measured with a Texture Analyzer. Butters were achieved with low-IA (1.5), high-IA (2.3), and bulk tank-IA (1.9). Corresponding values for Cheddar were 2.1, 3.4, and 2.4 and for provolone 1.6, 2.6, and 2.1. Panels detected no differences in flavor notes for the three IA types of butter. For Cheddar, sour and bitter flavor notes approached but did not achieve significance at 0.05. Provolone showed differences in buttery flavor, with both high- and low-IA being significantly less than bulk tank. High-IA butter was harder than low-IA butter as measured by penetration and creep. Similar differences were observed in the hardness of Cheddar and provolone cheeses, with high-IA being harder than low-IA. Seemingly, cows that give milk fat with a low IA will yield butter and cheeses with insignificant differences in flavor but slightly softer textures.

Key Words: Milk Fat, Butter, Cheese

ASAS/ADSA Physiology: General Physiology

1592 Influence of corticotropin-releasing hormone (CRH) on the expression of steroidogenic acute regulatory (StAR) protein in neonatal pigs derived by Caesarian section or natural birth. J.A. Carroll*[1], D. Alberts[2], D.J. Parzik[2], D.M. Stocco[2,3], and T.H. Welsh, Jr[2,3], [1]*Animal Physiology Research Unit, ARS-USDA, Columbia, MO,* [2]*Texas Tech University Health Science Center, Lubbock, TX,* [3]*Texas A&M University, College Station, TX.*

We previously reported that pigs born by Caesarian section (C-section) have greater basal and CRH-induced cortisol (CS) secretion compared to natural birth pigs. This study evaluated potential differences in StAR protein and cytochrome P450 side-chain cleavage enzyme (P450scc) protein expression in C-section pigs which may have contributed to this altered CS secretion. Eight crossbred sows were selected for the study (n = 4 natural birth and n = 4 C-section). Gestation length did not differ between natural birth and C-section pigs (113.6 ± .1 and 113.2 ± .3 d, respectively). Blood and tissue samples from 38 pigs were collected at birth. Remaining pigs were sustained with natural birth sows until 2wk of age (n = 39). At 2wk of age, pigs were non-surgically cannulated for blood sample collection to assess pituitary-adrenal responsiveness to porcine CRH (10 μg/kg). Blood samples were collected at -30, -15, 0, 5, 10, 20, 40, 60, and 90 min, with CRH or saline given at Time 0. Total RNA was isolated from the pituitary and adrenal glands to evaluate mRNAs specific for pro-opiomelanocortin (POMC) and for the ACTH receptor. Adrenocortical samples were used in Western blot procedures to determine StAR and P450scc protein content. While basal serum concentration of CS was not different at birth (P = .86), adrenocortical expression of StAR protein was lower (P < 0.0001) in C-section pigs as compared to natural birth pigs. Interestingly, serum ACTH (P = 0.0008) and ACTH receptor mRNA (tendency; P < 0.06) were greater in C-section pigs, suggesting a compensational effect due to the reduced expression of StAR protein in these pigs. A developmental decrease was observed in serum CS (P < 0.0001) and ACTH (P < 0.044), while a developmental increase was observed in POMC mRNA (P < 0.0001). ACTH receptor mRNA expression tended (P < 0.07) to decrease with age. Both, serum concentration of CS and StAR protein expression were increased following the CRH challenge (P < .05) whereas no change was detected in P450scc protein expression. Type of birth, age, and CRH challenge each influenced the correlations among various components of the HPA axis. These data suggest an important role for StAR and P450scc adrenocortical proteins in mediating developmental- and hormonal-induced changes in CS synthesis and secretion in the young pig.

Key Words: StAR protein, Cortisol, Pigs

1593 Hepatic corticosteroid-binding globulin (CBG) mRNA expression and plasma CBG levels in pigs in response to social and heat stress. J. Heo*[1], H. G. Kattesh[1], M. P. Roberts[1], R. L. Matteri[2], J. L. Morrow[3], and J. W. Dailey[3], [1]*University of Tennessee, Knoxville TN,* [2]*ARS-USDA, Columbia MO,* [3]*ARS-USDA, Lubbock TX.*

Plasma cortisol, CBG, hepatic CBG expression, and other physiological as well as behavioral measures of stress were studied in pigs in response to elevated temperature in conjunction with establishing social hierarchy. Twenty-four pigs (four pigs/litter) were weaned at 25 days of age and housed by litter for 2 wk at 23 ± 2°C. On d 0, animals were weighed and placed under general anesthesia for collection of blood (10 ml) and liver tissue (∼100 mg). On d 1, three pigs of similar weight (23 ± .9 kg) but from different litters were allotted to eight nursery pens within two environmentally controlled rooms (12 animals/room). One room was maintained at 23 ± 2°C (CONT) and the other at 33 ± 2°C (HEAT). On d 8-14 both rooms were maintained at 23 ± 2°C (REC). Animals were videotaped for 72 h beginning on d 1 and 8 to document behavioral changes in response to room temperature and to determine social order. Blood and liver tissue were collected again on d 7 and 14. Data was analyzed as a randomized complete block design using Proc Mixed procedure of SAS. Plasma haptoglobin increased (467.2 ± 122.5 vs 763.4 ± 112.9 ug/ml; d 0 vs d 7, P<.05) and cortisol and CBG decreased (9.92 vs 8.51 ± .83 ug/dl, 11.41 vs 9.93 ± 1.07 ug/ml; d 0 vs d 7, respectively, P<.05) in the HEAT group. Hepatic CBG mRNA level and neutrophil: lymphocyte ratio were not affected (P>.1) by treatment. HEAT pigs displayed increased (P<.01) drinking but reduced feeding (P<.01) and lying in contact with other pigs (P<.05) behaviors. ADG tended (P=.06) to be lower for HEAT (.64 ± .06 kg/d) compared to CONT (.82 ± .06 kg/d) pigs. During REC, HEAT pigs had similar (P>.1) ADG, plasma cortisol, CBG, haptoglobin, and drinking and feeding but increased (P<.01) lying with contact behaviors compared to CONT. Measured physiological and behavioral responses were not related to social status. These results indicate that reduced circulating levels of cortisol and CBG in pigs following 7-d exposure to elevated temperature may not be determined by hepatic CBG mRNA expression.

Key Words: Pig, Heat stress, CBG

1594 Cold-induced changes in brown adipose tissue (BAT) composition and iodothyronine 5'-deiodinase (5'D) activity in newborn Angus and Brahman calves. S.J. Falck*[1], G.E. Carstens[1], S. Kahl[2], S.R. Busch[1], L.J. Slay[1], C.D. Gilbert[1], and S.B. Smith[1], [1]*Texas A&M University, College Station, TX,* [2]*USDA, Agricultural Research Service, Beltsville, MD.*

We previously found that newborn Brahman **(BR)** calves generate less heat from BAT in response to a norepinephrine challenge than Angus **(AN)** calves even though BAT 5'D activities (converts thyroxine, T_4 to triiodothyronine, T_3) were higher in BR calves. The aim of this study was to further characterize 5'D in newborn calves, and to examine breed effects on cold-induced changes in BAT composition, 5'D activity, and plasma T_3 and T_4 levels. AN (n = 15) and BR (n = 20) calves were each assigned to 1 of 3 postnatal treatments: newborn **(N)**, cold **(C)** and warm **(W)** calves. Newborn calves were killed at 9 h of age, whereas, C and W calves were killed after 48 h of exposure to 4 and 20° C. Rectal temperature **(RT)** and blood samples were collected at 0, 12, 24 and 48 h, and plasma analyzed for T_3 and T_4. Deiodinating activity of BAT was determined by quantifying $^{125}I^-$ released from ^{125}I-labeled reverse-T_3 using assay conditions for type I **(5'D-I)** and type II **(5'D-II)**. RT were higher (P < .01) in AN calves at time 0 (38.8 vs 38.1 ± .16° C, but were similar to BR calves thereafter. BR calves had higher plasma T_3 (61%; P < .001) and T_4 (31%; P < .05) levels than AN calves, but plasma T_3 increased more due to cold in BR (62%; P < .001) than AN (10%; P = .80). Overall, plasma T_4 levels were 31% higher (P < .001) in C than W calves. Compared to AN calves, BR had less (P < .05) BAT lipid at birth (2.12 vs 1.33) and after C (1.95 vs .63), but similar amounts after W treatment (2.32 vs 1.89 ± .28 g/kg BW). Cytochrome c oxidase activity (μmol/min/g BAT) was unaffected by breed and was 68% higher in C than W calves. BAT 5'D-I activity was higher (P < .001) in BR calves at birth (.53 vs .95 ± .01 nmol I/h/mg protein), but not after C or W treatment. Cold decreased (P < .01) 5'D-I activity 29% in BR calves, but numerically increased 5'D-I in AN calves. 5'D-II activity was higher (P < .001) in BR calves (.29 vs .94 ± .12 pmol I/h/mg), but was unaffected by postnatal treatment. The results suggest that although BR calves mobilize BAT lipid more rapidly during cold exposure, this substrate is not effectively used to support BAT thermogenesis.

