W141 Effect of leukemia inhibitory factor on feed intake and body temperature in sheep. J. L. Sartin*[1], D. L. Marks[2], B. K. Whitlock[3], J. A. Daniel[4], and B. P. Steele[1], [1]*Auburn University*, *Auburn, AL,*[2]*Oregon Health Sciences University*, *Portland,* [3]*University of Tennessee*, *Knoxville,* [4]*Berry College*, *Mt Bery, GA*.

Leukemia inhibitory factor (LIF) has been suggested to function as a potent inhibitor of feed intake in rodents. These studies were designed to determine whether LIF was found in the ovine hypothalamus and whether LIF inhibited feed intake in sheep. Sheep hypothalami were used to clone LIF to indicate presence of the gene in the hypothalamus. The sequence was similar to published data. Another group of sheep were provided intracerebroventricular (ICV) cannulas and injected with doses of LIF at 250, 500, 1000 and 2500 ng per sheep, ICV. Feed intake was inhibited by the 1000 and 2500 ng dose (trt, $P < 0.0001$; time × trt, $P < 0.02$). All doses of LIF elevated temperature above 40 C, indicating a fever. In a second experiment, the sheep were injected ICV with 2500 ng LIF, and blood samples collected at 10 min intervals for 6 h for assay of luteinizing hormone (LH), growth hormone (GH) and 30 min interval samples assayed for glucose and free fatty acids. There was no effect of LIF on GH. There was no effect of trt for LH, but there was a time × trt interaction indicating reduced LH ($P < 0.0001$). There was an effect of trt and a time × trt interaction indicating elevated plasma free fatty acids ($P < 0.03$; 0.001) and glucose ($P < 0.006$; 0.0001), opposite to the effects of LIF in rodent models. The effects of LIF on feed intake and other parameters is similar to the effects of LPS and leads to a hypothesis that LIF expression in response to LPS may be a component of the mechanism for feed intake inhibition in disease.

Key Words: appetite, free fatty acids, luteinizing hormone

W142 Effects of late gestation metabolizable protein (MP) supplementation on ewe organ and blood parameters. T. J. Swanson*[1], L. A. Lekatz[1], T. L. Neville[1], M. L. Van Enom[2], C. S. Schauer[2], K. R. Maddock Carlin[1], C. J. Hammer[1], and K. A. Vonnahme[1], [1]*North Dakota State University*, *Fargo*, [2]*Hettinger Research Extension Center*, *Hettinger, ND*.

To examine the effects of maternal MP supplementation in late gestation on blood and organ parameters multiparous ewes (n = 45) were allotted randomly to one of 3 treatments, 75% (LOW), 100% (CON), or 125% (HIGH) of MP requirements fed from d 100 to d 130 of gestation. Blood samples were drawn before initiation of diets and before slaughter for chemistry panel analysis. Body measurements including loin eye area, back fat, and body wall thickness where taken using ultrasound before treatment initiation and before slaughter to examine changes in body condition. At d 130 ewes were slaughtered and dissected. Ewes carried singletons and twins therefore fetal number was included as a main effect. There was no effect of treatment or fetal number on final BW, loin eye area, back fat, body wall thickness, eviscerated body weight (EBW), or weights of blood, perirenal fat, thyroid, and adrenals ($P \geq 0.06$). There was a treatment effect on heart weight with CON being heavier than LOW and HIGH which were not different ($P \leq 0.01$). Kidney weight was also affected by treatment with LOW being lighter compared with HIGH and CON which were not different ($P \leq 0.01$). Ewes carrying twins had increased liver, mammary, and gravid uterus weights ($P \leq 0.03$). Ewes with singletons had increased lung weights compared with ewes carrying twins ($P \leq 0.03$). When organ weight was examined as a proportion of EBW (g/kg) there was no difference in heart, perirenal fat, kidney, lung, or thyroid masses ($P \geq 0.06$). Ewes carrying twins had increased blood, liver, mammary, and gravid uterus weights as a proportion of EBW ($P \leq 0.02$). Initial chemistry panel results showed no differences in parameters of interest. Treatment decreased aspartate aminotransferase and blood urea nitrogen ($P \leq 0.01$) in LOW ewes compared with HIGH and CON ewes which did not differ. Change in gamma-glutamyl transpeptidase was greater in ewes carrying twins ($P \leq 0.01$). Results indicate that litter size and dietary MP supplementation during late gestation alters ewe organ weights.

Key Words: gestation, metabolizable protein, organ weights

W143 Effect of to PFKM and TFDP2 gene expression on muscle growth in sheep. J. W. Buchanan*[1], M. L. Thonney[2], and R. G. Mateescu[1], [1]*Oklahoma State University*, *Stillwater*, [2]*Cornell University*, *Ithaca, New York*.

Muscle growth rate in response to testosterone is an important characteristic of meat producing species utilized in production agriculture. The onset of puberty and subsequent testosterone release in males causes differential gene expression within sexually dimorphic muscles, resulting in measurable hypertrophy. Phosphofructokinase muscle type (PFKM) and transcription factor Dp-2 (TFDP2) are 2 genes recently shown to have significantly higher transcript levels in callipyge lambs. PFKM is a kinase directly involved in glycolysis, and TFDP2 is a transcription factor that has been implicated in the initiation of the cell cycle as well as DNA replication. The objective of this project was to compare the expression of these 2 genes in 2 different skeletal muscles in sheep. Muscle samples were collected from 18 sets of twins, with one individual from each set castrated at birth. Each set of twins was randomly assigned to 4 groups corresponding to 77, 105, 133, and 161 d of age at slaughter. Total RNA was extracted from semitendinosus (non-sexually dimorphic) and splenius (sexually dimorphic) muscle samples collected from each individual. Two-step reverse transcription polymerase chain reaction (RT-PCR) utilizing oligo(dT) primers was used to synthesize complimentary DNA. A quantitative polymerase chain reaction (q-PCR) was then performed to quantify the amount of PFKM and TFDP2 mRNA transcript in the muscle samples. The difference in gene expression between the SP and ST muscles was analyzed using the General Linear Model procedure of SAS to determine the effect of sex, age class and their interaction. No statistically significant difference was found in PFKM or TFDP2 mRNA level between the 2 muscles in rams and wethers at any age. Identifying genes that control muscle hypertrophy has the potential to create increased gains and efficiency in populations utilizing a marker assisted selection program. Further expression analysis is needed to determine the response of other candidate genes controlling hypertrophy in sexually dimorphic muscles.

Key Words: gene expression, muscle growth, sheep

W144 Excessive maternal selenium intake induces inflammatory response in the ovine fetal gut. H. Wang*[1], J. Zhao[1], Y. Huang[1], X. Yan[1], A. Meyer[2], M. Du[1], K. Vonnahme[2], L. Reynolds[2], J. Caton[2], and M. J. Zhu[1], [1]*Department of Animal Science, University of Wyoming*, *Laramie,* [2]*Department of Animal Science, North Dakota State University*, *Fargo*.

Selenium (Se) is a coenzyme for glutathione peroxidase and thioredoxin reductase, and its dietary supplementation has anti-inflammatory effects.

Many areas of North Dakota and other states have soils enriched in Se, rendering sheep and cattle grazing in these areas ingesting excessive Se. The objective was to evaluate the effects of higher energy diets and elevated Se intake on inflammatory response in lamb ileal tissues, a major immune organ. Rambouillet ewes (age = 240 ± 17 d; initial BW = 52.1 ± 6.2 kg) were fed either a control diet (adequate Se: 11.5μg Se.kg $BW^{-1}.d^{-1}$) or a high Se diet (77.0 μg Se.kg $BW^{-1}.d^{-1}$) with Se provide as Se-enriched wheat millrun. On 40 d of gestation (dG) ewes in each Se group were assigned randomly to 100%, or 140% of dietary requirements except for Se. At parturition, offspring were removed before nursing and raised independent of the dams until necropsy at 20 d of age. Offspring ileal tissues were sampled for immunoblotting and qRT-PCR analyses. Control, high energy (COB) and control, high Se (SEC) had no major difference in inflammatory signaling compared with control, normal Se (CC). However, high inflammatory signaling was detected in high energy, high Se (SEOB) group, as shown by increased ($P < 0.05$) mRNA expression of tumor necrosis factor (TNF)α and chemotaxis interleukin (IL)8. Consistent with this, phosphorylation of c-Jun N-terminal kinase (JNK), a primary inflammatory signaling, was greater ($P < 0.05$) in ileal tissue from offspring of SEOB treated dams. Both mRNA and protein content of transforming growth factor (TGF) β was also greater ($P < 0.05$) in SEOB lambs. No difference in mRNA expression of IL6, CD14, IL1α and β, and toll-like receptor (TLR) 2 and 4 were observed. In conclusion, high Se or maternal gestational high energy diet had no major effects on inflammatory signaling, but combining high energy diet with excessive Se induced inflammatory response in offspring intestine.
INBRE P20RR016474; USDA-NRI 2008–35203–19084.

