

Late-Breaking Abstracts

LB1 Evidence for the presence of an autoimmune component to the chronic muscle wasting disease characteristic of calves infected with *Sarcocystis cruzi*. T. H. Elsasser*¹, S. Kahl¹, R. Fayer², and E. E. Connor¹, ¹USDA-ARS, Bovine Functional Genomics Laboratory, Beltsville, MD, ²USDA-ARS, Environmental Microbial and Food Safety Laboratory, Beltsville, MD.

Sarcocystis spp. infection often resolves in a progressive decline in muscle integrity. Causes for this have remained unresolved. Previously, we described the proinflammatory muscle protein nitration (PMPN) in calves (*ScI*) chronically infected with *Sarcocystis cruzi*. Knowing that instances of PMPN are associated with autoimmune muscular atrophy disorders in human, we hypothesized that calves infected with *S. cruzi* might develop autoantibodies consistent with the definition of autoimmune disease. To test this, 3-mo-old Holstein steers (n = 7) were infected with 250,000 *Sc* oocysts; plasma was collected before (d 0) and after infection (d 55) along with contemporary plasmas from non-infected steers (CTL, n = 3). Steers were euthanized on d 55. Samples of semitendinosus muscle were collected as a target for autoantibody reactivity (frozen sections, 6 µm) or homogenate preparation for activity measurement of the proinflammatory marker xanthine oxidase (XO, µU/mg of protein). For immunofluorescence (IF) detection, slide-mounted serial sections were fixed, permeabilized, blocked, and incubated (12 h, 4°C) with each animal's own d-0 and d-55 plasma (1:80, 1:160, 1:320 in PBS); autoantibody binding was visualized with Alexa₄₈₈-rabbit anti-bovine IgG. Mean (LSM ± SE) d-55 XO activity was 23.5 versus 129.7 (±17, $P < 0.001$) in CTL and *ScI*, respectively. The presence of plasma autoantibodies against CTL muscle was not evident with d-0-CTL, d-55-CTL, or d-0-*ScI* plasmas. All muscle samples from *ScI* contained cysts. Autoimmunoreactivity of *ScI* plasma was evident: 7/7 calves at 1:80, 4/7 at 1:160, and 2/7 at 1:320. The IF intensity declined across muscle subcomponents where the signal with cysts > smooth muscle > vascular endothelium > intrafiber proteins but co-localized prominently in vascular tissue with Complement C9. Only plasma from *ScI* had anti-nuclear antibodies when incubated with fixed cultured bovine epithelial cells and this signal declined ~70% with DNase pretreatment. The data are consistent with autoantibody-mediated tissue pathology affecting animal health in this disease paradigm.

Key Words: autoimmune disease, muscle, parasitic infection

LB2 Maternal nutrient restriction followed by realimentation during early to mid-gestation on hepatic steroid inactivation in beef cows. C. G. Hart*¹, L. E. Camacho², K. C. Swanson², K. A. Vonnahme², and C. O. Lemley¹ ¹Mississippi State University, Mississippi State, ²North Dakota State University, Fargo.

The objective was to examine the effect of early to mid-gestation maternal nutrient restriction followed by realimentation on hepatic steroid inactivation. On d 30 of pregnancy, cows were assigned to dietary treatments: control (C; 100% NRC; n = 18) and restricted (R; 60% NRC; n = 34). On d 85, cows were slaughtered (C, n = 6 and R, n = 6), remained on control (CC; n = 12) and restricted (RR; n = 12), or were realimented to control (RC; n = 11). On d 140, cows were slaughtered (CC, n = 6; RR, n = 6; RC, n = 5), remained on control (CCC, n = 6; RCC, n = 5), or were realimented to control (RRC, n = 6). On d 254, all remaining cows were slaughtered. Jugular blood samples were collected prior to slaughter. At slaughter, maternal liver samples were frozen for