Key Words: Brown Fat, 5'-Deiodinase, Thyroid Hormones

1595 Growth rates of Holstein heifers fed diets differing in amounts of protein, energy and protein:energy ratios and treated or not with bST. M. Liboni*, T.I. Belloso, M.S. Gulay, M.L. Schairer, M.J. Hayen, L.C. Teixeira, K.C. Bachman, and H.H. Head, *University of Florida*.

One hundred and twenty Holstein heifers were assigned to one of four diets (30/diet) and bST treatments in a 2x2x2 factorial arrangement of treatments beginning when they were 120 d of age. Diets were formulated to contain 14 or 19% CP with energy concentrations of 100% and 110% of daily requirements (NRC,1989) for each CP percentage. These diets were fed once daily in quantities to meet desired average daily weight gains (ADG) of 0.81 kg for diets with 14% CP and 1.09 kg for diets with 19% CP. The g of CP/Mcal ME for the four diets were held constant throughout the trial at 50, 55, 65 and 73, respectively. When heifers reached 341 Kg BW they then were transferred to the breeding herd. Heifers were raised in 8 groups of 10 heifers each and 2 groups of 20 heifers each. All heifers were weighed and height at withers measured when assigned to trial and then weekly thereafter. One-half of the heifers in each group was injected with bST (POSILAC ®, 500 mg bST/1.4 ml, Monsanto) biweekly in the following amounts: 0.2 ml up to 181 Kg BW, 0.3 ml between 182-273 Kg BW, and 0.4 ml above 273 Kg BW. These volumes provided about 5.1, 7.6 and 10.2 mg bST/d. Average daily weight gains (kg/d) of heifers on the four diets were 0.89 0.01, 0.97 0.01, 1.03 0.01 and 1.02 0.01. The ADG for the four diets differed (P<0.0001). At 341 Kg BW the average ages of heifers fed the four diets were: 365 4.63, 368 4.00, 337 4.00 and 356 4.00 d. Difference in the average age to reach 341 Kg was significant (P<0.0003). Average total height increases (AHI, cm) during the time period heifers were on experiment were 28.46 0.69, 31.79 0.70, 31.78 0.70 and 34.62 0.70, respectively. The AHI for the four diets differed (P<0.05). Overall, there were significant positive effects of bST on ADG (P<0.0129) and AHI (P<0.0898) for heifers fed the four diets. Injections of bST reduced the number of days for heifers to reach 341Kg for the two high energy diets compared to uninjected heifers fed the same diets (P<0.0611), but not for the lower energy diets. Considering the whole growth period, no significant effects of season of birth of heifers were detected on age to reach 341 Kg, ADG or AHI. No season of birth x bST interaction was detected for any growth measure.

Key Words: Heifer, Proten:energy, Growth

1596 Hepatic oxidative metabolism in lactating dairy cows is modulated by increasing doses of intravenous lipopolysaccharide. M. R. Waldron*[1], T. Nishida[1], B. J. Nonnecke[2], and T. R. Overton[1], [1]*Cornell University, Ithaca, NY,* [2]*National Animal Disease Center, USDA ARS, Ames, IA*.

The effect of intravenous lipopolysaccharide (LPS; *E. coli* O11:B4) infusion on liver metabolism in twelve multiparous midlactation cows (150-220 DIM) was investigated. A covariate liver sample was obtained via percutaneous trochar biopsy 7 d prior to LPS infusion. Liver slices were used *in vitro* to determine hepatic capacities for conversion of [1-^{14}C]propionate, [1-^{14}C]alanine, and [1-^{14}C](L+)-lactate to CO_2. Hepatic capacities for conversion of [1-^{14}C]palmitate to CO_2, acid soluble products (a proxy for ketone body production), and stored esterified products also were determined. Seven days later, cows were intravenously infused with either 0 (n=6), 1.0 (n=4), or 2.0 (n=2) μg LPS/kg BW dissolved in 100 ml sterile physiological saline during a 100-min infusion. Saline infused cows were pair-fed with LPS-infused cows during the infusion period to eliminate effects of feed intake with LPS treatment. Liver was biopsied at 4.5 h after the beginning of infusion and metabolic incubations were conducted as described for covariate samples. Differences in the capacities of liver from control and LPS-infused cows to convert [1-^{14}C]palmitate to CO_2, acid soluble products, and stored esterified products were not significant ($P > 0.20$). However, LPS infusion tended to increase the capacity of liver to convert [1-^{14}C]propionate (4.19, 4.23, 6.07 μmol/h x g wet weight; $P < 0.12$) and [1-^{14}C](L+)-lactate (0.66, 0.98, 1.00 μmol/h x g wet weight; $P < 0.06$) to CO_2 for 0, 1.0, and 2.0 μg LPS/kg BW, respectively. Differences in hepatic capacity to convert [1-^{14}C]alanine to CO_2 were not significant ($P > 0.20$). These data suggest that aspects of hepatic carbohydrate metabolism in lactating dairy cows are affected by the inflammatory response elicited by LPS infusion; however, effects of the inflammatory response on hepatic fatty acid metabolism in midlactation cows appear to be minimal.

Key Words: Liver Metabolism, Lipopolysaccharide, Immune System

1597 Circulating leukocyte populations, serum cytokines and plasma vitamins A and E in mid-lactation dairy cows infused with varied doses of lipopolysaccharide (LPS). B. J. Nonnecke[1], M. R. Waldron*[2], T. Nishida[2], T. R. Overton[2], and R. L. Horst[1], [1]*National Animal Disease Center (NADC), USDA ARS, Ames, IA,* [2]*Cornell University, Ithaca, NY*.

Four multiparous lactating cows were used in 4 x 4 Latin square design to assess effects of LPS (serotype *E. coli* 011:B4 at 0.0, 0.5, 1.0, 1.5 μg/kg BW) on blood leukocyte populations, cytokines (TNF-α and IFN-γ) and vitamins [retinol,RRR-α-tocopherol and β-carotene (provitamin A)]. LPS, in 100 ml of physiological saline, was infused IV at 1 ml/min over 100 min. Blood was collected by jugular catheter immediately before infusion (0h) and at 2 (leukocytes not sampled at this time), 4, 6, 24 and 48h. Samples were shipped overnight to the NADC for analysis. Blood leukocytes were phenotyped by flow cytometry and the cytokines and vitamins were quantified by ELISA and reverse-phase HPLC, respectively. LPS infusion caused a marked leukopenia at 4 and 6 h that was characterized by a reduction in the number of T cells ($CD3^+$, $CD4^+$, $CD8^+$ and $\gamma\delta TCR^+$ subsets), B cells, monocytes and cells expressing IL-2 receptors and MHC class II antigens. By $\geq$24h, values were comparable to preinfusion and control-cow values. Interestingly, percentages of B cells and $\gamma\delta TCR^+$, IL-$2r^+$ and MHC class II $antigen^+$ cells in leukocyte populations from LPS-treated cows were elevated at 4 and 6 h. Serum TNF-α was affected by infusion of LPS in a dose-dependent fashion, with maximal concentrations occurring at 2 h. TNF-α concentrations in these cows declined precipitously from 2 to 6 h and were not different from preinfusion values or those of control cows at $\geq$24h. Serum IFN-γ, extremely low throughout the experimental period, was unaffected by LPS. Plasma retinol, RRR-α-tocopherol, and β-carotene also were unaffected by LPS. These results and metabolic data presented in the companion poster suggest LPS-induced inflammation influences broad aspects of the immune system, affecting the metabolism of dairy cows.

Key Words: Lipopolysaccharide, Vitamins A and E, Immune Function

1598 Metabolic responses of lactating dairy cows to intravenous infusion of increasing amounts of lipopolysaccharide. M. R. Waldron*[1], T. Nishida[1], B. J. Nonnecke[2], and T. R. Overton[1], [1]*Cornell University, Ithaca, NY,* [2]*National Animal Disease Center, USDA ARS, Ames, IA*.

Four multiparous lactating cows (175-220 DIM) were used in a 4x4 Latin square design to assess the effects of increasing doses (0.0, 0.5, 1.0, 1.5 μg/kg BW) of lipopolysaccharide (LPS; *E. coli* 011:B4) on performance and blood metabolites. Treatments were dissolved in 100 ml of sterile saline and infused intravenously over a period of 100 min. Milk production was decreased linearly for 24 h after LPS infusion (34.9, 22.4, 22.0, and 16.5 kg; $P < 0.001$) and generally returned to preinfusion levels by the sixth milking following LPS infusion. The DMI also was decreased linearly for 24 h following LPS infusion (21.2, 17.6, 15.5, and 10.9 kg; $P < 0.01$) and regained preinfusion levels 3 d following infusion. Blood was sampled immediately before infusion (0 h), at 60-min intervals for 8 h, and at 24 and 48 h postinfusion. Reported means for hormone and metabolite concentrations with treatment by time effects are at the 3 h timepoint. Plasma insulin (1.54, 1.64, 2.37, and 3.88 ng/ml), glucagon (80.6, 204.0, 249.3, and 326.3 pg/ml), and cortisol (0.14, 1.16, 1.51, and 1.79 ng/ml) concentrations increased linearly following LPS infusion (treatment by time, $P < 0.01$). Plasma (L+)-lactate concentrations (5.83, 7.43, 7.95, and 6.63 mg/dl) increased following LPS infusion (treatment, $P < 0.001$). There was a linear decrease (treatment by time, $P < 0.01$) for plasma BHBA (9.8, 5.3, 4.4, and 2.9 mg/dl) after infusion. Plasma NEFA were increased by the 1.5 μg LPS/kg BW treatment (204, 185, 198, and 260 mg/dl for 0.0, 0.5, 1.0, 1.5 μg LPS/kg BW, respectively; $P = 0.037$). There was no effect of LPS infusion on plasma glucose concentrations ($P > 0.2$). These data suggest that the inflammatory response elicited by LPS has dose-dependent metabolic consequences that may be important for metabolic health in dairy cows.

