

but was higher in **AG** breed ($P < 0.001$). Romosinuano steers exhibit a lower sweat and respiratory rates than **AG** during HS, while maintaining the lower rectal temperature. Indices of heat loss used in

this study suggest that these avenues are not used to generate a lower rectal temperature seen in Romosinuano cattle during heat stress.

Key Words: Cattle, Heat, Tolerance

Animal Health - Livestock and Poultry: Bovine II

W10 Tumor necrosis factor- α (TNF- α), nitric oxide (NO), and xanthine oxidase (XO) responses to endotoxin (LPS) challenge in steers: Effect of progesterone (P4) and estradiol (E2) treatment. S. Kahl* and T. H. Elsasser, *USDA, Agricultural Research Service, Beltsville, MD.*

The severity of host response in some diseases differs between sexes and this dimorphism has been attributed to the immunomodulating effects of steroid hormones. In females, prevailing P4 or E2 concentration during different estrous cycle phases have been suggested to affect the immune responses to a disease stress. Our objective was to determine in steers the effect of P4 or E2 treatment on circulating concentrations of immune response mediators after two consecutive LPS challenges (LPS1 and LPS2, 6 d apart; 2.5 $\mu\text{g}/\text{kg}$ BW, i.v., *E. coli* 055:B5). Plasma concentrations of inflammatory initiation cytokine TNF- α , nitrate+nitrite (NOx, estimate of NO production), and XO activity (mediator of superoxide production), were measured. Twenty crossbred steers (392 ± 7 kg) were fed a forage-concentrate diet (15% CP) to appetite and assigned to control (C, $n = 7$), P4 ($n = 8$) or E2 ($n = 5$) treatment. Progesterone (1 mg/kg BW; i.m.) and 17 β -estradiol (E2, 2 mg/steer, i.m.) was injected 5, 3, and 1 d before LPS1 and LPS2. For each challenge, jugular blood samples were obtained at 0, 1, 2, 3, 4, 7, and 24 h relative to LPS injection. The primary response to LPS challenge was measured as area under the time \times concentration curve (AUC). Compared to C, P4 treatment decreased plasma TNF- α AUC after LPS1 (5.68 vs 8.15 ng/mL \times h, $P = 0.08$) and NOx AUC after LPS2 (32.3 vs 131.2 $\mu\text{M} \times$ h, $P < 0.05$). In contrast, E2 treatment augmented ($P < 0.01$) plasma TNF- α (14.66 vs 5.96 ng/mL \times h) and NOx (299.8 vs 131.2 $\mu\text{M} \times$ h) responses to LPS2. Plasma XO AUC was increased ($P < 0.01$) over C by E2 treatment after both LPS1 (406.8 vs 179.4 mU/mL \times h) and LPS2 (413.1 vs 156.5 mU/mL \times h). Results indicate that in cattle, circulating P4 and E2 may, respectively, attenuate or amplify the TNF- α response to LPS challenge as well as the subsequent responses of immune mediators (NO, XO) involved in oxidative damage to animal tissues.

Key Words: Endotoxin, Estradiol, Progesterone

W11 Prevalence of *Chlamydophila* spp. in randomly selected dairy farms in the western part of Germany. K. Kemmerling*¹, U. Mueller¹, M. Mielenz¹, K. Sachse², J. Winkelmann³, F. Jaeger⁴, and H. Sauerwein¹, ¹Institute of Animal Science, Physiology & Hygiene Group, University of Bonn, Germany, ²Federal Research Institute for Animal Health, Jena, Germany, ³North-Rhine-Westphalian Chamber of Agricultural Matters, Roleber, Germany, ⁴North-Rhine-Westphalian Ministry of Environment, Conservation, Agriculture and Consumers Protection (MUNLV), Duesseldorf, Germany.

Infections with intracellular bacteria of the genus *Chlamydophila* are associated with various symptoms such as infertility in cattle. Serological studies suggested a high level of exposure to *Chlamydophila*

spp. but systematic epidemiological investigations for the DNA-based detection of *Chlamydophila* spp. are only scarcely available. The objective of our study was to characterize the prevalence of *Chlamydophila* spp. in dairy cows in the western part of Germany (North-Rhine-Westphalia) since the epidemiological status of dairy cattle infection with *Chlamydophila* spp. in North-Rhine-Westphalia (NRW) was unknown. In total 100 dairy farms were randomly selected. For this purpose the dairy cow stocking rate in the different administrative districts of NRW was taken into account. Ten dairy cows per farm or at least 10% of the stand density per farm were sampled. For the detection of *Chlamydophila* spp. vaginal swabs from non-pregnant, early lactating dairy cows were analysed using an established highly sensitive genus specific real-time PCR. At present, samples from 80 dairy farms i.e. from 870 individual dairy cows have been analysed. Positive testings were observed in 61% of the farms and in 15% of the cows. The lower prevalence observed on a per cow basis might be explained by the discontinued shedding of the pathogen. Nevertheless, our results suggest that *Chlamydophila* spp. is widely spread in NRW. To evaluate the impact of *Chlamydophila* infections on herd health and fertility and to develop strategies to counteract *Chlamydophila* associated health disturbances, further investigations are needed.

Key Words: *Chlamydophila*, Dairy Cows, PCR

W12 Growth, health, and select immunologic and metabolic functions of preruminant calves housed in warm and cold environments. B. J. Nonnecke*¹, R. L. Horst¹, M. R. Foote², B. L. Miller³, T. E. Johnson³, and M. Fowler³, ¹National Animal Disease Center, Ames, IA, ²Iowa State University, Ames, ³Land O'Lakes Research Farm, Webster City, IA.