later determination of cytochrome P450 3A (CYP3A), 2C (CYP2C) and uridine diphosphate-glucuronosyltransferase (UGT) activities via luminogenic substrates. Serum concentrations of estradiol-17β (E2) and progesterone (P4) were measured via RIA. Gestational day effects were tested among C, CC, and CCC cows, and dietary treatment effects were tested within a given slaughter day. Data were analyzed using the mixed procedure of SAS. CYP3A activity tended to decrease ($P < 0.09$) on d 254 vs. d 85 and d 140 and did not differ across dietary treatments ($P > 0.25$). CYP2C activity tended to decrease ($P < 0.09$) in R vs. C and did not differ across gestational days ($P > 0.30$). UGT activity did not differ across gestational days ($P > 0.30$). UGT activity was increased ($P = 0.05$) in R vs. C and decreased ($P < 0.05$) in RR vs. RC and CC. Concentrations of E2 increased ($P < 0.01$) on d 254 vs. d 85 and d 140. On d 85, E2 concentrations were increased ($P < 0.05$) in R vs. C. On d 140, P4 concentrations were increased ($P < 0.05$) in RR vs. RC and CC. Concomitant changes occurred in hepatic CYP2C and CYP3A activities with serum concentrations of E2. An increase in P4 concentrations on d 140 was accompanied by a decrease in hepatic UGT activity in nutrient restricted cows. The current study highlights the relevance of steroid inactivation in modulating serum concentrations of E2 and P4 during pregnancy.

Key Words: cow, pregnancy, steroid

LB3 Zilpaterol hydrochloride increases carcass yield of fed steers by altering tissue proportions. T. J. McEvers*¹, N. D. May¹, J. A. Reed¹, L. J. Walter¹, J. P. Hutcheson², and T. E. Lawrence¹ ¹West Texas A&M University, Canyon, ²Merck Animal Health, Summit, NJ.

A randomized controlled study was conducted to understand body mass changes leading to increased carcass yield of calf-fed Holstein steers supplemented zilpaterol hydrochloride (ZH) as compared to those not supplemented ZH (CON). Cattle (ZH = 55; CON = 55) were harvested every 28 d (5 animals per diet per harvest) over a 308-d period. Non-carcass components [blood, hide, ears, nose pad, lips, tail, head, tongue, metatarsals/metacarpals, penis, esophagus, heart, lungs, trachea, liver, kidneys, KPH, pancreas, thymus gland, spleen, gastrointestinal tract, and digesta] were collected and weighed to the nearest 0.09 kg. Components were calculated as a percentage of empty body weight (EBW) using a mixed model with fixed effects of ZH treatment and days on feed and the random effect of pen. Cattle fed ZH had greater EBW than CON steers ($P = 0.02$; 623.1 vs. 605.4 kg). Comparison of external noncarcass components indicated cattle supplemented ZH had less hide ($P < 0.01$; 8.14 vs. 8.48%) than CON steers. Internal noncarcass components also differed; cattle fed ZH had less liver ($P < 0.01$; 1.37 vs. 1.48%), kidney ($P < 0.01$; 0.24 vs. 0.27%), and empty gastrointestinal tract ($P < 0.01$; 11.84 vs. 12.45%) than CON steers. Cattle fed ZH had greater hot carcass weight (HCW; $P < 0.01$; 431.6 vs. 410.1 kg). Reduction of external and internal noncarcass components coupled with increased HCW resulted in greater dressed carcass yield as a percentage of EBW ($P < 0.01$; 69.11 vs. 67.65%). These results indicate that cattle fed ZH experience reductions in noncarcass components totaling 1.09% of EBW. This information becomes useful in describing the repartitioning effects of ZH, which increased dressed yield as a percentage of EBW by 1.46%. We postulate that the reduction in noncarcass components explains 74.66% of the increase in carcass yield of calf-fed Holstein steers.