Key Words: selenium, ovine, inflammation

W145 Serum concentrations of ghrelin, IGF-I, and prolactin in Rambouillet lambs during the preweaning period. C. D. Felker*, M. J. Hendricks, K. A. Jurado, A. D. Stapp, L. E. Camacho, and D. M. Hallford, *New Mexico State University*, *Las Cruces*.

During a 60-d preweaning period in each of 2 yr, Rambouillet lambs (n = 75 and 52 for yr 1 and 2, respectively) were used to examine effects of sex (77 females, 50 males) and type of birth (TOB; 46 single, 81 multiple lambs) on serum concentrations of ghrelin (total), IGF-I, and prolactin (PRL). Lambs were born in mid-March of each year and were weighed on the day of birth (d 0; 5.1 ± 0.1 kg) and at weaning (60 d, 20.1 ± 0.4 kg). Serum was harvested by centrifugation from blood collected (jugular venipuncture) on d 1, 14, 28, 42, and at weaning. Data were examined by ANOVA for a randomized complete block design with a 2 (sex) x 2 (TOB) factorial arrangement, sampling day as a repeated measure, and year as the block. Males were heavier at birth than females ($P < 0.001$), but weaning weight and ADG were similar ($P > 0.35$). Single lambs were heavier ($P < 0.001$) at birth and weaning and had greater ADG ($P < 0.001$) than multiple lambs. Serum ghrelin was similar ($P > 0.24$) in male and female and in single and multiple lambs. However, ghrelin declined during the preweaning period with values of 557, 373, and 358 (±9) pg/mL (quadratic, $P < 0.001$) on d 1, 28, and 60, respectively. Serum PRL and IGF-I were influenced ($P < 0.05$) by sex × day and TOB × day interactions. Males and females had similar ($P = 0.23$) IGF-I on d 1; but on d 14, 28, 42, and at weaning, males had greater ($P < 0.05$) IGF-I than did females. Serum IGF-I was greater ($P < 0.02$) in single than in multiple lambs on all sampling days. Serum PRL was similar ($P > 0.10$) in male and female lambs throughout preweaning. However, single lambs had greater ($P < 0.001$) serum PRL concentrations than did multiple lambs on d 42 and at weaning. In general, PRL and IGF-I tended to increase in a quadratic ($P < 0.01$) fashion across the preweaning period. Correlation coefficients determined on d 60 between serum IGF-I, PRL, and ghrelin concentrations and preweaning ADG were 0.65, 0.44, and −0.26 ($P < 0.005$), respectively. Sex of lamb, type of birth, and age should be considered when evaluating serum hormone profiles in rapidly growing lambs.

Key Words: sheep, growth, hormones

W146 Patterns of fat growth in the primal cuts of beef composites. L. A. Goonewardene*[1], Z. Wang[2], R. W. Seneviratne[1], J. A. Basarab[1], J. Stewart-Smith[3], J. L. Aalhus[4], M. A. Price[2], and E. K. Okine[2], [1]*Alberta Agriculture and Rural Development*, *Edmonton, AB, Canada*, [2]*University of Alberta*, *Edmonton, AB, Canada*, [3]*Beefbooster Inc.*, *Calgary, AB, Canada*, [4]*Agriculture and Agri-Food Canada*, *Lacombe, AB, Canada*.

Beef composites (C) have combined favorable traits of pure breeds. The objective was to compare the growth rates (GR) and partitioning patterns of total fat (F), inter muscular (IM), body cavity (BC) and subcutaneous (SC) fat in 5 primal cuts of serially harvested Beefbooster composites (SM = C of small breeds n = 37, AH = C of Angus and Hereford n = 69 and GLC = C with Gelbvieh, Limousin or Charolais terminal sires n = 71) from 274 to 456 d (d) of age in relation to composite type and primal cut. Analysis of covariance obtained the slopes (GR/d) for total F, IM, BC and SC fat within each cut and C type. The GR of total fat deposition in AH was 24.7% higher ($P < 0.10$) than SM and 23.6% higher than GLC. The GR of total fat in the chuck was over 3 times higher than the loin and short loin and over twice the GR of the rib and round in each composite. The GR of IM fat in the chuck and rib was higher ($P < 0.10$) than BC and SC fat, but the GR of SC fat was higher than IM in the round and loin in each composite. The round and rib had similar GR for IM fat in SM and GLC but in the AH a 40.5% difference in GR for IM fat was observed. Pronounced differences were observed in the GR of SC fat between composites in the round and loin, and AH had higher ($P < 0.10$) GR compared with SM and GLC was in between. The difference in the fattening patterns in the 3 composites was most pronounced in the SC depot followed by the IM with little change in the BC depot. Selective breeding and the development of beef composites have primarily resulted in changes in SC fat partitioning in the 5 primal cuts and IM fat in the chuck. More fat trim at retail may be needed in the AH compared with SM and GLC.

Key Words: fat, growth, beef

W147 Prepartum supplementation in primiparous beef cows affected hepatic IGF-I mRNA expression in female calves. J. Laporta*[1], A. L. Astessiano[1], A. Scarsi[1], R. Pérez-Clariget[1], G. Quintans[2], and M. Carriquiry[1], [1]*School of Agronomy, UdelaR*, *Uruguay*, [2]*Instituto Nacional de Investigación Agropecuaria*, *Treinta y Tres, Uruguay*.

We examined the effect of supplementation during the last month of gestation of primiparous cows, on hepatic gene expression of growth hormone receptor (GHR), insulin–like growth factor–I (IGF–I) and IGF binding protein–3 (BP3) of their calves. The experiment was conducted in INIA Treinta y Tres (33°15′S 54°28′W) using 20 crossbred (Angus/Hereford) calves (n = 5 per treatment and sex). Cows, ranked by body weight (BW) and body condition score, were blocked by calving day and assigned randomly to control or supplement treatments. Supplemented cows were offered (1 kg/100 kg BW, 4.5 kg/d) a mix (67:33% as–fed basis; 16% CP, 11% ADF) of sorghum grain and protein concentrated from 33 ± 1.4 d prepartum until calving. Before, during and after the supplementation period, cows grazed together a native

pasture paddock (60 ha) with an average forage mass available of 1345 kg DM/ha (10.4% CP, 45.2% ADF). Calf BW, and liver samples were collected at weaning. Total RNA was extracted and mRNA abundance determined by SYBR Green RT–PCR in real time and normalized to hypoxanthine phosphoribosyl transferase (HPRT) expression. Means from a mixed model analysis were considered to differ when $P < 0.05$. Prepartum supplementation, calf sex, or their interaction did not afect BW at weaning (155 ± 11 kg at 180 ± 1.4 d). The expression of HPRT mRNA was not affected by supplementation, calf sex or their interaction. Although GHR, BP3 and IGF–I mRNA did not vary with supplementation, GHR expression tended ($P = 0.08$) to be greater and IGF–I mRNA was greater ($P = 0.04$) in female than male calves. There was an interaction ($P < 0.05$) between treatment and calf sex for IGF–I and BP3 mRNA, supplementation increased their expression only in females. Results suggest that prepartum supplementation during periods of limited forage availability of native pastures, through its effect in the fetal stage or the suckling period, could affect potential growth of female beef calves at weaning.

Key Words: liver, prepartum nutrition, RTP–CR

W148 Glucagon-like peptide 2 may mediate growth and development of the bovine gastrointestinal tract. E. E. Connor*[1], R. L. Baldwin[1], A. V. Capuco[1], C. M. Evock-Clover[1], S. E. Ellis[2], and K. S. Sciabica[3], [1]*USDA-ARS, BARC*, *Beltsville, MD*, [2]*Clemson University*, *Clemson, SC*, [3]*Beckman Coulter, Inc.*, *Brea, CA*.

