
To determine the elution pattern and electrophoretic characterization of granular-secretory fraction of the bovine fetal cotyledons, an experimental procedure was developed. A multi-protein complex extract of the granular-secretary fraction of the bovine fetal cotyledons, was frozen at -20°C, then it was centrifuged at 46,000 x g for 60 min in 4°C. After that, the supernatant was removed and saved by further use. The pellet was washed with a 0.05 M NH4HCO3 buffer solution at pH 7.8 and then it was centrifuged at 46,000 x g at 4°C for 60 min, the supernatant also was removed. Both supernatants were mixed and suspended together, then recovered protein was quantified reading in spectrophotometer at 595 nm. The suspension was applied to a 5 x 70 cm column of Sphadex G-75. The protein was eluted with a pH 7.8 buffer solution of 0.05M NH4HCO3 at 4°C. From the elution peak observed in the elution pattern, aliquots were obtained for electrophoretic characterization. The frictions with MW, were divided in aliquots of 50 mL or less. 0.125g of sacarose were added for each aliquot. Then were frozen at -20°C and lyophilized at 4 x 10⁻⁴ mbar. To characterize the electrophoretic pattern, SDS-PAGE with 12.5% gels were used. Protein recuperation of elution the process was 82.9%. The elution pattern showed three different peaks; the first one named peak A, has the 32.7% of the protein in 150 mL of eluent; in a second peak (B) 62% of protein was obtained in 165 mL of buffer; in a third peak (C), 4.8% of protein was obtained in 140 mL of eluent. The strip pattern of electrophoresis, produced three main regions, the first one grouped three proteins with the higher MW ranks from 42 to 95 kDa, and a second one constituted with proteins of intermediate MW (33.5 kDa); and third one grouping three proteins complex with the lower MW ranking from 20.5 to 28.5 kDa. It is concluded that the procedure described is able to produce partially purified placental bovine proteins usable in bioassays and ultrapurification process.

Key Words: Bovine, Placental , Protein Purification

1770 Granular-secretary fraction of the bovine fetal cotyledons: II. Effect on rate of growth of mice. F.G. Rios1, F.A. Nuez2, and R. Barajas1, 1FMVZ- Universidad Autonoma de Sinaloa. Culiacan, Sinaloa Mexico, 2FZ-Universidad Autonoma de Chihuahua.

There are many factors in animal tissues that promote animal growth. The purpose of this study is to evaluate the effect of placental bovine proteins usable in bioassays and ultrapurification process. We evaluated the possibility of using natural placental protein isolates as growth promoters. For this purpose, we observed the effect of placental bovine proteins obtained by ultrapurification from granular-secretory fraction of the bovine fetal cotyledons on growth of mice. The experiment was conducted in the Fernando M. V. Zepeda Research Center, FZUAS, Culiacan, Sinaloa, Mexico.

The null hypothesis is that the protein isolate does not affect the growth of mice. The hypothesis is tested by comparing the weight of mice that received the protein isolate with the weight of a control group that received a placebo. The results showed that the protein isolate had a significant effect on the growth of mice. The average weight of mice that received the protein isolate was significantly higher than the average weight of mice in the control group. These results suggest that placental bovine proteins can be used as growth promoters for mice.

Key Words: Bovine, Placental , Protein Purification
Previous work indicated that rainbow trout (and other fish) exhibit a tremendous growth response when administered exogenous GH. However, it is not known whether the fish is not characterized by the age of internal organs (empty body basis) was higher in the offspring of increasing growth rates in rainbow trout.

The influence of pST treatment of pregnant sows on live weight development, of nutrition, prior to and during gestation, on ovine fetal growth and development. Mature age Merino ewes (n=119) were stratified on liveweight and randomly allocated to 2 nutritional treatment groups: 30% roughage: 40% grain plus additives for 12 weeks prior to, and through- out pregnancy (p=0.44 and p=0.26 respectively). By day 133 fetal weight was significantly different between H (4.08±0.11kg) and L (3.57±0.15kg) fetuses (p<0.01). Singleton fetuses were heavier (4.20±0.11kg) vs twins (3.57±0.14kg) (p<0.01), while males (4.0±0.13kg) tended to be heavier than females (3.78±0.11kg) (p=0.18). However, there were no significant interactions between level of nutrition, litter size and sex. Liver, lungs and kidneys were heavier in day 133 H than L fetuses (p<0.05), while heart weight did not differ between treatments. These results indicate that the effect of nutrition, prior to and during gestation, on ovine early pregnancy was not of advantage for postnatal growth and carcass quality in the offspring.

**Key Words:** Somatotropin, Pig, Growth
fetal growth and development are not evident until the last trimester of pregnancy. However, the interaction between pre-natal nutrition and the expression of specific genes and implications for postnatal growth are yet to be determined.

Key Words: Sheep, Maternal nutrition, Fetal growth

1778 Stereoselectivity of porcine beta-adrenergic receptors for ractopamine isomers. J.D. Kissel¹, D.J. Smith², and S.E. Mills³, ¹Purdue University, ²USDA-ARS Fargo, ND.

Ractopamine is a β-adrenergic receptor (βAR) ligand that enhances protein gain in animals. The commercial product is a mixture of four stereoisomers. In rats, the levorotatory RR isomer was shown to be responsible for the growth response of ractopamine, consistent with known stereoselectivity.

β2AR were grown to confluence and cell membranes collected for centrifugation from lysed cells. Dissociation constants (Kd) were determined by competitive displacement of [125I]iodocyanopindolol binding by ractopamine isomers, and binding parameters were determined by non-linear regression. Membrane preparations were also used to quantify activation of AC; cAMP was measured by RIA after 10-min incubations with membranes and test ligands. Activation rates were expressed relative to isoproterenol. Experiments were replicated 3 to 5 times. The RR isomer had a high affinity for both β1- and β2AR (Kd of 25 and 29 nM, respectively); Kd estimates for the other stereoisomers were higher (RS = 463 and 78 nM, SR = 3,230 and 831 nM, SS = 16,600 and 3,530 nM for the β1- and β2AR, respectively). Isoproterenol stimulated AC activity 600% relative to basal rates with both βAR subtypes. Ractopamine stereoisomers did not significantly stimulate AC through the β1AR at moderate (near Kd) or high (10^-4M) concentrations. In contrast, the RR isomer increased AC activity 200 to 300% through the β2AR at moderate and high concentrations; RS and SR isomers elicited a 50 to 75% increase in AC, but the SS isomer was ineffective at the β2AR. The porcine β2AR exhibited the highest affinity for the RR isomer exhibiting the highest affinity for β1- and β2AR. In contrast, ractopamine stereoisomers seemed to be more effective at elicting cAMP responses from β2AR than β1AR.

Key Words: Pig, Receptor, Adrenergic

1779 Leptin in neonatal pigs: effects of oral versus intramuscular administration. N.C. Whitney¹, E.L. McFadin-Buff², P.R. Buff², and D.H. Keisler³, ¹University of Maryland-Eastern Shore, Princess Anne, MD, ²University of Maryland, Columbia, MO.

Leptin is found in milk and has been implicated in growth and development of neonates. Milk leptin levels are highest in the milk of sheep and swine during the first few days of lactation. This observation indicates that leptin may influence the order of lactation. This study was designed to investigate the role of leptin in milk oxidation (OX) and ketogenesis in pig liver. Glucagon (G); insulin (I) ratios of media were manipulated, to investigate effects of leptin with I and G in hepatocyte monolayers. Hepatocytes were isolated from 30-70kg barrows and seeded into pig tail collagen-coated T25 flasks. Monolayers were established in medium containing fetal calf serum and switched after 24h to serum-free media containing deoxymethasone (10^-6M) and 1% DMSO. To evaluate palmitate OX, a culture medium was formulated to maintain hepatocytes in an air environment (2.1 mixture of M199/HBSS and M199/EBSS containing 25mM HEPES). Cells were maintained in media containing 50ng/ml I, with G levels increasing from 0 to 500ng/ml. On day 4, medium containing 0.23mM 13C-palmitate was added to flasks and 6h gas samples were collected for determination of 13CO2 by IRMS. As the ratio of G:I decreased from 10:1 to 0.4:1, palmitate OX increased by 13.5% (P<.02), respectively. Meanwhile, cAMP level and IGF-I level increased by 20% (P<.02) and 25% (P<.02), respectively. The hormone sensitive lipid (HSL) total activity was increased (increased in I-treated monolayers in 25% (P<.02) in NMA treated pigs. The results indicated that adding of 50 mg/kg NMA to diet can induce the endocrine dramatic change in finishing pigs, further, inhibit the fat synthesis through suppressing fat synthetases and promote the fat degradation by elevating HSL activity in finishing pigs.

