

## Physiology and Endocrinology: Endocrinology/Metabolic Physiology

**W146 Glucose-dependent insulin response in dairy cows within segregating family structure is related to milk yield.** H. M. Hammon\*, O. Bellmann, J. Voigt, F. Schneider, and C. Kühn, *Research Institute for the Biology of Farm Animals (FBN), Dummerstorf, Germany.*

An experiment has been initiated using segregating F<sub>2</sub> offsprings from crosses of Charolais bulls and German Holstein cows to evaluate properties of nutrient transformation in cattle with respect to secretion type (German Holstein) and accretion type (Charolais). In 51 dairy cows of 5 F<sub>2</sub> families glucose-dependent insulin response was investigated during first lactation to test the hypotheses that insulin response segregates within the F<sub>2</sub> population and that the level of milk yield affect insulin response after glucose challenge. Cows received intravenous glucose infusions (1 g/kg BW<sup>0.75</sup>) 10 d before and 30 and 100 d after parturition. Blood samples were taken before and 7, 14, 21 and 28 min after glucose challenge, glucose (whole blood) and insulin plasma concentrations were measured, and areas under the concentration curve were calculated for glucose (AUC<sub>gluc</sub>) and insulin (AUC<sub>ins</sub>). The GLM and CORR procedures of SAS were used to evaluate data. Milk yield was low, ranging from 5.3 to 13.6 kg on d 30 and 0.4 to 10.4 kg on d 100 of lactation among F<sub>2</sub> families and showed significant differences among families. After glucose challenge glucose concentrations increased in all cows, but AUC<sub>gluc</sub> did not differ among families at any d. Insulin concentrations also increased after glucose challenge in all cows, showed no between-family differences for AUC<sub>ins</sub> before parturition, but AUC<sub>ins</sub> differed (P < 0.05) among groups on d 30 and 100 of lactation. Calculated correlations revealed a significant negative relationship between milk yield and AUC<sub>ins</sub> on d 30 and 100 of lactation. In conclusion, milk production of F<sub>2</sub> Charolais × German Holstein cross bred dairy cows was on a low level, but differed among families. Glucose-dependent insulin response also differed among families during lactation, but not during dry period, and was inversely related to milk production. This suggests that genetic capacity for milk production is related to peripheral insulin-dependent glucose metabolism in cows.

**Key Words:** Cow, Glucose metabolism, Cross breeding

**W147 Characterization of metabolic hormones and insulin sensitivity in transition dairy cows.** C. C. Stanley\*, C. C. Williams, D. T. Gantt, J. C. Roberts, and S. R. Adams, *LSU Agricultural Center, Baton Rouge.*

A study was conducted to characterize insulin sensitivity at the end of gestation and in early lactation by using the frequently sampled glucose tolerance test (FSIGT), insulin tolerance test (ITT), and hyperinsulinemic euglycemic clamp test (EC). The FSIGT with minimal model computer analysis (MM), the EC, and the ITT were performed in 5 Holstein cows at 7-10 days prior to anticipated calving date and again at 7-10 days after calving. During the FSIGT glucose was administered (0.3g/kg BW) followed 20 min later by insulin administration (0.03 IU/kg BW) through a jugular catheter. Blood samples were collected relative to glucose administration for a 3 hr period for measurement of plasma glucose and insulin concentrations which were used in the MM to determine glucose effectiveness (S<sub>G</sub>), insulin sensitivity (S<sub>I</sub>), and the acute insulin response relative to glucose administration (AIR<sub>G</sub>). The EC used a variable rate of glucose infusion to achieve euglycemia while infusing insulin at 6 mU/ kg •min<sup>-1</sup>. Blood was collected every 5 min, and glucose concentrations

were measured using a handheld glucometer. Glucose disposal rate (GDR) was calculated from the glucose infusion rate after euglycemia was achieved. The GDR was converted to S<sub>I</sub> by factoring the change in plasma insulin from basal to hyperinsulinemia and plasma glucose at euglycemia. Plasma was analyzed for glucagon, insulin, and glucose concentrations during the basal and steady states. The ITT consisted of bovine insulin administration (0.1 IU/kg of BW) through a jugular catheter. Samples were collected for plasma leptin, glucose, glucagon, and insulin analyses during the 3 hour test. There was a main effect of week for mean plasma glucagon concentrations (P < 0.05) with an increase in basal concentrations after calving. There were no effects (P > 0.05) of week for mean basal plasma insulin, glucose, or leptin concentrations or for GDR, S<sub>I</sub> EC, S<sub>I</sub> MM, S<sub>G</sub>, or AIR<sub>G</sub>. All tests reflected a numerical increase in insulin sensitivity after calving, although the responses indicated low insulin sensitivity both pre- and postpartum.

**Key Words:** Transition cows, Metabolic hormones, Insulin sensitivity

**W148 Bovine somatotropin and dietary fat enriched with omega-3 fatty acids in dairy cows: III. Postpartum ovarian activity.** M. Carriquiry\*, C. R. Dahlen, W. J. Weber, G. C. Lamb, and B. A. Crooker, *University of Minnesota, St. Paul.*

Multiparous cows (n=59) were blocked by expected calving date and previous 305ME and assigned randomly to a 2x2 factorial design to determine effects of bST (POSILAC<sup>®</sup>) and dietary fat on ovarian activity during the first 90 DIM. Isocaloric diets (1.98 Mcal NEL<sub>1x</sub>) that included whole, high-oil sunflower seeds (10% of dietary DM, SS) or a mixture of Alifet-High Energy<sup>®</sup> and Alifet-Repro<sup>®</sup> (3.4 and 1.5% of dietary DM, AF) were provided from calving. Cows received 0 or 500 mg bST (N, Y) every 10 d from 12 to 70 DIM and at 14 d-intervals thereafter. Follicular dynamics, luteal growth and development (transrectal ultrasonography) and plasma P4 concentrations were evaluated (3X/wk) from 15 to 90 DIM. Breeding started after 90 DIM. Means differed when P<0.05. Days to first ovulation (33.6 ± 1.4) and incidence of anovulation at 45 or 70 DIM did not differ among treatments but time to first ovulation was less variable for AFY. Intraovulatory intervals were similar among treatments (22.1 ± 0.9 d). Incidence of 2-wave cycles was greater for SSY (71.0%) and AFN (80.0%) but more 3-wave cycles occurred with AFY (83.3%). Growth rate of the ovulatory follicle was greater for AF than SS (1.9 vs 2.2 ± 0.11 mm/d) and diameter of ovulatory follicles was larger for AFN than the other treatments (17.9 vs 15.7 ± 0.7 mm). Multiple ovulations tended (P<0.10) to increase with bST. Maximal CL volume did not differ among treatments. Area under the P4 curve was reduced for SSY (63.2<sup>a</sup>, 48.1<sup>b</sup>, 55.5<sup>ab</sup>, and 61.4<sup>a</sup> ± 5.1 dxng/mL for SSN, SSY, AFN, and AFY). The number of class 1 (2 to 5 mm; decreased), class 2 (6 to 9 mm; increased), and class 3 (10 to 15 mm; tended to increase (P=0.07)) follicles were altered by bST. Class 2 follicles were reduced with AF. Neither diet nor bST affected the number of class 4 (>15 mm) or the mean size of class 2, class 3, and class 4 follicles. Initiation of bST at 12 DIM and dietary omega-3 fatty acids enhanced ovarian activity during the first 90 DIM and could benefit reproductive performance.

**Key Words:** bST, Omega-3 fatty acids, Follicular dynamics

**W149 The ontogeny of insulin-like growth factor-I (IGF-I) modulation of GH secretion from porcine anterior pituitary cells in culture.** C. R. Barb\* and G. J. Hausman, *USDA, ARS, Russell Research Center, Athens, GA.*

The ontogeny of IGF-I modulation of GH secretion from the anterior pituitary was studied. Anterior pituitary cells collected from 110 day old fetuses (n=4), 2- (n=7), 4- (n=8) and 6- (n=7) month old gilts were studied in primary culture. On day 4 of culture, 100,000 cells/well were challenged with 0.1, 10 or 1000 nM [Ala15]-hGRF-(1-29)NH<sub>2</sub>, or 0.01, 0.1, 1, 10, 30 nM IGF-I alone or in combinations with 1000 nM GRF. Secreted GH was measured at 4 h after treatment. Basal (control) GH secretion was 267 ± 46, 76 ± 8, 39 ± 3 and 40 ± 7 ng/well for fetal, 2-, 4- and 6 month old animals, respectively. Relative to control at 4 h, all doses of GRF increased (P < 0.0001) GH in fetal pituitary cultures. Only 1000 nM GRF increased (P < 0.001) GH secretion in pituitary cultures from 4- and 6-month old pigs whereas pituitary cultures from 2 month old pigs did not respond to GRF. All doses of IGF-I increased (P < 0.04) basal GH secretion except for 0.01 nM in fetal pituitary cultures. Only the 1, 10 and 30 nM IGF-I increased (P < 0.04) basal GH secretion in pituitary cultures from 6 month old gilts. All doses of IGF-I failed to affect GH secretion in pituitary cell cultures from 2- and 4- month old gilts. Only 0.1, 10 and 30 nM IGF-I enhanced (P < 0.01) GH response to 1000 nM GRF from fetal pituitary cells in cultures. In pituitaries from 2 month old pigs only 30 nM IGF-I enhanced (P < 0.02) the response to 1000 nM GRF. All doses of IGF-I failed to influence the GH response to 1000 nM GRF in pituitary cultures from 4- and 6 month old pigs. These results demonstrate that GRF and IGF-I modulation of GH secretion is age dependent and occurred independent of hypothalamic input.