Key Words: Lipopolysaccharide, Hormones, Metabolites

1599 Propionibacteria as a direct fed microbial: effects on the insulin-like growth factor system and reproduction in early postpartum dairy cows. C. C. Francisco*, D. N. Waldner, C. S. Chamberlain, and L. J. Spicer, *Oklahoma State University, Stillwater, OK.*

This study was designed to investigate the effects of supplementing a direct fed microbial, Propionibacteria culture (*P. acidipropionici*) to the diets of postpartum cows on plasma IGF-I and IGF-I binding protein (IGFBP) concentrations and reproduction during the first 12 wk of lactation. Pluriparous Holstein cows (n=19) were individually fed a TMR and randomly assigned to either a control or treatment group. Each of the treated cows (n=9) received 17 g of Propionibacteria culture daily from -2 to 12 wk postpartum. In blood samples collected twice a week, plasma IGF-I and IGFBP concentrations did not differ between the two groups of cows, however, IGF-I concentrations increased (P<0.001) by fourfold from 1 to 12 wk postpartum. For both groups of cows, plasma IGFBP-3 concentrations at wk 1 were 14% and 15% lower (P<0.001) than at wk 6 and 12, respectively. Plasma IGFBP-2 concentrations at wk 6 and 12 were 19% and 27% greater, respectively, than wk 1, and plasma IGFBP-5 concentrations were 32% and 33% greater (P<0.001) at wk 6 and 12, respectively, than wk 1. Propionibacteria-treated and control cows had similar concentrations of progesterone peak and area under the progesterone curve during first and second postpartum estrous cycles. Interval to first and second postpartum ovulations did not differ between treated and control cows. Feeding Propionibacteria to early lactating cows did not affect the changes in the plasma profiles of IGF-I and IGFBP nor did it affect reproductive function.

Key Words: Propionibacteria, Insulin-like Growth Factors, Reproduction

1600 Propionibacteria as a direct fed microbial: effects on energy balance, milk production, milk components, metabolic hormones and metabolites in early postpartum dairy cows. C. C. Francisco, D. N. Waldner*, C. S. Chamberlain, R. P. Wettemann, and L. J. Spicer, *Oklahoma State University, Stillwater, OK.*

The objective of this study was to determine the effects of directly feeding Propionibacteria culture (*P. acidipropionici*) on energy balance (EB), milk yield, metabolic hormones and metabolites in early lactating cows. Pluriparous Holstein cows (n=19) were individually fed a TMR and randomly assigned to either a control or treatment group. Each of the treated cows (n=9) received 17 g of Propionibacteria culture daily from -2 to 12 wk postpartum. Calculated EB of cows supplemented with Propionibacteria (-7.3±1.5 Mcal/d) tended to be greater (P<0.10) than control cows (-11.4±1.5 Mcal/d) during wk 1 of lactation. Fat-corrected milk and DMI were not affected by treatment. BCS at wk 4 and 10 postpartum did not differ (P>0.50) between groups. Cows fed Propionibacteria had greater (P<0.05) percentages of milk protein (5.0±0.15% vs. 4.0±0.14%) and SNF (10.5±0.15% vs. 9.7±0.14%) than control cows during wk 1 of lactation. Percentages of milk fat and lactose were not influenced by treatment. In plasma samples collected twice a week, NEFA concentrations at wk 1 were greater (P<0.01) in treated than control cows. Plasma leptin concentrations tended to be greater (P<0.10) in cows fed Propionibacteria (8.1±1 ng/mL) compared with control cows (5.3±1 ng/mL) during the 12 wk study and did not change with week postpartum. Plasma cholesterol and glucose concentrations were not affected by supplemental feeding of Propionibacteria but both metabolites increased (P<0.05) with week postpartum. Concentrations of plasma insulin were also unaffected by treatment and increased by about twofold (P<0.05) between wk 1 and 12. In conclusion, feeding Propionibacteria culture to early lactating dairy cows increased milk protein and SNF content, and increased plasma NEFA and leptin levels.

Key Words: Propionibacteria, Leptin, Nonesterified Fatty Acids

1601 Administration of bST elevates phosphoenolpyruvate carboxykinase mRNA in lactating dairy cows. J.C. Velez* and S.S. Donkin, *Purdue University West Lafayette, IN.*

Somatotropin (bST) administration increases milk production and a corresponding increase in hepatic glucose output is necessary to meet increased needs for mammary metabolism and lactose synthesis. The objective of this study was to determine the effects of bST on expression of pyruvate carboxylase (PC) and phosphoenolpyruvate carboxykinase (PEPCK), two enzymes that are critical to the synthesis of glucose in liver. Eighteen cows were randomly allocated to two treatment groups receiving either bST (Posilac®, Monsanto, St. Louis) or saline injections, over 14-day intervals during a 42-d period. Feed intake and milk production were recorded daily and milk composition was determined weekly. Liver biopsies and blood samples were obtained on day seven following the second and third injections. Blood samples were analyzed for non-esterified fatty acids (NEFA) and glucose. Total RNA was extracted from liver biopsy samples and used to determine the abundance of PC, PEPCK, and 18S mRNA using Northern blot analysis. Milk production and dry matter intake were increased (P<.05) for bST treated cows 38.6 vs. 33.8 1.4 kg/day and 27.2 vs. 25.8 0.5 kg/day, respectively. Plasma NEFA was increased due to bST treatment (111 vs. 170 14 μM). Plasma glucose was not affected by bST treatment (P >.05). Treatment with bST increased (P<. 08) PEPCK mRNA but not PC mRNA expression. The effect of bST was accentuated with repeated injection cycles (time effect, P <.05). The relative abundance of PEPCK during the third injection interval was elevated 18% compared to control samples. The magnitude of increase in PEPCK mRNA and changes in milk production are of a similar magnitude. The data indicate a slight increase in PEPCK due to bST which may reflect a greater potential for hepatic glucose production.

Key Words: Somatotropin, liver, mRNA expression

1602 Pyruvate carboxylase 5' untranslated region mRNA variants are heterogeneously expressed within and among bovine tissues. C. Agca* and S.S. Donkin, *Department of Animal Sciences, Purdue University, West Lafayette, IN 47907.*

Pyruvate carboxylase (PC) catalyzes the first regulated reaction in the conversion of pyruvate to glucose in gluconeogenic tissue and the use of acetyl groups for fatty acid synthesis in adipose tissue. Multiple transcripts have been identified for rat and human PC mRNA that are the products of a single gene, encode the same protein, but differ with respect to translational efficiencies and exhibit tissue specific expression patterns. Two bovine PC variants containing different 5' untranslated regions (UTR) of the mRNA were previously identified and reported by our laboratory. The objective of this study was to determine the presence of additional variants of bovine PC mRNA and their distribution among bovine tissues. Total RNA was extracted from bovine liver and used in 5' rapid amplification of cDNA ends and reverse transcription (RT) -PCR. Four new 5'UTR variants were identified which share regions of similarity ranging from 68 to 185 bp. In total, six unique 5'UTR have been identified, cloned and sequenced. The 5'UTR clones are designated as bPC5'**A**, bPC5'**B**, bPC5'**C**, bPC5'**D**, bPC5'**E**, and bPC5'**F** and contain 68, 253, 178, 89, 227, and 162 bp respectively. Northern blot analysis indicates that PC mRNA is more abundant in perirenal fat, omental fat, kidney, mammary tissue, brain and liver than muscle, heart and lung. The profile of 5'UTR variants, determined using RT-PCR and primers for the 5' UTR indicates that bPC5'**B**, bPC5'**C**, bPC5'**D**, and bPC5'**E** are present all tissues although bPC5'**D**, and bPC5'**E** are found in reduced quantities in heart, lung and muscle. The variants bPC5'**A** and bPC5'**F** are detectable only in adipose tissue, kidney and liver. The data demonstrate the complexity of expression of PC mRNA 5'UTR variant profiles in liver and indicate PC variant expression that is tissue specific. The exclusivity of bPC5'**A** and bPC5'**F** suggests control of the PC gene expression that is unique to gluconeogenic and lipogenic tissues.