The physiological response of the preruminant calf to cold-stress has not been studied extensively. This study examined effects of sustained environmental cold on growth performance and health of preruminant calves. Functional measures of energy metabolism, fat-soluble vitamin and mineral status, and immune competency were also evaluated. Holstein calves, 3 to 10d of age, were assigned randomly to warm or cold environments and kept in these environments for 7wk. Cold environment calves ($n=15$) were exposed to temperatures maintained as close to 2°C as possible. Frequent wetting of the environment and calves was used to augment effects of the cold environment. Warm environment calves ($n=14$) were maintained as close to 15°C as possible. Warm environment humidity was not manipulated. Preventative medications or vaccinations that might influence disease resistance were not administered. Non-medicated MR (20% CP and 20% fat fed at .45 kg/d) and calf starter (ad libitum) were fed to all calves. During the 7wk period, cold environment temperatures averaged almost 20°C lower ($P \leq .05$) than warm environment temperatures. Relative humidity averaged 10% higher ($P \leq .05$) in cold environment. Warm environment calves were healthier and needed less medical

intervention. Growth rate was unaffected ($P > .05$) by environmental temperature; however, cold environment calves consumed more ($P \leq .05$) starter at wk5, 6 and 7. Blood glucose concentrations were lower ($P \leq .05$) and NEFA concentrations were higher ($P \leq .05$) in cold environment calves, indicative of a state of mild negative energy balance. Serum tumor necrosis factor, interferon and fat-soluble vitamin concentrations as well as antibody responses induced by vaccination were not affected ($P > .05$) by sustained exposure to cold. These results suggest that environmental cold has minimal effect on growth but does compromise calf health.

Key Words: Preruminant Calf, Cold Stress, Calf Health

W13 Pasteurization of colostrum reduces the incidence of paratuberculosis in neonatal calves. J. R. Stabel*, *USDA-ARS-NADC, Ames, IA.*

Feeding colostrum from infected dams to neonatal calves is one mode of transmission of paratuberculosis (Johne's disease). Recent studies have demonstrated improved morbidity and mortality rates in calves fed colostrum replacers or pasteurized colostrum. In the present study, the potential benefits of feeding pasteurized colostrum was demonstrated in calves born to dams naturally infected with *Mycobacterium avium* subsp. *paratuberculosis*. Calves were separated at birth from their dams and randomly allocated into a group fed the dam's colostrum, followed by feeding the dam's milk for 3 weeks (DC; $n = 6$), or into a group fed pooled pasteurized colostrum and milk replacer (PC; $n = 5$). After 3 weeks, calves were then weaned onto calf starter and housed and fed in a similar manner throughout the rest of the study. Blood and fecal samples were taken at birth and monthly throughout the 12-month study. Calves were necropsied at the end of the study and tissues were cultured for *M. paratuberculosis*. Twenty-five tissues were taken at necropsy and cultured for *M. paratuberculosis*. Sixteen of the 25 tissues were positive across treatment groups, with 14 of 16 tissues positive for DC calves and 9 of 16 tissues positive for PC calves. The degree of colonization within a tissue was low and varied between calves even within a treatment group. Fecal shedding was minimal during the study with 2 calves from each treatment group shedding negligible amounts of *M. paratuberculosis*. As a measure of early immune response to infection, blood obtained from calves was stimulated in vitro with *M. paratuberculosis* antigen preparations and IFN-gamma secretion was measured. Antigen-specific IFN-gamma was consistently higher throughout the study in calves fed colostrum and milk from their infected dams (0.95 vs 0.43 Abs540nm). These results indicate that feeding a source of clean colostrum to neonatal calves may reduce their exposure to *M. paratuberculosis*.

Key Words: *Mycobacterium avium* subsp. *paratuberculosis*, Calves, Colostrum

W14 Effects of pre- and postpartum feeding fish meal on total leukocyte and differential counts in transition and early lactating cows. A. Heravi Moussavi*¹, M. Danesh Mesgaran¹, T. Vafa¹, and A. Soleimani², ¹*Center of Excellence for Animal Science, Ferdowsi University of Mashhad, Mashhad, Khorasan Razavy, Iran,* ²*Azad University of Kashmar, Kashmar, Khorasan Razavy, Iran.*

The study was designed to test the effects of dietary fish meal supplementation on total leukocyte and differential counts in transition and early lactating cows. From approximately 21 d before anticipated calving until 35 d postpartum, cows ($n = 10$; 5/treatment) were fed diets that were isoenergetic containing 0 (Control) or 3.5% and 1.95% fish meal during prepartum and postpartum, respectively. Holstein cows were blocked in pairs based on their previous 305 d milk, parity (2nd and 3rd to 5th) and expected calving dates. Cows within each block were randomly assigned to one of the two treatments. Blood samples were obtained by coccygeal venipuncture within one week before calving and weekly after calving using EDTA as the anticoagulant. In the blood samples total leukocyte and differential counts were determined. The data were analyzed using the MIXED model for a completely randomized design with repeated measures. The model included treatment, time and the interaction. Results showed that the total leukocytes were similar among groups (7296 and 8134 \pm 599 for control and supplemented groups, respectively). The effect of time was significant ($P < 0.05$) and the interaction of time and treatment was not significant. Percentage of neutrophils (42.95 and 53.77 \pm 4.3, respectively), lymphocytes (53.94 and 42.85 \pm 4.4, respectively), and monocytes (2.88 and 2.96 \pm 0.3, respectively) were all similar between control and supplemented groups. Except than monocytes percentage which was affected by time ($P < 0.001$), others were similar during the study. Results from this experiment demonstrate that dietary fish meal supplementation pre- and postpartum had no apparent effect on total leukocyte and differential counts in transition and early lactating dairy cows.