Glucagon-like peptide 2 (GLP-2), secreted by enteroendocrine cells, promotes growth, reduces apoptosis, and enhances blood flow, nutrient absorption, and barrier function in intestinal epithelium of monogastric species. Regulatory functions of GLP-2 in the ruminant gastrointestinal tract (GIT) are unknown. Our objectives were to characterize expression of GLP-2 pathway members throughout bovine GIT including proglucagon (GCG) mRNA, the parent peptide from which GLP-2 is derived through cleavage by prohormone convertase (PCK1), PCK1 mRNA, GLP-2 receptor (GLP2R) mRNA, and mRNA for dipeptidyl peptidase IV (DPP4), the enzyme that inactivates GLP-2. Gene expression was evaluated in rumen, reticulum, omasum, abomasum, duodenum, jejunum, ileum, cecum, and rectum collected at slaughter from prepubertal heifers, mature cows in early, mid, and late lactation, and non-lactating cows (n = 3/stage) by a multiplex gene expression profiling assay based on traditional RT-PCR that uses a universal priming strategy. Further, mRNA expression of 14 genes involved in nutrient transport, enzyme activity, blood flow, apoptosis, and proliferation were evaluated in the 9 GIT tissues for association with GCG and GLP2R mRNA. Results indicated that mRNA expression of GCG, PCK1, GLP2R, and DPP4 varies across the 9 GIT tissues ($P < 0.001$), with greatest expression in intestines, and generally non-detectable levels in forestomachs. Expression of DPP4 and GLP2R mRNA varied by developmental stage or lactational state ($P < 0.05$) in intestinal tissues. Expression of GCG or GLP2R mRNA was correlated with markers of proliferation, apoptosis, blood flow, enzyme activity, and urea transport, depending on tissue type, supporting involvement of GLP-2 in these processes in ruminants. Lastly, GLP2R protein was localized to cells lining the intestinal crypts by immunohistochemistry, consistent with distribution in monogastric species. Our findings support a functional role of GLP-2 in bovine GIT and its potential use to improve intestinal function and nutrient absorption in ruminants.

Key Words: cattle, gene expression, GLP-2

W149 Effects of maternal metabolizable protein supply on fetal organ weights. T. L. Neville*[1], L. A. Lekatz[1], T. J. Swanson[1], M. L. Van Emon[2], C. S. Schauer[2], K. R. Maddock Carlin[1], C. J. Hammer[1], and K. A. Vonnahme[1], [1]*Center for Nutrition and Pregnancy, Department of Animal Sciences, North Dakota State University*, *Fargo,* [2]*Hettinger Research Extension Center, North Dakota State University*, *Hettinger*.

Objectives were to investigate the effects of maternal MP supply during gestation on fetal target organ weights. Multiparous ewes (n = 45; 15/treatment) were fed an isocaloric and isonitrogenous diet that contained either 75% (LOW), 100% (MOD), or 125% (HIGH) of MP requirements from d 100 to 130 of gestation. At d 130 ewes were harvested and fetuses dissected. Ewes carried singletons and twins, therefore fetal number was included in the model. Fetal weight, curved crown rump, heart girth, eviscerated BW, brain, and several target tissues were not affected by maternal MP supply during gestation ($P \leq 0.06$). Fetal right ventricles (mm) from LOW ewes were thicker ($P = 0.05$) than MOD ewes with those from HIGH ewes being intermediate. Fetuses from HIGH and LOW ewes had thicker ($P = 0.01$) right ventricles than MOD ewes (0.095, 0.098, and 0.082 ± 0.004 mm/g brain weight for HIGH, LOW and MOD respectively). Fetuses from LOW ewes had lighter ($P = 0.01$) ovaries (g/g brain weight) than MOD and HIGH ewes. Singleton lambs from LOW ewes had lighter ($P = 0.02$) adrenal glands than singleton lambs from MOD ewes and twins from LOW ewes with all other lambs being intermediate. Total gastrointestinal tract was heavier ($P \leq 0.03$) in twin lambs from LOW ewes compared with singleton lambs from LOW and twins from HIGH ewes with all other lambs being intermediate. These data indicate maternal dietary MP during the last trimester impacts fetal organ growth near term.

Key Words: fetal lambs, metabolizable protein, organ weights

W150 Nutrient restriction from early to mid-gestation in the cow increases offspring adipocyte size at slaughter. C. B. Tousley[1], N. M. Long*[1,2], S. P. Ford[1,2], W. J. Means[2], B. W. Hess[2], and M. Du[2], [1]*Center for the Study of Fetal Programming*, *University of Wyoming, Laramie,* [2]*Depertment of Animal Science, University of Wyoming*, *Laramie*.

Multiparous Angus × Gelbvieh cows were used to evaluate effects of nutrient restriction on adipose tissue morphology of beef steers and heifers at standard production endpoints. At 45 d after AI from a single sire, pregnancy was confirmed, and cows (n = 8 per treatment) were allotted randomly to be individually fed a control diet (C,100% of NRC recommendations), nutrient restricted (NR,70% of C NEm), or NR + protein supplement (NRP,NR + protein to equal C metabolizable AA). At 185 d of gestation (dG), all cows were comingled and fed the C diet. Bull calves were castrated at birth and all calves weaned at 210 d of age, backgrounded for 28 d, and then placed in the feedlot. Steers and heifers were harvested on separate days at an average 12th rib fat thickness of 7.6 mm. Selected organ weights and carcass characteristics were determined. Adipose tissue from selected depots was collected and fixed in paraffin and adipocyte diameters determined by image analysis. There was no difference ($P < 0.23$) in BW or BCS between C, NRP, and NR cows at 45 dG, which averaged 489.74 ± 17.7 kg and 5.35 ± 0.13, respectively. At 185 dG, C and NRP cows had similar BW (566.1 ± 14.8 and 550.2 ± 14.8) and BCS (6.34 ± 0.27 and 5.59 ± 0.27), but BW (517.9 ± 14.8) and BCS (4.81 ± 0.27) of NR cows were less ($P < 0.05$). There were no treatment differences ($P < 0.26$) in live BW, carcass characteristics or organ weights of steers and heifers at slaughter. In contrast, average adipocyte diameters of NR offspring increased or tended to increase (Table 1) in finished steers and heifers, which could contribute to altered adiposity and metabolism in later life.

Table 1. Average adipocyte diameter (um) from selected depots of offspring (4 steers and 4 heifers per treatment) at slaughter

	Control	NRP	NR
Subcutaneous	74.5 ± 2.6[a,]	81.6 ± 2.5[ab]	86.9 ± 2.7[b]
Perirenal	113.5 ± 3.5[c]	115.6 ± 3.5[c]	124.69 ± 3.8[d]
Mesenteric	113.03 ± 3.8C[a]	121.37 ± 3.5[ab]	129.28± 3.5[b]
Omental	117.2 ± 4.5[a]	114.4 ± 4.2[a]	129.6 ± 4.3[b]

[a,b]Means differ $P \leq 0.05$; [c,d]Means differ $P < 0.10$.

Key Words: prenatal programming, adipocyte size, cows

W151 Two messenger RNA targets, Programmed Cell Death Protein 4 and Phosphatase and Tensin Homolog, of microRNA-21 are expressed in cultured bovine adipocytes. S. L. Pratt*, T. A. Burns, and S. K. Duckett, *Clemson University*, *Clemson, SC*.