Key Words: Pyruvate carboxylase, 5'UTR

1603 Differential relationships of metabolic hormones to growth and reproductive development in performance-tested Angus, Brangus, and Brahman bulls. M.G. Thomas*[1], R.M. Enns[2], D.M. Hallford[1], D.H. Keisler[3], B.S. Obeidat[1], C.D. Morrison[3], J.A. Hernandez[1], W.D. Bryant[1], R. Flores[1], and R. Lopez-Ordaz[1], [1]*New Mexico State University,* [2]*University of Arizona,* [3]*University of Missouri.*

Understanding mechanisms that regulate growth and reproduction are important for improving selection strategies in cattle. In this study, Angus, Brangus, and Brahman bulls (n = 7 per breed) of similar age were randomly selected from a group of 65 weanlings. Bulls were then evaluated for 112 d for concentrations of metabolic hormones and metabolites and growth and reproductive traits. Performance data and serum were collected on d 0, 28, 56, 84, and 112. Serum was also collected on d 50

to 59 and 103 to 112. These time periods were titled 56D and 112D periods. Angus bulls were heavier ($P < 0.05$) than Brangus and Brahman bulls on d 56, 84, and 112. Initial and final BW for Angus, Brangus, and Brahman bulls were 292.7, 260.6, 230.4, and 468.3, 435.6, 350.7 12 kg. Conversely, Brahman bulls had greater hip height ($P < 0.05$) than Brangus with Brangus being taller ($P < .05$) than Angus. Angus bulls had the greatest ($P < 0.05$) scrotal circumference (SC) and Brahman bulls the least. Mean SC across days was 31.5, 29.7, 25.0 .6 cm for the three breeds. Serum testosterone was greater ($P < 0.01$) in Angus and Brangus bulls (10.0 and 8.9 1.4 ng/mL) than in Brahman bulls (4.0 1.4 ng/mL) throughout the study. Serum concentrations of IGF-I and leptin were greater ($P \leq 0.059$) in Angus bulls on d 56, 84, and 112 than in Brangus and Brahman bulls. Serum concentrations of GH ($P < 0.08$) and glucose ($P < 0.03$) were greater in Brangus bulls relative to Angus and Brahman bulls throughout the study. Prediction analyses suggested that breed and serum concentration of leptin could be used to predict ($P \leq 0.08$) BW and SC ($R^2 > 0.82$) in the 56D and 112D periods. Leptin and breed were also useful in predicting ($P \leq 0.09$) serum concentration of GH and testosterone in the 112D period ($R^2 > 0.32$). Residual correlation with the effect of breed removed indicated that leptin was positively correlated ($r \geq 0.53$, $P < 0.05$) with both SC and serum testosterone. Angus and Brahman cattle differ in phenotype, level of adiposity, and rate of sexual maturity. Data herein suggest that these characteristics could be due to varying mechanisms by which metabolic hormones such as leptin, GH, and IGF-I are regulated Work supported by NIH-SCORE GMO8136 and NMAES 180674.

Key Words: Bulls, Breed, Leptin

1604 LH and leptin pulsatile secretions are independent in ewe lambs.

S.E. Recabarren, A. Lobos, C.A. Vilches*, and P. Munoz, *University of Concepcion, Chillan, Chile*.

It has been proposed that leptin may be a metabolic signal regulating the onset of puberty in females and that stimulates also the secretion of GnRH from the hypothalamus. The objective of this work was to recognize if the secretion of leptin depends on the LH secretion in prepubertal sheep.Spring-born Suffolk ewe lambs of 20 weeks of age(n=5) received i.m. a long acting GnRH agonist (Decapeptyl®). Treatment was repeated at 24 and 28 weeks of age. Control lambs(n=6) received the vehicle. Pulsatility of LH and leptin was studied at 20 (before Decapeptyl injection), 26 and 30 weeks of age. For that, blood samples were collected at 10-min intervals for 6 hours. LH and leptin were measured in all samples by RIA and pulsatile hormone secretion characteristics assessed by the Cluster program. To further characterize the synchrony between LH and leptin pulses, GnRH (10 ng/kg BW) was injected at 60-min intervals for 6 times in other 5 ewe lambs of the same ages. The LH secretion was disminished in the Decapeptyl treated lambs (D) and was lower than in the control lambs (C) at 26 and 30 weeks of age. The transversal mean of plasma leptin concentrations (ng/mL/6h) did not change in the C lambs. In the D and in the GnRH lambs, mean leptin concentrations were higher ($P \leq 0.05$) at 30 weeks of age than at 20-weeks or 26-weeks. No difference in mean leptin concentrations was observed between groups at any age, except between the C and D lambs at 26-weeks(2.2±0.12 vs 1.42±0.11 ng/mL/6h, $P \leq 0.05$). Amplitude of leptin pulses was higher at 30 weeks of age than at 20 and 26-weeks in the D and GnRH lambs. Between groups, amplitude was higher in the C lambs than in the D lambs (2.6±0.15 vs 1.8±0.13 ng/mL, $P \leq 0.05$)at 26 weeks of age. No change in leptin pulse frequency was observed within and between groups. No coincidence between LH and leptin pulses was observed in any group. Data suggest that pulsatile leptin secretion is independent of LH secretion in ewe lambs. Supported by Fondecyt grant 1990389

Key Words: LH, Leptin, Ewe lamb

1605 Effect of obesity and fasting on leptin secretion and message expression in ewes.

J. A. Daniel*[1], B. K. Whitlock[1], J. A. Baker[1], B. Steele[1], C. D. Morrison[2], D. H. Keisler[2], T. H. Elsasser[3], and J. L. Sartin[1], [1]*Auburn University, Auburn, AL,* [2]*University of Missouri, Columbia, MO,* [3]*USDA, Beltsville, MD*.

The study was designed to examine the production and secretion of leptin by fat. Ewes (thin to obese) were assigned to either fed (F) or short-term restricted (R) groups. In the first experiment, subcutaneous fat samples were collected from ewes under local anesthesia from just above the last rib. Samples were analyzed for leptin expression using Northern blotting. In a second experiment, plasma samples were collected for leptin analysis from ewes every 15 min for 24 h beginning 48 h after initiation of feed restriction. Plasma concentrations of insulin, glucose, and NEFA were determined every 6 h. Fat thickness over the last rib was determined by ultrasound (0.025 to 2.18 cm). Expression of leptin mRNA did not differ in F vs. R ewes ($P > 0.14$). Profiles of plasma concentrations of leptin were episodic in nature, and did not differ in a diurnal manner. F ewes had greater leptin mean, area under the curve, number of peaks, peak height, and nadir than R ewes ($P < 0.05$), as determined by CLUSTER analysis. Additionally, F ewes had a shorter interval between peaks ($P < 0.054$). F ewes also had greater concentrations of insulin ($P < 0.01$) and glucose ($P < 0.01$) and lower concentrations of free fatty acids ($P < 0.01$) than R ewes. Fat thickness was correlated with leptin mean ($r = 0.73$, $P < 0.02$), area under the curve ($r = 0.73$, $P < 0.02$), peak height ($r = 0.71$, $P < 0.03$), and nadir ($r = 0.76$, $P < 0.02$) in F ewes and with leptin mean ($r = 0.89$, $P < 0.01$), area under the curve ($r = 0.89$, $P < 0.01$), number of peaks ($r = 0.69$, $P < 0.03$), peak height ($r = 0.87$, $P < 0.01$), and nadir ($r = 0.86$, $P < 0.01$) in R ewes. Fat thickness was also correlated with concentrations of insulin ($r = 0.88$, $P < 0.01$) in R ewes. At the 6-hour time points, leptin was correlated with insulin in F ewes ($r = 0.51$, $P < 0.01$) and in R ewes ($r = 0.59$, $P < 0.01$). These data provide evidence that profiles of plasma concentrations of leptin are episodic in nature and influenced by nutritive state and fat thickness.

Key Words: Leptin, Sheep

1606 Intracerebroventricular melanin-concentrating hormone stimulates food intake in sheep.

B.K. Whitlock*[1], L.A. Daniel[1], D.F. Buxton[1], F.C. Buonomo[2], C.J. Dyer[2], and J.L. Sartin[1], [1]*Auburn University,* [2]*Monsanto Company*.

Melanin concentrating hormone (MCH) is a peptide that is present in the sheep hypothalamus and stimulates feeding when injected intracerebroventricularly (i.c.v.) into rat brains. To clarify its role as a regulator of food intake in ruminants, we investigated the effect of MCH injected i.c.v. in sheep. The doses of MCH used were derived from previous experiments in our lab with neuropeptide Y (NPY), a known appetite regulator in sheep. Six, castrate male sheep were satiated and then received one of six treatments [saline and either 0.243, 2.43 or 12.1 μg/kg MCH, or MCH + NPY (0.243 and 0.432 μg/kg, respectively)] injected i.c.v. over 30 s. Food intake was measured for 2 h before and at 2, 4, 6, 8, 12, and 24 h after i.c.v. injection. In the second experiment, the same sheep were satiated and received one of four treatments [saline, 0.243, or 2.43 μg/kg MCH, and NPY (0.432 μg/kg)] injected i.c.v. over 30 s, then infused i.c.v. hourly for 6 h (500 μl/h). Food intake was measured for 2 h before and at 2, 4, and 6 h. Serial blood samples were collected at 15-min intervals from 2 h to 6 h relative to i.c.v. injection. In the first experiment, all doses of MCH resulted in greater ($P< 0.05$) food intake than saline or NPY, while NPY did not ($P = 0.36$) affect food intake relative to controls. In the second experiment, food intake was increased ($P< 0.03$) in sheep given the higher dose of MCH and in NPY treated sheep. Concentrations of nonesterified fatty acids (NEFA) were not ($P = 0.47$) affected by treatment. We conclude that a single i.c.v. injection of MCH stimulates an increase in food intake in sheep. Moreover, i.c.v. infusion of both MCH and NPY increase food intake above saline but did not affect NEFA concentrations. Previous research has determined that MCH is present in food intake regulatory regions of the sheep hypothalamus and MCH can be altered by physiological states associated with appetite. In this study we found i.c.v. MCH to stimulate food intake in sheep. Hence we propose that MCH qualifies as an endogenous appetite regulator in sheep.