Key Words: Dairy Cows, Fish Meal, Leukocyte

W15 New intramammary infections during the dry period: The effect of short (30 days) vs. long (45 or 60 days) dry periods. A preliminary report. G. T. Church*¹, L. K. Fox¹, J. M. Gay¹, C. T. Gaskins¹, and C. S. Schneider², ¹*Washington State University, Pullman,* ²*University of Idaho, Moscow.*

Lactating dairy cattle from 3 herds in eastern Washington and 1 herd in Idaho were enrolled in a study to test the hypothesis that cows given a **short** (theoretical 30-d dry) dry period would have no more **new intramammary infections (IMI)** than cows given a **long** (theoretical 45-d or 60-d dry) dry period. Cows were assigned systematically without bias to treatments groups. One tube of either a commercial lactating (short) or dry-cow (long) formulation of cephalosporin was administered to each lactating quarter per treatment group. Duplicate quarter foremilk samples were aseptically collected prior to dry-off and single quarter foremilk samples were collected 2, 7, 14, and 21 days post partum. **Pathogens** were classified as major (coagulase positive staphylococci, streptococcal species, and coliform species) minor (coagulase negative staphylococci and corynebacterium species) or other (gram positive bacilli and arcanobacterium pyogenes). **Cured** mammary quarters were free of IMI by the pathogen isolated from milk before dry-off. **Not cured** mammary quarters had the same pathogen in milk before dry-off and after parturition. Mammary quarters with **new IMI** had a new or different pathogen in milk post partum as compared to the dry-off sample. It appears that there is approximately a 40% increase in new IMI's in long ($n=23$) vs. short ($n=14$) dry periods.

Table 1. The number of quarters that cured, did not cure, or acquired a new IMI through the dry period

Pathogen	Short Dry Period			Long Dry Period		
	Major	Minor	Other	Major	Minor	Other
Cured	1	42	2	3	31	3
Not Cured	–	15	–	–	16	–
New IMI	5	8	1	2	21	–

Key Words: New Intramammary Infection (IMI), Dry Period

W16 Muscarinic receptors in the bovine gastrointestinal tract: mRNA expression and receptor binding in healthy cows and in cows with cecal dilatation-dislocation. E. C. Ontsouka*, R. M. Bruckmaier, A. Steiner, and J. W. Blum, *University of Berne, Vetsuisse Faculty, Berne, Switzerland.*

Acetylcholine interacts with muscarinic receptors (M) to mediate gastrointestinal (GI) smooth muscle contractions. We have compared mRNA levels and binding sites of M1 to M5 in muscle tissues from the fundus abomasi, pylorus, ileum, cecum, proximal loop of the ascending colon (PLAC), and external loop of the spiral colon (ELSC) of healthy slaughter cows (n=7). Furthermore, we have compared mRNA levels of M2 and M3 in full-thickness biopsies from the ileum, cecum, PLAC and ELSC of healthy (n=7) and of cows with cecal dilatation-dislocation (CDD; n=7). The mRNA levels were measured by qRT-PCR. The inhibition of [3H]-QNB binding by M antagonists [atropine (M1-5), pirenzepine (M1), methoctramine (M2), 4-DAMP (M3), and tropicamide (M4)] served to identify receptor subtypes at the protein level. Maximal binding (Bmax) was determined during saturation binding with atropine as a competitor. Location and group differences of Bmax and mRNA levels were tested for significance by ANOVA. The mRNA levels of M1, M2, M3 and M5 represented 0.2, 48, 50, and 1.8%, respectively, of total M, whereas mRNA of M4 was not detected. The mRNA levels of M2 and M3 in the ileum were lower (P < 0.05) than in other GI-locations. Atropine and antagonists for M1, M2 and M3 inhibited [3H]-QNB binding according to a one- or a two-site receptor model. The M4 antagonist had no effect on binding. Bmax in the fundus, pylorus, and PLAC was lower (P < 0.05) than in the ELSC, and lower (P < 0.05) in the pylorus than in the ileum. In cows with CDD, mRNA of M2 (cecum, PLAC and ELSC) and M3 (all locations) were reduced by 47 to 67% and 73 to 85%, respectively, as compared to healthy cows. In conclusion, M2 and M3 appeared to be the most important M for GI motility in cows. The decreased levels observed in cows with CDD may indicate involvement of these two receptor subtypes in the motility disorder leading to CDD.

Key Words: Cattle, Motility Regulation, Receptor Expression

W17 mRNA expression of motility-mediating receptors from the abomasum to the spiral colon of healthy cows and of cows suffering from left-sided abomasal displacement. E. C. Ontsouka*, M. Niederberger, A. Steiner, R. M. Bruckmaier, and M. Meylan, *Vetsuisse Faculty, University of Berne, Berne, Switzerland.*

Dairy cows frequently suffer from left-sided displacement of the abomasum (LDA), which causes important economical losses. Although the symptoms of LDA are well known, the pathogenesis of the disease remains unclear. Motor functions of the gastrointestinal (GI) tract are tightly controlled by the enteric nervous system, modulated by the sympathetic and parasympathetic nervous systems, involving α - and β -adrenergic receptors (AR), as well as muscarinic receptors (M) on nerve terminals and smooth muscle cells in the wall of GI organs. In addition, a non-adrenergic and non-cholinergic pathway also influences GI motor functions via motilin receptors (MTL-R). To investigate if the expression of receptors mediating motility vary in the GI tract of dairy cows depending on their health status, we have compared mRNA levels of α 2AD-AR, β 2-AR, M2, M3, and MTL-R in full-thickness specimens from the abomasum (fundus abomasi, pylorus), duodenum, cecum and external loop of the spiral colon (ELSC) of freshly slaughtered healthy cows (H; n=8) and of cows with LDA (D; n=8). The mRNA levels were measured by qRT-PCR and normalized relative to GAPDH. Receptor mRNA levels were evaluated using the repeated procedure of mixed model (SAS). Differences between H and D within locations were investigated with the t-test. The mRNA levels of α 2AD-AR, β 2-AR, M2, M3, and MTL-R varied among GI locations (P < 0.05). The mRNA levels of all five receptors were lower (P < 0.05) in the duodenum of D than of H. In addition, the mRNA levels of β 2-AR were higher (P < 0.05) in the ELSC of D than of H. In conclusion, differences between H and D of mRNA levels for α 2AD-AR, β 2-AR, M2, M3 and MTL-R in the duodenum, suggests that LDA might be caused by a primary motility disturbances in this GI location rather than in the abomasum itself.

