

1769 Granular-secretory fraction of the bovine fetal cotyledons: I. Elution pattern and electrophoretic characterization. F.G. Rios¹ and F.A. Nuez², ¹FMVZ- Universidad Autonoma de Sinaloa, Culiacan, Sinaloa Mexico., ²FZ-Universidad Autonoma de Chihuahua..

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-secretory fraction of the bovine fetal cotyledons, was frozen at -20°C, then was centrifuged at 46,000 x g for 60 min at 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 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 fractions with MW, were divided in aliquots of 50 mL or less. 0.125g of saccharose were added for each aliquot. Then were frozen at -20 °C and lyophilized at -44 x 10⁻³ 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-secretory 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 mouse is an adequate animal model to carry out exploratory tests of these substances. Our objective was to determine if an extract of a granular-secretory fraction of bovine fetal cotyledons would promote rate of growth of mice. Sixty pre-pubertal 20-day-old mice (Balb/c strain; male and female; BW 11.4 g), were used in a complete randomized experiment. The animals were allocated individually in metabolic crates, with free access to feed and fresh water. After two days adaptation period, mice were divided into five groups (12 mice/group; six males and six females). The groups were randomly assigned to receive a daily intramuscular injection of various proteins. The treatments were: 1) No injection (control); 2) High Molecular Weight (42-95 kDa) placental protein isolate (HMWP); 3) Intermediate Molecular Weight (33.5 kDa) placental protein isolate (IMWP); 4) Low Molecular Weight (20-28.5 kDa) placental protein isolate (LMWP); and 5) Bovine somatotropin (bST). Body weight and feed intake were recorded daily for a 10 d experimental period. Rate of growth was calculated as average daily gain as percentage of final weight. Feed efficiency was calculated as weight gain divided by feed intake. The experiment was analyzed as a complete randomized design, with comparison of means performed by least significant difference. Feed intake, which averaged 4.12 g/d, was not affected (P>0.10) by treatments. The HMWP treatment improved (P<0.01) growth rate 10.6% (4.71 vs 4.26%/day). Feed efficiency was increased (P<0.01) 16.3% by HMWP (0.235 vs. 0.202 g of gain/g of feed). The other treatments had no effect (P>0.10). We conclude that high molecular weight proteins isolated from the granular-secretory fraction of the bovine fetal cotyledons improves growth in mice.

Key Words: Mice, Growth, Placental Proteins

1771 Tibial lesions in broiler chicks after feeding different dietary concentrations of calcium and ammonium chloride. I. B. Toure*, S. Weisbrode, and J. D. Latshaw, *The Ohio State University.*

Different concentrations of dietary calcium and ammonium chloride were used to influence the development of the tibial growth plate. Day-old male broiler chicks in battery pens were fed 1.0, 0.8 or 0.6% Ca and another diet of 1.0% Ca plus 0.5% NH₄Cl. All diets contained 0.47% NPP and 200 ICU of vitamin D per kg. At 10, 17 and 24 days, six chicks were randomly selected from each treatment. They were euthanized, and the left tibia was removed and cleaned of soft tissue. Calipers were used to measure the length of the bone and the diameter at mid-length. The metaphysis of each end was sectioned longitudinally to permit assessing the growth plate. Distance from growth plate to growth plate was recorded. The bones were fixed in 10% formalin, sectioned, and stained with hematoxylin and eosin. Prior to euthanasia at 24 days, blood was collected by heart puncture to provide plasma for blood chemistry profiles. Plasma from the 0.6% Ca chicks was significantly lower in Ca (8.2 mg/dL) than that of the other chicks (10.1 mg/dL). At 17 days, chicks fed 1.0% Ca had normal growth plates, tibial dyschondroplasia-like lesions (TDLL) were present in those fed 1.0% Ca + 0.5% NH₄Cl (6/6), 0.8% Ca chicks had TDLL (4/6), and 0.6% Ca chicks had rickets (3/6) or rickets at the distal end and TDLL at the proximal end (3/6). A marked proliferation of osteoprogenitor cells with the formation of spicules and congested vascular channels was noted in birds with the 1.0% Ca + 0.5% NH₄Cl treatment and was the predominant distinctive feature between lesions from that treatment and the 0.8% Ca treatment. We have hypothesized that the marked cellular proliferation could be due to bone morphogenic proteins that are released from bones under the influence of NH₄Cl induced acidosis.

Key Words: Calcium, Growth plate, Tibial dyschondroplasia

1772 Relationships between a single-point mutation in the chloride channel-1 gene and phenotypic responses in the Myotonic goat. B. L. Sayre*, S. Wildeus, M. P. L. Dismann, and J. R. Collins, *Virginia State University, Petersburg, VA.*

The Myotonic goat has congenital myotonia, a mutation in the chloride channel-1 (ClC-1) gene, often associated with muscle hypertrophy. This experiment determined relationships between the presence of a ClC-1 mutation and expression of myotonia in a herd of Myotonic goats. Myotonic does (n=63) were genotyped and phenotyped for presence of congenital myotonia. Spanish does (n=18) were used as negative controls. DNA was harvested from whole blood samples. A 177-bp ClC-1 fragment was amplified with PCR, and then digested with Mbo II. The presence of two (105 & 72 bp), three (72, 53, & 52 bp), or four (105, 72, 53, & 52 bp) fragments was used for classification of genotype as homozygous normal (NORM), homozygous mutated (MYO), or heterozygous (HET), respectively. Phenotype was determined as (1) the duration of muscle knots after a sharp strike with a percussion hammer (five repetitions; PH), (2) duration of myotonia (MD), and (3) severity of myotonia (Scored on a scale of 1-9; MS). After a 20 min rest, does had to jump over a step (approx. 45 cm), which induced myotonia in positive does for measurement of MD and MS. There was a positive correlation between genotype and phenotype (P<.001), and phenotypic measures were positively correlated to each other (P<.001). There were no differences (P>.1) in phenotypic measures between NORM and HET genotypes. MYO had greater (P<.001) MD (5.5 vs. 0.0 sec), MS (5.4 vs 1.0), and PH (4.0 vs.1.1 sec) than NORM and HET. Older MYO does (>2 yrs) had increased (P<.01) MD (6.8 vs. .9 sec), MS (6.6 vs. 1.8), and PH (4.8 vs. 1.5 sec) compared to younger does (<2 yrs). Myotonic goats homozygous for a mutated ClC-1 gene exhibited classic phenotypic responses for congenital myotonia, while heterozygous goats did not show signs of myotonia. In goats homozygous for the mutation, the phenotypic response is less apparent in younger animals than older animals. Future studies will link carcass characteristics to the genotype and phenotype relationships.