Key Words: Melanin concentrating hormone, Food intake, Sheep

1607 GHRH-receptor is essential to the regulation of GH by GHS in cultured rat pituitary cells.

Sang-gun Roh*[1], Chen Chen[2], Ki-choon Choi[1], Shin-ichi Sasaki[1], and Chang Yoon[3], [1]*Lab of Animal Molecular Physiology, Faculty of Agriculture, Shinshu University, Naganoken, JAPAN ,* [2]*Endocrine Cell Biology Group, Prince Henry's Institute of Medical Research, Melbourne, Australia,* [3]*Dept of Animal Science, Iksan College, Iksan, Korea*.

Growth hormone (GH) is secreted in a pulsatile manner from anterior pituitary glands. Secretion of GH is stimulated by GH-releasing hormone (GHRH) and inhibited by somatostatin. The cloning of receptor

for synthetic GH secretagogues (GHS) and the finding of endogenous ligand for GHS-receptor (GHS-R), ghrelin, strongly suggested the presence of another important regulator for the secretion of GH. To address the problem of specificity, we selectively inhibited GHRH-receptor (GHRH-R) and GHS-R gene expression using modified antisense oligonucleotides (ON). We hypothesized that selective inhibition of the GHRH-R and/or GHS-R would disturb the cross talk between GHRH and GHS. To test our hypothesis, we measured the secretion of GH induced by GHS and GHRH challenge (30 min) in cultured male rat pituitary cells in the presence of antisense 18mer phosphorothiate ON targeting on the initiation codon region of GHRH-R and GHS-R mRNA for 3 days. We found a significant decrease in GHRH-R and GHS-R mRNA levels in the corresponding antisense-treated groups compared with the control group and the sense-treated condition. Treatment with GHS-R antisense ON showed the normal GHRH-stimulated GH secretion compared with that in control, lipofectamine and sense-treated cells. In addition, GHRP-2 (peptide GHS)-induced GH secretion was not inhibited by GHS-R antisense ON treatment either. In the cultured cell exposed to the GHRH-R antisense ON, GHRH-stimulated GH secretion was significantly diminished. GHRP-2-induced GH secretion was, however, not affected by such an exposure to the GHRH-R antisense ON. These results indicate a critical importance of GHRH-R for the GH secretion induced by both GHRH and GHS whereas GHS-R is only responsible for the GHS-stimulated GH secretion in cultured rat pituitary cells.

Key Words: Growth hormone, Growth hormone secretagogues, Antisense oligonucleotide

1608 Effect of growth hormone releasing factor (GRF) on long form leptin receptor (Ob-Rl) expression in porcine anterior pituitary. J. Lin*[1], C. R. Barb[2], R. R. Kraeling[2], and G. B. Rampacek[1], [1]*University of Georgia, Athens,* [2]*USDA-ARS, Athens, GA.*

We previously demonstrated that the Ob-Rl is expressed in the hypothalamus and anterior pituitary of the pig. Moreover, leptin stimulated basal GH secretion, but suppressed the GH response to GRF in pig anterior pituitary cells in culture. Pituitary cells from six pigs, 180 to 200 days of age, were studied in primary culture to determine if GRF affects Ob-Rl expression. On day 4 of culture, 10^5 cells / well were challenged with either 10^{-6}, 10^{-7} or 10^{-8} M [Ala^{15}]-hGRF-(1-29)NH_2. Secretion of GH into the media and pituitary Ob-Rl mRNA expression were determined at 4 h after treatment. Media were analyzed for GH by RIA and total RNA were isolated from cells for Ob-Rl expression by semi- quantitative RT-PCR. Basal GH secretion was 32 ± 2 ng/mL (n=6 pituitaries). Relative to control at 4 h, 10^{-6},10^{-7} and 10^{-8} M GRF increased ($P<0.0001$) GH secretion by 151%, 129% and 120% and decreased ($P<0.05$) Ob-Rl expression by 32%, 50% and 38%, respectively. These results indicate that GRF directly modulates Ob-Rl expression at the level of the pituitary and may play a role in regulating pituitary sensitivity to leptin.

Key Words: Pig, Leptin receptor, GRF

1609 Sequence and distribution of a single cDNA encoding growth hormone-releasing hormone-like peptide and pituitary adenylate cyclase activating polypeptide in channel catfish. B. Small* and D. Nonneman, *USDA/ARS Catfish Genetics Research Unit, Stoneville, MS.*

Significant advances have been made in channel catfish nutrition, reproduction and disease research over the past 30 years; however, the mechanisms controlling catfish growth have received little attention. Two neuropeptides which have been implicated in the regulation of fish growth are growth hormone-releasing hormone (GHRH) and pituitary adenylate cyclase activating polypeptide (PACAP). This study reports the sequence and tissue distribution of a single cDNA encoding both GHRH-like peptide (GHRHLP) and PACAP in channel catfish. The GHRHLP/PACAP precursor cDNA was cloned from catfish hypothalamic tissue and a brain cDNA library. Two cDNA variants of the gene were identified as a result of alternative splicing, a long form encoding both GHRHLP and PACAP, and a short form encoding only PACAP. The 110 bp deletion in the short precursor cDNA corresponds to the excision of exon 4, which encodes 71% of the GHRHLP sequence. The complete cDNA sequence was composed of 375 bp of 5-untranslated region containing a polymorphic compound microsatellite (CT-CA). The coding region covered nucleotides 376-900, containing a signal peptide from 376-435 bp (aa 1-20), GHRHLP from 625-759 bp (aa 84-128) and PACAP from 765-879 bp (aa 130-168). The 3-untranslated region contained a compound microsatellite (GC-GA) beginning within the stop codon. The consensus polyadenylation signal was located from 1058-1063 bp. Catfish PACAP and GHRHLP sequences share 89.5 and 31.1% identity with human PACAP and GHRH, respectively. Catfish GHRHLP is 80% identical to carp native GHRHLP sequence. Both the long and short form of the GHRHLP/PACAP precursor cDNA were identified in catfish brain, pituitary, fat, gastrointestinal tract, muscle, ovary and testes by RT-PCR detection; however, neither variant was detected in gill, heart, kidney, liver, pancreas, skin or spleen. This is the first demonstration of mRNA expression for this gene in fat or skeletal muscle of fish. By characterizing the GHRHLP/PACAP gene and its distribution in channel catfish, we have developed essential tools to investigate the roles of these peptides in the regulation of catfish growth.

Key Words: Channel catfish, GHRH, PACAP

1610 Development of specific antibodies for the quantification of plasma insulin-like growth factor-binding protein-3 in cattle. R. Renaville*[1], C. Bertozzi[1], S. Hetzel[1], I. Parmentier[1], S. Fontaine[1], V. Haezebroeck[1], and D. Portetelle[1], [1]*Gembloux Agricultural University, Animal and microbial biology unit, Gembloux, Belgium.*

Insulin-like growth factor-I and -II (IGF-I, IGF-II) circulate in biological fluids bounds to at least six different IGF-binding proteins that regulate IGF bioactivity. The IGF-binding protein-3 is regulated by growth hormone, and its concentration depends on nutrition and physiological state. Unavailable from commercial societies, bovine IGFBP-3 has been previously purified by acid precipitation, molecular filtration and affinity chromatography from pre-colostrum collected 3-5 days before parturition. This preparation has been used to produce specific mouse monoclonal and rabbit polyclonal antibodies and to develop a radioimmunoassay to quantify IGFBP-3 in bovine blood samples. Using the polyclonal antiserum, parallel displacement curves showed strong cross-reactivity with bovine, ovine, rabbit and human plasma protein and no cross-reactivity with porcine, rat, horse, dromedary or chicken plasma. Addition of IGF-I to a control pool of bovine plasma did not significantly alter control IGFBP-3 values in a radioimmunoassay. Nycthemeral periods, determined for three young bulls bled on two occasions were stable throughout the day; two or three samples were sufficient to characterize the animal. Heifers treated with recombinant bovine somatotropin (bST) had significantly higher serum levels of IGFBP-3 than did control animals. Likewise, plasma IGFBP-3 concentrations were decreased in growing bulls treated with clenbuterol (a b-agonist) while these concentrations increased after corticoid (dexamethasone) injection. Furthermore, IGFBP-3 levels were dramatically decreased during the first postpartum weeks. This radioimmunoassay for bovine IGFBP-3, which enables quantitative assessment of IGFBP-3 concentration in cattle, confirmed the previous observations using the less precise Western ligand blotting method. This research was supported by grants of the Belgian Ministry of Small Enterprises, Traders and Agriculture (grant 5736A) and Ministre de la Rgion Wallonne, subvention First Spin-Off (grant 991/3972).