**Key Words:** Pituitary, GH, IGF-I

**W150 Effects of glutamine supplementation on lymphocyte subpopulations and proliferation in the peripheral blood supply of the transition dairy cow.** J. A. Woodward\*<sup>2</sup>, R. J. Christopherson<sup>1</sup>, C. J. Field<sup>1</sup>, S. Goruk<sup>1</sup>, G. Murdoch<sup>1</sup>, M. A. G. von Keyserlingk<sup>2</sup>, J. A. Bell<sup>1</sup>, and J. R. Thompson<sup>2</sup>, <sup>1</sup>*University of Alberta, Edmonton, Canada*, <sup>2</sup>*University of British Columbia, Vancouver, Canada.*

Most infectious and metabolic diseases occur in dairy cows in the weeks following calving, likely due to immunosuppression. Glutamine, the most abundant free amino acid in circulation, plays a key role in supporting immune cell proliferation and function in stressed animals. The objective of this study was to determine if supplementation with glutamine could reduce the degree of immunosuppression in transition cows. Twenty-eight cows received either a saline infusion (n=9) or a glutamine infusion of either 0.25 mmol•kg<sup>-0.75</sup>•h<sup>-1</sup>(n=10) or 0.50 mmol•kg<sup>-0.75</sup>•h<sup>-1</sup>(n=9). Infusions were given for 7 days, 8h•d<sup>-1</sup>, beginning the day of calving. Lymphocytes from the peripheral blood supply were isolated on days 0, 7, 14, and 21 following calving. Using immunofluorescence and flow cytometry the percentage of cells belonging to various lymphocyte subpopulations was determined. Ability of lymphocytes to proliferate when incubated with and without mitogens was determined by thymidine uptake. Animals supplemented with glutamine had significantly higher (P<0.05) percentage of naive T cells and B cells, as indicated by detection of CD62L marker. On day 21 glutamine treated animals had significantly higher (P<0.05) percentage of both CD62L positive cells and CD14 positive cells; CD14 is a marker of granulocytes and monocytes. Though there was no difference between treatments for the ability of the lymphocytes to respond to mitogens, unstimulated lymphocytes from control

animals had significantly higher thymidine uptake than unstimulated lymphocytes from glutamine supplemented cows, indicating a higher degree of immune activation in control animals. Together, the evidence indicates that the control animals have an activated immune system in comparison to glutamine treated animals, suggesting the immune system of the glutamine-treated animals was exposed to fewer antigens. Further research is necessary to determine by which mechanism, protective or otherwise, glutamine affects immune system activation in transition dairy cows.

**Key Words:** Glutamine, Immunosuppression, Dairy cattle

**W151 Leptin genotype influences lactation performance of Holstein cows.** J. E. P. Santos\*<sup>1</sup> and R. C. Chebel<sup>2</sup>, <sup>1</sup>*University of California Davis, Tulare*, <sup>2</sup>*University of Idaho, Caldwell.*

Objectives were to evaluate the relationships between leptin genotype and lactation performance in Holstein cows. Sequencing of DNA at the Exon-2 region of the leptin gene was performed in 814 lactating Holstein cows to determine the presence of single nucleotide polymorphism. Resulting genotypes were CC=282 (34.6%), CT=392 (48.2%), and TT=140 (17.2%). Cows were milked three times daily and milk yields were recorded for individual cows once monthly during the first 305 d in lactation by the official California DHIA milk test. Monthly milk samples were analyzed for somatic cell count (SCC), and concentrations of fat and true protein at the DHIA Laboratory in Tulare, CA. All cows received 500 mg of exogenous bovine somatotropin every 14 d, starting at 63 ± 3 d postpartum. Data were analyzed for repeated measures using the MIXED procedure of SAS controlling for genotype, month postpartum, parity and interactions. Single nucleotide polymorphism in the Exon-2 region affected (P=0.04) average daily milk yield (CC = 40.1 ± 0.26, CT = 41.0 ± 0.22, TT = 40.3 ± 0.37 kg/d) and 3.5% fat corrected milk (CC = 40.1 ± 0.25, CT = 41.0 ± 0.21, TT = 40.6 ± 0.36 kg/d). Concentration of fat in milk was not affected (P= 0.47) by leptin genotype (CC = 3.53 ± 0.03, CT = 3.54 ± 0.02, 3.58 ± 0.04 %), but daily fat yield tended (P=0.06) to be influenced by genotype and it was higher (P=0.02) for cows with the CT than CC genotype (CC = 1.40 ± 0.01, CT = 1.43 ± 0.01, TT = 1.43 ± 0.01 kg/d). Similarly, leptin genotype did not (P=0.76) affect concentration of true protein in milk (CC = 2.98 ± 0.01, CT = 2.99 ± 0.01, TT = 2.98 ± 0.01 %), but it influenced (P=0.006) the average daily protein yield (CC = 1.19 ± 0.01, CT = 1.21 ± 0.01, TT = 1.19 ± 0.01 kg/d). Finally, linear SCC score was not affected (P=0.21) by leptin genotype (CC = 2.20 ± 0.06, CT = 2.33 ± 0.05, TT = 2.26 ± 0.08). Our data suggest that single nucleotide polymorphism in the Exon-2 region of the leptin gene influences lactation performance, and CT cows produced more milk, 3.5% fat-corrected milk, true protein, and fat than CC cows.

**Key Words:** Leptin, Dairy cow, Milk production

**W152 Influence of maternal nutrition on placental vascularity and mRNA expression of angiogenic factors (AFs) and their receptors (AFRs) in adolescent sheep.** D. A. Redmer\*<sup>1,2</sup>, R. P. Aitken<sup>2</sup>, J. S. Milne<sup>2</sup>, M. L. Johnson<sup>1</sup>, D. Rouse<sup>1</sup>, P. P. Borowicz<sup>1</sup>, M. Borowicz<sup>1</sup>, K. C. Kraft<sup>1</sup>, L. P. Reynolds<sup>1</sup>, J. S. Luther<sup>1,2</sup>, and J. M. Wallace<sup>2</sup>, <sup>1</sup>*North Dakota State University, Fargo*, <sup>2</sup>*Rowett Research Institute, Aberdeen, Scotland, UK.*

Placental and fetal growth are compromised in rapidly growing adolescent ewes. Our aim was to determine the ontogeny of placental AF/AFR mRNA expression and vascular morphology throughout gestation. Singleton pregnancies (single sire) were established by

embryo transfer and then adolescent dams were offered a high (H, n=30) or control (C, n=34) nutrient intake. Whole placentomes, collected at Day (D) 50, 90 or 130 of gestation, were (a) frozen for quantitative real-time RT-PCR determination of placental AF/AFR mRNA expression or (b) perfusion-fixed and stained for quantification of vascular morphology. Placentome mass was independent of nutrition at D50 and 90, and by D130 both placentome (-46%) and fetal weight (-20%) were reduced in H pregnancies. Placentome mRNA expression of vascular endothelial growth factor (VEGF), angiopoietin (Ang)-1 and Ang-2, basic fibroblast growth factor (FGF-2), endothelial nitric oxide synthase (eNOS), and placental growth factor (PLGF), and AFRs VEGFR-1 (flt), VEGFR-2 (KDR), and FGFR-2 increased as gestation advanced ( $P<0.001$ ), whereas, hypoxia inducible factor (HIF1 $\alpha$ ), soluble guanylate cyclase (NO receptor) and neuropilin (NP)-2 declined ( $P<0.05$ ). H-intakes reduced NP-1 expression ( $P<0.04$ ) at each stage and increased eNOS at D130 ( $P<0.01$ ). Tie-2 (Ang receptor) expression was unaffected by stage or nutrition level. H-intakes reduced ( $P<0.004$ ) capillary area density and size in the cotyledon at D50, but thereafter placental vascular morphology per unit tissue was largely unaffected. Similarly, caruncular vascular morphology was independent of nutrition throughout. This study provides clues as to the AF/AFR's involved in the nutritional mediation of placental growth. As vascular morphology per unit placenta is largely unperturbed at D90 and 130, the data further suggest that it is the small size of the placenta and thus the reduction in total vascular volume that is the major limitation to fetal nutrient supply in late pregnancy.