Key Words: Bovine, Gastrointestinal Motility, Receptors

W18 The relationship between postpartum uterine bacterial infection (BI) and subclinical endometritis (SE). R. O. Gilbert*, N. R. Santos, K. N. Galvão, S. B. Brittin, and H. B. Roman, *Cornell University, Ithca, NY.*

The reported prevalence of SE exceeds reported prevalence of uterine BI after completion of postpartum involution. The objective was to investigate the relationship between uterine BI and SE. Samples for simultaneous uterine culture and cytology were obtained from 56 Holstein cows in a single herd at 0, 7, 21, 35 and 49 (\pm 3) DIM. Overall positive culture was 57%, 71%, 55%, 26% and 36%, respectively. On d 0 and 7 the commonest aerobic pathogens cultured were E. coli and Streptococcus spp and from d 21 Arcanobacterium pyogenes (Apyo) predominated. Amongst anaerobes, Clostridium perfringens was cultured frequently on d 0 and 7, but seldom thereafter. Prevotella melaninogenica (Pmel), and Fusobacterium necrophorum were cultured at low rates throughout. The median proportion of neutrophils (PMN) at each time point was 37%, 20%, 41%, 7% and 4% (P < 0.01). The proportion of other cell types at each time point did not differ with days postpartum. The median proportion of macrophages (LMN) was zero at each time point (range; 0 to 10%), and the median proportion of lymphocytes (SMN) was 2.0% (range; 0 to 13.5%). Bacterial isolates were not correlated to proportion of LMN or SMN. On d 0 there was a significant negative correlation between PMN proportion and bacterial isolates (aerobic, r = -0.46, P = 0.03; and anaerobic, r = -0.47, P = 0.07). PMN proportion at d 0 was also negatively correlated with PMN on d 49 (r = -0.48, P 0.01). This suggests that prompt PMN recruitment to the uterus after parturition limits bacterial infection and reduces the risk of later SE. The PMN proportion on d 49 was correlated to

the presence of Apyo ($r = 0.49$, $P < 0.01$) and anaerobes, particularly Pmel ($r = 0.44$, $P < 0.01$). In turn, the presence of these pathogens was affected by the presence of E.coli on d 7 ($r = 0.40$, $P < 0.01$). By multiple linear regression, the presence of Apyo or Pmel explained 48% of the variance in PMN on d 49 ($P < 0.01$). The proportion of PMN on d 49, previously shown to be significantly correlated to subsequent reproductive performance was correlated to uterine BI, which was more prevalent on d 35 and 49 than previously reported.

Key Words: Endometritis, Bacteria, Dairy Cow

W19 The recurrence of mycoplasma mastitis investigated by bulk tank analysis. V. Punyapornwithaya*, L. K. Fox, D. D. Hancock, and J. M. Gay, *Washington State University, Pullman.*

The objective of this study was to determine the epidemiology of the recurrence of mycoplasma investigated by culture of bulk milk. Fourty farms that had *Mycoplasma spp.* culture from bulk tank milk were investigated for 1 year by further monitoring bulk tanks to determine the prevalence of recurrence of this agent. Dairies had at least 5 bulk tank milk cultures during the study. Bulk milk samples collected within the same month from 10 farms after the first positive culture were evaluated for *Mycoplasma spp.* Milk samples were plated on mycoplasma agar and incubated at 37°C with 10% CO₂ for 7 days. The percentage of farms with a recurrence of mycoplasma mastitis was 57.5% ($n=23/40$). The mean number of recurrences within 1 year was 2.45. Bulk milk samples from 4 herds that were examined in the same month of a first positive culture of Mycoplasma spp. were negative at the second test, as opposed to the 6 herds also tested twice in the same month that remained positive. This study suggested that the prevalence of recurrence infection of mycoplasma mastitis was greater than 50%. Cultures of bulk tank milk can be used to monitor mycoplasma mastitis, and help dairy managers use it as a tool to control the disease.

Key Words: *Mycoplasma spp.*, Mastitis, Recurrence

W20 Use of a calcium bolus to improve calcium homeostasis after calving. J. D. Sampson*¹, J. N. Spain¹, L. Carstensen², and C. Jones³, ¹University of Missouri, Columbia, ²Boehringer Ingelheim Denmark A/S, Copenhagen O, Denmark, ³Boehringer Ingelheim Vetmedica, Inc., St. Joseph, MO.

Milk fever occurs in 5-10% of dairy cows, and the incidence of subclinical hypocalcemia is likely several times greater. Milk fever increases the risk of numerous periparturient disorders such as ketosis and dystocia. Providing periparturient cows with supplemental calcium may decrease the risk of hypocalcemia. The objective of this study was to investigate the effects of Bovicalc™, an oral calcium supplement, on calcium homeostasis during the first 24 h after calving. Multiparous Holsteins ($n=20$) were blocked by parity and day of calving and randomly assigned to one of two treatment groups. Control (C) cows received no calcium supplement. Treatment group (B) received one bolus immediately after calving and a second bolus 12 hours after calving. All cows were housed in a sand bedded free stall barn with free access to clipped pasture and fed a TMR formulated to meet NRC requirements. Prepartum blood samples were taken at approximately 24 and 48 hours prior to calving. Ionized blood calcium (iCa) was