Key Words: Endocrine Response, Fat Metabolism, NMA

1780 Studies on lipid metabolism in hepatocytes from growing pigs. T.J. Caperna¹, I. Fernandez-Figares¹, A.E. Shannon¹, and D. Wray-Cahen², ¹USDA, ARS, Beltsville, MD, ²FDA, Rockville, MD.

Two in vitro assay systems were developed to elucidate the role of leptin in lipid oxidation (OX) and ketogenesis in pig liver. Glucagon (G); insulin (I) ratios of media were manipulated, to investigate effects of leptin with I and G in hepatocyte monolayers. Hepatocytes were isolated from 30-70kg barrows and seeded into pig tail collagen-coated T25 flasks. Monolayers were established in medium containing fetal calf serum and switched after 24h to serum-free media containing deoxymethasone (10^-6M) and 1% DMSO. To evaluate palmitate OX, a culture medium was formulated to maintain hepatocytes in an air environment (2.1 mixture of M199/HBSS and M199/EBSS containing 25mM HEPES). Cells were maintained in media containing 50ng/ml I, with G levels increasing from 0 to 500ng/ml. On day 4, medium containing 0.23mM 13C-palmitate was added to flasks and 6h gas samples were collected for determination of 13CO2 by IRMS. As the ratio of G:I decreased from 10:1 to 0.4:1, palmitate OX increased by 13.5% (P<.02), respectively. Meanwhile, cAMP level and IGF-I level increased by 20% (P<.02) and 25% (P<.02), respectively. The hormone sensitive lipid (HSL) total activity was increased (increased in I-treated monolayers in 25% (P<.02) in NMA treated pigs. The results indicated that adding of 50 mg/kg NMA to diet can induce the endocrine dramatic change in finishing pigs, further, inhibit the fat synthesis through suppressing fat synthetases and promote the fat degradation by elevating HSL activity in finishing pigs.

Key Words: Endocrine Response, Fat Metabolism, NMA

1781 Endocrine response and fat metabolism change in finishing pigs treated with N-methyl-d,l-aspartate(NMA). Gang Xi*, Zirong Xu², and Ping Xiao², ¹University of Minnesota, St. Paul, MN, ²Zhejiang University, Hangzhou, China.

A trial was conducted to investigate the effect of the additional of NMA on several growth related hormones and fat metabolism in finishing pigs. A total of 84 cross bred finishing pigs (average initial BW of 56±.37 kg) were divided into 6 pens, 14 pigs per pen (7 gilts and 7 barrows per pen). 3 pens of pigs were fed with control diet (corn-soybean meal) and the others were fed control diet addition with 50 mg/kg NMA. During trial, all pigs were given free access to feed and water. After 44 days trial, 8 pigs from each treatment (4 gilts and 4 barrows, weight similar to average group weight, 86.94±.71 kg for control group, 90.55±.15 kg for NMA treated group ) were sacrificed to collect the sample of liver, longissimus muscle, subcutaneous fat (10th rib), and hypothalamus. Before the pigs were slaughtered, blood samples were collected and analyzed for serum leptin, cAMP, and IGF-I. Serum leptin concentrations (P<.001) leptin increased serum concentrations of leptin (P<.001) leptin increased serum concentrations of leptin over time, with significant increasing the highest serum leptin concentrations (P<.001). Serum leptin averaged 6.2 ± 4.9, 19.5 ± 4.15 and 107.1 ± 5.4 ng/ml for controls, O and I pigs, respectively. Leptin receptor mRNA (leptin receptor mRNA:28S rRNA) was not influenced by treatment and averaged 7.7 ± 2.2, 5.5 ± 2.1 and 6.3 ± 2.2 relative optical density units for C, O, and I treatments, respectively. Milk samples taken from the pigs after parturition were analyzed for leptin and averaged 98.9 ng/ml. It is concluded that oral administration of leptin can be absorbed to influence peripheral leptin levels. Leptin levels are highest in the milk of sheep and swine during the first 24 hours after parturition, and this study supports the hypothesis that leptin may play a role in the fetal to neonatal transition process.

Key Words: Leptin, pig milk

sensitive to hormonal manipulation, chronic exposure to rh-leptin did not directly influence lipid OX and metabolism. Supported in part by a Spanish Ministry of Education fellowship (IF-F).

**Key Words:** Ketogenesis, Lipid oxidation, Leptin

### 1782 Recruitment and differentiation of intramuscular preadipocytes in stromal-vascular (S-V) cell cultures derived from fetal pig semitendinosus muscles.

G.J. Hausman, R. Gaines, and S.P. Paulos, USDA ARS, Athens, GA.

Semitendinosus muscles in 105 day old fetuses contain a small number of intramuscular adipocytes. Therefore, we examined intramuscular preadipocyte development in S-V cell cultures derived from semitendinosus muscles from 105 day old fetuses. Both semitendinosus muscles were excised from 4-8 fetuses removed from each of four dams laparotomized at 105 days of gestation. All visible connective tissue was removed from the muscles prior to mincing and processing with a conventional collagenase digestion used to establish adipose tissue S-V cell cultures. Four muscle S-V cell cultures were established since muscles were pooled for each litter. After 1 hour in 10% fetal bovine serum (FBS) one-half of the muscle S-V cell cultures were rinsed to remove debris and insoluble muscle protein. Cultures were reacted for the AD-3 antibody, a preadipocyte marker. Only cultures containing large clusters of preadipocytes (AD-3+) were randomly distributed and not clustered after 1 day in FBS. After 4-5 days in 10% FBS muscle S-V cell cultures reached confluence with large clusters of preadipocytes. Treatment with insulin + dexamethasoneDEX(5) for 5 days after confluence did not increase preadipocyte number(P>0.05), but markedly increased preadipocyte size with no consistent change in lipid accretion. The proportion of large preadipocyte clusters (>3 cells) was similar before and after DEX + insulin treatment (75%±12% and 75%±9%,respectively) but was reduced (P<0.05) to 46±6% in cultures treated with insulin alone. Muscle S-V cell cultures seeded on laminin coated dishes contained only preadipocytes whereas laminin coated dishes contained preadipocytes and earlier precursor cells in previous studies of adipose tissue S-V cell cultures. Thus, preadipocyte precursor cells are not present in fetal muscle.

**Key Words:** preadipocytes, pig muscle, differentiation

### 1783 The effect of LXRα ligands on adipocyte differentiation.

T.D. Brandebourg* and C.Y. Hu, Oregon State University, Corvallis.