Key Words: IGFBP-3, Cattle, RIA

1611 Responses of Holstein cows to low dose of somatotropin (bST) prepartum and postpartum. M. S. Gulay*, J Hayen, and H. H. Head, *University of Florida, Gainesville, FL.*

Objectives were to evaluate effects of a low dose of bST injected pre- and postpartum on BCS, BW, MY and IGF-I, INS and metabolites in plasma during experiment carried out during two consecutive years. Holstein cows (194) were assigned randomly to one of two groups [Control (C)=99 vs. Injected (I) =95 cows/group]. Biweekly injections of bST (POSILAC®) were prepartum (starting 21 d before expected calving) and through 60 d postpartum (C vs. I; 0 vs. 10.2 mg bST/d). After 60 d, all cows received a full dose of bST. IGF-I, INS, NEFA and glucose were measured in plasma from 82 of the cows (yr1) and BCS and BW of 112 cows (yr2); no blood samples were collected (yr2). MY of all cows were merged and analyzed using mathematical models that included main effects of treatment (bST), calving month, the 2-way interaction, and cow nested in treatment-calving month as error term. bST prepartum resulted in higher mean concentrations of INS (C vs. I; 0.650.037 vs. 0.780.042 ng/ml, $P<0.05$) and numerically greater NEFA (236.916.4 vs.,

281.616.4 μeq/l, P< 0.13). bST postpartum did not affect mean concentrations of glucose (66.410.84 vs. 66.740.84 mg/dl) or IGF-I (165.94.5 vs. 168.04.5 ng/ml. Postpartum treatment resulted in increased concentrations of NEFA (331.2 21.2 vs 410.6 20.8 μeq/l, P<0.02). No effects of bST treatment were observed on mean concentrations of INS (0.55 0.02 vs. 0.53 0.02 ng/ml), IGF-I (110.57.3 vs. 115.37.1 ng/ml), or glucose (66.131.68 vs. 64.451.65 mg/dl). Mean BCS did not differ prepartum, (3.730.02 vs. 3.760.02) around parturition (3.430.02 vs. 3.520.02),or postpartum (3.240.02 vs. 3.320.02). Although mean BW (kg) did not differ during prepartum (689.25.3 vs.722.83.3, P<0.12) and around calving (653.43.8 vs.664.55.1, P<0.45), bST injected group maintained BW better postpartum (618.53.6 vs.645.83.3, P<0.07). Greater MY was observed for bST-injected group during first 30-d (33.32 vs. 35.95 kg/d,P<0.06), and 60-d (36.92 vs. 39.42 kg/d, P<0.07) of lactation. No difference in MY was observed during the 100-d period (38.09 vs. 39.72 kg/d, P<0.22), which included full dose of bST (60d-100d; 39.72 vs. 40.30 kg/d, P<0.70). No adverse effects of bST treatment were observed during either the pre- or postpartum periods.

Key Words: Transition period, bST, Lactation

1612 Induction of lactation during winter and summer seasons in non-pregnant reproductive cull Holstein cows. M. Chahine*[1], W. J. Weber[1], J. K. Reneau[1], B. A. Crooker[1], T. H. Klusmeyer[2], M. F. McGrath[2], E. A. Reed[2], and J. L. Vicini[2], [1]*University of Minnesota, St. Paul,* [2]*Monsanto Animal Agriculture Group, St. Louis, MO.*

Effect of season on induction of lactation was assessed with multiparous cows induced in February (FEB, n=15) or June (JUN, n=18). Treatments started on day 0 and consisted of twice daily injections of 17β-estradiol (0.1 mg/kg BW/d) and progesterone (0.25 mg/kg BW/d) for 7 d, twice daily mammary massage (2-3 min/gland) for the next 6 d, and an injection of dexamethasone (0.05 mg/kg BW/d) on day 13 of study. Cows received POSILAC 1 STEP® (500 mg bST) on day 0, 10, 20, and 30 of study and at 14 d intervals thereafter. Milking (3x/d) commenced on day 14 of study and continued for 122 d. Samples from each milking on a single day in weeks 1, 2, 3, 4, 8, 12 and 16 of lactation were collected for analyses. Effect of season was determined using GLM of SAS. Cows were non-responsive and removed if they failed to produce $\geq$ 9.1 kg of milk on a single day by 24 d of lactation. Induction was successful if at least one daily total milk yield was $\geq$ 13.6 kg by 50 d of lactation. Two cows failed to respond in each season and one JUN cow responded but was removed for health reasons on day 12 of lactation. Thus, induction was successful in 13 of 15 (86.7%) FEB cows and 15 of 18 (83.3%) JUN cows. Of these cows, one FEB and two JUN cows were removed prior to day 70 of lactation for health reasons. Shape of the lactation curve was similar for both seasons. Total 122 d yields of actual milk (2966, 2729 kg), 3.5% FCM (3077, 2964 kg), and 4.0% SCM (2875, 2722 kg) did not differ between seasons. Daily yield per cow averaged 25.0 kg milk, 27.6 kg FCM, and 25.5 kg SCM. Milk composition did not differ between FEB and JUN cows and averaged 4.2% fat, 3.4% protein, 4.9% lactose and 174 x 10^3 SCC/ml. In this study, induction was successful in 85% of the cows and milk yield was not affected by season.

Key Words: Induced Lactation, bST, Season

1613 Reduced milk ejection as a consequence of chronic treatment with exogenous oxytocin in cows. R. M. Bruckmaier*, *Institute of Physiology, Techn. Univ. Munich-Weihenstpehan, Freising, Germany.*

Oxytocin (OT), released from the posterior pituitary in response to mechanical teat stimulation, causes myoepithelial contraction and hence alveolar milk ejection during milking. In dairy practice, exogenous OT is often administered before milking for therapy of disturbed milk ejection due to lacking or reduced OT release. The objective of this study was to test the hypothesis that chronic OT administration at each milking causes reduced milk ejection after withdrawal of exogenous OT. Experiments were performed with twenty-one healthy Brown Swiss dairy cows with normal milk ejection during machine milking at 5 a.m. and 4 p.m. One min before the start of each milking cows were injected i.m. either with 50 I.U. of synthetic OT (n = 7; OT-G) or with an equal volume of 0.9 % NaCl (n =7; NaCl-G) for three weeks. Control animals (n = 7; CO-G) did not receive daily pre-milking injections. At evening milkings on d 0, 7, 14, and 21, no pre-milking injections were performed and residual milk was obtained by i.v. injection of 10 I.U. OT at the end of milking. In addition, residual milk was obtained at the end of evening milking on d 1, 8, 15 and 22, when each group received a pre-milking injection of 50 I.U. OT i.m. Total milk yield as a sum of spontaneously removed milk (including stripping) and residual milk was set 100 %. On d 0, 952, 903 and 942 % of milk were spontaneously obtained before removal of residual milk in groups OT-G, NaCl-G and CO-G, resp. Spontaneously removed milk was reduced in OT-G on d 7, 14, and 21 (p<0.05) to 825, 816 and 814 %, resp., but was not altered in NaCl-G and CO-G. Pre-milking OT injection on d 1 did not significantly increase the spontaneously removed milk fraction (961, 941 and 971 % in OT-G, NaCl-G and CO-G, resp.). Similar results as on d 1 were obtained on d 8, 15 and 22. In conclusion, chronic OT administration caused a rapid decline of spontaneous milk ejection. Repeated NaCl injection did not reduce milk ejection thus excluding stress effects of the injection procedure. As the response to exogenous OT remained unchanged throughout the experiment, the diminished milk ejection after chronic OT treatment is rather due to reduced OT release than to OT-receptor down regulation.

Key Words: Milk Ejection, Cow, Chronic Oxytocin Treatment

1614 Gene expression of immunologically relevant factors in blood cells, milk cells and mammary tissue of cows. R. M. Bruckmaier*, S. L. Wittmann, H. H. D. Meyer, and M. W. Pfaffl, *Institute of Physiology, Techn. Univ. Munich-Weihenstephan, Freising, Germany.*

Tumor necrosis factor a (TNFa) and eicosanoids are involved in the mammary gland's immune response. Lactoferrin (Lf) has bacteriostatic effects. The goal of this study was to localize and quantify the synthesis of these factors in milk, blood and mammary tissue cells of cows exhibiting distinct levels of somatic cell counts (SCC) while lacking clinical signs of mastitis. mRNA expression of TNFa, Lf and key enzymes of leukotriene and prostaglandin synthesis, 5-lipoxygenase (5-LO) and cyclooxygenase (COX)-1 and COX-2, resp., was determined in Brown Swiss cows. Control animals (CTR; n=8) had < 150,000 cells/ml of milk in all quarters (CQ) while cows with partially elevated SCC (PE; n=7) had minimum one quarter > 150,000 (HQ) and one quarter < 150,000 cells/ml (LQ). Milk of one CQ of CTR and one HQ and LQ of PE was collected and a blood sample was taken. In addition, mammary tissue samples were obtained from the respective quarters. Total RNA was isolated, reverse transcription and quantitative real-time PCR were performed. TNFa and COX-2 were predominantly expressed in milk cells, 5-LO in blood cells whereas Lf was mainly found in mammary tissue. COX-1 was expressed at similar levels in all cell/tissue types tested. None of the parameters investigated was significantly different between CTR and PE in blood cells (TNFa: 1.00.3 and 0.90.3; COX-1: 0.80.3 and 1.00.3; COX-2: 0.60.2 and 1.91.1; 5-LO: 3.21.7 and 1.00.2; Lf: 0.10.05 and 0.20.1 molecules/cell, resp.). TNFa mRNA expression was elevated in HQ as compared to LQ in mammary tissue (1246 and 277 molecules/mg, resp.; P<0.05). COX-2 mRNA expression was higher in milk cells of HQ than LQ (24.25.8 and 12.94.6 molecules/cell, resp.; P<0.05). 5-LO expression tended to be elevated in milk cells and Lf in mammary tissue of HQ as compared to LQ. No difference was found between CQ and LQ in milk cells and in mammary tissue for any of the parameters tested. In conclusion, the immunological activity of the mammary gland is quarter specific. Besides mammary tissue, somatic milk cells seem to be an important source of factors involved in chemical signalling during immune response.