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**Key Words:** Angiogenesis, Placenta, Nutrition

**W153 Propionate infusion alters G protein-coupled receptor GPR41 mRNA expression and the leptin system in goats.** M. Mielenz, C. Seybold, and H. Sauerwein\*, *University of Bonn, Germany.*

The importance of the GPR41 for the regulation of the leptin system in ruminants is not yet clarified. The aim of this study was to compare GPR41 mRNA expression in two different fat depots and to test the effects of intravenously infused propionate on GPR41 mRNA, serum leptin and its mRNA as well as leptin receptor mRNA in goats. In mice, GPR41 is recognized to be regulated by short chain fatty acids (SCFAs) with propionate being the most potent ligand. Propionate up-regulates leptin synthesis in this species. In contrast to monogastric mammals, ruminants almost entirely depend on SCFAs as energy source, but the reports about the relevance of propionate for leptin secretion in these species are controversial. Castrated male goats (Deutsche-Edelziege, 10 to 12 months old) were allocated to infusions through jugular catheters after an overnight fast. They received propionate infusions (96  $\mu\text{mol/kg}^*\text{min}$ ; n=8) or NaCl-solution (equivalent Na-concentration; n=5). After recording leptin baseline concentrations for 1h, infusions were carried out for 260 min. Blood samples were collected in 10 to 15 min intervals and analyzed for leptin by ELISA. The mRNAs of leptin, leptin receptor (long form) and GPR41 from subcutaneous adipose tissue (SAT) and perirenal adipose tissue (PAT) were quantified by real-time RT-PCR after euthanasia (propionate group: n=4; NaCl group: n=5). Repeated measures analysis of serum leptin values showed an increase in the propionate versus the control group ( $p<0.05$ ). Estimation of the mRNA expression data indicated a tendency for an increase of leptin mRNA in PAT ( $p=0.081$ )

but not in SAT ( $p=0.106$ ). The leptin receptor mRNA decreased in PAT ( $p=0.042$ ) and in SAT, the latter decrease, however, did not reach the level of significance ( $p=0.286$ ). GPR41 mRNA was elevated in SAT ( $p=0.029$ ) but not in PAT ( $p=0.756$ ). In conclusion, we established the expression of GPR41 in adipose tissue of a ruminant species; the effects of the propionate challenge indicate that GPR41 is involved in mediating propionate effects on the leptin system, at least at the provocative dosage tested.

**Key Words:** Ruminant, GPR41, Propionate

**W154 Evaluation of the adaptive capability in dairy cows during the periparturient period.** S. Hachenberg, C. Weinkauff, S. Hiss, U. Müller, and H. Sauerwein\*, *University of Bonn, Germany.*

The transition of pregnancy to lactation with the concomitant negative energy balance during early lactation requires substantial adaptive performance of the cow. Based on parameters of fat mobilization, we aimed to evaluate the adaptive status in dairy cows during the periparturient period. Blood samples were collected weekly from 4 wk ante partum to 12 wk post partum (p.p.) from 38 high yielding Holstein Frisian cows. For differentiating "adaptive performance satisfying" versus "adaptive performance limited" (AP-ltd), non-esterified free fatty acids (NEFA), beta-hydroxybutyrate, insulin-like growth factor-1 (IGF-1) and leptin were determined in serum and BCS was recorded. The animals were classified by using different thresholds and/or parameter combinations to determine which parameter will yield maximal consent with the other parameters to qualify only one or few parameters for field applications (GLM; SPPS). Health status was characterized using the concentrations of haptoglobin (Hp), the numbers of leukocytes and neutrophils as well as oxidative stress (dROM test) in blood. Liver impact was evaluated via glutamate-dehydrogenase (GLDH) activity in blood. From 7 criteria of classification, most information on fat mobilization was obtained from NEFA ( $>0.5$  mM in wk 1 p.p.) as well as IGF-1 ( $<39$  ng/mL in wk 1 p.p.) threshold values, and were combined as the criterion NEFA+IGF-1. Using this, the classification was matching in 27 animals. Meaningful conclusions about the relation between adaptive, health and liver status could be made on the basis of NEFA- and NEFA+IGF-1-classification. In the "AP-ltd" animals (NEFA; n=17 and NEFA+IGF-1; n=13), increased GLDH activities ( $p<0.05$ ) and reduced leukocyte numbers ( $p<0.001$ ) were determined. No differences were detected for neutrophil numbers, oxidative stress and concentration of Hp. In conclusion, routine determination of IGF-1 seems not justified to determine adaptive status. Instead, NEFA values  $>0.5$  mM during the first wk of lactation were considered as the most suitable parameter for identifying limited adaptive performance.

**Key Words:** Dairy cow, NEFA, Transition

**W155 Hot season and BCS affect leptin secretion of periparturient dairy cows.** U. Bernabucci\*, N. Lacetera<sup>1</sup>, L. Basiricò<sup>1</sup>, B. Ronchi<sup>1</sup>, P. Morera<sup>1</sup>, E. Seren<sup>2</sup>, and A. Nardone<sup>1</sup>, <sup>1</sup>DiPA, *Università della Tuscia, Italia*, <sup>2</sup>DiMorfiPA, *Università di Bologna, Italia*.

The study was carried out to ascertain the effects of the hot season and BCS on leptin secretion in transition dairy cows. Twenty-four Holstein cows were utilized in the study. Twelve cows (SP cows) gave birth in spring (28 March to 30 April, 2003). The remaining 12 cows (SU cows) gave birth in summer (15 June to 2 July, 2003). The two groups were balanced for parity and body condition score (BCS), and were fed the same rations. The 24 cows were selected on a BCS basis: cows with BCS  $<2.5$ , BCS from 2.6 to 3.5, and BCS  $>3.5$  were assigned



to low (LBCS), medium (MBCS), and high (HBCS) BCS groups, respectively. From -32 to 38 d relative to calving BCS was registered, and blood samples were taken weekly. Plasma leptin was determined. Data were analyzed using the Mixed Procedure of SAS. During SP, either the day (9-20 h) or the night (21-8 h) temperature-humidity index (THI) was below the upper critical THI (72). During SU, the mean daily THI values were  $79.5 \pm 2.9$  during the day, and  $70.1 \pm 4.7$  during the night. Furthermore, during SU, three heat waves occurred. The first heat wave lasted 5, the second 6, and the third 15 d. BCS began to decrease approximately 15 d before calving in SU cows, whereas in SP cows BCS started to decline after calving. Plasma leptin decreased ( $P < 0.05$ ) approaching to calving both in SU and SP cows. The HBCS cows showed higher ( $P < 0.05$ ) BCS reduction than LBCS and MBCS groups both during SU and SP. Compared with SP cows, plasma leptin was higher ( $P < 0.01$ ) in SU cows. In SU cows only, either before or after calving, a negative relationship ( $P < 0.01$ ) was found between plasma leptin and BCS, and HBCS cows had lower ( $P < 0.001$ ) plasma leptin compared with their LBCS and MBCS counterparts. On the contrary, SP-HBCS cows had higher ( $P < 0.01$ ) plasma leptin when compared with their LBCS and MBCS counterparts. Results reported herein, indicate that hot season is responsible for increased leptin secretion, and for changes into the relationships between body condition and leptin secretion in periparturient dairy cows.

**Key Words:** Leptin, Heat stress, Dairy cow

**W156 Detection of photonic emissions with varying concentrations of *Salmonella typhimurium*-lux through porcine intestinal tissue: A comparison of two photonic imaging systems.** K. Moulton<sup>\*1</sup>, E. Williams<sup>1</sup>, P. Ryan<sup>1</sup>, D. Moore<sup>1</sup>, S. Kim<sup>1</sup>, D. Lay<sup>2</sup>, and S. Willard<sup>1</sup>, <sup>1</sup>Mississippi State University, Mississippi State, <sup>2</sup>USDA-ARS Livestock Behavior Research Unit, West Lafayette, IN.