We recently demonstrated that ligand activation of LXRα, an orphan receptor expressed in adipose tissue, negatively regulates adipocyte differentiation. Administration of an LXRα agonist inhibited differentiation of 3T3-L1 cells while geranylgeraniol (GG), a metabolite of mevalonate that antagonizes LXRα, blunted the inhibitory effect of the LXRα agonist. The objective of this study was to extend those results by evaluating the effect of several LXRα ligands ([22(R)‑hydroxycholesterol (22R), 22(S)‑hydroxycholesterol (22S), 20(S)‑hydroxycholesterol (20S), 25‑hydroxycholesterol (25OH)],) on the differentiation of 3T3-L1 preadipocytes. Cells were grown to confluence (d -2) and differentiation was induced on d 0. Cells were treated with either ligand or carrier (ETOH) from d 0 to d 7. In separate experiments, either individual LXRα ligands or cholesterol (Chol), which neither activates nor binds LXRα, were administered at concentrations of 2.5, 5, 10, or 20 mM. Differentiation was evaluated by measuring sn‑glycerol‑3‑phosphate dehydrogenase (GPDH; EC 1.1.1.8) activity on d 8. Independent experiments were performed on duplicate wells where n=4 for 22R, 20S, 25OH and Chol and n=2 for 22S. 25OH significantly decreased GPDH activity (nmol/(min*mg protein) versus controls by 42% at 2.5 mM (p<0.001), 53% at 5 mM (p<0.01), 61% at 10 mM (p<0.001) and 66% at 20 mM (p<0.001). Administration of 22R decreased GPDH activity by 9% at 2.5 mM (p<0.05), 17% at 5 mM (p<0.01), 35% at 10 mM (p<0.001) and 41% at 20 mM. Administration of 20S decreased GPDH activity by 12% at 2.5 mM (p<0.001), 20% at 5 mM (p<0.01), 27% at 10 mM (p<0.01) and 57% at 20 mM (p<0.001). Conversely, administration of 22S, which is known to bind LXRα with high affinity, failed to significantly alter GPDH activity at any concentration. As expected, Chol did not affect GPDH activity at any concentration tested. These results further support a role for LXRα in the regulation of adipocyte differentiation. 25OH was most efficacious while 22R and 20S decreased GPDH activity with similar efficacies. Importantly, 22S represents a potential LXRα antagonist that may prove useful in subsequent studies of the role of LXRα as a regulator of adipocyte differentiation.

**Key Words:** Adipose Tissue, Differentiation, LXR

### 1784 Hormonal regulation of postnatal chicken preadipocyte differentiation in vitro.

T. G. Ramsay* and R. W. Rosebrough, USDA-ARS, Beltsville, MD.

The present study was designed to develop a chronic culture system from the stromal vascular fraction of chicken adipose tissue for use in identifying hormones or peptides that promote adipocyte formation. Abdominal adipose tissue was excised from 2-4 week old male broilers by sterile dissection. The stromal vascular cell fraction from the adipose tissue was isolated by collagenase digestion, filtration, and subsequent centrifugation. These preadipocytes were seeded in six well culture plates and proliferated to confluence in 10% fetal bovine serum in DMEM/F12 (50:50) medium. At confluence, experiments were initiated to determine hormonal requirements for differentiation in the presence of 2.5% or 10% chicken serum. Isobutylmethylxanthine (10 mM) in combination with 1 mM dexamethasone could not promote differentiation, as determined by the expression of citrate lyase (CL) and sn-glycerol-3-phosphate dehydrogenase (GPDH) relative to lactate dehydrogenase. Insulin (100 nM) stimulated expression of CL and GPDH (P<0.05) in the presence of 2.5% chicken serum, but not with 10% chicken serum. Triiodothyronine (1 nM) and IGF-I (100 ng/ml) had no effect on differentiation. Dexamethasone (1 nM) stimulated differentiation in 2.5% or 10% chicken serum (P<0.05). The combination of insulin and triiodothyronine stimulated differentiation (P<0.05) but the effect was no greater than insulin alone in medium containing 2.5% chicken serum. Insulin, dexamethasone and 2.5% chicken serum synergistically stimulated differentiation and can replace 10% chicken serum in culture. Development of a culture system that only requires low serum concentrations for stimulating adipocyte formation may permit identification of important regulatory hormones for differentiation.

**Key Words:** Preadipocyte, Differentiation, Chicken

### 1785 Effects of dietary protein on the endogenous calpain/calpastatin proteolytic system in canine skeletal muscle.

E. E. Heilman¹, E. H. Lonergan¹, S. M. Lonergan¹, and G. M. Davenport²,¹ Iowa State University, Ames, IA. The Iams Company, Lewisburg, OH.

The cysteine proteinases µ- and m-calpain along with their inhibitor, calpastatin, and possibly skeletal muscle specific p94, have been hypothesized to play a role in skeletal muscle protein degradation. Previous studies have indicated a nutritional influence on calpastatin. Our working hypothesis is that protein nutrition can influence regulation of the calpain system in muscle. Our objectives were to determine the effects of dietary protein on the expression of calpastatin and p94 in canine skeletal muscle. A biopsy was taken from the semitendinosus of 56 dogs prior to and after 12 weeks on their respective diets. This experimental design allowed us to examine change within individual dogs. Our study consisted of 8 diets with 7 dogs per diet. Diets 1-4 were 12% total protein and contained ratios of chicken to corn gluten protein of 100:0, 67:33, 33:67, and 0:100%, respectively. Diets 5-8 were 28% total protein with identical protein ratios to diets 1-4. We examined these differences qualitatively using SDS-PAGE and immunoblotting, and quantitatively with densitometric analysis. Western blots were examined using an anti-calpastatin antibody (MA945, Affinity Bioreagents). p94 blots were examined with an anti-p94 antibody (NCL-CALP-12A2, Novocas- trolabs). The majority of our calpastatin blots showed an expression of three distinct calpastatin bands, the uppermost appearing at approximately 110 kDa. Diet 5 resulted in an increase in the expression of the 110 kDa calpastatin band. A significant difference (P<0.05) was obtained from comparison of the ratio of relative intensity in all three bands when comparing diet 5 (100:0) to diet 8 (0:100). Our results showed no treatment differences in detection of p94. Our calpastatin data suggest that dogs fed a diet containing a higher percentage of chicken protein may have a greater potential to regulate calpain-mediated degradation of muscle protein.

**Key Words:** p94, calpastatin, canine
The objective of this study was to determine if a one step-program for dairy heifers during the transition period. Nineteen Holstein heifers averaging 770 kg of body weight were randomly divided into two treat- ments. The treatments were imposed at 95 d of gestation. The control group (CON) was fed a diet containing 14% crude protein and 22.5 Mcal of metabolizable energy (ME) per d for the entire 180 d of the trial. The treatment group (TRT) was fed a diet containing 18.5% crude protein and 14.5 Mcal of ME per d until d 185 of gestation, then the diet was changed to 14% crude protein and 29.2 Mcal of ME per d for the re- alimentation period. Heifers were weighed for three consecutive d at the start, at 185 d of gestation and after calving. Body weights were not different at 185 d of gestation (P = 0.3), or at calving (P = 0.71). Blood was drawn from the heifers around parturition on d -13, -10, -7, -1, -2, -1, 0 (within 3 h of calving), 0.5, 1, 2, 3, 5, 7, 10, 13 to monitor various metabolites, white blood cell counts, and lymphocyte popula- tions (CD3, CD4, CD8, and gamma/delta T-cells). Blood glucose levels before calving were higher in the TRT group (P = 0.08), but were not affected after calving. Insulin levels were increased in the TRT group before calving (P = 0.03), but they were not statistically different after calving. Triglycerides were increased before calving and after calving in the TRT group (P = 0.06, after P = 0.01). There were no differ- ences in non-esterified fatty acid concentrations before or after calving. White blood cell counts did not differ between treatments before or after parturition. The results support our hypothesis that compensatory growth during the last trimester of gestation improves metabolic status of prepartum heifers.

Key Words: Heifer, Transition, Compensatory growth, Blood metabolites

1789 Effects of added rumen undegraded protein and bovine somatotropin administration on skeletal growth rates in prepubertal dairy heifers. U. Moeller5, E. G. Dahl1, E. K. Duffey-Tower1, A. V. Capuco5, and R. A. Erdman1, 1University of Maryland, College Park., 2USDA-ARS, Beltsville, MD.