measured using an IDEXX Vet Stat Analyzer. Cows with an iCa level of 1.10 mmol/L or less were included in the study. Blood and urine samples were collected at 0, 1, 6, 12, 13, and 24 hours postpartum. Blood was analyzed for iCa and pH. Urinary pH was measured. Rectal temperatures and clinical scores for appetite, ataxia, fecal consistency, and muscle tremors were recorded at 0, 6, 12, and 24 hours postpartum. iCa levels were not different between treatment groups (1.24 vs. 1.22 and 1.19 and 1.18 mmol/L for C and B at -48 and -24 h, respectively. iCa levels were similar at calving (0h, 0.95 vs. 0.94 for C and B, respectively, $P=0.84$). iCa levels differed due to treatment over time ($P=0.02$). iCa were significantly higher in cows given the bolus at 1 and 13 hour. Urine pH was different by treatment ($P=0.002$) and by treatment over time ($P=0.003$). Urine pH decreased in cows given B from 7.58(0h) to 6.79 (24h) compared to 8.00 (0h) to 8.09 (24h) for control cows. Blood pH was not different ($P=0.27$). Calcium supplementation with Bovicalc™ after calving decreased urine pH and increased iCa levels compared to control cows.

Key Words: Milk Fever, Calcium, Dairy

W21 Dietary fish oil does not impact the response of early lactating cows to an endotoxic mastitis challenge. M. K. Yelle*, D. W. Kim, E. J. DePeters, and M. A. Ballou, *University of California, Davis.*

Cows in early lactation are more susceptible to the deleterious local and systemic effects of the acute phase response. Supplementing fish oil in many animal models attenuated the acute phase response. The objective was to determine the effects of supplementing fish oil during the peripartum period on the pathophysiological response to an intramammary LPS challenge. 42 multiparous, Holstein cows were completely randomized to one of three treatments at 3 weeks prepartum. Treatments were no supplemental lipid or supplemental lipid, 250 g (prepartum) or 1% of the previous day's intake (postpartum), from either Energy Booster® (EB) or fish oil (FO). Supplemental lipid was fed as a bolus prior to the AM feeding. At 7 DIM cows supplemented with lipid were infused with 100 µg of E coli LPS into one rear quarter; cows not supplemented with lipid served as un-infused controls. DMI was measured daily and clinical measures taken at 0, 1, 2, 3, 4, 5, 6, 8, 12, 24, and 72 h. Blood samples were collected at 0, 2, 4, 6, 8, 12, 24, and 72 h for biochemical analyses. Milk samples were collected before and from the 6 milkings following the LPS challenge. All data will be reported as EB versus FO respectively. For EB and FO, LPS caused a significant increase in both rectal temperature and heart rate, which peaked at 6 h. Total white blood cell counts decreased following LPS, reaching nadir after 6 h. Supplemental lipid source had no effect on any clinical response. Feed intake significantly decreased the day LPS was infused (16.6 and 17.0%). LPS increased serum glucose levels and caused a significant biphasic decrease in NEFA, reaching nadir at 4 and 8 h and returning to baseline levels within 24 h. Milk production in the infused quarter decreased dramatically over the first two milkings (33.9 and 41.8%) and returned to baseline by the 5th milking. Control quarters also decreased and reached nadir at the 1st milking (15.4 and 12.9%) but quickly recovered to baseline production by the 2nd milking. Supplementing fish oil had no ameliorative effect on either the local or systemic acute phase response of early lactating, Holstein cows.

Key Words: Endotoxin, Mastitis, Transition

W22 *Escherichia coli* lipopolysaccharide upregulates the expression of both toll like receptor 4 and 2 (TLR4 and TLR2) in cultured bovine mammary epithelial cells. E. M. Ibeagha-Awemu^{*1}, J.-W. Lee², A. E. Ibeagha¹, D. D. Bannerman³, M. J. Paape³, and X. Zhao¹, ¹McGill University, Ste Anne De Bellevue, Quebec, Canada, ²National Pingtung University of Science and Technology, Neipu, Pingtung, Taiwan, ³United States Department of Agriculture, Beltsville, MD.

Bovine mammary epithelial cells contribute to the innate immune response to intramammary infection. Their ability to mount such a response is dependent upon mammary epithelial recognition of the invading pathogen by specialized receptors. Toll-like receptor 4 (TLR4) is one such receptor that recognizes and is specifically activated by lipopolysaccharide (LPS), a component of the outer envelope of gram-negative bacteria. Recently, it was shown that *Staphylococcus aureus*, a gram-positive bacteria, initiated the upregulation of both TLR2 and TLR4 in the bovine mammary gland. Because the mammary gland is known to elicit a robust innate immune response to *Escherichia coli*, we hypothesized that LPS could similarly induce upregulation of TLR4. To evaluate this, MAC-T cells (a bovine mammary epithelial cell line) were incubated in the presence or absence of 1 µg/ml LPS for 24 hrs. Expression of TLR2 and TLR4 were analyzed at both the mRNA and protein levels by quantitative real time PCR and flow cytometry, respectively. The mRNA for both receptors in treated cells was upregulated by 2.0 (TLR4) or 2.2 (TLR2) fold ($P < 0.01$) in comparison to untreated cells. Similarly, specific antibodies against TLR2 and TLR4 detected the increased surface expression of the toll proteins. The mean channel fluorescence in treated cells as compared to untreated cells was 1224 vs. 554 for TLR4 and 3394 vs. 1671 for TLR2, respectively. These results demonstrate that LPS upregulates both TLR4 and TLR2, similar to that reported with ligands from *S. aureus*. This suggests that the bovine mammary epithelium possesses the necessary immune repertoires required to mount a robust defense against *E. coli*. This may also indicate a positive adaptation by the mammary epithelial cells to effectively deal with different types of mastitis pathogens.

Key Words: Lipopolysaccharide, TLR4 and TLR2, Mastitis

W23 Effect of supplementation with a *Bacillus*-based direct-fed microbial on calf growth, *Clostridium perfringens* shedding, and incidence of scours. C. Wehnes^{*1}, E. Davis¹, K. Novak¹, V. Patskevich¹, T. Rehberger¹, D. Shields², and J. Coalson², ¹Agtech Products, Inc., Waukesha, WI, ²Merrick's, Inc., Union Center, WI.