The objective was to correlate photonic emissions with concentration of *Salmonella typhimurium* (*S. typh-lux*; transformed with plasmid pAK1-lux) using the Berthold/NightOwl (BNO) and Stanford Photonic (SP) imaging systems in porcine intestinal tissues. Porcine small and large intestines were obtained after harvest and cleaned of digestive material. Two intestinal segments were imaged per session and 18 images were collected in both the SP and BNO imaging systems (1 to 60 s and 3 to 300 s, respectively). Varying concentrations of bacteria were used in small and large intestine studies, ranging from  $5.7 \times 10^5$  to  $8.0 \times 10^7$  CFU/ml. For each experiment, one segment was empty for a background count and one segment was injected with *S. typh-lux* broth; the difference of which was used in analysis. Throughout imaging (up to 6 replicates) *S. typh-lux* broth was serially diluted, plated and 24 h later counted to determine concentrations of bacteria imaged. Images from the SP and BNO systems were analyzed using Image J (NIH) and WinLight software, respectively. Data analysis included correlations between imaging systems, photonic emissions and bacterial concentrations. From small intestines, photonic emissions were positively correlated with bacterial concentration in SP ( $R=0.80$ ) and BNO ( $R=0.60$ ;  $P < 0.05$ ) systems. The correlation between imaging systems in small intestines relative to photonic emissions was 0.73 ( $P < 0.05$ ). From large intestines, photonic emissions were positively correlated with bacterial concentration in SP ( $R=0.52$ ) and BNO ( $R=0.60$ ;  $P < 0.05$ ) systems. The correlation between imaging systems in large intestines relative to photonic emissions was 0.65 ( $P < 0.05$ ). These data indicate that concentrations of *S. typh-lux* are highly correlated with photonic emissions, and may be detected through porcine intestinal tissues using either SP or BNO imaging systems. [USDA-NRI grant #

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**Key Words:** Biophotonics, *Salmonella*, Porcine intestines

**W157 Modulation of bovine hepatic ApoB100, ApoE and MTP gene expression by fatty acids.** J. A. A. Pires<sup>\*2</sup>, D. Pirazzi<sup>1</sup>, D. G. Mashek<sup>2</sup>, L. Basirico<sup>1</sup>, S. J. Bertics<sup>2</sup>, R. R. Grummer<sup>2</sup>, and U. Bernabucci<sup>1</sup>, <sup>1</sup>Università della Tuscia, Viterbo, Italy, <sup>2</sup>University of Wisconsin, Madison.

The objective was to investigate the effect of different fatty acids (FA) on the gene expression of ApoB100, ApoE, and microsomal triglyceride transfer protein (MTP), which are involved in the VLDL synthesis and secretion by bovine hepatocytes. Hepatocytes were isolated from three 7 to 10 d old male Holstein calves, cultured in monolayer, and treated for 48 h (from 20 to 68 h after seeding) with either no FA added (control) or 0.1, 0.2 and 1.0 mM of C16:0, C18:0, C18:1, C18:2, C18:3, C20:5, C22:6, or a physiological FA mix (NEFA; 15% C18:0, 30% C16:0, 45% C18:1, 5% C16:1, and 5% C18:2). Gene expression was quantified by Real-Time PCR, using ubiquitin as internal control. Data were analyzed by the Mixed Procedure of SAS using a model with fixed effect of treatment and random effect of calf. Contrasts were 1) linear and 2) quadratic concentration effects of single FA treatments; 3) linear and 4) quadratic concentration of NEFA treatment (positive control); 5) saturated and mono-unsaturated vs. polyunsaturated (PUFA); 6) C18 PUFA vs. long chain PUFA; 7) omega-6 vs. omega-3; 8) C20:5 vs. C22:6. Increasing the length and degree of unsaturation of FA down-regulated gene expression of ApoB and MTP (contrasts 5, 6, 7;  $P < 0.05$ ) and ApoE (contrasts 6, 7, 8;  $P < 0.01$ ). Increasing the concentration of single FA down-regulated gene expression of ApoE and MTP (contrast 1;  $P < 0.05$ ), but not of ApoB. Increasing NEFA concentration up-regulated gene expression of ApoB100 (contrast 3;  $P < 0.01$ ), ApoE (contrasts 3, 4;  $P < 0.05$ ) and MTP (contrast 4;  $P < 0.001$ ). These results show that FA modulate the gene expression of proteins involved in VLDL metabolism. Additionally, concentration effects of the NEFA and single FA treatments differed; for most single FA treatments, increasing concentration down-regulated gene expression while increasing the concentration of NEFA mix, which reflects the FA profile *in vivo*, up-regulated gene expression.

**Key Words:** Fatty acids, Hepatic apolipoprotein, Bovine

**W158 Characterization of bovine granulocyte chemotactic protein-2 in mammary glands with *Escherichia coli* mastitis.** J.-W. Lee<sup>\*1</sup> and X. Zhao<sup>2</sup>, <sup>1</sup>National Pingtung University of Science and Technology, Neipu, Pingtung, Taiwan, <sup>2</sup>McGill University, Ste-Anne-de-Bellevue, Quebec, Canada.

Neutrophils, migrating from the blood into to the milk in response to inflammatory signals, are considered as the first line of defense for eliminating invading bacteria in the mammary glands. The promptness of neutrophil recruitment is very crucial to the outcome of mastitis. By using the Differential Display (DD)-PCR technique, we previously demonstrated that the expression of a bovine chemokine gene, granulocyte chemotactic protein-2 (GCP-2), is upregulated in bovine mammary epithelial cells stimulated with the lipoteichoic acid, but not the peptidoglycan, from *Staphylococcus aureus*. In the present study, 6 dairy cows with clinical *Escherichia coli* (*E. coli*) mastitis were used to investigate the involvement of GCP-2 in infections induced by

Gram-negative bacteria. Biopsy samples and milk somatic cells were collected from the quarters with or without clinical *E. coli* mastitis in an animal. The expression of GCP-2 gene was analyzed by real-time PCR and calibrated with the expression of a house-keeping gene ( $\beta$ -actin). The expression of GCP-2 gene is significantly ( $P < 0.05$ ) increased in tissues ( $15.8 \pm 5.6$  fold) and milk somatic cells ( $17.39 \pm 2.8$  fold) collected from the quarter with clinical *E. coli* mastitis in comparison with the control quarter in the same animal. The preliminary results suggest that bovine GCP-2 might be another chemoattractant, in addition to interleukin (IL)-8 and C5a, produced by mammary cells during mastitis. Further research is required to investigate whether GCP-2 has a direct effect on recruiting leukocytes, such as neutrophils, to the site of infection.

**Key Words:** Mastitis, Chemoattractant, Neutrophil

**W159 Somatotropic axis components in and growth rates of IGF-I divergent Angus heifers receiving exogenous bovine (b) ST.** K. J. Steinman<sup>\*1</sup>, T. A. Hoagland<sup>1</sup>, M. E. Davis<sup>2</sup>, and S. A. Zinn<sup>1</sup>, <sup>1</sup>University of Connecticut, Storrs, <sup>2</sup>The Ohio State University, Columbus.

To determine growth rate and measure components of the somatotropic axis, 30 Angus heifers divergently selected for greater (H;  $n=14$ ) or lesser (L;  $n=16$ ) IGF-I concentrations were treated with exogenous bST [single-injection in tail head, 500 mg (Posilac);  $n=22$ ] or no bST (control;  $n=8$ ). Animals were grouped into pens with 3 or 4 H heifers and 4 L heifers per pen. Blood samples ( $N=3$ , 10 mL) were collected at 30 min intervals by jugular venipuncture from a jugular vein on d -6, -4, -2, 0, 1, 3, 5, and 7 relative to bST administration. Sampling regimen was conducted at approximately 320, 370, and 420 d of age (d 0). Animals were weighed weekly. Serum concentrations of ST and IGF-I were quantified by RIA, and IGFBP-2 and -3 concentrations were determined by Western ligand blot. Initially, H heifers were heavier ( $P=0.008$ ;  $265.1 \pm 7.6$  kg) than L IGF heifers ( $237.4 \pm 7.4$  kg) and this difference was maintained throughout the experiment ( $P=0.05$ ). However, ADG was not different ( $P=0.89$ ) between H ( $0.6 \pm 0.03$  kg) and L ( $0.6 \pm 0.03$  kg) IGF heifers, nor did bST-treatment influence ADG ( $P=0.95$ ). Before bST treatment, no differences were observed in ST ( $P=0.10$ ;  $13.9 \pm 2.6$  vs.  $18.8 \pm 2.5$  ng/mL), IGF ( $P=0.34$ ;  $134.4 \pm 20.8$  vs.  $106.3 \pm 20.3$  ng/mL), or IGFBP-3 ( $P=0.27$ ;  $60.4 \pm 4.3$  vs.  $52.3 \pm 4.3$  AU) in H and L IGF heifers, respectively. Concentrations of IGFBP-2 in L heifers tended to be greater than in H animals ( $P=0.08$ ;  $41.1 \pm 2.0$  vs.  $35.9 \pm 2.1$ ). Following bST-treatment, ST ( $35.8 \pm 3.6$  ng/mL), IGF-I ( $216.6 \pm 14.9$  ng/mL) and IGFBP-3 ( $86.0 \pm 3.0$  AU) increased ( $P=0.001$ ) compared with controls, but there were no differences ( $P \geq 0.14$ ) in ST, IGF-I or IGFBP-2 between genetic groups. Following bST treatment, IGFBP-3 was increased on d 5 and 7 in H compared with L ( $P < 0.05$ ;  $95.2 \pm 5.7$  vs.  $76.7 \pm 5.7$  AU) IGF heifers. Although these heifers came from IGF-I divergent genetic lines, no differences due to genetic line in basal or bST-induced IGF-I, ST or IGFBP-2 concentrations were observed. However, the increase in IGFBP-3 in H IGF heifers may account for increased BW in this genetic group.