The objective of this study was to test effects of added rumen un- degraded protein (RUP) and recombinant bST administration on skeletal growth in dairy heifers from 90 days of age until onset of puberty. Fifty Holstein heifers (90 days of age) were used in the experiment and were randomly assigned to one of four treatment groups. Treatments con- sisted of added dietary RUP (52%, DM basis) and 0.1 mg/kg BW/d recombinant bST applied in a 2 x 2 factorial design. Weekly blood pro- gesterone concentrations, measured beginning at 180 d, were used to determine onset of puberty. Body weight (BW), wither height (WH), and hip height (HH) were measured every 2 weeks. Average age at puberty (314 ± 23 d) was not affected by treatment (P > .05). Daily growth rates for BW, HH and WH were increased by both RUP and bST alone and these effects were additive in the combination of RUP and bST (RUP+bST) were additive. The combination of bST and RUP resulted in 42.8 kg, 2.9, and 2.9 cm increases, respectively in 315 d final BW, WH and HH as compared to Controls. Across treatments, average daily BW gain increased from 647 g/d at 105 d to 1330 g/d at 315 d of age while WH and HH rates decreased from 0.15 and 0.16 cm/d at 105 d to .13 and 0.13 cm/d at 315 d of age, respectively. After 200 to 210 d of age, the combination of bST and RUP was the only treatment that significantly increased rates of BW gain compared to Controls. Skele- tal growth rates were increased by added dietary RUP between 90 and 200 d of age, but not later. Conversely, the effects of bST on skeletal growth rates were small at the early ages but increased as the heifers approached puberty. These results suggest that the potentiation of growth during the early post-weaning period, whereas circulating bST was more important during the time just prior to puberty.

Key Words: Myostatin, Inclusion body, Purification


The objective of this study was to determine if a one-step-program for dairy heifers during the transition period. Nineteen Holstein heifers averaging 770 kg of body weight were randomly divided into two treat- ments. The treatments were imposed at 95 d of gestation. The control group (CON) was fed a diet containing 14% crude protein and 22.5 Mcal of metabolizable energy (ME) per d for the entire 180 d of the trial. The treatment group (TRT) was fed a diet containing 18.5% crude protein and 14.5 Mcal of ME per d until d 185 of gestation, then the diet was changed to 14% crude protein and 29.2 Mcal of ME per d for the re- alimentation period. Heifers were weighed for three consecutive d at the start, at 185 d of gestation and after calving. Body weights were not different at 185 d of gestation (P = 0.3), or at calving (P = 0.71). Blood was drawn from the heifers around parturition on d -13, -10, -7, -5, -3, -2, -1, 0 (within 3 h of calving), 0.5, 1, 2, 3, 5, 7, 10, 13 to monitor various metabolites, white blood cell counts, and lymphocyte popula- tions (CD3, CD4, CD8, and gamma/delta T-cells). Blood glucose levels before calving were higher in the TRT group (P = 0.08), but were not affected after calving. Insulin levels were increased in the TRT group before calving (P = 0.03), but they were not statistically different after calving. Triglycerides were increased before calving and after calving in the TRT group (P = 0.06, after P = 0.01). There were no differ- ences in non-esterified fatty acid concentrations before or after calving. White blood cell counts did not differ between treatments before or after parturition. The results support our hypothesis that compensatory growth during the last trimester of gestation improves metabolic status of prepartum heifers.

Key Words: Heifer, Transition, Compensatory growth, Blood metabolites

1787 Solubilization and purification of a recombinant chicken myostatin expressed as inclusion bodies in E. coli. Y. S. Kim*, K. S. Baek, and M. A. Dunn1, 1University of Hawaii, Honolulu, HI, 2National Livestock Research Institute, Namwon, Korea.

The objective of this study was to solubilize and purify a recombinant chicken myostatin fragment expressed as inclusion bodies in E. coli culture. Plasmids containing a 369 bp C-terminal fragment of chicken myo- statin were transformed into expression competent E. coli, followed by IPTG-induced protein expression. Inclusion bodies from the cells were isolated and washed to homogeneity. An average of 80 mg of inclusion body proteins was produced per L of culture. The myostatin inclusion bodies were solubilized in 50 mM Tris buffer containing 8 M urea, 5 mM EDTA, and 200 mM mercaptoethanol. The effect of buffer pH (8, 9, 10, and 11) and incubation condition (30 min boiling and overnight incubation at 25°C with mild shaking) on inclusion body solubilization and purification was investigated using a combination of centrifugation, gel filtration, and SDS-PAGE. When the extent of solubilization was examined by the formation of precipitates after centrifugation at 11,000 g for 10 min, all the above conditions solubilized inclusion bodies at 3 mg/mL. Significant breakdown of recombinant myostatin was observed during the solubilization at pH 10 and 11 in both incubation condi- tions, but not at pH 8 and 9. A significant proportion of recombinant myostatin stayed as heterogeneous multimer forms in the solubilized in- clusion bodies, and increasing pH favored monomer formation. Thus, 30 min boiling or 25°C overnight incubation at pH 9 provided the opti- mum condition for inclusion body solubilization, and the proportion of monomer was approximately 50% in these conditions. Solubilized inclu- sion body proteins were refolded using a slow dialysis process by stepwise addition of Tris buffer containing no urea or mercaptoethanol. When the dialyzed inclusion body solutions were subjected to gel filtration, the myostatin monomer fraction was less than 1% of the total protein. This result demonstrates that heterogenous multimer formation had occurred during the dialysis process. Also, it was demonstrated that buffer pH and incubation conditions are important factors affecting solubilization of inclusion bodies.

Key Words: Myostatin, Inclusion body, Purification

1786 Growth of myoblasts derived from genetically different mice, pigs, and cattle. C. Rehfeldt*, G. Nurnberg, U.K. Zettl, E. Mitz, M. Wittstock, U. Renne, J.H. Papstein, and K. Endler. 1Research Institute for the Biology of Farm Animals, Dum- merstorf, Germany, 2Rostock University, Rostock, Germany.

The objective of this study was to investigate the influence of long-term selection for different growth traits on intrinsic differences in myoblast proliferation activity and susceptibility to serum deprivation. Myoblasts were isolated from lines of mice and bovines of different ages (d 4 to 6 of cultivation (17 to 30%), DNA levels did not clearly differ. Serum deprivation by changing to 1% FBS during exponential growth as examined in the murine cell lines responses being higher in all long-term selected lines. In part, this was due to apoptosis as examined with DU-Ks and DU-6 cells. Higher per- centages of apoptotic cells were found in cultures of DU-Ks (P < 0.001) and DU-6 (P < 0.0001) cells cultivated in 1% as compared with 10% FBS. The results suggest that long-term growth selection is capable to induce intrinsic changes in myoblasts that determine muscle growth.

Key Words: Growth, Myoblast, Selection
The objective of this study was to test effects of added rumen undergraded protein (RUP) and recombinant bST administration on body composition. In a companion growth experiment, added RUP increased rates of BW and skeletal growth early (<200 d age) while bST increased BW and skeletal growth rates at the end of the experiment (>200 d age). Twenty-four Holstein heifers housed and fed with animals in the growth experiment were randomly assigned to 1 of 4 treatment groups beginning at 90 d of age. Treatments consisted of added dietary RUP (+2%, DM basis) and 0.1 mg/kg BW/d recombinant bST applied in a 2 x 2 factorial design. Twelve heifers, 3 from each treatment group, were slaughtered at 5 mo and another twelve at 10 mo of age. Body weight at 90 d of age was used as a covariate in the statistical analysis to adjust for differences in chemical composition. Across treatments, empty body fat and energy increased with age from 9.1% to 13.9%, and 1.87 to 2.31 mc/kg at 5 and 10 mo of age (P < 0.0001), respectively. There were no significant effects of age on empty body ash and protein content. Across slaughter ages, bST increased ash (P < 0.06), tended to increase protein (P < 0.13) and decreased body fat content (P < 0.05). At 10 mo of age, EB weight was increased by bST, RUP, and bST plus RUP by 23.6, 11.3, and 20.8 kg, respectively. The amounts of body ash (P < 0.04) and protein (P < 0.07) were increased by bST whereas amounts of body fat were not changed at 10 mo of age. In summary, added RUP did not influence body chemical composition while bST altered body composition at 10 mo by redirecting increased growth towards skeletal development as shown by increased ash and protein deposition.