Key Words: Mammary Gland, Immunology, Chemical Signalling

1615 Effect of intramammary infusion of Escherichia coli endotoxin on ovulation in lactating dairy cows. A. M. Nugent, T. B. Hatler, S. H. Hayes, S. C. Kiggins, and W. J. Silvia*, *University of Kentucky, Lexington.*

The purpose of this experiment was to determine if intramammary inflammation during the periovulatory period in lactating dairy cows affects the occurrence of ovulation. Ten lactating, cyclic, Holstein dairy cows received 2 injections of prostaglandin F2a, at 11-day intervals, to synchronize luteolysis. The day of the second injection was designated as Day 0. Ovulation was anticipated on days 3-5. Beginning on day 1, cows received intramammary infusions of either Escherichia coli endotoxin (10 ug; n=5) or infusion vehicle (controls; n=5) into 2 quarters immediately after milking. The same quarters were infused after each milking through day 4. Venous blood samples were collected daily

from day -1 through 13 for determination of progesterone to monitor luteolysis and formation of the new corpus luteum. Samples were also collected at 4-h intervals (days -1 to 2), then at 2-h intervals (days 2 to 5) to measure concentrations of luteinizing hormone (LH). Ovaries were examined ultrasonographicly on days -1 through 5 and on day 12 to monitor follicular growth and formation of the corpus luteum. Collectively, these observations were used to determine if and when ovulation occurred. Intramammary infusion of E. coli endotoxin induced an immediate increase in somatic cell numbers in treated quarters. However, this treatment had no effect on the occurrence or timing of ovulation. Four of 5 cows in each treatment group ovulated. Preovulatory surges of LH were detected within the intensive bleeding periods for 3 cows in each treatment group. The magnitude of the LH surge was reduced in cows receiving endotoxin. Supported by KABA/Select Sires and the KY Agr. Expt. Station.

Key Words: ovulation, luteinizing hormone, endotoxin

1616 Effects of N-nitro-arginine on blood flow and nutrient uptake in the mammary glands of dairy cows. T. G. Madsen*[1], D. R. Trout[2], S. Cieslar[3], N. G. Purdie[4], M. O. Nielsen[1], and J. P. Cant[3], [1]*Department of Anatomy and Physiology, The Royal Veterinary and Agricultural University, Denmark,* [3]*Department of Animal and Poultry Science, University of Guelph, Canada,* [2]*Department of Clinical Studies, University of Guelph, Canada,* [4]*School of Land and Food Sciences, University of Queensland, Australia.*

Nitric oxide (NO) has been shown to be a potent vasodilator in many tissues and previous research demonstrated that the arginine analogue N-nitro-arginine, which blocks NO synthesis from L-arginine, decreased mammary blood flow (MBF) in dairy goats. The objective of this study was to determine if N-nitro-arginine has the same effect on MBF in dairy cows and, if so, how changes in blood flow affect mammary metabolite uptakes, i.e. arterio-venous (AV) difference multiplied by MBF. Two multiparous Holstein cows in mid-lactation were fed and milked twice daily at 8:00 and 18:00 h. Between morning and evening milking on two consecutive days, saline or a mixture of amino acids (40 g/h) was infused for 9 h into the external iliac artery supplying one udder half. At 15:00 h N-nitro-arginine was added to the infusion at 25 mg/min for 60 min. Blood samples were taken approximately 25 min before and 10, 30, 50, 90 and 140 min after start of N-nitro-arginine infusion for estimation of MBF using the para-amino hippuric acid dilution technique. Blood flow decreased approximately 30 % during infusion of N-nitro-arginine and remained low for at least one hour after the infusion stopped. AV differences for acetate, β-hydroxy butyrate (BHB) and triacylglycerol (TAG) increased when the blood flow dropped; extraction efficiency increased 13, 38 and 14 %, respectively, for these three nutrients. Non-esterified fatty acid AV differences correlated negatively with AV differences of TAG. In conclusion, it is possible to decrease blood flow to the mammary glands of dairy cows by close arterial infusion of a NO synthesis blocker. The mammary glands compensated for the decreased nutrient supply resulting from the lowered blood flow by increasing the efficiency of nutrient extraction from blood.

Key Words: Blood flow, Nitric Oxide, Arginine analogue

1617 Effects of an induced mammogenesis and lactogenesis in sheep on the mRNA expression levels of immune globulin receptors (FcRn; pIGR) and zona occludens proteins (ZO1; ZO2; ZO3). MW Pfaffl*, A Dzidic, P Rojas, RM Bruckmaier, and D Schams, *Institute of Physiology, Technical University of Munich, Freising-Weihenstephan, Germany.*

Mammary immune response is mediated mainly through phagocytotic leucocytes but to some extent by secreted immune globulins A (IGA) and G (IGG). Tight junction (TJ) barrier, built by zona occludens proteins 1–3 (ZO1–3), limit the movement of immunoactive leucocytes from the blood circulation into milk. During pregnancy TJs are leaky, undergoing a closure shortly before parturition to remain impermeable during lactation. IGG and IGA are transported transcellularly into secretion by IGG receptor (FcRn) and polymeric IG receptor (pIGR), respectively. Influence of steriods, dexamethason (DEX) and prolactin (PRL), to induce mammogenesis (MG) and lactogenesis (LG), and effects on the expression profile were investigated in sheep. MG was induced by a frequent estrogen and progesterone treatment (day 1–29), and LG via DEX and manual stimulation (day 30–37). In one group PRL secretion was inhibited by bromocryptin (BR+, n=4) and compared with BR- (n=4) and a non treated control (K, n=4). Sheep were slaughtered (day 38) and mammary tissue was sampled for RNA extraction. Expression studies were done in real-time RT-PCR and each sample was normalised to internal ubiquitin expression. Relative expression levels of BR+ and BR- were compared with the control group (K=1). MG and LG could be induced and histology showed an alveolar development in BR- higher than in BR+, and a white secret could be retrieved by hand milking with physiological concentrations of fat, protein, lactose and somatic cells. PRL concentrations were successfully suppressed in BR+. Expression data showed no significant changes in pIGR, ZO1 and ZO3. FcRn and ZO2 expression were significantly elevated in BR+ and BR-. Only a trend of higher pIGR and ZO3 expression could be shown in BR- compared to BR+. In conclusion, the PRL inhibition and desired blockage of LG through BR had no effects in sheep. FcRn and ZO2 concentrations could be up-regulated, influenced by steroids and DEX, resulting in a higher IGG concentration in colostrum milk and impermeable TJ. No effects of BR treatment could be observed.

	FcRn	pIGR	ZO1	ZO2	ZO3
BR-	3.40±2.97 p=0.02	2.57±2.50 p=0.07	1.04±0.31 p=0.74	2.42±0.40 p≤0.001	2.17±1.45 p=0.07
BR+	4.27±3.28 p=0.008	1.11±0.81 p=0.67	1.03±0.27 p=0.73	2.02±0.21 p≤0.001	1.70±1.10 p=0.14
K	1	1	1	1	1

Key Words: Induced Mammogenesis and Lactogenesis, IG receptors, Zona occludens proteins, Relative expression in real-time RT-PCR

1618 Effect of chromium-methionine level supplementation on inmune response of bull calves recently arrived to feedlot. L. Almeida*[1] and R. Barajas[1], [1]*FMVZ-Universidad Autonoma de Sinaloa.*

To determinate the effect of chromium-methionine level supplementation on immune response of calves recently arrived to feedlot, twenty bull calves (7/8 Brahman; 167 kg) were used in a 28 days complete randomized design experiment. After 300 km truck transportation the animals were randomized assigned (five by treatment) to receive one of four diets in that consist the treatments: 1)Diet 30:70 roughage:concentrate with 15% CP and 1.4 Mcal NEm/kg (control); 2)Diet similar to control, but supplemented with 0.4 ppm of chromium from chromium-methionine (Cr-Met; Microplex^{TM}, Zinpro, CO); 3)Diet supplemented with 0.7 ppm of Cr from Cr-Met; and 4)Diet supplemented with 1.0 ppm of Cr from Cr-Met. The animals were allocated in ground pens with central shade, fed twice a day with permanent access to fresh tap water. Next day to arrived, animals were dewormed, vaccined again IBR, PI3, BRSV,BVD, Clostridium, and Pasteurella; weighed, tagged, and ear hormone implanted. Blood samples were taken from jugular vein at start the experiment, and in days 7, 14 and 28. Cortisol, creatinin, glucose, globulin G, globulin M, and aspartate aminotransferase enzyme were determinate. After seven days, chromium supplementation tended (P<0.09) to diminished in 60% blood cortisol concentration (3.6 vs. 1.4 μg/dL), and at 14 days 1.0 ppm of Cr reduce (P<0.04) cortisol (2.8 vs. 0.2 μg/dL). Chromium 1.0 ppm increased (P<0.03) immunoglobulin G levels in days 14 and 28. Immunoglobulin M tended (P<0.14) to be increased by 1.0 ppm Cr supplement. Addition of 0.7 and 1.0 ppm of chromium tended (p<0.07) to increase glucose level in day 28. Aspartate aminotransferase enzyme concentrations tended to decrease (P<0.07) in day 7 with 1.0 ppm Cr treatment. Creatinin was not affected (P>0.20) by treatments. It is concluded that chromium-methionine supplementation improve the immune status of bull calves recently arrived to feedlot and that 1 ppm of Cr appears to be eligible level.