**Key Words:** Beef cattle, Insulin-like growth factor, Somatotropin

**W160 Relative quantification of ghrelin mRNA in Holstein calves.** S. L. Greenwood<sup>\*</sup>, D. R. Glimm, A. F. Keating, N. S. Beswick, and J. J. Kennelly, University of Alberta, Edmonton, Canada.

The discovery of the novel orexigenic peptide ghrelin in monogastrics has stimulated research regarding ghrelin's involvement in feed intake

in ruminants. Though ghrelin mRNA expression is primarily found in the stomach, mRNA expression has also been observed in various other non-digestive tissues in monogastrics. The objectives of this study were to examine ghrelin mRNA expression in calf tissues, including various non-gastrointestinal tissues, and determine the influence of adrenocorticotrophin hormone (ACTH) injection on ghrelin, leptin and stearoyl-CoA desaturase (SCD) mRNA expression. Twenty-one different tissue types were collected from 6-month-old male Holstein calves ( $n=12$ ) after euthanasia. The calves were used in a stress response study and were injected with either saline or ACTH. The calves (3/group) were then euthanized at 15, 50 or 100 minutes following injection of 0.56 IU/kg ACTH or immediately following saline injection. RNA was isolated using the TRIzol method, and mRNA expression was determined using Real-time PCR. Cyclophilin was used as the control gene. Ghrelin, leptin and SCD mRNA expression was determined relative to cyclophilin expression and averaged within treatment groups. Ghrelin mRNA expression was most abundant in the abomasum in all treatment groups. However, ghrelin mRNA expression was also observed in the lung, spleen, kidney, aorta, endocardium, testis, epididymis, gall bladder, duodenum, jejunum and ileum. These findings are the first to indicate that ghrelin mRNA expression is present in non-gastrointestinal tissues in ruminants and could provide insight into possible endocrine and/or paracrine roles of ghrelin.

**Key Words:** Ghrelin, Adrenocorticotrophin hormone

**W161 Transcriptional profiling of hepatic constitutive androstane receptor and target genes in relation to boar taint compounds in backfat of pigs.** D. L. Greger<sup>\*1</sup>, C. Morel<sup>3</sup>, G. Bee<sup>2</sup>, S. Ampuero<sup>2</sup>, C. R. Baumrucker<sup>1</sup>, and J. W. Blum<sup>3</sup>, <sup>1</sup>Pennsylvania State University, University Park, <sup>2</sup>Agroscope Liebefeld-Posieux, Posieux, Switzerland, <sup>3</sup>University of Bern, Switzerland.

The primary causes of off-odors in pork from uncastrated male pigs, a phenomenon commonly known as "boar taint", are the presence of pheromonal androstene steroids as well as skatole and indole in adipose tissue. The latter compounds are products of microbial tryptophan metabolism. Hepatic cytochromes p450 (CYP) catalyze diverse reactions that are involved in the metabolism of compounds that cause boar taint. Constitutive androstane receptor (CAR) is a nuclear receptor (NR) known to regulate transcription of many hepatic genes including CYP genes. The objective of this investigation was to determine whether CAR mRNA expression is related to expression of hepatic CYPs and associated with concentrations of boar taint compounds. Liver samples from market weight pigs were collected at slaughter and total RNA extracted from 18 intact males and 8 castrates. Real-time reverse-transcription polymerase chain reaction (RT-PCR) was performed to quantify relative mRNA abundance for NR and selected target genes. Concentrations of androstenone, skatole and indole in backfat were determined by HPLC. Hepatic expression of CAR was positively correlated ( $P < 0.05$ ) with mRNA expression of PXR (pregnane X receptor), CYP2E1, CYP2A6, CYP1A2, and uridine glucuronosyl transferase 1A1 (UGT1A1). CAR mRNA abundance was negatively correlated with skatole ( $r = -0.41$ ;  $P < 0.05$ ) and indole ( $r = -0.64$ ;  $P < 0.01$ ); CYP2B22 mRNA abundance was negatively correlated ( $P < 0.05$ ) with indole ( $r = -0.49$ ;  $P < 0.05$ ) concentrations in backfat. Abundance of PXR mRNA was negatively correlated with indole concentrations ( $r = -0.41$ ;  $P < 0.05$ ), but was not associated with skatole level in backfat ( $P > 0.10$ ). These results suggest that CAR plays an important role in regulating transcription of hepatic

genes involved in metabolism of compounds that cause boar taint in intact male pigs.

**Key Words:** Boar taint, Constitutive androstane receptor, Liver

**W162 Expression of beta-adrenergic receptors in adipose tissue of Holstein dairy cattle.** J. Sumner\* and J. McNamara, *Washington State University, Pullman.*

The objective was to determine the expression of beta-adrenergic receptors in adipose tissue of Holstein dairy cattle. Activity of hormone sensitive lipase and responsiveness to beta-adrenergic stimulation increases during the transition period; and the adaptation varies among cattle with different milk production ability. However, expression of beta-adrenergic receptor subtypes and their potential role in control of lipolysis is unknown for dairy cattle. Therefore, twenty Holstein dairy cattle were grouped by lactation number (1, 2, and 3 or more). Mean 305ME was 13350 kg (SE 423); 3.6% fat and 3.3% protein. 305ME for 1st, 2nd and higher parity cows were 14936, 12791, and 11326 (SE 931). Fat averaged 3.55 and protein 3.3 %. Day 30 postpartum production was 39.3 kg/d (SE 3.4) with 3.6% fat and 2.7% protein and d 90 postpartum production was 48.8 kg/d (SE 1.5) with 3.4% fat and 2.9% protein. Body weight and BCS, measured at -30, 30, 90 and 270 days around calving averaged 682, 598, 634 and 638 kg and 3.3, 2.3, 2.5, and 2.7 BCS units. Subcutaneous adipose tissue was biopsied from the tailhead region. Duplicate samples were extracted for RNA and tissue was also incubated to measure basal and stimulated lipolysis. The RNA was used to synthesize cDNA, and using forward and reverse primers of CCCAGGCACCGAAAACCT and TCCCTTGTAATCAATGCTATCA, we determined that the beta-2-adrenergic receptor is expressed in Holstein adipose tissue at all time points measured during late pregnancy and lactation, by PCR and agarose gel analysis as well as real time RT-PCR. Beta-3 receptors were also measured using forward and reverse primers of AGGCAACCTGCTGGTAATCG and GTCACGAACACGTTG-GTCATG, and these were also expressed, with qualitative assessment suggesting at a lower amount. This is the first demonstration of expression of beta-receptor subtypes in Holstein adipose tissue.

**Key Words:** Beta-adrenergic receptors, Lipolysis, Transition dairy cattle

**W163 Evaluation of reproduction and blood metabolites in heifers fed dried distillers grains plus solubles or soybean hulls during late gestation.** C. L. Engel\*, H. H. Patterson, B. L. Perry, and G. A. Perry, *South Dakota State University, Brookings.*

Dried distillers grains plus solubles (DDGS) contain both undegradable intake protein and fat, which have both been shown to increase reproduction when supplemented to primiparous heifers. The mechanisms leading to enhanced reproduction when fat or UIP are supplemented have not been fully defined. The objective of this experiment was to evaluate DDGS or soybean hulls (SBH) during the last trimester of gestation on reproductive efficiency. Ninety-five cross bred heifers were stratified by expected calving date, body weight, BCS, and randomly assigned to DDGS or SBH (n = 6 pens per treatment). Diets were developed to meet the nutrient requirements at d 240 of gestation and were limit fed during the last trimester of gestation until parturition. Blood samples were collected prior to calving and once per week following calving for 4 weeks. There was no effect of treatment on BCS at calving ( $P = 0.29$ ;  $5.9 \pm 0.07$ ). Treatment had no effect on circulating concentrations of GH ( $P = 0.51$ )

or IGF-1 ( $P = 0.18$ ). However, time influenced both GH ( $P < 0.01$ ) and IGF-1 ( $P < 0.01$ ). Circulating concentrations of GH were elevated at calving ( $14.4 \pm 1.5$  ng/mL) and decreased by 4 d after calving ( $9.0 \pm 1.6$  ng/mL). Circulating concentrations of IGF-1 rose for the first 2 d following calving ( $57.1 \pm 5.2$  ng/mL) and then decreased through d 6 ( $30.5 \pm 5.6$  ng/mL). At the start of the breeding season there was no difference between DDGS and SBH in the percent of heifers that had initiated estrous cycles ( $P = 0.75$ ; 74% and 70%; respectively). There was a tendency ( $P = 0.11$ ) for more DDGS treated animals to become pregnant during a 64 d natural service breeding season compared to SBH treated animals (92% vs 80%; respectively). However, there was no difference in the distribution of pregnancies ( $P = 0.30$ ) between treatments. In summary, DDGS and SBH fed during the last trimester of pregnancy to heifers had similar effects on GH and IGF-1, but DDGS tended to improve pregnancy rates during a defined breeding season.