### Key Words:
- bST, RUP, Skeletal growth

**1790 Effects of added rumen undergraded protein and bovine somatotropin administration on organ and tissue weights in prepuberal dairy heifers. U. Moallin1, G. E. Dahl2, A. V. Capuco2, R. L. Baldwin2, and R. A. Erdman2.**

Twenty-four dairy heifers were used to determine the effects of added rumen undergraded protein (RUP) and recombinant bST administration from 3 to 10 months of age on organ and tissue weights. The heifers (90 days of age) were blocked randomly into 2x2 factorial trial, which consisted of added dietary RUP (+2%, DM basis) and 0.1 mg/kg BW per day of recombinant bovine somatotropin (bST). Twelve heifers, 3 from each group were slaughtered at 5 mo of age and another twelve at 10 mo of age (estimated onset of puberty). Weights of rumen and intestinal tract components including digesta fill along with liver, heart, lungs, spleen and other organs at the time of slaughter. Body weight at 90 d of age was used as a covariate in the statistical analysis to adjust for differences in initial tissue weights. At 5 mo, bST increased the average liver weight by 28.4% compared to Controls and when expressed as a fraction of EBW by 10% (P < 0.05). Rumen undegraded protein increased the empty rumen + reticulum and abomasum weights by 24.4% and 11.5% and as a fraction of EBW by 16% (P < 0.01) and 6% (P < 0.10) compared with controls, respectively. Rumen digesta fill was increased by 27.3% by added RUP (P < 0.05). This enlargement of the anterior digestive tract is consistent with the 10% increased DMI across slaughter ages, bST increased the average weights of heart (28%, P < 0.05), 10% (NS), and 5% (NS), respectively. In addition, at 10 mo, omasum, small and large intestine were increased by 22.3%, 22.1% and 22.3%, respectively, by bST administration. These results indicate that administration of bST to growing heifers altered relative organ growth, in a manner that would support increased metabolic activity associated with partitioning of nutrients toward increased protein deposition. In comparison, the added RUP effects were shown only through increased mass of the digestive tract and digesta fill.

### Key Words:
- bST, RUP, Organ weights


The study included 115 single-born crossbred Black-and-White heifers that were sampled at the age of 2, 21, 42, 90, 200, and 440 days. At sampling, the heifers were weighed, excepting the day 21 of birth. There were found numerous significant partial correlation coefficients between concentration of serum immunoglobulins and growth rate traits, that were most informative for immunoglobulins determined on the days 2 and 90 after birth. Serum somatotropin concentration measured on the day 2 was significantly correlated with body weight at the age of 200 days (r = 0.21; P < 0.05), and body weight gains measured in different periods within the first 200 days of life (r from 0.21 to 0.24; P < 0.05). All correlation coefficients between concentration of serum immunoglobulins determined at the 90 days of age and body weight gains measured beginning from the day 2 or 42 after birth were negative and greater than 0.4 (P < 0.0001), ranging from -0.42 (42-200 days) to -0.56 (2-90 days). It is concluded, that in growing dairy heifers passive immunity is associated positively with body weight gains during the first 200 days after birth, and that growth rate is associated negatively with immunity acquired actively during the post-natal period of life.

### Key Words:
- Heifers, Serum immunoglobulins, Growth rate

**1792 Relationships between concentration of serum immunoglobulins and growth rate of dairy heifers. W. Jarmuz1, I. Szelag1, and R. Skrzypek1.**

The objective of this study was to test effects of added rumen undergraded protein (RUP) and recombinant bST administration on body composition. In a companion growth experiment, added RUP increased rates of BW and skeletal growth early (< 200 d age) while bST increased BW and skeletal growth rates at the end of the experiment (> 200 d age). Twenty-four Holstein heifers housed and fed with animals in the growth experiment were randomly assigned to 1 of 4 treatment groups beginning at 90 d of age. Treatments consisted of added dietary RUP (+2%, DM basis) and 0.1 mg/kg BW/d recombinant bST applied in a 2 x 2 factorial design. Twelve heifers, 3 from each treatment group, were slaughtered at 5 mo and another twelve at 10 mo of age. Body weight at 90 d of age was used as a covariate in the statistical analysis to adjust for differences in chemical composition. Across treatments, empty body fat and energy increased with age from 9.1% to 13.9%, and 1.87 to 2.31 mc/kg at 5 and 10 mo of age (P < 0.0001), respectively. There were no significant effects of age on empty body ash and protein content. Across slaughter ages, bST increased ash (P < 0.06), tended to increase protein (P < 0.13) and decreased body fat content (P < 0.05). At 10 mo of age, EB weight was increased by bST, RUP, and bST plus RUP by 23.6, 11.3, and 20.8 kg, respectively. The amounts of body ash (P < 0.04) and protein (P < 0.07) were increased by bST whereas amounts of body fat were not changed at 10 mo of age. In summary, added RUP did not influence body chemical composition while bST altered body composition at 10 mo by redirecting increased growth towards skeletal development as shown by increased ash and protein deposition.

### Key Words:
- bST, RUP, Body composition
natural cow-calf systems, feeding frequencies may also affect postnatal development. In this study growth performance and metabolic and endocrine traits in calves fed C (3 L on d 1, 4 L on d 2, and 5 L on d 3) and milk (5 - 11 L from d 4 up to d 28) by a computer-programmed automate that allowed frequent daily intakes (GrA; n = 7) were compared with calves twice daily pair-fed by bucket (GrB; n = 7). Blood samples were taken after birth on d 1, 2, 14, 21, and 28 before feed intake, on d 3 before and 1, 2, 4, 6 and 8 h after feed intake, and on d 7 every 20 min for 8 h. Plasma concentrations of total protein (TP), urea, glucose, and triglycerides (TG) were measured photometrically. Plasma concentrations of insulin, glucagon, growth hormone (GH), insulin-like growth factor (IGF-I) and thyroid hormones were measured by RIA. Data were evaluated by analysis of variance using a mixed model with different feeding frequencies and time as fixed effects and the individual calves as random effects. Weight gains did not differ between groups. Plasma TP was higher from d 14 to 28 in GrB than GrA. Plasma glucose increased postprandially (d 3 and 7) in GrB, remained unchanged in GrA, and was higher postprandially (on d 7) in GrA than in GrB. Preprandial plasma TG (on d 7) were higher and mean postprandial concentrations (d 7) tended to be higher in GrA than in GrB. Insulin concentrations (d 7) tended to be higher in GrA than in GrB. During an 8-h period GH (d 7) was higher from 120 to 140 min, but was lower from 240 to 360 min after feeding in GrA than in GrB. Plasma IGF-I was higher in GrA than in GrB on d 7, 14 and 28. In conclusion, feeding at high frequency with an automate transiently changed some metabolic and endocrine traits, but had no significant effects on growth performance during the first 4 wk of life.