Electrolytes are common and effective treatments for dehydration in calves; however, electrolytes do not treat the primary cause of dehydration, bacterial and viral associated scours. The objective of this study was to evaluate the efficacy of the addition of a *Bacillus* direct-fed microbial (DFM) to an electrolyte as therapy for scours. The DFM was evaluated based on calf performance and fecal *C. perfringens* shedding. In October 2006, 65 Holstein bull calves were placed in outdoor hutches and 21 calves were randomly selected to determine preliminary *C. perfringens* fecal shedding. Calves were assigned to three treatments based on the presence of scours: non-scouring, electrolyte, and electrolyte+DFM. Scouring calves received electrolyte for a mandatory two days. Fecal samples were collected

on d 1, 3, 7, 14, 21, 28, and 42 post-placement and enumerated for *C. perfringens*. *C. perfringens* colonies were harvested and DNA was extracted. Putative *C. perfringens* colonies were confirmed as *C. perfringens* via multiplex Polymerase Chain Reaction (mPCR) for the four major *C. perfringens* toxin genes (α , β , ϵ , and ι). randomly amplified polymorphic dna pcr (rapd pcr) was performed to characterize *C. perfringens* isolates. By d 14, 49 of the 51 scouring calves were treated. Although results indicate that electrolyte+DFM calves began the trial with significantly greater ($P = 0.01$) populations of fecal *C. perfringens*, by d 7 the electrolyte+DFM significantly reduced ($P = 0.002$) populations of fecal *C. perfringens* compared to electrolyte alone. Electrolyte+DFM calves had significantly higher ($P = 0.02$) ADG than both non-scouring and electrolyte treated calves in wk 1. Gain:feed was significantly greater ($P = 0.04$) in electrolyte+DFM calves than calves administered electrolyte and non-scouring treatments during wk 8. Results indicate that supplementation of electrolyte+DFM reduced *C. perfringens* fecal shedding and improved gain during wk 1. The DFM may have other ancillary benefits after feeding has ended, as evidenced by improved gain:feed in wk 8.

Key Words: Probiotic, Bovine, Electrolyte

W24 Prevalence, etiology and antimicrobial sensibility of subclinical mastitis. M. C. Rubio Robles^{*}, M. A. Luque, R. Verdugo, R. Chin, R. Félix, E. Hernández^{*}, T. Leal, and J. Mena, *Universidad Autonoma de Sinaloa, Culiacan, Sinaloa, Mexico.*

Mastitis is an inflammation of the mammary gland, the subclinical mastitis is characterized not to cause a visually inflammation of the mammary gland, nor macroscopic changes in milk, being necessary for their diagnosis tests that allow to detect the involved microorganisms. both clinical and subclinical forms of mastitis cause a reduction in milk quality and quantity and is recognized as a cause of major economic loss to the dairy industry. The objective of this study was to determine the prevalence of subclinical mastitis and its causal agents in familiar dairy stables, in San Jose of the Brecha, located in 137 km Culiacán-Guasave highway; municipality of Guasave, Sinaloa, México. where sampled 24(100%) cows of American Swiss Brown with Brahman, 3 to 8 childbirths and weights of 350 to 400 kg, fed at prairie, including all the cows of milks, except less than 15 days of lactation and more than 7 months of gestation; using the California mastitis test, resulted 100% of the dairy stables presence of subclinical mastitis; 62.5%(15) of the cows was positive, and 37.5%(9) negative. In the positive cows 26(27.08%) of the quarters were infected with some degree of mastitis, 57.69%(15) degree 1, 34.61%(9) degree 2 and 7.69%(2) degree 3. The positive samples were analysed with bacteriological and antibiograms in the Bacteriology laboratory of the FMVZ-UAS. showed 18.18%(2) development of *Staphylococcus* spp, rest 81.1%(9) was negative. In antimicrobial sensitivity *Staphylococcus* spp was resistant to the Enoxacin and the Dicloxacillin, very susceptible and moderately susceptible to penicillin and the anetilmicin. it is concluded that the prevalence of subclinical mastitis in San Jose of the Brecha is present in all the familiar dairy stables and a considerable proportion in cows, which can trigger another type of mastitis (clinical) producing lost economic considerable the producers.

Key Words: Mastitis, Dairy Cattle, Prevalence

W25 A cross-sectional survey of *Salmonella* serotypes from dairies with a history of Salmonellosis in the Great Lakes Region of the United States. C. Wehnes*, V. Patskevich, K. Mertz, and T. Rehberger, *Agtech Products, Inc., Waukesha, WI.*

Characterization of *Salmonella* shedding to identify cattle and herd associated risk factors has been well researched; however, there is a paucity of data characterizing *Salmonella* strains across dairies. For these reasons, a cross-sectional survey of *Salmonella* on dairies with a history of Salmonellosis was performed with the objectives of assessing *Salmonella* prevalence, serotype, and strain diversity. From June to December 2006, fecal samples from cows and calves as well as bedding samples from cow, calf, and sick cow pens were collected from eight independent dairy farms experiencing Salmonellosis. Each sample was cultured for *Salmonella*; putative colonies were confirmed as *Salmonella* via a latex agglutination assay (Remel, Lenexa, KS). DNA was extracted from *Salmonella* positive colonies and Randomly Amplified Polymorphic DNA Polymerase Chain Reaction (RAPD PCR) was performed. RAPD PCR products were analyzed using BioNumerics software (Applied Maths Inc. Austin, TX) to assess *Salmonella* diversity. Of the 196 fecal and 104 bedding samples collected, 42 (21%) and 37 (36%) contained *Salmonella* respectively. Serogroups found included B (13%), C (52%), D (3%), and E/G (32%). A total of 342 isolated *Salmonella* colonies were collected throughout the study. The results of the *Salmonella* genotypic diversity survey indicate that the strain richness (S) and evenness (E_D) for the 342 *Salmonella* isolates was 56 and 0.35 respectively at 80% similarity using the Pearson similarity coefficient with the unweighted pair group method using arithmetic averages (UPGMA). The reciprocal of the Simpson Index ($1/D$) was 19 and D_{max} was 56. The RAPD PCR results indicate that these farms contained *Salmonella* strains with little genotypic diversity within each farm. Conversely, the genotypic diversity between farms is large with few overlapping strains present. Future research should focus on further characterizing *Salmonella* from dairies as a means to understand the basis of farm specific isolates.