Key Words: beef, carcass transfer, zilpaterol

LB4 Effects of glucuronic acid and *N*-acetyl-D-glucosamine on the in vitro fertilization of porcine oocytes. K. Schmidt*, K. Dalton, C. Durfey, K. Lemon, A. Mello, and B. D. Whitaker, *University of Findlay, Findlay, OH*.

Pig oocytes fertilized in vitro experience high polyspermic penetration rates due to inadequate cortical granule exocytosis. The objective was to minimize polyspermic penetration by increasing the perivitelline space (PVS) thickness through supplementation of its components, glucuronic acid (GA) and *N*-acetyl-D-glucosamine (GlcNAc) during maturation. Oocytes ($n = 1,000$) were supplemented during the last 24 h of maturation with either 0.01 mM GA, 0.01 mM GlcNAc, 0.01 mM GA and GlcNAc, or 0.005 mM GA and GlcNAc and then evaluated for zona pellucida and PVS thickness. Intracellular glutathione concentrations were determined after maturation in a portion of the oocytes ($n = 300$) and the remaining oocytes were fertilized. At 12 h post-fertilization oocytes ($n = 300$) were evaluated for fertilization and cortical granule characteristics and the remaining putative embryos were cultured and evaluated for cleavage and blastocyst formation at 48 h and 144 h postfertilization. There were no significant differences between the treatments for zona pellucida thickness, intracellular glutathione concentrations, penetration rates, or male pronuclear formation. The PVS thickness was significantly thicker ($P < 0.05$) in all treatments compared to the control ($4.45 \pm 0.71 \mu\text{m}$). Oocytes supplemented with 0.01 mM GA ($9.62 \pm 0.87 \mu\text{m}$) or 0.01 mM GA and GlcNAc ($9.65 \pm 0.91 \mu\text{m}$) had significantly thicker PVS ($P < 0.05$) than 0.01 mM GlcNAc ($7.57 \pm 1.16 \mu\text{m}$) or 0.005 mM GA and GlcNAc supplemented oocytes ($5.99 \pm 0.82 \mu\text{m}$). Oocytes supplemented with 0.01 mM GA or 0.01 mM GA and GlcNAc had significantly more cortical granule exocytosis ($P < 0.05$) compared with the other treatments. Oocytes supplemented with GA had significantly lower incidences ($P < 0.05$) of polyspermic penetration compared with the control ($32.00 \pm 4.80\%$) or 0.01 mM GlcNAc ($40.00 \pm 7.93\%$) and significantly higher rates ($P < 0.05$) of cleavage and blastocyst formation by 48 and 144 h postfertilization. These observations indicate that supplementing GA during oocyte maturation decreases polyspermic penetration by increasing PVS thickness and cortical granule exocytosis in pigs.

Key Words: cortical granules, polyspermy, pig

LB5 Direct use of MACE EBV in the Walloon single-step Bayesian genomic evaluation system. J. Vandenplas^{1,2}, F. Colinet¹, P. Faux¹, S. Vanderick¹, and N. Gengler*¹ ¹*ULg-GxABT, Gembloux, Belgium*, ²*FNRS, Brussels, Belgium*.

Single-step genomic evaluations (ssGBLUP) should reduce potential biases in the estimation of genomically enhanced breeding values (GEBV) by the simultaneous combination of genomic, pedigree and phenotypic information, also because fewer approximations are made than in multi-step methods. However, most current genomic evaluation systems are multi-step, relying heavily on the use of multiple across-country evaluation (MACE) results as the primary source of foreign phenotypic information. Recently a need was identified to develop direct use of MACE estimated breeding values (MACE EBV) in ssGBLUP. Therefore, the aim of this report is to show the development and practical use of an innovative method that considers simultaneously all available genotype, pedigree, local, and foreign information in a genomic evaluation system. The developed method is a Bayesian approach associated with ssGBLUP. The method allows a correct propagation of information and avoids multiple considerations of contributions. It also allows adding and subtracting contributions from different information sources; for example, to avoid double counting of local information already contributing to MACE EBV. Another advantage of this Bayesian