Adipogenesis is regulated in part by post-transcriptional gene regulation through small non-coding inhibitory RNA, microRNA (miRNA), via translational repression or RNA interference. We have shown miR-21 expression to increase in adipocytes compared with preadipocytes as differentiation progresses, and this miRNA is known to promote cell survival and hypertrophic growth, in part, by the translational inhibition of Programmed Cell Death Protein 4 (PDCD4) and Phosphatase and Tensin Homolog (PTEN). Neither the protein nor message for PDCD4 or PTEN has been reported to be expressed in, or involved in adipogenesis/adipocyte function. Our objective was to detect and quantify the mRNA for bovine PDCD4 and PTEN in primary bovine cell cultures. Bovine preadipocyte cell lines generated from subcutaneous adipose tissue obtained from 18 mo old Angus steers were cultured to confluence and held 2 d in Dulbecco's modified eagles medium (DMEM) containing 10% fetal calf serum (FCS), and 2X antibiotiotic/antimycotic (AB/AM). Cells were differentiated using a 2- step process by culture for 2-d in DMEM containing 5% FCS, 2X AB/AM, insulin at 2.5 ug/ml, 0.25 uM dexamethasone, 5 uM troglitasone and 0.5 mM isobutylmethylxanthine (IBMX) for 2-d followed by a 4-d or 10-d incubation in DMEM containing 5% FCS, 2X AB/AM, insulin, and troglitasone. tcRNA was isolated for each cell line at 2-d confluence (Control), and after 2-d (D2), 4-d (D6) and 10-d (D12) of differentiation. tcRNA was used in real time qRT-PCR to determine expression levels for the respective message for bovine PTEN and PDCD4. Expression levels for PTEN and PDCD4 across all days ranged from −0.8- to 1.0-fold compared with Control ($P > 0.05$). In contrast, miR-21 was previously shown to be upregulated during the same stages of adipogenesis. We hypothesize that miR-21 downregulates the proteins PDCD4 and PTEN by translational inhibition during adipogenesis allowing cells to survive and undergo hypertrophic growth required for lipid filling.

Key Words: miRNA, adipogenesis, qRT-PCR

W152 MicroRNA-21 and its messenger RNA targets Programmed Cell Death Protein 4 and Phosphatase and Tensin Homolog are expressed in bovine adipose tissue. S. L. Pratt*, E. Curry, T. A. Burns, and S. K. Duckett, *Clemson University*, *Clemson, SC*.

MicroRNA (miRNA) are small non-coding RNA that regulate adipocyte function, both in vivo and in vitro by translational repression or RNA interference of targeted messenger RNA. miR-21 expression increases in adipocytes compared with preadipocytes, and this miRNA promotes cell survival and hypertrophic growth, in part, by the translational inhibition of Programmed Cell Death Protein 4 (PDCD4) and Phosphatase and Tensin Homolog (PTEN). Neither PDCD4 nor PTEN has been reported to be expressed in, or involved in adipocyte function. Our objectives were to detect and quantify 1) miR-21 and 2) PDCD4 and PTEN in adipose tissue isolated from Angus steers (289 kg) finished on pasture only (PA; n = 6) or on a high-concentrate diet (C; 85% concentrate/15% roughage; n = 7). Fat samples (s.c.; 5 g per sample) were removed from the tail head area of each carcass at slaughter, rinsed with sterile saline, frozen in liquid nitrogen and stored at −80°C. Protein extracts were subjected to SDS-PAGE, Western blotting and immunodetection with PTEN and PDCD4 antisera. tcRNA was used in real time qRT-PCR to determine expression levels for miR-21, PTEN, and PDCD4. Western blotting and immunodetection with antisera to PTEN and PDCD4 detected proteins ~50 and 60 kDa, respectively. Message levels for PTEN and PDCD4 increased 3.2- and 6.7-fold, respectively in PA compared with C treatment ($P < 0.05$); in contrast miR-21 expression was repressed in PA compared with C ($P < 0.05$). These data show that miR-21, PTEN and PDCD4 are expressed in bovine adipose tissue. Message level for PTEN and PDCD4 in mature adipose tissue may be regulated at the transcriptional level, and low levels of concentrate in the diet may increase the PTEN and PDCD4 message under chronic conditions which would stimulate increased adipocyte cell mortality and turnover. PTEN and PDCD4 protein expression may be increased due to increased message levels and to the decrease in miR-21 expression which negatively regulates the translation of both messages.

Key Words: miRNA, adipose tissue, qRT-PCR

W153 Both growth hormone and signal transducer and activator of transcription 5b inhibit glycerol-3-phosphate dehydrogenase activity and CCAAT/enhancer binding protein α mRNA expression in differentiating bovine preadipocytes. L. Zhao*, B. A. Corl, and H. Jiang, *Virginia Polytechnic Institute and State University*, *Blacksburg*.

It has been long known that growth hormone (GH) inhibits adipogenesis in various animals, including cattle. However, the underlying mechanism remains poorly understood. The objective of this study was to determine if GH inhibits bovine adipogenesis by activating the signal transducer and activator of transcription 5b (STAT5b), a transcription factor known to be activated by GH in many tissues including the adipose tissue. Two studies were conducted to achieve this objective. In one study, preadipocytes from adipose tissue explants of cattle were induced to differentiate in the presence or absence of 10 ng/mL GH and 100 ng/mL GH for 6 d before differentiation assessment. In the other study, the preadipocytes were induced to differentiate in the presence of 10 or 100 multiplicities of infectivity (MOI) of adenovirus expressing constitutively active STAT5b (STAT5bCA) or β-galactosidase (LacZ) as a control for 6 d before differentiation assessment. Differentiation was assessed by measuring glycerol-3-phosphate dehydrogenase (G3PDH) activity, and mRNA expression of CCAAT/enhancer binding protein α (CEBPα), peroxisome proliferator-activated receptor γ (PPARγ), and fatty acid binding protein 4 (FABP4). Both concentrations of GH inhibited G3PDH activity ($P < 0.05$, n = 5) and decreased mRNA expression of CEBPα compared with the no-GH control ($P < 0.05$, n = 5). Neither GH concentration altered mRNA expression of PPARγ or FABP4 compared with the no-GH control. The STAT5bCA adenovirus at 100 MOI inhibited the G3PDH activity ($P < 0.05$, n = 4) and decreased mRNA expression of CEBPα compared with the LacZ adenovirus or the no-adenovirus control ($P < 0.05$, n = 4). The STAT5bCA adenovirus did not change mRNA expression of PPARγ or FABP4. In summary, both GH and STAT5bCA overexpression inhibited G3PDH activity and CEBPα mRNA expression in differentiating bovine preadipocytes.

Key Words: growth hormone, STAT5b, bovine preadipocyte

W154 Primary preadipocytes can be isolated, propagated, and differentiated from bovine intermusclar fat harvested 48 h postmortem. T. A. Burns*, S. K. Duckett, and S. L. Pratt, *Clemson University*, *Clemson, SC*.

Traditionally, primary cell cultures are isolated from tissues collected at slaughter using collagenase digestion or tissue explantation methods. Although we have previously been successful isolating and differentiating bovine preadipocytes from slaughtered cattle, the abattoir is not a conducive environment for aseptic tissue collection. Therefore, the objective of this study was to isolate, propagate, and successfully differentiate bovine preadipocytes collected from refrigerated rib sections 48 h postmortem. Sections of intermuscular fat were excised and minced using sterile instruments, rinsed with Hanks' Balanced Salt Solution (HBSS), and digested in 25 mL HBSS containing 2 mg/mL collagenase, type I, and 40 mg/mL bovine serum albumin under constant shaking at 37°C for 120 min. Cells were plated at 1×10^4 cells/cm^2, passaged every 2–4 d when 60% confluent, and were frozen after 4 passages. To test a cell line's ability to differentiate, cells were thawed, passaged 3 times, and seeded in wells at 1×10^5 cells/cm^2. Cells were allowed to reach confluence, held for 2 d, and differentiated on d 0 with Dulbecco's modified eagles medium containing 10% fetal calf serum, 2X antibiotic/antimycotic, insulin at 2.5 μg/mL, 0.25 μ*M* dexamethasone, 20 μ*M* troglitasone, 0.5 m*M* isobutylmethylxanthine, and 10 m*M* acetate. Undifferentiated cells were harvested on d 2 (CON) and treated cells were harvested on d 2 (D2), 6 (D6), and 12 (D12) for fatty acids and RNA. Gross morphology of differentiating cells displayed characteristic shape change from fibroblastic to spherical. Using GLM procedure of SAS, duplicate fatty acid data were analyzed across 3 cell lines by time. Total fatty acids were not different between CON and D2, but increased ($P < 0.05$) on D6 and D12 compared with CON. On a percentage basis, C16:0, C18:0, and C18:2 decreased ($P < 0.05$) and C18:1c9 increased ($P < 0.05$) by D12. In addition, differentiation-associated genes were increased ($P < 0.05$) in treated cells. This evidence suggests that cells harvested 48 h postmortem are capable of propagating and differentiating for use in in vitro studies.

