase in milk after LPS challenge. The increase of antibodies against BTV in milk suggests that the primary mechanism of IgG transfer is opening of the blood-milk barrier and not specifically targeted to mastitis pathogens. In conclusion, opening of the blood-milk barrier during mammary inflammation increases the presence of blood constituents including antibodies in milk, and this does not appear to be a specific immune function response.

Key Words: blood-milk barrier, mastitis, antibody

668 Characterizing the temporal and seasonal pattern of plasma lipopolysaccharide binding protein during the transition period. A. Nayeri*¹, N. C. Upah¹, M. V. Sanz-Fernandez¹, E. Sucu^{1,2}, A. L. Gabler¹, R. L. Boddicker¹, D. B. Snider¹, J. M. Defrain³, and L. H. Baumgard¹, ¹Iowa State University, Ames, ²Uludag University, Turkey, ³Zinpro Corporation, Eden Prairie, MN.

During the periparturient period and heat stress (HS), dairy cows are more susceptible to rumen acidosis, which may compromise the gastrointestinal tract's (GIT) barrier function. Infiltrating lipopolysaccharide (LPS) is bound by LPS binding protein (LBP), but the role of LBP in dairy cow physiology is not well known. To investigate this, production data and blood samples were collected on lactating Holstein cows (n = 40) housed in a freestall barn and fed individually from -21 to 21 DIM. Two analyses were conducted: 1) good vs. poor transition cows and 2) winter vs. summer (July 2011) calving cows. It is important to note that data from 5 of the 40 cows were used as part of the winter and good transition analyses. Poor transition cows (n = 7) were defined as those treated for ketosis within 22 d after parturition. Conversely, good transition cows (n = 9) were void of a diagnosed health problem during the transition period. Cows included in the winter (n = 24) vs. summer (n = 5) analysis did not experience a diagnosed health event. Irrespective of classification, plasma LBP levels were lowest before calving and peaked at 3 DIM then slowly returned to pre-parturition concentrations by 21 DIM. Overall, poor transitioning cows had increased (21%, $P = 0.04$) circulating LBP compared with good transition cows and the difference was most pronounced (>2 fold) on 3 DIM. Nonesterified fatty acid and BHBA levels were not different between good and poor transitioning cows. However, we observed strong correlations between peak LBP and the transition success measures, milk yield slope ($r = -0.58$, $P < 0.05$) and BHBA area under the curve ($r = 0.72$, $P < 0.01$). Interestingly, winter calving cows had 52% ($P = 0.05$) greater LBP than summer calving cows and the difference was most pronounced (82%) at 3 DIM. In these cows, peak LBP was also negatively associated with average milk yield ($r = -0.49$, $P < 0.01$) and d 1-21 dry matter intake slope ($r = -0.57$, $P < 0.05$). Taken together, we hypothesize that circulating LBP concentration may play a role in determining seasonal and transition period success, or at least is closely associated with periparturient performance.

Key Words: dairy cow, LPS, transition

669 Effect of dry period length on rumen adaptation in dairy cows. R. M. A. Goselink¹, J. T. Schonewille², G. van Duinkerken¹, and A. T. M. van Knegsel³, ¹Wageningen UR Livestock Research, Lelystad, the Netherlands, ²Utrecht University, Utrecht, the Netherlands, ³Wageningen University, Wageningen, the Netherlands.