approach is that it creates a model equivalent to a complete one directly combining all available information without any additional deregression steps. The approach was set up using 27,376 Holstein animals, 11,550 with a Walloon EBV and 1,345 bulls with MACE EBV. A total of 1,351 cows and bulls contributed to the genomic relationship matrix that was combined with the pedigree-based numerator relationship matrix. Phenotypic information was added through Walloon EBV and MACE EBV. Local information also included in MACE EBV was discounted for by subtracting in the Bayesian integration process the local EBV sent to Interbull. This genomic evaluation system passed the Interbull GEBV tests for milk, fat and protein yields and the majority of type traits in February 2013. This approach has the potential to improve current genomic prediction strategies also in beef and in other species (e.g., swine).

Key Words: Bayesian integration, MACE, genomic prediction

LB6 Genetic mechanisms that contribute to differences in beef tenderness following electrical stimulation. R. N. Vaughn*, A. K. Torres, K. J. Kochan, R. K. Miller, C. A. Gill, A. D. Herring, D. G. Riley, J. O. Sanders, J. W. Savell, T. H. Welsh, and P. K. Riggs, *Texas A&M University, College Station*.

Beef tenderness is valued by consumers and influenced by both environmental and genetic factors. Postmortem treatment by electrical stimulation (ES) increases tenderness and reduces, but does not eliminate, variation among carcasses. The purpose of this study was to examine genetic factors that influence tenderness, particularly in post-ES beef. Skeletal muscle samples were collected immediately after slaughter from crossbred F₂ Nellore-Angus steers. Warner-Bratzler shear force (WBSF) measurements following 14 d of aging were used as an objective measure of tenderness. Microarray analysis of samples from 48 steers, chosen for divergent response to ES, was used to identify networks of genes with significantly different gene expression between tenderness groups, and to identify significantly enriched signaling pathways. In addition, SNP haplotype blocks encompassing genes of interest were constructed to examine parent and breed of origin effects. The extracellular matrix (ECM) and focal adhesion pathways were enriched in the microarray assay. From this pathway, a total of 40 genes were assayed by qRT-PCR. Several genes in the integrin family were upregulated in the group that responded well to ES compared with the group that responded poorly. Through haplotype analysis, we found that breed and parent of origin had an effect on tenderness. Breed of origin of integrin alpha-6 (*ITGA6*) corresponded to a 0.15-kg difference in ES residual tenderness values when inherited maternally ($P = 0.03$). The gene fibronectin 1 (*FNI*) had a difference in ES residual tenderness of 0.23 kg for different paternally inherited haplotypes ($P < 0.01$). Also, *ITGA6* protein expression was closely related to mRNA expression in the subset analyzed by Western blot (gene and protein expression levels were both 1.8-fold higher in the tender group than the low; $P = 0.04$ and $P = 0.02$ respectively). This approach identified a network and biological mechanism associated with tenderness not previously established. These results also suggest that targeting components of the ECM represents a novel area of research for improving tenderness.

Key Words: tenderness, beef, electrical stimulation

LB7 A new inline device for predicting individual cow somatic cell count using ATP measurement technology. B. W. Woodward*¹, A. J. Seykora², and T. Koopman³, ¹*NextGen, Lawrenceville, GA*, ²*University of Minnesota, St. Paul*, ³*Isogen Animal Care, De Meern, the Netherlands*.

