

Animal Health: Reproductive health and acute immune responses

20 Efficacy of nonsteroidal antiinflammatory drugs for the treatment of acute puerperal metritis in dairy cows. Alina Pohl and Wolfgang Heuwieser*, *Clinic of Animal Reproduction, Freie Universität Berlin, Berlin, Germany.*

Acute puerperal metritis (APM) in dairy cows is often treated with antibiotics. An increasing antibiotic resistance is well documented and associated with decreasing clinical efficacy, animal welfare and economic consequences. Hence, there is a significant need to encourage prudent use of antibiotics and alternative therapies to antibiotics. The objective of this study was to compare the efficacies of ketoprofen and ceftiofur for the treatment of APM. Between June 2013 and February 2015, a total of 610 dairy cows from 6 farms in Germany were enrolled. Inclusion criteria was a rectal temperature (RT) $\geq 39.5^{\circ}\text{C}$ and a reddish-brown fetid vaginal discharge within the first 10 DIM. Cows meeting the inclusion criteria were randomly allocated to treatment with ketoprofen (3 mg/kg of BW, $n = 300$) or treatment with ceftiofur (1 mg/kg of BW, $n = 310$), both on 3 consecutive days. Rectal temperature was recorded daily for a period of 7 d after enrollment. Cows that showed RT $\geq 39.5^{\circ}\text{C}$ on d 4 to 7 after inclusion received an extended treatment with ceftiofur for 3 (ketoprofen group) or 2 (ceftiofur group) more days. Between 21 and 34 DIM, cows were examined with the Metrichheck device and vaginal discharge was categorized on a 5-point scale according to the presence of pus. A total of 51 cows (34 from ketoprofen group, 17 from ceftiofur group) were excluded from analysis due to concurrent disease ($n = 15$), additional medication of APM ($n = 11$), and due to missing protocol compliance ($n = 25$). Cows of the ketoprofen group (54%) received an extended treatment more often than cows of the ceftiofur group (30%; $P < 0.01$). Occurrence of purulent vaginal discharge was similar for both treatment groups (ketoprofen: 57.8%, ceftiofur: 55.3%, $P = 0.62$). More than half of the cows initially treated with ketoprofen needed ceftiofur treatment. However, there is potential of reducing antibiotic use by utilizing ketoprofen for the treatment of APM as up 46% of cows treated with ketoprofen did not need extended treatment. Animal welfare and economic aspect need to be considered.

Key Words: acute metritis, treatment, antibiotic

21 Intrauterine cephalosporin infusion is associated with better reproduction performance in cows with purulent vaginal discharge and cytological endometritis. José Denis-Robichaud*¹ and Jocelyn Dubuc², ¹*Department of Population Medicine, University of Guelph, Guelph, Ontario, Canada,* ²*Faculté de Médecine Vétérinaire, Université de Montréal, Saint-Hyacinthe, Québec, Canada.*

The objectives of this study were to quantify the effect of an intrauterine infusion of cephalosporin on the reproductive performance at first service of postpartum dairy cows affected by purulent vaginal discharge (PVD) or cytological endometritis (ENDO) using different diagnostic strategies and to determine if the presence of prolonged anovulation would influence the magnitude of treatment benefit. A total of 2,259 Holstein cows in 28 herds were enrolled in a randomized clinical trial. At 35 (± 7) DIM, cows were diagnosed for PVD (purulent vaginal discharge or worse using the metrichheck device) and ENDO ($\geq 6\%$ polymorphonuclear cells using the cytobrush technique or at least small amounts of leukocytes using the leukocyte esterase colorimetric test). Regardless of reproductive tract disease status, cows were randomly assigned to receive an intrauterine cephalosporin infusion or no treatment. Serum progesterone

was measured at 35 and 49 (± 7) DIM (14 d apart); cows were considered to have prolonged anovulation if progesterone was < 1 ng/mL at both times. Statistical analyses were conducted using multivariable mixed logistic regression models adjusted for confounders and herd clustering effect. Intrauterine cephalosporin treatment was associated with an increased first service pregnancy risk in cows diagnosed with PVD (no treatment: 15.4%; treatment: 31.4%; $P < 0.05$) and ENDO (cytobrush: no treatment: 16.2%, treatment: 24.4%, $P < 0.05$; leukocyte esterase: no treatment: 15.8%; treatment: 25.1%, $P < 0.05$), but not in cows unaffected by any form of reproductive tract disease (no treatment: 34.8%; treatment: 32.6%; $P = 0.5$). The effect of cephalosporin treatment in anovular cows (no treatment: 21.0%; treatment: 26.4%; $P = 0.26$) was numerically lower than in cyclic cows (no treatment: 22.7%; treatment: 34.1%; $P < 0.05$). Overall, an intrauterine infusion of cephalosporin improved first service pregnancy risk in cows with postpartum reproductive tract disease and this effect was influenced by postpartum anovulation status.