Key Words: Neonatal Calves, Feeding Frequency, Metabolites and Hormones


Developmental anomalies, as well as high rates of fetal and postnatal mortality have been reported in somatic-cell cloned cattle. Our objective was to determine if the development of the somatotropic axis in somatic clones (clones) was similar to AI produced heifers (controls). We compared the changes in Growth Hormone (GH), Insulin-like Growth Factor (IGF-I) -1 and IGF Binding Proteins (BP) -2 and -3 of 4 clones generated from a 13-year-old cow with 4 age matched controls from 5 to 15 mo of age. Once a month for 7 mo, serum samples were collected every 30 min for 6 h. Animals were also challenged with GH Releasing Factor (GRF; 3 µg/100kg BW) and Somatostatin (SRIF; 1.87 and 5 µg/100kg BW) at mo 10. Concentrations of GH for clones and controls were compared with 4 post-pubertal heifers (PP; 11 mo of age). Averaged across all time points, concentrations of GH were not different between clones (7.29 ± 96 ng/mL) and controls (5.50 ± 89 ng/mL). However, there was a decline in GH over time in controls, but not in clones (p < .01). GH concentrations in PP animals were less than controls in mo 1 (p < .01) and less than clones in mo 2 (p = .05). When PP, clones and controls were compared at similar ages, concentrations of GH were not different between the three groups. GRF-induced GH secretion was greater in clones than controls (p < .02). SRIF (1.87 µg) inhibition of GRF-induced GH was less (p < .01) in clones than controls. IGF-I concentrations of clones and controls paralleled each other over time. However, overall concentrations of IGF-I were less in clones than controls (203.7 ± 13.8 vs 306.4 ± 13.1 ng/mL). BP-3 was greater in controls than clones (85 ± 3.7 vs 70 ± 3.7%), but did not change over time for either group. BP-2 did not change over time and average concentrations were not different between clones and controls. Although, there were some differences in measures of the somatotropic axis between these clones and these age-matched controls, values of each variable measured were within reported ranges for cattle of similar ages, indicating that these clones have normal development, in terms of the somatotropic axis. However, further studies are required using clones derived from different cell types and from different donor animals to validate this conclusion.

Key Words: Somatic Cell Derived Clones, Growth Hormone, Insulin-like Growth Factor-1

1979 Feed Intake Patterns, Metabolic and Endocrine Traits, and Growth Performance During the First Month of Life of Calves Provided Restricted or Unlimited Amounts of Colostrum and Milk with an Automate. J.W. Blum*, A. Nussbaum, G. Schiessler, and H.M. Hammon, University of Berne, Switzerland.

Amounts of colostrum (C) and milk intake greatly influence postnatal development of calves. Automatic feeding systems allow calves to drink high amounts of C and milk already during wk 1 of life. High C and milk intakes in GrL were accompanied by moderate metabolic and endocrine changes. High weight gains in wk 1 could not be maintained up to 1 month.

Key Words: Feeding intensity, Growth performance, Metabolites and Hormones

1979 Glucose metabolism in Holstein and Jersey calves fed milk replacer once versus twice daily. C. M. Cheatham*, C. C. Williams1, J. M. Fernandez1, W. A. Nipper1, H. G. Bateman1, J. C. Lovejoy2, D. T. Gantt3, L. R. Gentry3, and G.E. Goodier1, 1Louisiana State University Agricultural Center, Baton Rouge, LA, 2Pennington Biomedical Research Center, Louisiana State University, Baton Rouge, LA.

Eighteen Holstein and fifteen Jersey heifer calves were fed milk replacer (MR) once (1x) or twice (2x) daily to determine effects of two feeding systems on glucose metabolism during the neonatal period. From birth to 5d of age all calves were fed colostrum or whole milk. From d5 to d6 of age, all calves were fed MR 2x. Beginning on d7 of age, calves fed 2x were offered 15g MR DM/kg birth weight reconstituted to 15% DM. Calves fed 1x were offered 15g MR DM/kg birth weight reconstituted to 21% DM. At 5 wk, MR was decreased by 50% for all calves followed by abrupt weaning at 6 wk. Calves were allowed free access to grass and water throughout the experiment. At weeks 3, 6, and 10 minimal Model intravenous glucose tolerance tests were performed to assess glucose effectiveness (Sg), an estimate of insulin-independent glucose disappearance; insulin sensitivity (S1), an estimate of insulin-dependent glucose disappearance; and acute insulin response (AIRglucose), insulin secretion relative to glucose administration. Of the three parameters measured, Jersey calves differed from Holsteins only by having greater S1 (P < 0.05). The S1 and Sg were similar for calves regardless of MR feeding frequency (P > 0.10). However, S1 decreased with age for all calves (P < 0.05) while AIRglucose increased with age (P < 0.05) in calves fed MR 2x. AIRglucose was greater than in calves fed MR1x (P < 0.05). These data suggest that feeding calves MR 1x versus 2x did not significantly affect glucose metabolism during the neonatal period.

Key Words: Calves, Glucose metabolism, Insulin sensitivity

1979 Post-weaning growth of cattle destined for Japanese and Korean markets: Relationships between growth during backgrounding and intramuscular fat percentage (IMF%) at slaughter. S. A. Hardin,1,2 P. L. Greenwood1,2, and V. H. Oddy1,4, 1 Cooperative Research Centre for Cattle and Beef Quality, Armidale, Australia, 2NSW Agriculture, Beef Industry Centre, University of New England, Armidale, NSW 2351, Australia, 3NSW Agriculture, Tamworth Centre for Crop Improvement, Tamworth, NSW 2340, Australia, 4Meat and Livestock Australia, 165 Walker Street, North Sydney, NSW 2000, Australia.

Effects of growth rate during backgrounding (weaning to commencement of finishing) on intramuscular fat percentage (IMF%) at slaughter of beef cattle destined for Japanese or Korean markets were assessed. Cattle were grazed on temperate perennial pastures on the Northern Tablelands of NSW, Australia (4 experimental years; years 1 to 4, n=329, 273, 290 and 107, respectively). Angus, Hereford, Murray Grey and Shorthorn steers were grown either without supplementation or with supplementary feeding of high protein pellets or with access to a forage crop to an overall group mean target LW of 400 kg. They were finished either on pasture or in the feedlot for Korean (average LW 520 kg) or Japanese (average LW 600 kg) market weights. IMF% was measured on a sample of m. longissimus at the 12/13th rib using near infrared spectroscopy. The variance in IMF% was analysed for each year by fitting a mixed model; the terms were market category, finishing system, market category x finishing system, breed, carcass weight and backgrounding growth rate. All terms contributed significantly to IMF% (P<0.05). The proportions of total variation in IMF% accounted for across the years were: market (9% to 22%), breed (7% to 14%), carcass weight (1% to 8%), market category x finishing system (9% to 9%), finishing system (16% to 31%), backgrounding growth rate (7% to 6.1%) and unexplained (residual) variation (30% to 63%). The results show that backgrounding growth rate had a small positive effect on IMF%, and that finishing system had a greater effect on IMF%. However, the high degree of unexplained variation supports the need for further investigations into factors affecting IMF% at different market weights.

**Key Words:** Cattle, growth rate, intramuscular fat

1800 Effect of Synovex-S® on pituitary-thyroid axis response to challenge with a combination of thyrotropin releasing hormone (TRH) and growth hormone releasing hormone (GHRH) in beef steers. S. Kahl*, T. S. Rumsey, and T.H. Elsasser, USDA, Agricultural Research Service, Beltsville, MD.

Thyroid status is an important regulator of growth. Studies have suggested that thyroid hormones may be involved in the mechanism of action of estrogen-based growth promoters in steers. This study evaluated the effect of Synovex-S® ear implants (SYN, 20 mg estradiol benzoate + 200 mg progesterone) on basal and TRH/GHRH-stimulated plasma concentrations of thyrotropin (TSH), thyroxine (T4), and triiodothyronine (T3), and on hepatic and pituitary 5’-deiodinase (5’D) activity. Sixteen crossbred steers (404 ± 13 kg) were fed individually a 30:70 silage concentrate diet (17.8% CP, 2.87 Mcal ME/kg DM) to gain 1.2 kg BW daily and assigned to no implantation (control) or implantation (n = 8). Two days before and 14, 28, 42, and 56 d after implantation, all steers were challenged (i.v.) with a combination of TRH + GHRH (1.0 + 0.1 μg/kg BW, respectively). For each challenge jugular blood samples were obtained at 0, 10, 20, 30, 40, 60, 120, 240, and 360 min after challenge for TSH analysis and at 0, 1, 2, 4, 6, 8, 12, and 24 h after challenge for T4 and T3 analyses. Primary response to challenge was measured as area under the time × concentration curve (AUC). Liver and pituitary samples were collected at slaughter 5 d after the last challenge. Compared to control steers, SYN did not affect (P > 0.05): (a) basal plasma concentrations of TSH, T4, and T3, (b) TSH and T3 responses to TRH + GHRH challenge at any time after implantation, and (c) 5’D activity in liver (type-I) and pituitary (type-II). However, SYN increased T4 response to each TRH + GHRH challenge (742 vs 589 ng/mL × h, SEM = 52, P < 0.05). Results indicate that the primary action of SYN on the pituitary-thyroid axis in steers is greater sensitivity of the thyroid gland to TSH stimulation resulting in increased T4 secretion.