The occurrence and treatment of mastitis in US dairy herds is reported to cost as much as \$2 billion annually. Part of the challenge in managing this disease is the difficulty in diagnosing those cows with subclinical mastitis. This difficulty stems from the lack of availability, speed, cost, and/or adoption of equipment to individually measure somatic cell count (SCC) for cows at the time of milking, especially during the transition period when the incidence of mastitis is at its highest. Mastiline is a new inline device that provides SCC values by sampling milk from each individual cow directly from the flow before it reaches the main milk line. Approximately 120 μ L of milk is drawn into the device where proven ATP measurement technology is used to measure the number of live somatic cells in less than 2.5 min, in contrast to the Fossomatic method that measures live and dead cells from samples sent to laboratories. Considerable research over the last 20 yr supports ATP measurement as a reliable indicator of SCC in milk. Inside the device, the milk sample is mixed with a specially designed nontoxic reagent that breaks open the somatic cells to release the ATP, which is then measured and the results converted into SCC. Initial laboratory testing of the Mastiline device involved spiking full-fat samples taken from retail pasteurized milk with 50,000, 200,000, and 500,000 cells/mL, as well as raw milk samples from individual cows. All samples were measured by the Mastiline device 3 or 4 times. The coefficient of variation (CV) for SCC was $\leq 9.32\%$ for each retail and raw milk sample repeatedly measured by the Mastiline device. The CV for retail and raw milk samples with Mastiline SCC between 100,000 and 500,000 cells/mL was $\leq 4.30\%$. The raw milk samples were also measured via Fossomatic. Regressing the Fossomatic and spiked SCC values on the Mastiline SCC values yielded a regression value of 1.22 ± 0.06 and an R^2 value of 0.95. Additional data are currently being collected on commercial European dairy farms, and a summary of these data will be presented.

Key Words: ATP, subclinical mastitis, somatic cell count

LB8 Conjoint regulation of glucagon secretion via insulin and glucose plasma concentrations in dairy cows M. Zarrin^{*1,2}, O. Wellnitz¹, H. A. van Dorland¹, and R. M. Bruckmaier¹, ¹*Veterinary*

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Insulin and glucagon are glucoregulatory hormones that contribute to glucose homeostasis. Insulin is generally accepted as a suppressor of glucagon secretion, whereas the effects of insulin in interaction with glucose concentration on glucagon release have not been shown in dairy cows, to our knowledge. The objective was to investigate effects of long-term (48 h) elevated insulin concentrations on glucagon secretion during simultaneous hypoglycemia or euglycemia in mid-lactating dairy cows. Dairy cows were randomly assigned to 3 treatment groups: (1) an intravenous insulin infusion (HypoG, $n = 5$) to decrease plasma glucose concentration (2.5 mmol/L), (2) a hyperinsulinemic euglycemic clamp to maintain plasma glucose concentration at pre-infusion level to study effects of insulin at simultaneously normal glucose concentration (EuG, $n = 6$), and (3) a 0.9% saline solution infusion (Control, $n = 8$). Glucose was measured in blood at 5-min intervals to allow adjustments of glucose infusion rate. Insulin and glucagon blood concentrations were analyzed in hourly samples. Area under the curve was evaluated by ANOVA with treatment as fixed effect. Data are presented as means \pm SEM. Insulin infusion (0.6 mU/kg/min) increased plasma insulin concentration in HypoG (41.9 ± 8.1 mU/L), and EuG (57.8 ± 7.8 mU/L) compared with the control group (13.9 ± 1.1 mU/L; $P < 0.01$). Plasma glucose concentration decreased to 2.25 ± 0.1 mmol/L in HypoG ($P < 0.01$) and remained unchanged in EuG (3.8 ± 0.2 mmol/L) and control group (4.1 ± 0.1). Plasma glucagon concentration decreased in EuG (88.0 ± 20.0 pg/mL; $P < 0.05$) and increased in HypoG (133.6 ± 5.6 pg/mL; $P < 0.01$) compared with the control group (108.4 ± 4.2 pg/mL). The results show that intravenous insulin infusion induces glucagon secretion during hypoglycemia, while the same insulin infusion reduces glucagon secretion at simultaneously normal glucose concentration. These results are in contrast to previous reports that suggested a general inhibitory effect of insulin on glucagon secretion, and it can be assumed that both insulin and glucose are conjoint regulatory factors of glucagon secretion in dairy cows.

Key Words: glucagon, insulin, glucose