**Key Words:** DDGS, Gestation, Pregnancy

**W164 Responses of tissues to epinephrine as affected by homeorhetic state in dairy cattle.** K. L. Smith\*, A. K. Rauf, B. C. Benefield, A. W. Bell, and T. R. Overton, *Cornell University, Ithaca, NY.*

In vivo epinephrine (EPI) challenges were used to evaluate changes in peripheral tissue responses to catecholamines in the late prepartum (PRE), early postpartum (POST) and midlactation (ML) periods in Holstein cows. Periparturient (n=6) cows were studied during 4 periods (-30 to -26 d and -12 to -8 d PRE; 3 to 7 d and 26 to 30 d POST) and ML (n=5) cows were studied on d 148 to 152 POST. During each period, cows were administered a randomized sequence of 5 doses of EPI (0.2, 0.4, 0.8, 1.2 and 1.6  $\mu\text{g/kg BW}$ ) by i.v challenge. Blood was sampled frequently via jugular catheter from 30 min pre-challenge to 2 h post-challenge. Plasma NEFA and glucose responses to 0.8 and 1.6  $\mu\text{g/kg BW}$  of EPI were used to estimate tissue sensitivity and responsiveness. Both plasma glucose ( $\text{mg/dl} \cdot \text{min}^{-1}$ ) and NEFA ( $\mu\text{Eq/L} \cdot \text{min}^{-1}$ ) area under the curve (AUC) increased dose-dependently following EPI challenge. Periparturient cows had larger NEFA response during POST than PRE for both 0.8 and 1.6  $\mu\text{g/kg BW}$  of EPI, suggesting both greater sensitivity and responsiveness of adipose tissue to EPI during POST. Periparturient cows had larger glucose responses to both doses only at d 26 to 30 POST. Responses of NEFA for cows studied in ML were intermediate compared to those during PRE and POST. Responses of glucose for cows studied during ML were comparable to those at d 26 to 30 POST. Overall, responses of peripheral tissues to EPI were greater during lactation than during late pregnancy; those differences were more pronounced for release of NEFA from adipose tissue.

**Table 1. Responses by day relative to parturition**

	-30 to -26	-12 to -8	+3 to +7	+26 to +30	ML	SE
NEFA AUC						
0.8 $\mu\text{g/kg BW}$	579 <sup>b</sup>	525 <sup>b</sup>	1893 <sup>a</sup>	1725 <sup>a</sup>	1228 <sup>ab</sup>	380
1.6 $\mu\text{g/kg BW}$	837 <sup>bc</sup>	718 <sup>c</sup>	1937 <sup>a</sup>	2146 <sup>a</sup>	1672 <sup>ab</sup>	371
Glucose AUC						
0.8 $\mu\text{g/kg BW}$	280 <sup>b</sup>	319 <sup>b</sup>	186 <sup>b</sup>	555 <sup>a</sup>	632 <sup>a</sup>	78
1.6 $\mu\text{g/kg BW}$	538 <sup>bc</sup>	446 <sup>c</sup>	512 <sup>b</sup>	851 <sup>a</sup>	780 <sup>ab</sup>	98

Means within the same row with different superscripts differ,  $P < 0.05$

**Key Words:** Transition cow, Epinephrine, Homeorhesis



**W165 Responses of tissues to insulin as affected by homeorhetic state in dairy cattle.** K. L. Smith\*, A. K. Rauf, B. C. Benefield, A. W. Bell, and T. R. Overton, *Cornell University, Ithaca, NY.*

In vivo insulin (INS) challenges were used to evaluate peripheral tissue responses to insulin in the late prepartum (PRE), early postpartum (POST) and midlactation (ML) periods in Holstein cows. Periparturient (n=7) cows were studied during 4 periods (-30 to -26 d and -12 to -8 d PRE; 3 to 7 d and 26 to 30 d POST) and ML (n=5) cows were studied from d 148 to 152 DIM. During each period, cows were administered a randomized sequence of 5 doses of INS (0.2, 0.4, 0.8, 1.2 and 1.6 µg/kg BW) by i.v. challenge conducted once daily. Blood was sampled frequently via jugular catheter from 30 min pre-challenge to 2 h post-challenge. Plasma glucose and insulin responses to 0.8 and 1.6 µg INS/kg BW were used to estimate tissue sensitivity and responsiveness, respectively. Plasma glucose (mg/dl\*min<sup>-1</sup>) area under the curve (AUC) decreased and plasma insulin (ng/ml\*min<sup>-1</sup>) AUC increased dose-dependently following INS challenge. Plasma glucose AUC after 0.8 µg INS/kg BW was lower during PRE than during d 3 to 7 POST and ML; however, glucose AUC following 1.6 µg INS/kg BW was similar among PRE and POST and lower than ML. Plasma insulin AUC after 0.8 µg INS/kg BW was greater during d -12 to -8, d 3 to 7, and d 26 to 30 compared with d -30 to -26 and ML and was greatest after 1.6 µg INS/kg BW during d -12 to -8 and intermediate during d 3 to 7 POST compared to other study periods. Collectively, these data suggest that peripheral tissue responses to INS are greatest during ML and least during the immediate prepartum period.

**Table 1. Responses by day relative to parturition**

	-30 to -26	-12 to -8	+3 to +7	+26 to +30	ML	SE
Glucose AUC						
0.8 µg/kg BW	148 <sup>c</sup>	194 <sup>bc</sup>	238 <sup>a</sup>	211 <sup>ab</sup>	240 <sup>a</sup>	17
1.6 µg/kg BW	249 <sup>b</sup>	254 <sup>b</sup>	256 <sup>b</sup>	246 <sup>b</sup>	358 <sup>a</sup>	35
Insulin AUC						
0.8 µg/kg BW	74 <sup>b</sup>	155 <sup>a</sup>	136 <sup>a</sup>	124 <sup>a</sup>	53 <sup>b</sup>	18
1.6 µg/kg BW	190 <sup>b</sup>	269 <sup>a</sup>	223 <sup>ab</sup>	196 <sup>b</sup>	172 <sup>b</sup>	28

Means within the same row with different superscripts differ,  $P < 0.05$

**Key Words:** Transition cow, Insulin, Homeorthesis

**W166 Acute and chronic reduction in plasma progesterone concentrations after feed intake in dairy cows.** R. M. Santos\*, G. C. Perez, D. G. B. Demetrio, A. B. B. Maciel, and J. L. M. Vasconcelos, *FMVZ-UNESP, Botucatu, SP, Brazil.*

This study tested the hypothesis that an increase in feed intake (FI) decreases plasma progesterone (P4) acutely (5h after FI) and chronically (12h after FI) in cows treated with intravaginal P4 device (CIDR<sup>®</sup>, 1,9g), without a CL (Exp.1) or during the estrous cycle, with a CL (Exp.2). In Exp.1, non-lactating Holstein cows (n=21) were synchronized with Ovsynch protocol. At day 7 after the 2nd GnRH all cows received a PGF2α injection, to regress the CL (95%; 20/21), and were divided (Fatorial 2x2) according to number of CIDR (1 vs. 2) and FI (2 vs. 8 kg of concentrate, offered twice a day, for 14 days). To evaluate the acute effects of FI on P4, blood samples were collected every 15min for 6h at day 14. In Exp.2, non-lactating Holstein cows (n=7), were synchronized with the same protocol, and at day of the 2nd GnRH divided in 2 groups: 8 kg (n=7) or 2 kg (n=4) of concentrate,

with one replicate. In both experiments, to evaluate the chronic effects of FI on P4, daily blood samples were collected, 12h after FI. Plasma P4 was determined by RIA. The data were analyzed by PROC MIXED of SAS. For Exp.1, there was an interaction ( $P < .01$ ) between number of CIDR, FI and day on P4 at 12h after FI. Cows receiving 1CIDR and 8 kg of concentrate had lowest P4 and cows receiving 2CIDR and 2 kg of concentrate had highest P4. On day 14, was detected the interaction ( $P < .01$ ) between FI and moment of the blood sample on P4, showing the acute effect of FI on P4. Cows receiving 8 kg of concentrate had higher decrease in P4. In Exp.2, there was an interaction ( $P < .05$ ) between FI and day on P4 at 12h after FI. P4 decreased more for the group that received 8 kg concentrate. The effects of FI on P4 at 12h after FI, shows the chronic effect of FI on exogenous (Exp. 1) and endogenous (Exp. 2) P4. The acute and chronic reduction on P4 by FI in cows is probably due to an increase in P4 metabolism. These effects of FI on P4 could explain the decrease in fertility in lactating dairy cows.