Key Words: Chromium, Supplementation, Bull calves

1619 The effect of equi-molar dietary betaine and choline additions on liver, plasma and gut of pig. K. Tiihonen*[1], S. Peuranen[1], H. Siljander-Rasi[2], and H.P. Simmins[3], [1]*Danisco Cultor Innovation Center, Kantvik, Finland,* [2]*Agricultural Research Centre of Finland, Hyvink, Finland,* [3]*Finnfeeds International Ltd., Marlborough, Wilts, UK.*

The mode of action of dietary betaine and choline in pigs were studied. In broiler chicks dietary betaine is twice as efficient as equi-molar dietary

choline for increasing liver betaine levels but in pigs that has not been studied previously. Individually penned Finnish Landrace and Yorkshire pigs (30 kg; n=70) were fed basal diet with no added betaine or choline, the basal diet supplemented with 250, 500 or 1000 mg/kg of betaine (Betafin® S1), or with a similar molar amount of choline (578, 1155 or 2310 mg/kg of choline chloride). The net energy content of the maize-soybean meal basal diet was diluted with oat hull meal (100 g/kg) and it contained 8.55 MJ/kg NE, 155 g/kg crude protein and 7.4, 4.4 and 4.3 g/kg digestible lysine, threonine and methionine+cystine, respectively. The pigs were on restricted diet, 1.5-3.0 kg feed/d. The experiment lasted 75 days. Daily weight gain improved linearly ($p \leq 0.01$) with increasing dietary betaine. The liver betaine level increased linearly with dietary betaine addition ($p \leq 0.05$). The additives had no significant effect ($p \geq 0.10$) on plasma homocysteine levels. Addition of betaine tended to improve the tensile strength of the proximal ileum linearly ($p=0.07$). Choline additions increased plasma carnitine linearly ($p \leq 0.01$) but had no effect on the pig performance, liver betaine or gut tensile strength. The results show that dietary betaine addition increased liver betaine and improved the daily weight gain of pigs on restricted diets with diluted energy concentration whereas choline addition increased plasma carnitine but had no effect on performance.

Betaine, mg/kg	0	250	500	1000	0	0	0	
Choline chloride, mg/kg	0	0	0	0	578	1155	2310	SEM
Daily weight gain, g	883	879	943	969	897	909	906	22.2
Liver betaine, mg/g	0.038	0.041	0.051	0.061	0.037	0.044	0.042	0.007
Plasma carnitine, mg/l	1.26	1.39	1.41	1.30	1.31	1.37	2.80	0.20
Gut tensile strength, kg	2.10	2.44	2.14	2.80	2.41	2.52	2.16	0.26

Key Words: Betaine, Choline, Pig

PSA Physiology: Cardiopulmonary, Immune, and Other Physiology

1620 Differences of autonomic nervous system activity in high and low body weight-selected chickens. A. Y. Kuo*[1], J. C. Lee[2], P. B. Siegel[1], and D. M. Denbow[1], [1]*Virginia Tech, Blacksburg,* [2]*VA-MD Regional Veterinary College, Blacksburg.*

This study was to investigate whether there are differences in the autonomic nervous system (ANS) function of chickens from lines selected or high (HWS) or low body-weight (LWS). The cardiovascular response to pharmacological agents was used as an indicator of ANS response. Ten birds from each line and sex were used. Catheters were introduced into the left brachial artery and vein, and connected to a MP100-BIOPAC system to record blood pressure (BP) and heart rate (HR). Birds were injected with phenylephrine, atropine, propranolol, and tetraethylammonium chloride (TEAC). Data were analyzed using ANOVA; significant differences imply $P \leq .05$. The LWS birds exhibited a greater increase in BP and less increase in HR than the HWS birds following atropine. The response to atropine showed a line and sex interaction, in which male birds had a greater increase in HR than females, and LWS females had a lower increase in HR than the HWS females. Injection of phenylephrine following pretreatment with atropine caused a baroreceptor reflex in which males showed a greater decrease in HR than females. In response to the beta-adrenergic receptor blocker propranolol, females displayed a greater decrease in BP than males, and LWS birds had a greater decrease in HR than HWS birds. In response to the autonomic ganglionic blocker TEAC, BP and HR were decreased equally in both lines. The percentage of adrenal and sympathetic impact on regulation of HR showed that LWS females required greater adrenal activity than the other birds. While change in the HR to BP ratio in response to phenylephrine was different between lines, the response was not different when phenylephrine was given following atropine. These results suggest that a higher parasympathetic nervous system tone is present in the HWS, and a higher sympathetic nervous system tone is present in the LWS than HWS birds. It is suggested that differences between the lines could be at the level of the adrenal gland.

Key Words: Autonomic nervous system, Blood pressure, Chickens

1621 Hemodynamic Responses of Broiler Pulmonary Vasculature to Intravenously Infused Serotonin. M. E. Chapman* and R. F. Wideman, *University of Arkansas, Fayetteville, AR, USA.*

Serotonin (5-hydroxytryptamine, 5HT) is a potent pulmonary vasoconstrictor actively accumulated by mammalian platelets and avian thrombocytes, and released into the plasma during platelet or thrombocyte aggregation. 5HT has been implicated in the mechanisms responsible for pulmonary hypertension in several human and animal studies. However, the role of 5HT in pulmonary hypertension syndrome (PHS, ascites) in broilers previously had not been evaluated. In the present study we evaluated the pulmonary hemodynamic responses of broilers to intravenous infusions of 5HT dissolved in 2.5% mannitol solution (carrier vehicle). Carrier vehicle infusion alone had no influence on any of the hemodynamic variables. 5HT infusion triggered rapid increases in pulmonary arterial pressure to approximately 50% above pre-infusion baseline values, accompanied by decreases in mean systemic arterial pressure and cardiac output. The peak pulmonary arterial pressure response occurred within approximately 70 s after the start of 5HT infusion, and remained elevated above baseline values over the course of a 10-minute infusion period. Pulmonary arterial pressure, mean systemic arterial pressure and cardiac output returned to pre-infusion baseline values upon cessation of 5HT infusion. Pulmonary hypertensive responses were associated with increased pulmonary vascular resistance (pulmonary vasoconstriction). The peak pulmonary arterial pressure attainable was inadequate to propel the normal cardiac output through the elevated pulmonary vascular resistance. Consequently, the impeded venous return to the left ventricle caused dependant reductions in stroke volume, cardiac output, and mean systemic arterial pressure. Reductions in cardiac output were associated with reductions in stroke volume but not heart rate. Any factor that reduces the pulmonary vascular capacity or increases the pulmonary vascular resistance theoretically can increase the incidence of PHS. The present study provides direct evidence that 5HT can trigger pulmonary vasoconstriction and pulmonary hypertension in broilers.

Key Words: Serotonin, Broiler, Hypertension

1622 Pulmonary Wedge Pressures Confirm Pulmonary Hypertension in Broilers is Initiated by an Excessive Pulmonary Arterial Resistance. M. E. Chapman* and R. F. Wideman, *University of Arkansas, Fayetteville, AR, USA.*.

High retrograde pressure through the pulmonary venous system caused by failure of the left ventricle or left atrio-ventricular valve may result in the elevated pulmonary arterial pressure and right ventricular hypertrophy associated with pulmonary hypertension syndrome (PHS; ascites) in broilers. Unanaesthetized male broilers from an ascites-resistant line, the base population from which the resistant line was derived, and a separate unselected line were used to determine whether changes in wedge pressure are predictive of differences in the pulmonary arterial pressure of clinically healthy and pre-ascitic broilers. Venous, right atrial, right ventricular, pulmonary arterial, and wedge pressures were obtained by inserting a catheter into a wing vein and progressively advancing the catheter into a pulmonary branch artery until the catheter tip became wedged in and occluded the flow through a terminal artery. Mean right ventricular and pulmonary arterial pressures were lower in the resistant line than in the base population, but wedge pressures did not differ between the resistant, base, and unselected lines. Right:total ventricular weight ratios (RV:TV) and the percentage saturation of hemoglobin with oxygen in arterial blood ranged in value from 0.18 to 0.44 and 65 to 96%, respectively. Wedge pressure, however, remained similar when pre-ascitic broilers with high RV:TV values and low oximetry values were compared with clinically healthy broilers. In all birds, whether healthy or showing pre-ascitic characteristics, the wedge pressure was slightly