Key Words: preadipocyte, isolation, differentiation

W155 *Trans*-10, *cis*-12 conjugated linoleic acid induces adipogenic gene expression in single and co-cultures of bovine preadipocytes and myoblasts. S. H. Choi*[1], K. Y. Chung[2], G. Go[1], C. W. Choi[3], B. J. Johnson[2], and S. B. Smith[1], [1]*Department of Animal Science, Texas A&M University*, *College Station,* [2]*Department of Animal and Food Science, Texas Tech University*, *Lubbock,* [3]*National Institute of Animal Science*, *Suwon, Gyunggi, Korea*.

A limited number of studies have investigated the differentiation of bovine adipocytes and myoblasts in single cultures, and there is even less information about the interaction between adipocytes and myoblastic cells in a coculture system. We hypothesized that preadipocyte differentiation would be depressed by differentiating myoblasts, where preadipocytes would promote adipogenic gene expression in myoblasts in a coculture system. We also determined the effects of arginine, a biological precursor of nitric oxide, and/or *trans*-10, *cis*-12 conjugated linoleic acid (CLA) on adipogenic gene expression of bovine preadipocytes and myoblasts. Muscle-derived satellite cells and preadipocyte were isolated from 14-mo old crossbred steers. Both bovine satellite (BSC) and preadipocytes (PA) cells were cultured with 10% FBS/DMEM and 1% antibiotics during the proliferation period (3 d). After proliferation, BSC and PA were treated with 3% of horse serum (HS) DMEM and 5% FBS/DMEM with antibiotics respectively, for 4 d. Finally, single or combined BSC and PA were cultured with 40 μ*M* *trans*-10, *cis*-12 CLA and/or 5 m*M* arginine for 2 h. Cocultured PA had significantly less AMPK ($P = 0.04$) and SCD ($P = 0.04$) gene expressive than single-cultured PA. Arginine stimulated, and CLA depressed SCD gene expression. Arginine significantly stimulated PPARγ gene expression ($P = 0.06$) in cocultured PA compared with other treatments. SCD gene expression was significantly increased by arginine in both single- ($P < 0.001$) and cocultured ($P < 0.01$) PA. Conversely, C/EBP-β gene expression was significantly enhanced by coculture in BSC in the absence or presence of arginine or CLA. PPARγ gene expression tended to be increased by coculture in BSC. We conclude that arginine and CLA had similar effects in single and cocultured PA and BSC, but the effects were stronger in cocultured cells.

Key Words: co-culture, preadipocytes, satellite cells

W156 Growth hormone stimulates liver expression of fibroblast growth factor 21 mRNA in cattle. J. Yu*[1,2], A. Wang[2], S. Eleswarapu[2], and H. Jiang[2], [1]*Sichuan Agricultural University*, *Yaan, Sichuan, China*, [2]*Virginia Polytechnic Institute and State University*, *Blacksburg*.

Fibroblast growth factor 21 (FGF21) is a novel endocrine regulator of glucose homeostasis, lipid metabolism, insulin sensitivity, and obesity. In addition, FGF21 is known to inhibit growth hormone (GH)-induced signaling through the signal transducer and activator of transcription 5 (STAT5). In the body, FGF21 mRNA is expressed predominantly in the liver. In this study, we tested the hypothesis that FGF21 expression in the liver is induced by GH. Nonlactating and nonpregnant cows were injected subcutaneously with 500 mg of recombinant bovine GH in a slow-release formula. Liver biopsy and blood samples were collected from each cow one week before and 24 h after the injection. Liver FGF21 mRNA abundance after GH injection was 21 times that before the injection ($P < 0.05$, n = 5), as measured by real-time RT-PCR. The GH injection tended to increase serum concentration of FGF21 ($P = 0.09$, n = 5), as measured by an enzyme-linked immunosorbent assay. The bovine FGF21 gene promoter contains 3 putative STAT5 binding sites. Electrophoretic mobility shift assays showed that all of them could bind to the STAT5 protein from bovine liver. Chromatin immunoprecipitation assays demonstrated that GH injection stimulated binding of STAT5 to the FGF21 promoter in the liver ($P < 0.05$, n = 4). Co-transfection analyses indicated that reporter gene expression from the bovine FGF21 promoter could be induced by GH in a STAT5-dependent manner. Taken together, these data suggest that GH stimulates expression of FGF21 mRNA in cattle and that the stimulation may be mediated by the transcription factor STAT5.

Key Words: growth hormone, fibroblast growth factor 21, STAT5

W157 Abundance of growth hormone secreteagogue receptor and PPARy2 in longissimus dorsi of beef cattle. C. L. Delvaux*, J. S. Jennings, and A. E. Wertz-Lutz, *South Dakota State University*, *Brookings*.

Objectives of this experiment were to determine the presence and abundance of growth hormone secreteagogue receptor (GHS-R) and PPARy2 in longissimus dorsi of beef cattle fed to achieve differences in composition of gain. Beef steers (n = 72) of similar age, weight (292 ± 1.44 kg), and genetic background were used. At trial initiation (d 0), 8 steers were harvested. The remaining 64 steers were allotted, by weight, to pen, and treatment was assigned randomly. Treatments were 1) 60% forage; 40% concentrate diet fed during the growing period (0–112 d) followed by 10% forage; 90% concentrate diet during the finishing period (113–209 d) (GRW-FNSH) or 2) 10% forage; 90% concentrate

diet fed for the duration of the experiment (0–209 d) (FNSH-FNSH). Steers were allowed ad libitum consumption regardless of dietary treatment. Eight steers per treatment were harvested on d 88, 116, 165, and 209. Tissue samples from the longissimus dorsi were collected 48 h subsequent to harvest and frozen. Protein separation and quantification was determined using SDS-PAGE and Western blotting techniques. Abundance of GHS-R and PPARy2 was quantified using the LI-COR system and standardized to β-Actin. Protein abundance data were analyzed statistically using the MIXED procedure of SAS evaluating diet, age at harvest, and their interaction. Differences in composition of gain have been reported previously. Abundance of GHS-R differed ($P < 0.001$) as a result of age at harvest, and dietary treatment tended ($P = 0.11$) to influence GHS-R abundance. Differences in GHS-R abundance relative to age at harvest, and potentially as a result of differential growth, warrants further investigation. The PPARy2 was not sufficiently abundant in longissimus dorsi and could not be quantified. The PPARy2 is exclusively associated with adipose tissue, and therefore not expected to be abundant in longissimus dorsi tissue. Degradation of PPARy2 also may have occurred as samples were not collected until 48 h postmortem.

Key Words: growth hormone secreteagogue receptor, PPARy2, longissimus dorsi

W158 Effect of estradiol-17β on protein synthesis and degradation rates in fused bovine satellite cell cultures. E. Kamanga-Sollo, M. E. White*, M. R. Hathaway, W. J. Weber, and W. R. Dayton, *University of Minnesota*, *St. Paul*.

Although androgenic and estrogenic steroids are widely used to enhance muscle growth and increase feed efficiency in feedlot cattle, their mechanism of action is not well understood. While in vivo studies indicate that Estradiol-17β (E2) affects muscle protein synthesis and/or degradation, in vitro results are inconsistent. We have examined the effects of E2 treatment on protein synthesis and degradation rates in fused bovine satellite cell (BSC) cultures. Additionally, to learn more about the mechanisms involved in E2-enhanced muscle growth, we have examined the effects of compounds that interfere with binding of E2 or IGF-I to their respective receptors on E2-induced alterations in protein synthesis and degradation rates in BSC cultures. Treatment of fused BSC cultures with E2 results in a concentration-dependent increase ($P < 0.05$) in protein synthesis rate and a decrease ($P < 0.05$) in protein degradation rate. The pure estrogen antagonist ICI 182 780 suppresses ($P < 0.05$) E2 -induced alterations in protein synthesis and degradation in fused BSC cultures. The G-protein coupled receptor (GPR)-30 agonist, G1, does not affect either synthesis or degradation rate establishing that GPR30 does not play a role in E2-induced alterations in protein synthesis or degradation. JB1, a competitive inhibitor of IGF-I binding to the Type 1 insulin-like growth factor receptor (IGFR-1), suppresses ($P < 0.05$) E2-induced alterations in protein synthesis and degradation. In summary our data show that E2 treatment directly alters both protein synthesis and degradation rates in fused BSC cultures via mechanisms involving both the classical ER and IGFR-1.