Key Words: dairy cows, endometritis, treatment

22 Potential role of lactic acid bacteria in the regulation of *Escherichia coli* infection and inflammation of the bovine endometrium. Sandra Genís*¹, Àlex Bach^{1,2}, Francesc Fàbregas¹, Marta Terré¹, and Anna Arís¹, ¹*Department of Ruminant Production, Institut de Recerca i Tecnologia Agroalimentàries (IRTA), Torre Marimon, Caldes de Montbui, Barcelona, Spain,* ²*Institució Catalana de Recerca i Estudis Avançats (ICREA), Barcelona, Spain.*

About 40% of dairy cattle develop uterine disease at postpartum, causing infertility. Some studies indicate that uterine infection, predominantly by *Escherichia coli* in the first week postpartum, is associated with metritis. Metritis is an inflammation of the uterus in which the cow fails to completely clear bacterial contaminants and reducing postpartum inflammatory processes. The aim of this study was to evaluate the potential role of 4 lactic acid bacteria (LAB; *Lactobacillus rhamnosus*, *Pediococcus acidilactici*, *Lactobacillus reuteri*, and *Lactobacillus sakei*) in the modulation of *Escherichia coli* infection and its respective inflammation response by endometrial cells. Primary endometrial epithelial cells were isolated from a fresh endometrium of a healthy cow and cultured at 8×10^4 cells/well in 24-well plates to evaluate the effects of LAB at 3 different doses. Cell extracts were obtained with TriZol. Pro-inflammatory status was assessed by qPCR quantification of IL8, IL1 β , TNF α , and IL6 gene expression. Internalization of *Escherichia coli* was determined by direct enumeration on MacConkey agar plates. Data were normalized and analyzed by an ANOVA. *Lactobacillus sakei* and *Lactobacillus reuteri* had a positive effect preventing *E. coli* infection (87% and 78% respectively, $P < 0.001$) but were associated with a dose-variable effect on tissular inflammation that could further exacerbate the pro-inflammatory status of the endometrium. *Pediococcus acidilactici* clearly decreased ($P < 0.001$) *E. coli* internalization up to an 83% reduction and slightly reduced the inflammation (up to 3.2-fold in IL1 β expression, $P < 0.001$). However, the pro-inflammatory cytokines IL8 and IL1 β decreased significantly ($P < 0.001$) up to 85.1 and 5.2 folds, respectively, in the presence of *L. rhamnosus*. In conclusion, these results demonstrate a clear beneficial effect of *P. acidilactici* and *L. rhamnosus* in the modulation of endometrial infection and inflammation in cattle.

Key Words: *Escherichia coli*, LAB bacteria, metritis

23 Uterine microbiota from calving until establishment of metritis in dairy cows. Soo Jin Jeon^{*2}, Achilles Vieira-Neto¹, Mohanathas Gobikrushanth^{2,6}, Rodolfo Daetz², Rodolfo Mingoti¹, Ana Carolina Parize², Sabrina Freitas², Antonio Nelson da Costa⁵, Rodrigo Bicalho³, Svetlana Lima³, Kwang C. Jeong^{1,4}, and Klíbs N. Galvão², ¹Department of Animal Sciences, University of Florida, Gainesville, FL, ²Department of Large Animal Clinical Sciences, University of Florida, Gainesville, FL, ³Department of Population Medicine and Diagnostic Sciences, Cornell University, Ithaca, NY, ⁴Emerging Pathogens Institute, University of Florida, Gainesville, FL, ⁵Departamento de Zootecnia, Universidade Federal do Ceará, Fortaleza, Ceara, Brazil, ⁶Department of Agricultural, Food and Nutritional Science, University of Alberta, Alberta, Canada.