The objective of this study was to evaluate the effect of length of dry period on rumen papillae dimensions of dairy cows around parturition. Twelve rumen-cannulated Holstein dairy cows were assigned to a dry period length of 60 (G60), 30 (G30) or 0 (G0) days. The experiment started 60 d before expected calving date and lasted until wk 8 postpartum for each individual cow. Cows received ad libitum forage mixtures of corn silage, grass silage and straw (containing 12% CP, 5.3 MJ NEL/kg DM for dry cows and 15% CP, 6.4 MJ NEL/kg DM for lactating cows). Concentrates were fed individually starting 10 d antepartum with 1.0 kg/d, gradually increasing up to 8.5 kg/d at d17 postpartum. Milk yield and DMI were recorded daily. The rumen contents of each cow were evacuated at wk-9, wk-6, wk-2, d3, d7, d14, d28 and d56 relative to parturition. At each time, biopsies were taken to determine papillae dimensions at 3 locations: the right dorsal sac cranially of the dorsal coronary groove (DS), the right wall of the caudodorsal blind sac (DB) and the ventral wall of the caudoventral blind sac (VB). Treatment effects were tested by mixed model analysis using REML procedure. Data are presented as means \pm SE. Mean DMI postpartum was 21.2, 22.2 and 23.7 (± 0.6 kg/d) for G60, G0 and G30 respectively ($P < 0.05$). Average daily milk yield postpartum was significantly lower for G0 ($P < 0.05$) compared with G30 or G60 (35.1 vs. 41.4 and 41.8 \pm 3.1 kg/d, respectively). For cows with a dry period (G30 and G60), papillae surface area decreased prepartum ($P < 0.05$). More importantly, during the first 2 weeks postpartum the papillae growth was significantly delayed for G60 compared with G30; mean papilla surface was 42.5, 58.6 and 60.6 (± 7.1 , $P < 0.05$) mm² for G60, G30 and G0 respectively. Irrespective of treatment, rumen papillae were largest at VB and smallest at DS and papilla surface increased approximately 50% postpartum ($P < 0.05$). In conclusion, a shortened dry period of 30 vs. 60 d increases papillae surface in the first weeks of lactation and herewith may improve absorption capacity of volatile fatty acids, without a negative effect on milk yield as found with 0 d dry.

Key Words: dairy cow, rumen papillae, dry period length

670 Sodium salicylate administration during the first 7 days of lactation has effects that extend through the entire lactation in dairy cattle. J. K. Farney¹, L. K. Mamedova¹, J. F. Coetzee², J. E. Minton¹, and B. J. Bradford¹, ¹Kansas State University, Manhattan, ²Iowa State University, Ames.

Inflammation has been proposed as a contributor to fatty liver disease, and this disorder has negative effects on milk production in dairy cattle. Our objective was to determine if administration of sodium salicylate (SS), a non-steroidal anti-inflammatory drug (NSAID), in the first week of lactation changes whole-lactation productivity and retention in the herd. At calving, 78 cows ($n = 39$ primiparous [1P]; $n = 24$ 2nd lactation [2P]; $n = 15 \geq 3$ lactations [3P]) were alternately assigned to either control (CON) or SS treatment for 7 d postpartum. SS treatment was administered via individual water bowls at a concentration of 1.68 g/L, delivering a mean of 113.5 \pm 6.8 g SS/d during the 7 d of treatment. Cows were followed through the lactation by monthly milk yield and component testing, and the effects of treatment on the risk of leaving the herd and on 305-d milk, fat, and protein yields were determined by Fisher's exact test and mixed model analysis, respectively. Predicted transmitting abilities (PTA) for milk, fat, and protein yields were not influenced by treatment or treatment by parity interaction (all $P > 0.17$);

nevertheless, PTA values were included as covariates in the statistical models. Treatment by parity interactions were detected for 305-d milk and fat yields, and a tendency for an interaction was detected for protein yield. Milk yield was 2,469 \pm 646 kg greater over the lactation in 3P SS cows compared with 3P controls (21% increase, $P < 0.01$), but milk yield tended to be 980 \pm 375 kg less for 1P SS cows compared with 1P controls (8% decrease, $P = 0.07$); no effects were detected in 2P cows. Furthermore, 3P SS cows produced 130 \pm 23 kg more milk fat over the lactation (30% increase, $P < 0.001$) and tended to produce 49 \pm 18 kg more protein (14% increase, $P = 0.06$). No effects on fat or protein yield were detected for 1P or 2P. A treatment by parity interaction was observed for the risk of leaving the herd. First parity cows treated with SS cows tended to have greater risk of leaving the herd than controls (30% vs. 6% risk, $P < 0.10$). However, treatment did not alter herd retention in 2P or 3P groups, and SS had no effect on the risk of leaving the herd overall ($P = 0.59$). Results indicate that sodium salicylate has long-term effects on lactation of aged cows, particularly on fat metabolism, but has potential negative effects for primiparous cows.

Key Words: inflammation, milk production, non-steroidal anti-inflammatory drug

671 Responses to a nutritional challenge in early and late lactation. N. C. Friggens^{1,2}, C. Duvaux-Ponter^{1,2}, J. Tessier^{1,2}, and P. Schmidely^{1,2}, ¹INRA UMR 791 Modélisation Systémique Appliquée aux Ruminants, Paris, France, ²AgroParisTech UMR 791 Modélisation Systémique Appliquée aux Ruminants, Paris, France.