Key Words: estradiol-17β, satellite cells, protein turnover

W159 Effect of trenbolone acetate on protein synthesis and degradation rates in fused bovine satellite cell cultures. E. Kamanga-Sollo, M. E. White*, M. R. Hathaway, W. J. Weber, and W. R. Dayton, *University of Minnesota*, *St. Paul*.

Although androgenic and estrogenic steroids are widely used to enhance muscle growth and increase feed efficiency in feedlot cattle, their mechanism of action is not well understood. While in vivo studies have indicated that androgens affect protein synthesis and/or protein degradation rate in muscle, results from in vitro studies have been inconsistent. Consequently, we have examined the effects of trenbolone acetate (TBA), a synthetic androgen, on protein synthesis and protein degradation rates in fused bovine satellite cell (BSC) cultures. Additionally, we have examined the effects of compounds that interfere with binding of TBA or insulin-like growth factor (IGF)-I to their respective receptors on TBA-induced alterations in protein synthesis and protein degradation rates in BSC cultures. Treatment of fused BSC cultures with TBA results in a concentration-dependent increase ($P < 0.05$) in protein synthesis rate and a decrease ($P < 0.05$) in protein degradation rate, establishing that TBA has a direct effect these parameters. Flutamide, a compound that prevents androgen binding to the androgen receptor, suppresses ($P < 0.05$) TBA -induced alterations in protein synthesis and degradation in fused BSC cultures, indicating the androgen receptor is involved. JB1, a competitive inhibitor of IGF-I binding to the Type 1 IGF receptor (IGFR-1), suppresses ($P < 0.05$) TBA-induced alterations in protein synthesis and degradation indicating that this receptor also is involved in the actions of TBA on both synthesis and degradation. In summary our data show that TBA acts directly to alter both protein synthesis and degradation rates in fused BSC cultures via mechanisms involving both the androgen receptor and IGFR-1.

Key Words: trenbolone acetate, satellite cells, protein turnover

W160 Zilpaterol HCl enhances adenosine monophosphate-activated protein kinase α (AMPKα) expression in bovine skeletal muscle. R. J. Tokach*, K. Y. Chung, and B. J. Johnson, *Texas Tech University*, *Lubbock*.

Zilpaterol HCl (ZH), a β-2 adrenergic agonist, has been used for enhancing muscle growth in feedlot cattle. The aim of these in vivo and in vitro experiments was to determine the effect of ZH on changes in enzymes and growth factors important in skeletal muscle growth. We hypothesized that AMPKα expression increased in animals or cells treated with ZH. Thirteen month old calf-fed Holstein steers implanted with an estrogen-based implant were used in randomized complete block design with a 2 × 2 factorial arrangement of treatments. Main effects included; ZH 0 or 20 d before slaughter and either a 3-d or 10-d withdrawal period (WD). Semimembranosus muscle (SM) was collected within 10 min of harvest for analysis of AMPKα and IGF-I protein and mRNA abundance. Western blot analysis revealed that the protein content of AMPKα in muscle from ZH-treated cattle with a 3-d withdrawal increased ($P < 0.05$) as compared with the control group. There was no significant difference of AMPKα in muscle from the ZH-treated cattle with a 3-d withdrawal as compared with the ZH-treated cattle with a 10-d withdrawal. Real-time quantitative PCR was used to measure the relative level of mRNA. The mRNA for AMPKα tended to increase in muscle from ZH-treated cattle with a 3-d withdrawal ($P = 0.12$) as compared with the control group. Withdrawal time resulted in a decrease in IGF-I mRNA. Primary cultures of bovine satellite cells (BSC) isolated from the SM were harvested after 120 h of either ZH, ZH antagonist (ICI-118,551), or ZH plus ICI-118,551 exposure for AMPKα and IGF-I protein and mRNA analysis. AMPKα abundance increased in BSC treated with ZH as compared with the control, ICI-118,551, and ZH plus ICI-118,551 treated BSC. IGF-I mRNA relative units (3.7 ± 0.6) increased in BSC treated with ZH as compared with the control (0.8 ± 0.4) and the addition of ICI-118,551 (0.9 ± 0.2) ameliorated these changes ($P < 0.05$). These data indicate that ZH alters mRNA and protein concentrations of AMPKα in SM, and these effects may be mediated through the β 2 adrenergic receptor.

Key Words: AMPKα, bovine satellite cells, zilpaterol hydrochloride

W161 Steroidal implants and zilpaterol HCl alter serum urea-N and NEFA responses in finishing beef steers. S. L. Parr*[1], M. L. Galyean[1], K. Y. Chung[1], J. P. Hutcheson[2], and B. J. Johnson[1], [1]*Texas Tech University*, *Lubbock,* [2]*Intervet / Schering-Plough Animal Health*, *De Soto, KS*.

British × Continental steers (n = 168; 7 pens/treatment; initial BW = 362 kg) were used to evaluate the effect of dose of trenbolone acetate (TBA) and estradiol-17β (E_2) and feeding of zilpaterol hydrochloride (ZH) on serum urea-N (SUN) and NEFA concentrations. A randomized complete block design was used with a 3 × 2 factorial arrangement of treatments. Main effects were implant (no implant [NI]; Revalor-S [REV-S; 120 mg TBA + 24 mg E_2]; and Revalor-XS [REV-X; 200 mg TBA + 40 mg E_2]) and ZH (0 or 8.3 mg/kg of DM for 20 d with a 3-d withdrawal before harvest). Blood was drawn from 2 steers per pen (n = 84/d) on d −1, 2, 6, 13, 27, 55, 83, 111, and 131 relative to implanting and d −1, 11, and 19 relative to ZH supplementation. Steers were fed for 153 or 174 d depending on block. Overall ADG (1.4, 1.7, and 1.8 kg for NI, REV-S and REV-X, respectively) and G:F (0.16, 0.18 and 0.19) were increased ($P < 0.05$) as TBA and E_2 dose increased. Carcass-adjusted ADG (1.5 vs. 1.7 kg for 0 and 8.3 mg/kg ZH respectively) and G:F (0.17 vs. 0.19) were increased ($P < 0.05$) by ZH. Serum urea-N increased ($P < 0.05$) over time (no implant × day interaction, $P = 0.16$). Implanting decreased ($P < 0.05$) SUN from d 2 through d 131 and SUN tended ($P = 0.08$) to be less for REV-S than REV-X at d 13 but was otherwise not different among implants. Feeding ZH decreased ($P < 0.05$) SUN. Serum NEFA concentrations were not affected by implants ($P = 0.44$). There was a day × ZH interaction ($P = 0.06$) for NEFA; steers not fed ZH had similar ($P > 0.10$) NEFA concentrations on d −1, 11, and 19, whereas ZH steers had increased ($P < 0.01$) NEFA concentrations at d 11 of ZH feeding. We conclude that implanting decreased SUN but a greater dose of TBA and E_2 did not result in further changes in SUN. Moreover, feeding ZH affected steer metabolism by decreasing SUN and increasing serum NEFA. These metabolic responses during ZH may aid in explaining steer performance and carcass responses to ZH.

Key Words: beef steers, estradiol-17β, zilpaterol hydrochloride

W162 Canonical relationships of body shape of grazing bulls under tropical conditions. H. J. Fernandes*[1], L. O. Tedeschi[3], M. F. Paulino[2], M. O. Porto[2], and L. M. Paiva[1], [1]*State University of Mato Grosso do Sul*, *Aquidauana, Brazil*, [2]*Federal University of Viçosa*, *Viçosa, MG, Brazil*, [3]*Texas A&M University*, *College Station*.

The objective of this study was to analyze and compare the changes of body shape, using body measurements (BM) of Nellore crossbred bulls (n = 20) under grazing conditions using canonical equations. Animals were grazing *Brachiaria decumbens* Stapf and received either mineral (control; C) or 3 concentrate supplementation programs (different protein patterns). Animals in the programs T1, T2, and T3 received 1.2 kg of a concentrate containing 32, 25, and 9% of CP during the nursing phase, 1.5 kg of isonitrogenous concentrate (35% CP) with 8, 4, and 0% of urea during the dry season, and 2.0 kg of isonitrogenous concentrate (27% CP) with 6, 3, and 0% of urea during the rainy season; respectively. The BM were taken every 28 d and included hooks width, pins width, pelvic girdle length, rump height, abdomen width, body length, height at withers, and rib depth. The first and the second canonical variables for each phase and for the entire experimental period was developed using all BM. The most important BM to explain the differences in animal′s body shape were rump height, height at withers, pins width, and rib depth for the nursing phase; rib depth, body length, abdomen width, and pelvic girdle length for the dry season; rump height, body length, abdomen width, and hooks width for the rainy season; and rib depth, pins width, rump height, and height at withers for the complete experimental period. Dietary supplementation did not affect the animal′s body shape in the nursing phase. Animals in the C program had heavier hindquarter during the dry season, and during the rainy season they were higher, longer, and wider than animals receiving concentrate supplementations. The animals in the T1 program tended to be higher and longer than animals in the other supplementation programs (T2 and T3). The results of the complete experimental period indicated animals in the C program tended to be shorter and wider, suggesting animals with smaller mature size likely due to malnutrition. Further studies should evaluate the relationship between body shapes and variations in body composition of the animals.