Key Words: Dairy, Diversity, Prevalence

W26 Correlating body weight and temperature changes after antibiotic treatment of morbid stocker calves with health and growth performance throughout the receiving phase. S. Behrends*, E. B. Kegley, and J. A. Hornsby, *University of Arkansas, Fayetteville.*

Records from market stressed bull and steer calves ($n = 383$) initially weighing 237 ± 46 kg were studied to identify an easily obtainable measurement that could be taken after an antibiotic treatment (AT) and used as a prognostic tool for calves affected by bovine respiratory disease (BRD). Calves were evaluated for signs of BRD and assigned a critical illness score of 1 (normal) to 5 (moribund). Based on these evaluations calves ≥ 2 were pulled. Calves that exhibited a rectal temperature of $\geq 40^\circ\text{C}$ were weighed and administered an AT. Rectal temperatures and body weights (BW) were taken 2 to 3 d after each treatment (according to product label). Calves that continued to exhibit a rectal temperature of $\geq 40^\circ\text{C}$ were administered another AT. Calves were treated with up to 3 antibiotics and were considered chronic if they did not respond (temperature $< 40^\circ\text{C}$) to the third treatment. A negative correlation existed between calves administered one AT and BW on d 7 and 42 ($r = -0.15$; $P < 0.01$ and $r = -0.24$; $P < 0.0001$

respectively). Body weights on d 15 and 42 were negatively correlated with whether calves were administered a second ($r = -0.14$; $P = 0.02$ and -0.23 ; $P < 0.0001$, respectively) and third ($r = -0.13$; $P = 0.03$ and $r = -0.23$; $P = 0.0001$, respectively) AT. Body weight change during the AT period (between initial pull and recheck) was negatively correlated with the need for a second and third AT ($r = -0.14$; $P = 0.03$ and $r = -0.29$; $P = 0.005$, respectively). Rectal temperature change (between initial pull and recheck) during the first AT period was correlated with those calves that were treated a second and third time ($r = 0.36$; $P < 0.0001$ and $r = -0.33$; $P < 0.0001$). Weight and temperature change during AT appear to both be useful indicators of potential outcomes for calves that are affected by BRD.

Key Words: Stocker Calves, Bovine Respiratory Disease, Antibiotic Treatment

W27 Feeding unprotected fish oil 3 weeks prepartum alters the fatty acid composition of plasma in both the cow and calf at parturition, but had no effect on bactericidal or cytokine function. M. A. Ballou*, R. C. Gomes, and E. J. DePeters, *University of California, Davis.*

Previous research from our lab demonstrated that supplementing a milk replacer with fish oil attenuated the acute phase response in calves; however it took weeks to months of fish oil (FO) supplementation before docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA) reached a new steady state, respectively. The objectives were to determine whether feeding a rumen unprotected FO would alter the fatty acid (FA) composition of plasma from both the cow and calf at parturition; as well as evaluate bactericidal and cytokine function. 30 Holstein cows were completely randomized to one of two treatments at 3 wk prepartum. Treatments were either 250 g of (1) Energy Booster® (EB) or (2) FO fed as a bolus prior to the AM feeding. Plasma was sampled from each cow at enrollment and from the cow and calf on the day of parturition for FA analyses of plasma phospholipids. At enrollment and parturition peripheral blood mononuclear cells (PBMC) were isolated from the cow and cultured with phytohemagglutinin; IFN- γ and TNF α were quantified. Whole blood bactericidal capacity against 3 microorganisms was evaluated at enrollment, parturition, and +21 d. In the calf, the whole blood bactericidal assay was measured 2 h after parturition, +1, and +21 d. In the cow FO increased the concentrations of EPA (3.38 vs. 0.62 g/100g FA), and DHA (2.74 vs. 0.29 g/100gFA) when compared to EB. Furthermore, FO increased EPA (0.81 vs. 0.39 g/100g FA) and DHA (2.09 vs. 1.12 g/100g FA) concentrations in the calf at calving. FO had no effect on the arachidonic acid concentration in the cow and calf. FO had no effect on the production of IFN- γ and TNF α from PBMC cell cultures. Furthermore FO had no effect on the bactericidal capacity of blood from either the cow or calf against any of the microorganisms. Supplementing the close-up diet with FO dramatically altered the FA composition of plasma from the cow and her calf at parturition. However, the plasma concentrations of EPA and DHA in the calf were below what can be acquired by supplementing the calf during the neonatal period.

Key Words: Immune, Fish Oil, Fatty Acid

W28 Relationship of plasma immunoglobulin G concentrations to temperament and growth performance. K. R. Parker^{*1}, S. T. Willard², R. D. Randel³, T. H. Welsh, Jr.⁴, and R. C. Vann¹, ¹MAFES-Brown Loam Experiment Station, Raymond, MS, ²Mississippi State University, Starkville, ³Texas A&M University Agricultural Research & Extension Center, Overton, ⁴Texas A&M University, College Station.