Objective was to characterize the progression of uterine microbiota from calving until establishment of metritis. Uterine swabs were collected at 0 (20 min from calving), 2, 4, 6, and 8 d postpartum (DPP) from 92 cows. Twelve cows were diagnosed with metritis at 4, 6, or 8 DPP (6 ± 2 DPP), and 12 healthy cows were selected for comparison. Swabs (n = 72) collected at 0, 2, and 6 ± 2 DPP were used for metagenomic sequencing of 16S rRNA gene on the Illumina MiSeq platform. A heat map showed that uterine microbiota was established in cows shortly after calving. The microbiota changed rapidly from 0 to 6 ± 2 DPP by decreasing the abundance of Proteobacteria and increasing the abundance of Bacteroidetes and Fusobacteria. At 6 ± 2 DPP, abundance of Bacteroidetes was significantly higher in metritic cows than healthy cows ($P < 0.01$). Although most genera were shared, healthy and metritic cows could be discriminated based on relative abundance at 0, 2, and 6 ± 2 DPP using discriminant analysis ($P < 0.01$). Also, discriminant analysis showed that *Bacteroides*, *Filifactor*, *Porphyromonas*, *Fusobacterium*, and *Arcanobacterium/Trueperella* were important predictors of metritis. Furthermore, *Bacteroides* and *Fusobacterium* were significantly correlated with uterine discharge score ($r_s = 0.51$ and $r_s = 0.49$, respectively; $P = 0.02$). Likewise, at species level, *Bacteroides heparinolyticus* and *Fusobacterium necrophorum* were the main bacteria for the development of metritis because they were both prevalent (16.8% and 20.2% in metritic cows; 7.0% and 15.8% in healthy cows) and correlated with uterine discharge score ($r_s = 0.42$ and $r_s = 0.42$, respectively; $P \leq 0.05$). In addition, there was a second (*Fusobacterium gonidiaformans*, *Helicoccus ovis*, and *Filifactor villosus*) and third (*Bacteroides pyogenes*, *Porphyromonas levii* and others) line of bacteria that acted synergistically with the main bacteria causing metritis.

Key Words: dairy cow, metritis, 16S metagenomics

25 Association among health and fertility and survival of high-producing dairy cows in three geographic regions of Chile. Pablo Pinedo^{*1,2}, Pedro Melendez³, Sushil Paudyal⁴, Felipe Arias⁵, Ricardo Krauss⁵, Hernando Lopez⁶, Alejandro Luco⁵, and Cristian Vegara^{5,6}, ¹Texas A&M AgriLife Research, Amarillo, TX, ²Department of Veterinary Pathobiology, College of Veterinary Medicine & Biomedical Sciences, Texas A&M University System, College Station, TX, ³Department of Veterinary Medicine and Surgery, College of Veterinary Medicine, University of Missouri-Columbia, Columbia, MO, ⁴West Texas A&M University, Canyon, TX, ⁵ABS Chile Ltda., Santiago, Chile, ⁶ABS Global Inc., DeForest, WI.

The objective was to analyze the association between disease occurrence during early lactation and reproductive performance and survival of dairy cows in high-producing herds, under different management practices in 3 geographic regions of Chile. Data included 30,157 lactation records of