Key Words: biometrics, cattle, growth

W163 Comparison of mathematical functions to describe the growth of grazing bulls in tropical conditions. H. J. Fernandes*[1], L. O. Tedeschi[2], M. F. Paulino[3], A. G. Silva[3], and L. M. Paiva[1], [1]*State University of Mato Grosso do Sul*, *Aquidauana, Brazil*, [2]*Texas A&M University*, *College Station*, [3]*Federal University of Viçosa*, *Viçosa, MG, Brazil*.

The availability of forage throughout the year is not constant and it can change the pattern of growth of grazing animals. The goal of this study was to evaluate different mathematical functions to characterize the growth of grazing Nellore crossbred bulls (n = 20) under tropical conditions. The animals had initial and final BW of 129 ± 28.1 and 405 ± 62.0 kg, initial age of 130 ± 30 d, and grazed *Brachiaria decumbens* Stapf during 420 d. BW was recorded every 28 d. Five mathematical functions (multiphase, linear, logarithmic, Gompertz, and Logistic) were used to describe the BW of the bulls. The multiphasic equation used 3 phases: 2 linear phases in which the difference was the ADG and a third phase represented by a Logistic model. The time points (thresholds) between these phases were assumed to be parameters of the function. The assessment of the adequacy of the mathematical functions was performed using the coefficient of determination, the simultaneous *F*-test of identity of the parameters, the concordance correlation coefficient, and the square root of the mean square error of prediction (MSEP). The analysis of the paired MSEP and the difference of Akaike′s Information Criterion (AIC) between mathematical functions were used to compare accuracy and precision of the functions. Adequacy of the mathematical functions (Table 1), the paired MSEP and the difference of AIC indicated the multiphasic function was more accurate and precise than the other functions in describing the growth of grazing beef cattle in tropical conditions. This was likely because of the effect of the dry season on the growth pattern of the animals. Other multiphasic functions should be evaluated to define the most appropriate equation for each production condition.

Table 1. Adequacy statistics of the functions[1]

Functions	r^2	P- value	CCC	RMSEP
Multiphase	0.993	0.617	0.996	6.83
Linear	0.961	0.678	0.980	16.1
Gompertz	0.916	< 0.001	0.945	25.0
Logarithmic	0.902	< 0.001	0.936	26.7
Logistic	0.861	< 0.001	0.886	34.0

[1]r^2 = coefficient of determination, *P* – value = probability of the *F*-test for intercept equal to zero and slope equal to one, CCC = concordance correlation coefficient, RMSEP = square root of the MSEP.

Key Words: beef cattle, grazing, seasonality

W164 Expression of specific genes regulating mammary growth in pre-pubertal Holstein heifers. F. Soberon*, M. J. Meyer, and M. E. Van Amburgh, *Cornell University*, *Ithaca, NY*.

In a previous study, we demonstrated that calves fed greater nutrients from milk replacer from birth to 100 kg BW had significantly increased mammary epithelial cell proliferation compared with calves fed a more restricted diet. Additional work has demonstrated the presence of a putative stem cell population that has been considered nutritionally responsive and might account for the observed cell proliferation. One objective of this study was to identify genetic markers of stem cells associated with the proliferation. Further, changes in mammary growth rate occur from birth to puberty, such as the change from allometric to isometric growth during the peri-pubertal period. The second objective of this study was to evaluate genes that might be responsible for signaling this change in mammary growth from an allometric to an isometric rate. Heifers (n = 66) reared on one of 2 dietary treatments (TRT), restricted (R) 650 g/d or elevated 950 g/d gain, were harvested at 100, 150, 200, 250, 300, or 350 kg BW. Mammary samples (n = 5 to 8) were excised from the mid parenchyma tissue (MPT) and extra parenchyma tissue (EPT), snap frozen and stored at −80°C. Gene expression in mammary tissue was determined by RT-PCR. Data were analyzed using a mixed model with 18S RNA as a covariate. Telomerase transcriptase was used as a marker for stem cells. Further SERPINB5 (maspin) was analyzed as a marker of myoepithelial cell development and a possible signal for changes in growth rate of the mammary gland. Telomerase expression in EPT was lowest ($P < 0.05$) at 100 kg BW for both treatments and tended ($P < 0.1$) to decrease in R at 350 kg BW. Telomerase expression did not differ between TRT in MPT but was lower at 100 and 150 kg BW ($P < 0.05$). Expression of SERPINB5 was significantly increased in EPT at 250 kg BW for both TRT whereas SERPINB5 expression in MPT remained constant among all weight groups. We suggest that SERPINB5 signals the end of allometric growth by arresting the effect of estrogen in the cells extending into the fat pad, but has no effect on the developed parenchymal tissue.

Key Words: mammary growth, calves, serpinb5

W165 Effects of meal timing on anabolic hormone status and energy metabolism in neonatal Holstein calves. K. C. Simon, C. C. Williams*, L. R. Gentry, B. F. Jenny, R. M. Doescher, and A. H. Dolejsiova, *LSU AgCenter*, *Baton Rouge, LA*.

A study was conducted to determine effects of feeding time on metabolic hormone secretion, growth, and energy metabolism in neonatal dairy calves. Twelve Holstein bull calves were randomly assigned to 1 of 2 dietary treatments which included milk replacer fed at a fixed meal time (REG) or a varied meal time (IRR). Body weights were measured every 2 wk from birth to 8 weeks. Serial blood collections were conducted biweekly from wk 2 through 8 at 0530, at 0, 15, 30, 45, 60, 75, 90, 105, 120, 135, and 150 min relative to regularly scheduled feeding time. Plasma was analyzed for ghrelin, leptin, growth hormone (GH), and insulin-like growth factor-1 (IGF-1). Rumen fluid was collected biweekly from wk 2 through 8 for analysis of volatile fatty acids (VFA) and pH. Treatment did not affect ($P > 0.10$) body weight or average daily intake. Mean plasma ghrelin, leptin, GH, and IGF-1 concentrations were not affected ($P > 0.10$) by treatment. A treatment by week interaction was observed ($P < 0.05$) for total plasma ghrelin concentrations, with total plasma ghrelin concentrations higher at wk 2 and 4 in REG calves. Total plasma ghrelin concentrations decreased ($P < 0.05$) in all calves as they aged. A treatment by time interaction was observed ($P < 0.10$) for IGF-1, and a treatment by week by time interaction was observed ($P < 0.10$) for GH and IGF-1. Growth hormone decreased as calves aged, while IGF-1 increased. There was no treatment effect ($P > 0.10$) on rumen VFA concentrations, but acetate, propionate, and butyrate increased ($P < 0.05$) with age. No treatment effect was observed ($P > 0.10$) on rumen pH. At wk 4 and 8, intravenous glucose tolerance tests were performed to assess glucose half life ($T_{1/2}$), glucose clearance rate (k), and insulin concentrations. REG calves had greater ($P < 0.05$) $T_{1/2}$ and lower ($P < 0.05$) k. Peak insulin concentrations were higher ($P < 0.05$) for REG calves. These data suggest that feeding time does not affect overall growth and feed intake but does have an effect on some of the regulatory mechanisms that control them.

Key Words: dairy calves, feeding times, energy metabolism

W166 Effect of supplementing fatty acids to prepartum Holstein cows and milk replacer enriched with linoleic acid on calf performance. M. Garcia*, L. F. Greco, M. G. Favoreto, R. S. Marsola, L. T. Martins, D. Wang, W. W. Thatcher, J. E. P. Santos, and C. R. Staples, *University of Florida*, *Gainesville*.