**Key Words:** Feed intake, Progesterone, Dairy cow

**W167 Effects of Posilac on immune and endocrine responses of channel catfish challenged with *Edwardsiella ictaluri*.** B. Peterson\*, B. Small, and A. Bilodeau, *USDA-ARS Catfish Genetics Research Unit, Stoneville, MS.*

Research was conducted to examine the effects of recombinant bovine somatotropin (rbST, Posilac) on immune and endocrine responses to channel catfish challenged with *Edwardsiella ictaluri* (*E. ictaluri*). Four hundred and eighty fish ( $11.7 \pm 1.0$  g) were randomly assigned to two treatments with six replicates each: 1.) sham-exposed (one needle puncture per week for 3 wks and then challenged with *E. ictaluri*) and 2) rbST-exposed (Posilac, injected at 30 mg/g body weight per week for 3 wks and then challenged with *E. ictaluri*). Fish were sampled on d (1, 4, 8, or 14) (d 0 = *E. ictaluri* challenge). Non-exposed groups (d 0 controls) of fish were sampled on d 0. On each of the sample days, liver, kidney spleen, gut, and blood were collected. During the three-week period prior to challenge, rbST-treated fish gained 19% more ( $P < 0.01$ ) weight. Levels of bacteria were higher ( $P < 0.05$ ) on d 4 and 8 in rbST-exposed fish but were similar on d 14 compared to sham-exposed. Plasma levels of lysozyme were elevated ( $P < 0.01$ ) in rbST-exposed fish on d 4, 8, and 14 compared to sham-exposed fish. Compared to d 0 controls, IGF-I levels decreased ( $P < 0.05$ ) in challenged fish while levels were similar ( $P > 0.10$ ) between treatments throughout the study. Similarly, abundance of GH receptor (GHR) mRNA tended to decrease ( $P = 0.06$ ) in liver of challenged fish while levels were similar ( $P > 0.10$ ) between treatments in the spleen, kidney, liver, and gut throughout the study. Abundance of toll like receptor 5 (TLR5) mRNA increased ( $P < 0.05$ ) in liver of fish challenged with bacteria compared to d 0 controls while levels were similar ( $P > 0.10$ ) between treatments in the spleen, kidney, liver, and gut throughout the study. Mortality was higher in rbST-exposed fish compared to sham-exposed fish. An increase in lysozyme in rbST-exposed fish may reflect increases in bacterial levels of *E. ictaluri*. A decrease in GHR mRNA and plasma IGF-I suggests a down regulation of the somatotrophic axis in response to disease. While TLR5 mRNA increased in response to disease, there was no apparent effect of exogenous rbST.

**Key Words:** rbST, GHR, TLR5

**W168 Effects of milking frequency in early lactation on prolactin and growth hormone release and on milk production throughout lactation.** E. A. Albers, C. C. Williams\*, C. F. Hutchison, D. T. Gantt, C. Leonardi, L. R. Gentry, and C. C. Stanley, *LSU Agricultural Center, Baton Rouge, LA.*

Twenty-five multiparous Holstein cows were assigned to one of two treatments to study the response of increased milking frequency during early lactation on milk production and hormonal concentrations. Treatments consisted of milking 4 times per day (4X; n = 12) or 2 times per day (2X; n = 13) for the first 21 days of lactation. Cows in the 4X group were milked at the beginning of the normal 2X milking time and again after the remaining herd had been milked (2 to 3 hours later). After 21 days, all cows were returned to the 2X milking schedule. Milk yield and component analyses were obtained from records generated through the monthly DHIA testing procedures. On days 9 and 21 of lactation, fifteen cows (4X, n = 7; 2X, n = 8) were fitted with indwelling jugular catheters and allowed approximately 1 hour of rest prior to sample collection. Blood samples were collected at 30 minute intervals for 8 hours beginning two hours prior to afternoon milking. Plasma was analyzed for prolactin (PRL) and growth hormone (GH) by radioimmunoassay. There was no effect ( $P < 0.05$ ) of treatment on mean GH concentrations. However, GH concentrations differed during the 8 hour sampling period on both days, with GH concentrations being greater ( $P < 0.05$ ) on day 9. There was a treatment by time interaction ( $P < 0.01$ ) for PRL concentrations on days 9 and 21, with PRL being greater in cows milked 4X/d. Milk production, energy corrected milk (ECM) and fat corrected milk (FCM) were decreased ( $P < 0.05$ ) in cows milked 4X. No effect of treatment was observed ( $P > 0.05$ ) for percent milk protein or percent milk fat. These data indicate that increased milking frequency during the transition period did not improve milk production in these dairy cows.

**Key Words:** Milking frequency, Milk production, Hormones

**W169 Evaluation of an early marker of failed pregnancy: Changes in expression of Mx2 mRNA in peripheral blood mononuclear cells in dairy heifers of different reproductive statuses.** J. L. Stevenson\*<sup>1</sup>, R. C. Chebel<sup>1</sup>, J. C. Dalton<sup>2</sup>, T. L. Ott<sup>3</sup>, C. Gifford<sup>3</sup>, and K. Racicot<sup>3</sup>, <sup>1</sup>University of Idaho, Caldwell, <sup>2</sup>University of Idaho, Caldwell, <sup>3</sup>University of Idaho, Moscow.

The objective of the present study was to evaluate the correlation between reproductive status and steady-state levels of Mx2 mRNA in peripheral blood mononuclear cells of dairy heifers. Holstein heifers (n = 257), 13 ± 1 mo of age, were assigned to be inseminated (BRED; n = 209) or not (NON-BRED; n = 48) after the completion of a synchronization protocol. All heifers received a CIDR insert for 7 d and at CIDR removal heifers received one injection of PGF2a (study d 0). Heifers from the BRED group were inseminated upon detection of estrus from study d 0 to 3, and those not inseminated received a GnRH injection concomitant with fixed time AI 72 h after PGF2a injection (study d 3). All heifers from the NON-BRED group received a GnRH injection on study d 2 to induce ovulation and were not inseminated. Blood samples collected on study d 2 and 20 were used to determine steady-state levels of Mx2 mRNA using quantitative real-time PCR, and fold increase in Mx2 mRNA levels from study d 2 to 20 was calculated. Pregnancy was diagnosed at 30 and 65 d after AI. Data were analyzed using GLM procedure of SAS. Pregnancy rate at 30 d and 65 d after AI were 49.6% and 43.7%, respectively, and pregnancy loss between 30 d and 65 d after AI was 11.9%. Heifers diagnosed as

pregnant on d 30 (NON-BRED = 1.61 ± 0.41, BRED/Non-pregnant = 2.03 ± 0.34, BRED/Pregnant = 2.98 ± 0.34 fold) and 65 after AI (NON-BRED = 1.61 ± 0.41, BRED/Non-pregnant = 2.12 ± 0.32, BRED/Pregnant = 2.99 ± 0.36 fold) had greatest ( $P = 0.03$ ) Mx2 mRNA fold change between study d 2 and 20. There was no difference ( $P = 0.94$ ) in Mx2 mRNA fold increase between pregnant heifers that experienced or did not experience fetal loss between 30 and 65 d after AI. The range of Mx2 mRNA fold increase was as follow: NON-BRED = 0.05-13.02; BRED/Non-pregnant = 0.05-13.08; BRED/Pregnant = 0.15-14.32; and Bred/Aborted = 0.19-6.73. The present study indicates that increased levels of Mx2 mRNA in peripheral blood mononuclear cells are related to pregnancy at 30 and 60 d after AI in dairy heifers.

**Key Words:** Dairy heifers, Gene expression, Reproductive status

**W170 Influence of breed type and temperament on anatomic and endocrinologic parameters of the bovine hypothalamic-pituitary-adrenal (HPA) axis.** K. O. Curley, Jr.\*<sup>1</sup>, J. Lyons<sup>1</sup>, M. S. Brown<sup>2</sup>, T. E. Lawrence<sup>2</sup>, J. A. Carroll<sup>3</sup>, R. C. Vann<sup>4</sup>, S. T. Willard<sup>5</sup>, T. H. Welsh, Jr.<sup>1</sup>, and R. D. Randel<sup>6</sup>, <sup>1</sup>Texas Agricultural Experiment Station, College Station, <sup>2</sup>West Texas A&M University, Canyon, <sup>3</sup>Livestock Issues Research Unit, ARS-USDA, Lubbock, TX, <sup>4</sup>Brown Loam Experiment Station, Raymond, MS, <sup>5</sup>Mississippi State University, Mississippi State, <sup>6</sup>Texas Agricultural Experiment Station, Overton.