This study was designed to assess the relationships between immune function, innate temperament and growth performance. Blood samples and BW were taken from spring born calves (n = 196) at 24h, 48h, 14d, and 84d post-calving. At birth calves were assigned a calf vigor score, calving ease score and dams were given a BCS according to Beef Improvement Federation guidelines (2002). Plasma was harvested via centrifugation and concentrations of immunoglobulin G (IgG) were determined by ELISA. Temperament data were recorded in the form of pen scores (PS) and exit velocity (EV). These measures were taken along with BW at 28d pre-weaning, weaning, 28d post-weaning, 56d post-weaning and as yearlings. Overall temperament scores (TPS) were assigned to each animal by averaging PS and EV over the four time periods. Statistical analyses were performed using the Proc Mixed procedure of SAS and where appropriate repeated measures analyses were used. Calves were ranked based on their TPS as follows: calves 1 SD above the mean were considered temperamental (T, n=26), calves 1 SD below the mean were considered calm (C, n=36), all other calves were considered intermediate (I, n=127). Calves were classified based on their IgG concentration with calves 1 SD lower than the mean ranked low (L, n=3), calves 1 SD above the mean ranked high (H, n=27), and remaining calves ranked moderate (M, n=159). Heavier calves at birth ($P \leq 0.05$) had lower IgG at 24h and 48h. Calves with higher IgG at 48h exhibit greater ($P = 0.062$) ADG between weaning and 56d. Calves with higher IgG concentrations did not experience calving difficulties ($P \leq 0.001$). Scrotal circumference (SC) at weaning and 56d post-weaning were higher ($P \leq 0.03$) for calves with greater 48h IgG concentrations and were higher in calves classified as calm. Calves with higher immune classification had improved growth performance after weaning. These results suggest that IgG classification could be useful in predicting growth performance after weaning.

Key Words: Immunoglobulin, Beef Calves, Temperament

Breeding and Genetics - Livestock and Poultry III

W30 The genes commonly expressed at early embryonic stages in mammals. C. Y. Lien^{*}, E.-C. Lin, C. C. Hsu, S. T. Ding, and W. T. K. Cheng, National Taiwan University, Taipei, Taiwan.

The early stages of embryogenesis are critical for mammalian embryo development. Several key developmental events occurred in these stages, such as cell growth, migration, differentiation, and morphogenesis. In spite of the importance occurring in the early embryogenesis, limited information has been provided by previous studies. The UniGene database in the National Center for Biotechnology Information (NCBI) was designed to collect sequences of expressed sequences tags (ESTs) and mRNA to provide many sets of transcript sequences from the same transcription locus. The purpose of this study is to utilize the UniGene database to screen those commonly expressed genes with related functions at early embryonic stages in mammals. Those unigene entries of embryos before implantation collected from *Bos taurus* (Bt), *Mus musculus* (Mm) and *Sus scrofa* (Ssc) were 1,727, 13,923 and 3,982, respectively. The unigene sequences of Mm (13,923)

W29 Impact of entomopathogenic fungus *Metarhizium anisopliae* on cattle naturally infested by adult *Haematobia irritans* in temperate Mexico. E. Maldonado-Simán¹, R. D. Améndola-Massiotti^{*1}, E. Galindo-Velasco², C. A. Angel-Sahagún², L. Bermúdez-Villanueva¹, and R. Lezama-Gutiérrez², ¹Universidad Autónoma Chapingo, Chapingo, México, ²Universidad de Colima, Tecomán, Colima, México.

Farmers tend to control horn fly (*Haematobia irritans* L.) with heavy pesticide applications which increase the risks of insecticide residues in products and the environment; therefore, biological pest control is considered to be ecologically safer. The University of Colima, Mexico recently developed a biopesticide based on the spores of the fungus *Metarhizium anisopliae* Ma34 as a natural pathogen. No studies have tested on the field the action of this entomopathogenic fungi on the control of *H. irritans*; therefore, the aim of this study was to assess the efficacy of *M. anisopliae* on the control of *H. irritans* in naturally infested cattle. The study took place in the Experimental Station of the University of Chapingo (temperate central area of the country). Twenty two grazing Holstein cows were randomly allocated into two groups of the same size, which grazed in different paddocks. Cows in the treated group were sprayed every seven days with a conidial suspension of *M. anisopliae* (strain Ma34) at a concentration of 1×10^8 conidia/ml. Eight applications took place during the experimental period. The other group remained as untreated control. The horn fly burdens were recorded on days 0, 1, 2, 3, 4, 5, 6 and 7 post-treatment. Cows sprayed with *M. anisopliae* showed fly counts 28% lower than the untreated control ($P < 0.05$). Since the experiment took place during the second half of the fall, horn fly counts became lower through time (from week one to week eight) in both treatments, being reduced from 156 to 24 flies per cow in the sprayed cows and from 216 to 37 flies per cow in the untreated cows; the reduction due to the application remained fairly constant. This result shows that *M. anisopliae* (strain Ma34) was effective in the control of adult *H. irritans* and might be a promising substitute of chemical pesticides.

Key Words: Hyphomycetes, Biological Control, Horn Fly

were used to search against (blastn) the unigene sequence sets of Bt (1,727) and Ssc (3,982) to obtain 972 and 2,039 commonly expressed genes (bit score ≥ 200), respectively. However, there were 417 unigene entries commonly expressed in all these three species. The 417 commonly expressed genes annotated to have certain gene identity in public databases for Bt, Mm and Ssc were 339, 361 and 151, respectively. Furthermore, those commonly expressed unigenes of Mm were annotated their functional classification using Gene Ontology terms into three categories: biological process (P), molecular function (F) and cellular component (C). The results showed the number of commonly expressed genes with functional annotation were 160 (P), 169 (F) and 153 (C), respectively. The commonly expressed genes and their related functions might be annotated using alignment tool among sequence databases of different species. The analytical procedure in this study would assist animal scientists to screen the commonly expressed genes with fundamental roles and functions.

Key Words: Early Embryonic Stages, Commonly Expressed Genes, Mammals