Characterizing robustness at the level of the individual animal would be valuable for refining management and selection strategies. Accordingly, to explore differences in adaptive capacity we studied responses of animals to a short-term nutritional challenge. Measurements of performance, milk composition and plasma metabolites were made in 16 dairy goats exposed to a 2-d nutritional challenge (underfeeding), at 2 different stages of lactation. Each challenge consisted of a 1-wk control period with standard TMR, 2 d of straw feeding, and a 1-wk recovery period on the TMR. All feeds were offered ad libitum. The 1st challenge was in late lactation (mean DIM, intake, milk yield: 280, 2.55 kg/d, 1.85 kg/d, respectively), the 2nd challenge was early in the following lactation (mean DIM, intake, milk yield: 26, 3.07 kg/d, 3.99 kg/d, respectively). Relative to the control periods, the challenges (means in late and early lactation, respectively) resulted in large drops in DMI (-1.78, -2.48 kg/d) and milk yield (-1.32, -2.46 kg/d) with increases in milk fat (+3.71, +4.65%) and milk protein (+0.91, +0.55%). For intake and milk yield, the size of the drop was strongly related to the pre-challenge values indicating that these 2 measures were tightly clamped by the challenge. For all other measures (milk fat, milk protein, NEFA, BHB, glucose, urea, insulin) the pre-challenge values had no significant effect, except for BHB in early lactation. Significant within animal repeatability between lactation stages in size of response for intake, milk yield, plasma urea, and BW were found (i.e., significant regression slopes; early vs late). Responses in milk fat, milk protein and the plasma metabolites (other than urea) were not significantly related, indicating that the metabolic adaptations to nutritional challenge vary not only between animals but also between stages of lactation. This was confirmed by principle components analysis which revealed different response patterns between individuals with regard to energy partitioning in milk. In conclusion, there is significant variation between individuals in response to a nutritional challenge and this can be used to develop a quantitative description of adaptive capacity.

Key Words: robustness, individual variation, ruminant

672 Supranutritional levels of antioxidants maintains feed intake and reduces heat stress in sheep. S. Chauhan^{1,2}, P. Celi³, B. Leury², and F. Dunshea^{*2}, ¹CSK, HP Agriculture University, Palampur, Kangra, India, ²The University of Melbourne, Parkville, Victoria, Australia, ³The University of Sydney, Sydney, Australia.

The present study was undertaken to investigate the impact of heat stress and dietary antioxidant supplementation on the oxidative status and physiology of sheep. Twenty-four Merino × Poll Dorset crossbred sheep were housed in one of 2 climatic chambers and offered either a control (10 IU Vitamin E and 0.24 mg Selenium/kg DM) or high antioxidant [100 IU Vitamin E and 1.20 mg Se (as SelPlex)/kg DM] diet. The sheep were subjected to 2 thermal treatments [Thermoneutral (TN): 18 to 21°C and 26–30% relative humidity and Heat Stress (HS): 28–40°C and 40–50% relative humidity] for 2 weeks in a single reversal design. After one week of dietary treatment, animals in one chamber were subjected to HS for one week, with the temperature being increased to 40°C between 9.00 h to 17.00 h and then maintained at 28°C overnight.

Those sheep in the TN group were maintained at 18–21°C. Physiological parameters (respiration rate, rectal temperature, skin temperature and heart rate) were recorded 4 times a day (9.00 h, 13.00 h, 17.00 h and 21.00 h and blood samples were collected on d 1 and 7 of heat treatment at 0hr, 8hr and 12hrs of start of heat. The temperature treatments were then reversed. The data was analyzed by undertaking REML (restricted minimum likelihood) variance component analysis using GenStat for windows (12th edition). All the physiological parameters recorded were elevated significantly ($P < 0.001$) by heat treatment. Respiration rate was reduced by antioxidant supplementation as indicated by a diet x temperature x time interaction ($P = 0.010$). There was 13% decline ($P = 0.014$) in feed intake of the control animals during heat stress whereas feed intake was maintained in sheep supplemented with antioxidants. Therefore, these data demonstrated that the negative effect of heat stress on feed intake and respiration rate can be reversed when sheep receive dietary antioxidant supplementation.

Key Words: heat stress, antioxidants, sheep