The aim of this study was to evaluate supplementing linoleic acid (LA) to cows during the last 2 mo of pregnancy and their calves from birth to 60 d of age on calf performance. Cows (n = 96) were fed diets formulated to supply minimum amounts of LA and supplemented without fat, with saturated fatty acids (SFA; Energy Booster 100, MSC) at 1.75% of dietary DM, or with Ca salts of unsaturated fatty acids enriched in LA (UFA; Megalac-R, Church and Dwight, Co.) at 2% of dietary DM. Within 2 h of birth, calves were fed 4 L of colostrum from their own dam or from a dam of the same dietary treatment using an esophageal feeder. Calves were blocked by gender and dam diet and assigned randomly to receive a milk replacer (MR) with low (0.56% LA) or high concentration of LA (1.78% LA, DM basis) for 60 d. Milk replacer (20% fat and 29% CP) was fed twice daily at 6.7 g of fat per kg of metabolic BW and amounts were adjusted weekly. A single grain mix of minimum LA concentration (18.7% CP and 4.2% fat) was offered in ad libitum amounts starting at 31 d of age. Calves fed the high-LA MR had greater ($P < 0.05$) ADG (0.50 vs. 0.45 kg/d) and efficiency of gain (0.63 vs. 0.58 kg of gain per kg of DMI) than calves fed the low-LA MR over the entire period. These benefits occurred during the period of feeding MR alone and during the period of feeding MR and grain mix. Calves born from cows fed SFA prepartum had better ($P = 0.01$) ADG than calves born from cows fed UFA (0.50 vs. 0.45 kg/d due to a tendency ($P = 0.07$) to consume more DM (1.45 vs. 1.40% of BW. Calves fed the high-LA MR had a lower ($P < 0.01$) mean concentration of plasma BHBA (0.9 vs. 1.4 mg/dL). Calves born from dams fed fat tended ($P = 0.07$) to have a greater concentration of BHBA (1.2 vs. 1.0 mg/dL). Mean concentration of plasma NEFA was not affected by MR or prepartum dam diet (190 mEq/L). Increasing average intake of LA from 2.8 to 8.8 g/d from the MR improved calf performance.

Key Words: calves, milk replacer, linoleic acid

W167 The effect of automated feeder system feeding curves (dilution/weaning age) on growth and health of calves fed milk replacer. T. J. Earleywine*, B. L. Miller, and T. E. Johnson, *Land O'Lakes, Inc.*, *Webster City, IA*.

Forty-seven (47) Holstein bull calves with an average initial weight of 43.7 kg were employed in a 49-d trial to evaluate 2 feeding curves on an automated milk replacer (MR) dispensing unit (Förster Technik, Vario Milk Powder Feeder). Calves were allotted to treatment based upon weight and blood gamma globulin status. Calves assigned to both curves were fed a 25% protein/20% fat MR powder to provide equal amounts (24.6 kg DM) through weaning. MR contained milk, soy and

plasma protein sources. Calves were fed either a 12% solids solution MR providing on average 676 g DM per d for 6 wk (12sol/6wk) or a 15% solids solution MR providing on average 822 g DM per d for 5 wk (15sol/5 wk). Calves were on test through 7 wk to limit the effect of weaning age on growth parameters. MR was medicated with 0.28 g neomycin and 0.14 g oxytetracycline/kg. Calf starter (20% CP as-fed) was fed throughout this 49 d trial. Calves were housed by treatment in 2 group pens. There was a trend for increased total gain ($P = 0.08$) for the 15%/5 week treatment. MR intakes and health data were similar ($P \geq 0.37$). Scour score is a 1 - 4 scale (1 = normal, 2 = loose, 3 = water separation, 4 = 3 with severe dehydration). Respiratory score equals days treated with antibiotics for respiratory infection. Data were analyzed by the MIXED procedure in SAS.

Table 1. Feeding Curve

Item	12 sol/6wk	15sol/5wk	P-value	CV
n	23	24		
MR (DM), kg	21.5	21.7	0.60	6.68
BW gain, kg	20.2	23.9	0.08	32.41
Scour score (4 wk)	1.28	1.23	0.37	14.65
Respiratory score (7 wk)	2.12	1.57	0.40	121.68

Key Words: calf, milk replacer, automated feeder

W168 The effect of automated feeder system feeding curves (weaning age) on growth and health of calves fed milk replacer. T. J. Earleywine*, B. L. Miller, and T. E. Johnson, *Land O'Lakes, Inc.*, *Webster City, IA*.

Thirty-six (36) Holstein bull calves with an average initial weight of 43.9 kg were employed in an 84-d trial to evaluate 2 feeding curves on an automated milk replacer (MR) dispensing unit (Förster Technik, Vario Milk Powder Feeder). Calves were allotted to treatment based upon weight and blood gamma globulin status (calf is experimental unit). Calves assigned to both curves were fed a 28% protein / 20% fat MR powder (15% solution) to provide equal amounts (49.1 kg DM) through weaning at 8 or 7 wk. 8-wk weaned calves were offered on average 1021 g MR daily. 7-wk weaned calves were offered on average 1262 g MR/d. MR was medicated with 0.22 g neomycin:0.11 g oxytetracycline/kg. Calf starter (22% CP as-fed) was fed throughout this 84 d trial. As calves were housed by treatment in 2 group pens, starter intake could not be statistically analyzed. 7-wk weaned calves had strong trends for increases in total 8 wk weight gain ($P = 0.06$) and in 12 wk weight gain ($P = 0.08$). Hip height had a strong trend for increase ($P = 0.07$) and heart girth gain was increased ($P = 0.03$) for calves assigned to the 7-wk feeding curve. Data were analyzed by the MIXED procedure in SAS.

Table 1. Weaning age

Item	8 Weeks	7 Weeks	P-value	CV
MR (DM), kg	48.3	46.7	0.01	3.82
Starter (DM, 12 wk), kg	123.4	148.5	- -	- -
BW gain (8 wk), kg	43.3	49.4	0.06	19.88
BW gain (12 wk), kg	80.3	90.0	0.08	18.69
Scour score (4 wk)	1.14	1.19	0.22	11.39
Respiratory score (12 wk)	2.35	1.05	0.25	199.22
Hip height gain, cm	16.35	18.42	0.07	19.00
Heart girth gain, cm	28.15	31.58	0.03	15.47

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W169 Strategies for feeding full potential rates of calf milk replacer: Two feedings daily and weaned at 7 weeks vs. three feedings daily and weaned at 6 weeks. B. L. Miller*, T. J. Earleywine, and T. E. Johnson, *Land O'Lakes, Inc.*, *Webster City, IA*.

Thirty-eight (38) Holstein bull calves with an average initial weight of 47.9 kg were employed in an 83-d trial to evaluate 2 strategies for feeding full potential rates of milk replacer (MR). The strategies evaluated included: 2 feedings daily with calves weaned at 7 wk (2X/7wk) or 3 feedings daily with calves weaned at 6 wk (3X/6wk). Calves were allotted to treatment based upon weight and blood gamma globulin status. Calves were fed a milk replacer (28% protein / 20% fat) to provide equal total amounts (48.2 kg) of DM per strategy. Actual consumption of MR by treatment is provided. 2X/7wk calves were fed 568 g of MR per feeding or 1135 g of MR daily. 3X/6wk calves received 454 g of MR per feeding or 1362 g of MR daily. During the final wk, 2X/7wk and 3X/6wk calves were offered 568 or 454 g of MR respectively in one feeding (amount = to one feeding during the prior week). MR contained all milk protein and was medicated with 0.22 g neomycin and 0.11 g oxytetracycline/kg. Calf starter (22% CP as fed) was fed throughout this trial. Total gain and body measurements were improved for calves assigned to the 3X/6wk strategy. Data were analyzed by the MIXED procedure in SAS.

Table 1. Strategy

Item	2X/7wk	3X/6wk	P-value	CV
BW gain (7 wk), kg	33.2	36.3	0.6000	18.62
BW gain (12 wk), kg	67.8	77.2	0.0046	13.23
MR (DM), kg	41.8	44.9	0.0025	6.88
Hip height, cm	96.9	99.1	0.0182	2.73
Heart girth, cm	105.8	109.4	0.0004	2.60
Body volume gain, 1	174.2	198.2	0.0040	12.81

Key Words: calf, milk replacer, feedings