cows calving between January, 2013 and June 2014, maintained under different production systems: Dry lot (DL; n = 3,881 cows in 6 herds); freestall (FS; n = 21,421 cows in 13 herds); grazing (G; n = 1,211 cows in 3 herds); and freestall/grazing (FG; n = 4,244 cows in 5 herds). Cows were also classified according to geographic location: central area (C, n = 6,375 cows); south-central area (SC, n = 17,073 cows); and south area (S, n = 7,309 cows). Logistic regression and ANOVA were used for the analyses (PROC GLIMMIX and PROC GLM, SAS), considering cow as the experimental unit. Covariables offered to the models included lactation number, cow, and herd milk yield. Average milk yield (305 ME) per cow was 11,793, 12,632, and 6,876 kg for C, CS, and S regions, respectively and 10,603, 12,270, 6,899, and 9,261 kg for DL, FS, G, and FG systems. The odds (95% CI) of pregnancy at 150 DIM (P150) and the odds of survival until 150 DIM (S150) for cows that had at least 1 disease event within 50 DIM were 0.82 (0.77–0.88) times the odds of pregnancy and 0.21 (0.17–0.26) times the odds of survival for healthy cows. The odds (95% CI) of P150 for cows under grazing systems were 1.48 (1.24–1.77), 1.07 (0.9–1.28), and 1.29 (1.09–1.52) times the odds of P150 for cows under DL, FG, and F systems, respectively. The odds (95% CI) of S150 for cows under grazing were 5.15 (3.0–8.8), 3.18 (1.88–5.40), and 2.0 (1.19–3.33) times the odds of S150 for cows under DL, FG, and F systems. The odds (95% CI) of P150 and S150 for cows located in the S area were 1.03 (0.95–1.11), and 1.11 (1.03–1.19) times the odds of P150 for cows in C, and SC areas and 1.42 (1.23–1.66), and 1.37 (1.20–1.60) times the odds of S150 for cows in C, and SC areas. Data suggested that cow's health status, geographic location, and production system had a significant association with reproductive performance and survival in this population of Chilean high-producing cows.

Key Words: dairy, health, fertility

26 Bovine viral diarrhea diagnostic testing results in the Intermountain West—Comparison between test methods, age, sex and beef versus dairy breeds. David J. Wilson^{*}, Thomas J. Baldwin, E. Jane Kelly, Arnaud VanWettere, and Gordon Hullinger, *Utah State University, Logan, UT.*

Bovine viral diarrhea (BVD) is an important cause of respiratory, gastrointestinal, and reproductive disease in cattle. The objectives were to calculate prevalence of BVD (“detected” test results) among all bovine samples tested for BVD at the Utah Veterinary Diagnostic Laboratory from 2009 to 2013, and to compare results by sex, age, dairy vs. beef breeds (if provided) of the cattle, and BVD test methods. For necropsied animals, most were tested by Ag capture ELISA, but fetuses were usually tested by PCR. Cattle were mainly from Utah, but also from surrounding states in the Intermountain West of the United States. This was not a planned experiment, but Chi-squared was used to test for significant differences in BVD prevalence between age, sex, breed and test methods. BVD was detected in 105/8,975 samples (1.2%), including 22/180 necropsies (12.2%). Test methods and results are given in Table 1. There were no significant differences in BVD detection by age or sex. Dairy breeds ($P = 0.07$), all necropsied animals, and those tested with PCR were significantly more likely to test as “detected” with BVD. The overall prevalence of >1% and the 8% prevalence in aborted fetuses demonstrates that despite the low reported prevalence of persistently infected cattle, BVD remains an important bovine disease.

Table 1 (Abstr. 26). Results by BVD test method, by age, sex, or breed, and by dairy versus beef breeds

Item	Tested, no.	BVD detected, no. (%)
BVD test method		
Ag Capture ELISA	7,692	79 (1.0)
Serum ELISA	1,195	19 (1.6)
PCR	88	7 (8.0)*
Necropsy (all methods) ¹	180	22 (12.2)*
Age, sex or breed		
Male	215	5 (2.3) ²
Female	382	9 (2.4) ²
Fetus	36	3 (8.3) ^{2,3}
Calf	579	29 (5.0) ^{2,4}
Immature	183	4 (2.2) ^{2,5}
Adult	75	4 (5.3) ^{2,6}
Dairy ⁷	754	25 (3.3) [†]
Beef ⁸	1,600	26 (1.6) [†]

¹Subset of above 3 categories.

²NSD among sex, age, or by test method within sex, age or breed.

³All 3 by PCR at necropsy.

⁴Twenty-seven by Ag Capture ELISA, including 16 of 114 necropsies (14.0%).

⁵Three by serum ELISA, 1 by Ag Capture ELISA, of 6 necropsies (16.7%).

⁶Two by serum ELISA, 2 by Ag Capture ELISA, of 24 necropsies (8.3%).

⁷95.0% Holsteins, 3.8% Jerseys.

⁸60.0% Angus, 22.8% "Beef."

*Higher prevalence of "BVD detected" results, $P < 0.0001$.

[†] $P = 0.07$.