**Key Words:** Growth promoters, Thyroid hormones, Thyrotropin
1801 Performance, carcass characteristics and plasma levels of thyroid hormones and insulin like growth factor-I in feedlot intact crossbred (Bos taurus × Bos indicus) Brazilian Superyoung System. 1 O Cheng*, 1 J. A. Ferro, 2 A. C. Silveira 3, L. R. Furlan 3, M. D. B. Arrigoni 3, H. N. Oliveira 1, M. I. T. Ferro, and M. Macari 2. 1UNESP - Botucatu, SP/Brazil, 2UNESP - Jaboticabal, SP/Brazil.

The objective of this study was to evaluate the performance, carcass characteristics and the levels of metabolic hormones in male and female young cattle of different genetic groups kept in feedlots and slaughtered at 12 months of age. Two-hundred male and female young cattle, crossbred from five breeds (Charolais, Gembivie, Aberdeen Angus, Hereford and Simmental) and Simmental × Nellore crossbred females, were weaned at seven months of age and kept in feedlots for 180 days in a completely randomized design. Diets were formulated according to the age (growth and finishing phases) and weight of animals. The performance was evaluated by weight gain and slaughter weight. The plasma levels of thyroid hormones were evaluated by EIA and the levels of IGF-I by RIA. Hereford animals had a higher slaughter weight when compared to the other breeds (p<0.01). Males had better performance than females for all characteristics evaluated (p<0.01). Ultrasonography measurements showed a higher marbling rate in Aberdeen Angus animals, as well as lower hindlimb percentage and chilling losses, which resulted in a higher fat index in the carcass (p<0.01). Aberdeen Angus animals had higher triiodothyronine (T3) levels and higher IGF-I levels during finishing phase as compared to the other breeds (p<0.01). Correlations between T3 and subcutaneous fat (r=0.76, p<0.01) and marbling (r=0.79, p<0.01) were significant as well as between IGF-I and subcutaneous fat (r=0.67, p<0.01) and marbling (r=0.59, p<0.01). Among the breeds considered in this study, Aberdeen Angus seems to be the most adequate to be used in crossings for intensive meat production like the Brazilian Superyoung System.

Key Words: Crossbred cattle, Meat production, Plasma hormones

1802 Effects of estradiol administration and level of protein intake on nitrogen metabolism and insulin-like growth factor-1 (IGF-1) gene expression in muscle in growing steers. O Cheng*1, W Knaus1, M Boehm1, and D Beermann1,2, 1Cornell University, 2University of Nebraska at Lincoln.

Modulation of IGF-1 expression in muscle was assessed using a ribonuclease assay developed to quantify the abundance of IGF-1 mRNA in ovine and bovine tissues. Four Holstein steers weighing 250 kg were fed a low-protein diet (7.6% CP) or a diet supplemented with urea to meet the ruminal requirement for N (12.2% CP) and administered twice daily subcutaneous injections of estradiol-17 beta (500 micrograms at 12-hr intervals) or excipient using a 4 x 4 Latin Square design. Daily N retention increased from 10 to 18 g/d and plasma urea nitrogen (PUN) increased from 3.6 to 7.8 mg/dL (both P<0.05) when the higher protein diet was fed. The higher level of protein intake increased IGF-1 mRNA abundance in the semimembranosus muscle to 189% of control levels (P<0.05), and circulating IGF-1 concentration was not altered. Estradiol administration increased plasma estradiol 42% (P<0.05). In skeletal muscle growth in steers may be controlled through autocrine or paracrine influence of IGF-1 mRNA abundance. The short-term twice-daily estradiol administration failed to enhance N balance, suggesting that either a longer treatment period or a higher level of protein intake is necessary for estradiol to exert its anabolic effect in growing Holstein steers.

Key Words: Muscle growth, IGF-1, estradiol

1803 Temporal effects of daily estradiol administration on nitrogen metabolism and insulin-like growth factor-1 (IGF-1) gene expression in liver and skeletal muscle in growing lambs. O Cheng*1, M. Boehm1, and D Beermann1,2, 1Cornell University, 2University of Nebraska at Lincoln.

Twelve Suffolk-sired crossbred wether lambs weighing 23 kg were fed a diet containing 67% barley, 15% soy hulls and 13.5% soybean meal (15.6% CP). The objective was to assess the temporal effects of subcutaneous administration of 175 micromoles of 17beta-estradiol on plasma estradiol, urea nitrogen (N) and IGF-1 concentrations, on daily N balance, and IGF-1 gene expression in liver and skeletal muscle. A ribonuclease assay was developed to quantify IGF-1 mRNA abundance in muscle and liver RNA samples. Daily N balance and jugular blood samples were collected from all lambs over a 7-day control period. During the following 7-day estradiol treatment period two lambs were immunized each day at 1, 2, 3, 4, 5, and 7 days for liver and muscle sample collection. Daily N balance samples were collected from each lamb until removed for tissue sample collection. Subcutaneous administration of estradiol increased plasma estradiol concentration 17-fold (from 50 to 450 pg/ml) within 15 min of injection and maintained concentrations at 300 pg/ml at 75 min and 150 pg/ml 300 min after administration (P<0.01). Estradiol administration did not alter daily fecal and urinary N excretion, daily N balance, biological value, or PUN concentrations (all P>0.05). Likewise, circulating IGF-1 concentration, and liver, semitendinosus and longissimus muscle IGF-1 mRNA abundance were not altered at any of the treatment intervals (all P>0.05). The lack of anabolic response to estradiol administration was unexpected. Control period variable means for N balance (11 g/day), biological value (0.59) and PUN concentrations (15 mg/dL) were higher than expected. These data suggest that the lambs were receiving adequate protein intake and may have been too close to their genetic potential for rate of protein gain, or that the treatment period was too short for a response to estradiol administration to be observed.

Key Words: Estradiol, N balance, IGF-1 expression


Twenty-two young intact male pigs were used in an 18-week growth assay to determine the effects of immunization against LHRH on growth performance, sex characteristics, and meat quality. At 20 kg BW and 8 weeks later, control animals (C) were injected with vehicle only, and the remaining males were immunized with an anti-LHRH peptide vaccine at either 40 (L) or 100 (H) µg per injection. Compared with control intact males, immunized pigs tended (P<1) to have high ADG (for C, L, H: 698, 765, 837 g/d) and ADFI (1.78, 1.92, 2.14 kg/d) with no changes in F/G ratio (2.55, 2.55, 2.58). Greater backfat (2.17, 2.65, 2.91 cm) and loin eye area (45.9, 44.8, 56.2 cm2) were detected in pigs treated with anti-LHRH vaccine at 100 µg level (P<0.05). Tenderness and juiciness of logissimus muscle chops did not differ among groups by sensory panel evaluations, but chops from immunized animals had less off-flavor (P<0.1) than intact controls. Genital tract weights (testes, epididymes, seminal vesicles, and prostate glands), measured at slaughter, were significantly (P<0.001) decreased by immunization, and both doses of anti-LHRH vaccine had similar efficacy. Consistently, plasma testosterone concentrations were completely inhibited (P<0.001) by anti-LHRH vaccine. The present results demonstrate that anti-LHRH immunization is an effective means to inhibit sexual development and to reduce incidence of boar taint for intact male pigs.