Temperament has been shown to affect bovine adrenal cortical and medullary stress responses. The biological mechanisms responsible for increased secretion of glucocorticoids and catecholamines in beef cattle with poor temperament remain undetermined. The objective of this study was to compare anatomic and endocrinologic parameters of the hypothalamic-pituitary-adrenal (HPA) axis within and across temperament and breed type. Exit velocity (EV), the rate at which the cattle exited the squeeze chute and traversed a fixed distance (1.83 m), was used to identify the 10 calmest (C) and 10 most temperamental (T) weaned calves from both Brahman (B) and Angus (A) herds. The steers were fed to a similar rib fat thickness at a feedyard in Canyon, TX. Blood samples were obtained via coccygeal venipuncture on d113 of the feeding period. Serum concentrations of cortisol (CS), and plasma concentrations of epinephrine (EPI) and norepinephrine (NOR) were determined by RIA and EIA, respectively. Temperament influenced CS ( $P = 0.001$ ; C = 16.98 ± 1.8, T = 31.07 ± 3.7 ng/mL), EPI ( $P = 0.025$ ; C = 136.71 ± 17.5, T = 490 ± 159.2 pg/mL), and NOR ( $P = 0.044$ ; C = 307.23 ± 22.9, T = 1119.83 ± 427.6 pg/mL). However, no effects of breed or breed by temperament interaction were observed for these endocrine parameters. At the time of harvest, left adrenal glands and pituitaries were collected so that adrenal cross-sectional areas and pituitary weights could be analyzed. There were no effects of temperament or breed by temperament interactions observed for the anatomical parameters. However, breed influenced adrenal area ( $P = 0.022$ ; A = 121.37 ± 4.4, B = 106.53 ± 4.8 mm<sup>2</sup>) as well as cortical area ( $P = 0.043$ ; A = 91.06 ± 3.1, B = 80.53 ± 4.2 mm<sup>2</sup>). Concerning the pituitary gland, breed influenced ( $P < 0.001$ ) both whole pituitary weight (A = 2.11 ± 0.1, B = 1.27 ± 0.04 g) and anterior pituitary weight (A = 1.65 ± 0.07, B = 0.97 ± 0.07 g). Breed type influenced anatomic parameters of the HPA axis whereas temperament influenced endocrinologic function of the adrenal cortex and medulla of beef steers.

**Key Words:** Temperament, Adrenal gland, Pituitary gland



**W171 Identification of growth hormone-regulated genes in the bovine liver by a microarray analysis.** H. Jiang\* and S. Eleswarapu, *Virginia Polytechnic Institute and State University, Blacksburg.*

The objective of this study was to identify the genes that are regulated by growth hormone (GH) in the bovine liver. Liver total RNA from three cows before and 7 days after administration of 500 mg recombinant bovine GH in a slow-release formula were subject to a microarray analysis, using the Affymetrix GeneChip® Bovine Genome Arrays that contained 23,000 bovine transcripts. On average, 6,837 (or 30%) of the transcripts were detected in the RNA samples. Analysis of the microarray data using GeneSifter revealed a set of 418 transcripts that were at least 1.5-fold different ( $P < 0.05$ ) in abundance between pre- and post-GH liver samples. Among the 418 transcripts, 392

displayed increased whereas 26 showed decreased expression after GH administration. These differentially expressed transcripts included insulin-like growth factor-I (IGF-I), acid-labile subunit of the IGF-binding protein complex, and hepatocyte nuclear factor 3 gamma, genes that were previously known to increase their expression in response to GH treatment, and IGF binding protein-1 (IGFBP-1), whose expression was previously known to be decreased by GH administration. The pre- and post-GH differences in the expression of these four transcripts were further confirmed by ribonuclease protection assays. The majority of the 418 differentially expressed transcripts were not known to respond to GH treatment before; they therefore represent new candidate genes that mediate GH action in the bovine liver.

**Key Words:** Growth Hormone, Microarray, Cattle

## Production, Management and the Environment III

**W172 Effectiveness of ocular thermography for the determination of body temperature in livestock: A multi-species analysis.** S. Willard\*<sup>1</sup>, P. Ryan<sup>1</sup>, D. Sykes<sup>1</sup>, M. Crenshaw<sup>1</sup>, R. Vann<sup>2</sup>, R. Randel<sup>3</sup>, T. Welsh<sup>3</sup>, S. Bowers<sup>1</sup>, M. Jones<sup>1</sup>, and A. Chromiak<sup>1</sup>, <sup>1</sup>Mississippi State University, Mississippi State, <sup>2</sup>Brown Loam Experiment Station, Raymond, MS, <sup>3</sup>Texas Agricultural Experiment Station, Overton and College Station, TX.

The eye may represent a measurement location from which an assessment of body temperature could be obtained. Applications for such measurements, if effective, include the non-invasive acquisition of body temperatures in a variety of species or as a rapid test for identifying sick (febrile) animals. The objectives of this study were to randomly sample livestock to determine whether digital infrared thermal imaging (DITI) of the eye is correlated with rectal temperature (RT). Total numbers of observations by species or breed were as follows: beef steers ( $n=127$ ), dairy cattle ( $n=180$ ;  $n=150$  cows and 30 calves), horses ( $n=119$ ;  $n=60$  mares and 30 foals) and gilts ( $n=120$ ). Regression analysis and associative correlations between maximum eye temperature (MET) and RT were determined. MET was lower ( $P < 0.05$ ) than RT in beef steers, dairy cattle, mares and gilts, but did not differ ( $P > 0.10$ ) for foals. Beef steers: measurements from two studies indicated high correlations between MET and RT ( $R=0.80$  and  $0.78$ ;  $P < 0.01$ ), yet when studies were combined the correlation was moderate ( $R=0.55$ ;  $P < 0.01$ ). Dairy cattle: cows exhibited a lower correlation ( $R=0.31$ ;  $P < 0.01$ ) than calves ( $R=0.52$ ;  $P < 0.01$ ), and when combined the correlation was lower ( $R=0.29$ ;  $P < 0.01$ ). Horses: mares exhibited a moderate correlation ( $R=0.41$ ;  $P < 0.05$ ) between MET and RT, however for foals correlations were not significant ( $R=0.16$ ;  $P=0.22$ ). When mares and foals were combined the correlation was moderately strong across all horses ( $R=0.65$ ;  $P < 0.01$ ). Gilts: the relationship between MET and RT was not significant ( $R=-0.06$ ;  $P=0.50$ ). The dynamic range of RT and MET obtained was narrow for dairy cattle (1.3 and 3.1 °C) and gilts (1.2 and 4.4 °C) and broader for beef steers (3.7 and 6.1 °C) and horses (2.2 and 5.6 °C), respectively. These data indicate that the effectiveness of ocular thermography as an assessment of body temperature may be species, breed and age dependent, and, when significant, were generally moderately correlated with RT overall. Nevertheless, ocular DITI may have application in screening for animals that are lower or higher than an expected normal range.

**Key Words:** Thermography, Eye, Temperature

**W173 Description and summarization of reticular core-body temperatures obtained from an automatic temperature recording system.** J. M. Bewley\*<sup>1</sup>, D. C. Batson<sup>2</sup>, and M. M. Schutz<sup>1</sup>, <sup>1</sup>Purdue University, West Lafayette, IN, <sup>2</sup>MaGiiX Inc., Post Falls, ID.

Automatic temperature recording may be used for dairy management and allow early detection of disease, estrus, heat stress, and the onset of calving. The MaGiiX™ Cattle Temperature Monitoring System (CTMS, MaGiiX Inc., Post Falls, ID) utilizes a passive bolus equipped with a temperature sensor, a panel reader placed at a parlor entrance or exit to query the bolus, and a software package to collect, analyze, and view data. The biologically inert bolus resides in the cow's reticulum and is queried each time the cow passes the reader (e.g. two or three times per day after milking). Ambient temperature (AT) and humidity (AH) are also measured for each observation. Reticular temperatures (RT) were monitored for a period of ten months (3/20/2005-1/24/2006) for a moderate-sized dairy in Montana milking 3 times daily. Unadjusted mean RT for 37,684 observations during this period was 103.0°F ( $\pm 0.9$ ). A mixed model including the effects of milking, date, AT, AH, and random cow, was used to compare the effects of morning, afternoon, or evening milking on RT. The RT differed by milking ( $P < 0.001$ ) with LS Mean RT of 103.0°F ( $\pm 0.04$ ), 102.8°F ( $\pm 0.04$ ), and 103.2°F ( $\pm 0.04$ ) for morning, afternoon, and evening milkings, respectively. The RT was weakly correlated with AT ( $r=0.050$ ,  $P < 0.001$ ), temperature humidity index ( $r=0.051$ ,  $P < 0.001$ ), and AH ( $r=-0.023$ ,  $P < 0.001$ ). Interpreting high RT to identify cows for further examination or monitoring is problematic. Strictly designating cows with RT greater than 103.5°F, 104.0°F, or 104.5°F as being elevated resulted in 19.0%, 6.7%, and 2.6% of cows identified, respectively. Contrastingly, 1.6% of cows were identified as having a high RT using a cut-off of +2 standard deviations while only 0.4% were identified using a cut-off of +3 standard deviations. Within milking variation (SD) of RT was correlated ( $p < 0.01$ ) with within milking, average AT ( $r=0.103$ ), and AH ( $r=-0.226$ ), and within milking SD of AT ( $r=0.127$ ).

**Key Words:** Temperature monitoring, Disease detection, Biosensors