Key Words: BVD, bovine, prevalence

27 Prewaning plane of nutrition and *Mannheimia haemolytica* dose influence inflammatory responses to a combined bovine herpesvirus-1 and *Mannheimia haemolytica* challenge in postweaned Holstein calves. K. P. Sharon^{*1,2}, Y. L. Liang¹, N. C. Burdick Sanchez², J. A. Carroll², P. R. Broadway², and M. A. Ballou¹, ¹Department of Animal and Food Sciences, Texas Tech University, Lubbock, TX, ²USDA-ARS, Livestock Issues Research Unit, Lubbock, TX.

To determine whether previous plane of milk replacer nutrition (PON) and *M. haemolytica* (MH) dose influences inflammatory responses to a combined viral-bacterial respiratory challenge, Holstein calves (1d of age; $n = 30$) were assigned to treatments in a 2×3 factorial with preweaned PON and dose of MH as main effects ($n = 5$ /treatment). Calves were fed either a low (LPN; $n = 15$) or a high PON (HPN; $n = 15$) from birth through weaning. Calves fed the LPN were fed 445 g DM/d of milk replacer until weaning, and HPN calves were fed 830g DM/d of milk replacer from 1 to 10d and 1080g from 11d until weaning. Calf starter and water were offered ad libitum. Calves were step-down weaned beginning at 54d and moved into an enclosed barn at 70d. Indwelling rectal temperature (RT) devices and jugular catheters were inserted at 80d. Calves were challenged with 1.5×10^8 PFU/mL/nostril of bovine herpesvirus-1 (BHV-1) at 81d and with 10^6 , 10^7 , or 10^8 cfu of MH at 84d. Blood samples were collected at varying intervals respective to BHV-1 and MH challenges. There was a PON \times MH interaction ($P < 0.01$) on RT, where calves receiving 10^8 MH had the greatest increase among HPN calves, but the least among LPN calves. Haptoglobin (Hp) concentrations were greater ($P = 0.04$) in LPN vs HPN calves after BHV-1 challenge. There was a time \times MH interaction ($P < 0.01$) for Hp concentrations after the MH challenge, where calves receiving 10^7 MH had the greatest Hp concentrations at 24 h, and calves receiving 10^8 MH the greatest at 144h. During the BHV-1 challenge, neutrophil concentrations (PMN) and change in total leukocytes were greater ($P <$

0.01) among LPN vs HPN calves. After the MH challenge PMN were greatest ($P \leq 0.05$) in calves receiving 10^8 MH at 5 and 24h. During the MH challenge calves receiving 10^8 MH had the greatest ($P < 0.05$) IL-6 and TNF α concentrations. These data demonstrate that LPN calves responded more severely to a combined viral-bacterial respiratory challenge and greater doses of MH increase the acute inflammatory response and prolong inflammation.

Key Words: health, nutrition, respiratory

28 Plasma leptin concentrations are increased during a vaccine-induced acute-phase response in beef cattle. Rodrigo Marques^{*1}, Reinaldo Cooke¹, Murilo Rodrigues¹, Bruno Cappellozza¹, Sergio Arispe², and David Bohnert¹, ¹EOARC, Oregon State University, Burns, OR, ²Malheur County Extension, Oregon State University, Ontario, OR.

The objective of this experiment was to evaluate if a vaccine-induced acute-phase reaction also results in increased plasma leptin concentration, which would explain a potential DMI decrease in vaccinated beef cattle. Eighteen yearling Angus \times Hereford heifers were ranked by BW and allocated to 2 groups, which were randomly assigned to a crossover design containing 2 periods of 7 d and the following treatments on d 0 of each period: (1) vaccination against *Mannheimia haemolytica*, bovine rhinotracheitis virus, and bovine viral diarrhea Types 1 and 2 viruses (VAC; BoviShield One Shot; Zoetis, Florham Park, NJ), or (2) saline-injected control (CON). Heifers were maintained in individual pens, offered grass hay for ad libitum consumption, in addition to 3.5 kg/d (DM basis) of a corn-based supplement. During Period 1, hay and concentrate intake were evaluated daily. During Period 2, blood samples were collected before (-2 and 0 h) and at 2, 4, 6, 8, 12, 16, 24, 36, 48, 60, 72, 96, 120, 144, and 168 h after treatment administration. All samples were analyzed for plasma haptoglobin concentration. Samples collected from 0 to 48 h were also analyzed for plasma cortisol, insulin and leptin concentrations. Plasma variables were analyzed using the mean result from samples collected at -2 and 0 h as covariate. Treatment \times day interactions were detected ($P < 0.01$) for hay and total DMI, given that these parameters were reduced ($P \leq 0.05$) in VAC compared with CON on d 0 and 1. Treatment \times hour interactions were detected ($P \leq 0.02$) for all plasma variables. Plasma cortisol concentrations were greater ($P \leq 0.05$) in VAC compared with CON from 2 to 16 h. Plasma insulin concentrations were greater ($P \leq 0.05$) in VAC compared with CON from 4 and 16 h. Plasma leptin concentrations were greater ($P \leq 0.03$) in VAC compared with CON from 6 to 16 h. Plasma haptoglobin concentrations were greater ($P \leq 0.02$) in VAC compared with CON from 16 to 120 h. In conclusion, plasma leptin concentration was increased during a vaccine-induced acute-phase reaction, which may explain the decrease in DMI observed herein in vaccinated cattle.