Key Words: Pigs, Immunocastration, LHRH
60 weanling CD rats (5 wk old; 75 g BW) were euthanized and anterior pituitary glands removed. Individual cells were dispersed with dispase and cells were maintained at 37°C (humidified atmosphere; 95% air: 5% CO2) in Dulbecco modified Eagle’s medium for 24 h. Cells were then treated with one of the following (2 wells /treatment; replicated in 3 experiments): 1. controls; 2. 10 nM T3; 3. 50 M DTPA; 4. 10 nM T3 plus 50 M DTPA; and 5. 10 nM T3, 50 M DTPA and 40 M zinc sulfate. Cells were incubated for 48 h prior to RNA extraction. Reverse transcription (RT) PCR, with specific primers for GH and thyroid stimulating hormone (TSH), was used to measure mRNA levels. Comparisons with control gene-specific primers RPL-32 and β-actin were used to calculate mRNA level of GH and TSH, respectively. To evaluate RT-PCR, preliminary experiments were performed with GHI cells. Results with RT-PCR and the GH primer paralleled previous results using Northern analysis, showing that expression DTPA and T3 stimulated GH mRNA expression. TSH was measured to confirm the viability of the anterior pituitary cells. As expected, T3 inhibited TSH mRNA expression 50% compared to controls. Expression of TSH mRNA was greater in DTPA treated cells compared with controls (p<0.01), but was lowered by combination with T3 and by combination with T3 and zinc. These results confirm that the anterior pituitary cells are viable and responsive. In these same cells DTPA alone reduced GH mRNA levels by 25% (p=0.03). However, in contrast to GH3 cells, DTPA did not stimulate GH mRNA levels in T3-treated pituitary cells (p=0.18). In conclusion, while zinc chelation increases TSH mRNA expression or absence of T3, it does not affect the GH mRNA in primary rat anterior pituitary cells in the same manner as GH3 cells.

Key Words: Rat, Growth hormone, Thyroid hormone

1808 Effects of dietary conjugated linoleic acid (CLA) on the composition and function of peripheral blood mononuclear leukocyte populations in heifer calves. J.M. Smith1, B.J. Nonnecke2, M.E. Van Amburgh1, B.A. Pesch1, and J.A. Harp1, 1 Cornell University, Ithaca, NY, 2 National Animal Disease Center (NADC), USDA, ARS, Ames, IA.

At approximately 2.5 months of age, heifer calves, raised and housed at the Cornell University Dairy Teaching and Research Facility, began receiving a diet formulated to support 1 kg/d gain (controls, n = 6) or a diet containing protected CLA (treated, n = 6). Total CLA was included in the diet at 1% of DM intake. The CLA-supplemented diet was formulated to support the same levels of ME and MP allowable gain as the control diet. Peripheral blood was collected at the initiation of the study (100 kg BW) and at 6.5 (200 kg BW) and 9.5 (300 kg BW) months of age. Anti-coagulated blood was maintained at room temperature and shipped overnight to the NADC where the composition and function of circulating mononuclear leukocyte (PBML) populations were evaluated. The composition of PBML populations was evaluated by flow cytometry. The total number of PBML was unaffected (P > 0.05) by dietary treatment or age. Percentages of CD4+ T cells (and CD4+, CD8+, and γδ T cell subsets) and B cells in the PBML population were unaffected (P > 0.05) by dietary CLA; however, the proportion of T cells did increase (P < 0.01) with age. Percentages of PBML expressing activation antigens (i.e. MHC class II antigen and interleukin-2 receptor) were also unaffected by dietary CLA or age. Leukocyte function was evaluated in vitro by measuring interferon-γ (IFN-γ), nitric oxide, and tumor necrosis factor-α secretion in PBML cultures, both unstimulated and mitogen-stimulated (i.e. with pokeweed mitogen (PWM), concanavalin A, and phytohemagglutinin-P). In general, dietary CLA did not affect these functions. The only exception was greater IFN-γ secretion by PWM-stimulated cells from heifers supplemented with CLA. Although these data suggest dietary CLA had minimal effect on the composition and function of PBML from healthy calves, additional research is needed to determine if dietary CLA would benefit calves experimentally or naturally infected with pathogens causing significant morbidity or mortality in the field.

Key Words: Conjugated linoleic acid, Immune function, Dairy heifers

1807 Feeding conjugated linoleic acid to reduce the impact of an infectious disease challenge in growing swine. J.A. Brown*, G.W. Almond, S.A. Mathews, W.T. Oliver, and R.J. Harrell, North Carolina State University, Raleigh, NC.

Respiratory diseases, particularly in the grow-finish phase, account for considerable economic loss in the swine industry. Studies in chucks and rostans have shown dietary conjugated linoleic acid (CLA) reduced the catabolic effects of a noninfectious inflammatory challenge. Our objective was to maintain performance and(or) reduce the duration of lower performance during an infectious disease challenge of porcine reproductive and respiratory syndrome virus (PRRSV) and M. hyopneumoniae (M. hyo).) by supplementing diets with CLA. Pigs were weaned from the sow at 12 days of age and reared in isolated facilities to ensure high health status. At 26.7±0.6 kg BW, 16 barrows were moved to metabolism cages and randomly assigned to a 2x2 factorial arrangement: diet (0 or 2% CLA-60) and disease challenge (uninfected or infected with PRRSV and M. hyo.). Pigs were allowed a 2 week adjustment period to their respective diet prior to infection. Pigs were then inoculated with PRRSV and M. hyo., and control pigs were given sterile media. Blood samples were taken weekly, and total urine and feces were collected during the third week post infection. Pigs were euthanized at a constant BW of 63.4±1.1 kg. PRRSV titers were not present initially, and only infected pigs were positive at the conclusion of the trial. In addition, only infected pigs had lung lesions typical of M. hyo. No differences in average daily feed intake, efficiency of gain, N-retention, plasma urea nitrogen, or total blood protein were found (P>0.11). Infected pigs had lower blood albumin (P<0.03) and tended to have reduced ADG (P<0.06) compared to uninfected pigs, but dietary CLA did not attenuate the reduced growth performance (P>0.20). Results thus far suggest that CLA does not attenuate the reduced growth performance associated with an infectious disease challenge. However, beneficial effects of CLA may have been compromised by the lack of severity of the disease challenge.

Key Words: Swine, Conjugated linoleic acid, Disease

AMSAS/ASAS Meat Science and Muscle Biology

1808 Prediction of the fat content of pork carcasses based on cross-sectional region analysis of dual energy X-ray absorptiometry scans. A. D. Mitchell*, A. M. Scholz1, and V. G. Purde1, 1 USDA, Agricultural Research Service, Beltsville, MD, 2 Ludwig Maximilians University-Munich, Oberschleisheim, Germany.

Dual energy X-ray absorptiometry (DXA) can be used to measure pork carcass composition by performing a total scan of the half-carcass. The scan can be analyzed for total or regional fat, lean, and bone mineral content, but is too slow for on-line slaughter application. The purpose of this study was to determine the feasibility of predicting carcass composition based on a single cross-sectional measurement. A total of 252 right half-carcasses (42.7 ± 5.2 kg) were scanned by DXA. The DXA scans were analyzed for percentage fat in the entire half-carcass as well as the shoulder, ham, loin, and side regions. A total of 14 cross-sections (57.6 mm wide) were analyzed: 6 in the shoulder/thoracic region, 3 in the loin region, and 5 in the ham region. Regression analysis was used to compare the DXA fat percentage measurements in the total carcass with those of the various regions. The mean fat content of the half-carcasses was 24.1 ± 7.0%; shoulder region, 23.8 ± 6.7%; ham region 22.9 ± 6.7%; loin region, 23.7 ± 7.6%; and the side region 27.9 ± 7.6%. The correlation (R2) between the fat content of a single cross-sectional slice and total fat content ranged from 0.908 to 0.976. The highest correlations were in the area of the last ribs. Based on previous results, it is estimated that a single slice could predict the percentage of carcass fat by chemical analysis with an R2 of 0.80. The highest correlations between single cross-section and region analysis were: shoulder, 0.978; ham, 0.972; loin, 0.973; and side, 0.959. These results indicate that carcass fat percentage can be measured by performing a single-pass cross-sectional scan that would be compatible with on-line processing.

Key Words: Carcass Composition, DXA, Swine