Key Words: cattle, acute-phase reaction, leptin

29 Effect of an early-life LPS challenge on a subsequent LPS challenge in Holstein bull calves. Aimee L. Benjamin^{*1}, Filiz T. Korkmaz¹, Theodore Elsasser², and David E. Kerr¹, ¹University of Vermont, Burlington, VT, ²USDA-ARS, Beltsville, MD.

The variable innate immune response between animals can be due to a variety of causes. Infections during the neonatal period could affect an animal's innate response phenotype. Ten Holstein calves were used to investigate the sustained effects of neonatal LPS exposure on the innate immune system. At 8d of age, 5 calves received an IV infusion of LPS (0.5 μ g/kg), while the remaining 5 received saline. A subsequent dose of

LPS (0.25 µg/kg) was given to all 10 calves at 30 d of age to determine if there was any lasting effects from the first LPS challenge. Skin biopsies were collected from all calves at 22 d of age to isolate dermal fibroblasts for a cell model to explore genetic and/or epigenetic changes that may have resulted from the early life LPS treatment. Two hours after the first challenge, LPS-treated calves had greater ($P < 0.05$) plasma IL-6 (14.4 ± 2.8 vs. 0.6 ± 0.3 ng/mL) and TNF-α (3.2 ± 1.0 vs. 0.2 ± 0.1 ng/mL) compared to calves that had received saline, respectively. Response curves of these plasma cytokines from measurements at 0, 1, 2, 3, 5, and 7 h following the second LPS challenge were similar ($P > 0.05$) between groups. Rectal temperatures were not significantly different between the treatment groups in either challenge. Dermal fibroblasts isolated from LPS- or saline-treated calves produced similar ($P > 0.05$) levels of IL-6 (318.4 ± 96.8 vs. 359.3 ± 54.6 pg/mL) and IL-8 (183.6 ± 33.8 vs. 157.1 ± 11.7 pg/mL), respectively, in response to a 24h LPS challenge (500 ng/mL). Although the fibroblasts produced much greater levels of IL-6 (5178.6 ± 637.9 vs. 5353.4 ± 853.1 pg/mL) and IL-8 (1554.2 ± 96.8 vs. 1596.9 ± 93.4 pg/mL), respectively, in response to a 24h challenge with IL-1b (10 ng/mL), there was no early-life treatment effect. There was substantial inter-animal variation in clinical symptoms following the LPS challenges, but the early-life exposure to LPS at 8d of age did not clearly influence the response to a subsequent LPS challenge in 30-d-old calves, or affect the ability of fibroblasts to respond in vitro to LPS or IL-1b. More work is needed to determine how early-life exposure to infection may influence an animal's innate immune response.

Key Words: neonatal, variation

30 ACTH-test reactivity affect disposition for storage of fat depots in dairy cows during the transition period. Lena Ruda¹, Claudia Raschka¹, Lea Fieguth¹, Asako Kinoshita¹, Anja Schacht¹, Marion Piechotta¹, Korinna Huber², Akos Kenez², Ulrich Meyer³, Sven Dänicke³, and Juergen Rehage^{*1}, ¹*Clinic for Cattle, University of Veterinary Medicine Hannover, Hannover, Germany*, ²*Department of Physiology, University of Veterinary Medicine Hannover, Hannover, Germany*, ³*Institute of Animal Nutrition, Friedrich-Loeffler-Institute, Braunschweig, Germany*.

It is generally recommended to adjust feeding requirements to avoid overconditioning of dairy cows at calving because high body condition is a major risk factor for ketosis in early lactation. However, besides feeding and environmental factors individual disposition for high body condition appears to have also a genetic and epigenetic background. The individual reactivity of the hypothalamic-pituitary-adrenal axis (HPA-axis) and the release of cortisol as tested in the ACTH-test varies considerably between cows. Thus, the aim of the study was to test the relationship between HPA-axis reactivity and body condition in dairy cows during the transition period. In 22 pluriparous German HF dairy cows, kept in freestalls with cubicles and fed a TMR based on grass and corn silage and concentrate, an ACTH challenge was performed on d 100 postpartum (pp). Before and after injection of ACTH (80µg) cortisol concentrations were measured in blood samples taken in short-term intervals from indwelling jugular vein catheters from which baseline and peak concentrations and AUC were assessed. Results were used in a regression analysis (SAS statistical package) and correlated with sonographically estimated subcutaneous fat mass (SC fat) assessed at d -42 before parturition, and at d 1, d 21, d 100 pp. Cows were also classified

according the AUC cortisol results into high (HR; n = 7), intermediate (IR; n = 8), and low (LR; n = 7) responder and fat mass was tested for differences by ANOVA. Significant linear negative correlations were found between AUC and peak cortisol concentrations from ACTH challenge and SC fat at d -42, d 1, d 21 and d 100. In HR compared with LR cows in average SC fat was significantly less at d -42, d 1, d 21 and d 100 and gain before and loss of SC fat after parturition was lower. Cows with high reactivity of the HPA-axis are leaner during the whole transition period, gain less before and lose less SC fat after parturition. Dynamics in body condition appears to be closely correlated with HPA axis reactivity during the transition period in dairy cows. This work was funded by the German Research Foundation.

Key Words: ACTH-test, BCS, dairy cows

31 Use of an ex vivo/in vitro laminitis model to elucidate the role of endotoxins during equine and bovine laminitis. Nicole Reisinger^{*1}, Simone Schaumberger², and Gerd Schatzmayr¹, ¹*Biomim Research Center, Tulln, Austria*, ²*Biomim Holding GmbH, Herzogenburg, Austria*.

Laminitis is one of the most common causes for lameness in horses and ruminants. The pathology of laminitis is still not fully understood. As it is a multifactorial disease, several substances and toxins such as endotoxins are discussed as possible trigger factors. The aim of our study was to test the influence of endotoxins on the lamellar integrity of hoof explants. Furthermore, the potential difference of effects of endotoxins on the lamellar integrity of hooves from horses and ruminants was investigated. Explants from hooves (n = 3) and claws (n = 3) were cultivated at 37°C and 5% CO₂ with D-MEM as culture medium. Lipopolysaccharides (LPS) of *Escherichia coli* O55:B5 were added to the equine [0, 2.5, 10, 100 µg/mL] and bovine [0, 1, 10, 100 µg/mL] explants for 24 h. After incubation, explants were tested for their integrity by measuring the force [Newton], which is needed to separate the explants (= separation force). Therefore, the explants were connected to a force transducer with clamps. Viability of explants was tested with the water-soluble tetrazolium (WST-1) assay. There was no effect on separation force when equine explants were incubated with 2.5 µg/mL LPS. Separation force of equine explants incubated with 10 and 100 µg/mL LPS was significantly decreased ($P < 0.005$) by 45% and 49%, respectively, compared with control explants. Similar to equine explants, there was no effect on separation force when bovine explants were incubated with 1 µg/mL LPS. Separation force of bovine explants incubated with 10 and 100 µg/mL LPS was significantly decreased ($P < 0.005$) compared with control explants. A reduction by 50% and 65% for LPS was observed, respectively. All explants were viable after 24 h incubation. In our study a concentration dependent reduction of separation force of explants incubated with LPS was observed. Similar effects of LPS were observed in explants from both species. Although laminitis has never been induced by endotoxins alone in animal experiments, our data suggest that endotoxins might play an important role during the onset of laminitis. The presented model could be used to test other potential trigger factors of laminitis and the interaction between these factors.

Key Words: laminitis, endotoxins, in vitro