Key Words: Myotonic goat, Myotonia, Chloride Channel-1

1773 Effect of Somatostatin-14 (SS-14) and Passive Immunization Against SS-14 on Circulating Levels of Growth Hormone (GH) in Rainbow Trout (*Oncorhynchus mykiss*). B. C. Peterson*, P. R. Simpson, R. W. Hardy, T. L. Ott, A. Ahmadzadeh, and G. T. Schelling, ¹University of Idaho, Moscow, ID/USA.

Previous work indicated that rainbow trout (and other fish) exhibit a tremendous growth response when administered exogenous GH. However, regulation of growth hormone secretion in fish is not well characterized. The objectives of this study were to further examine endogenous GH secretion by determining (1) circulating concentrations of GH in resting fish, (2) the effect of SS-14 on GH release, and (3) the effect of passive immunization against SS-14 on GH release in rainbow trout. In the first study, 10 fish (665 g) were bled from the caudal vessel every 20 min for 4 h to assess the pattern of GH release. Any value of GH greater than 2 SD above the mean, followed by at least 2 values of lesser concentration, was considered a pulse. No pulses were detected in any of the 10 fish and serum GH averaged 4.0 ± 2.6 ng/ml. In the second study, 10 fish (550 g) were used to assess the effect of administration of SS-14 on GH release. Fish received an i.p. injection of 5 ng/g BW of SS-14 or saline. Serial blood samples were taken for 4 h starting 20 min pre-treatment. Results of the study indicated SS-14 decreased ($P = 0.09$) GH levels (1.9 ± 1.7 ng/ml vs 6.4 ± 1.5 ng/ml) compared to control fish. In the third study, 20 fish (450 g) were used to assess the effects of passive immunization against SS-14 on GH release. Fish were selected randomly to receive an i.p. injection of SS-14 anti-serum diluted 1:25 in saline or control serum diluted 1:25 in saline. Fish passively immunized against SS-14 increased ($P < 0.01$) GH levels (12.9 ± 1.0 ng/ml vs 4.9 ± 1.1 ng/ml) when compared to control fish. These results indicate GH in rainbow trout is released in an asynchronous fashion with no evidence of pulsatility and the hypothalamic hormone SS-14 apparently regulates GH secretion similarly in rainbow trout as it does in mammals. Elevation of GH by immunizing against SS-14 may provide another approach of increasing growth rates in rainbow trout.

Key Words: Somatostatin-14, GH, Rainbow Trout

1774 WITHDRAWN , .

1775 Growth and carcass quality of offspring in response to somatotropin (pST) treatment of sows during early gestation. G. Kuhn, C. Rehfeldt*, G. Nrnberg, and K. Ender, *Research Institute for the Biology of Farm Animals, Dummerstorf, Germany.*

The objective of this study (two experiments) was to investigate the influence of pST treatment of pregnant sows on live weight development, compositional characteristics, and meat quality of their offspring. Sixteen sows received daily i.m. injections of 6 mg pST from d 10 to 27 after artificial insemination (pST). Sixteen sows received a placebo (control). Three neonates of the highest (HW), middle (MW), and lowest (LW) birth weights were selected from each litter for dissection into body tissues followed by chemical analysis. The remaining piglets were allocated to the three birth weight classes (HW, MW, and LW) by the help of frequency distribution and reared up to 182 days of age. After slaughter, carcass composition and meat quality of the pigs were examined. All data were statistically analyzed using the mixed model of analysis of variance. The average birth weight of the piglets from treated sows was lower ($P = 0.11$) compared with control piglets with HW neonates being most different ($P = 0.01$). Treatment of sows with pST influenced the body composition of the newborn piglets by increasing the percentages of internal organs ($P = 0.01$) and skin ($P = 0.02$). No differences in protein and lipid content of the piglet body were apparent between the two groups. Postnatal daily gains during various growth phases were not significantly affected, although live weights were lower until d 49 of age due to pST treatment ($P < 0.05$). At slaughter, the percentage of internal organs (empty body basis) was higher in the offspring of treated sows than in corresponding controls ($P < 0.05$). In longissimus muscle, drip loss and intramuscular lipid content were increased due to pST ($P < 0.05$). Moreover, the pH₄₅-value and the brightness of longissimus muscle tended to be affected by pST treatment ($P = 0.08$) to an undesired direction. In conclusion, pST treatment of sows during

early pregnancy was not of advantage for postnatal growth and carcass quality in the offspring.

Key Words: Somatotropin, Pig, Growth

1776 Effect of oxytocin (OT) on hourly milk secretion in gilts with mastitis. R. S. Kensinger*, D. M. Sanzotti, A. L. Magliaro, A. C. W. Kauf, and L. C. Griel, Jr., *Penn State University.*

The objective of the study was to determine if OT treatment (20 IU given IM) on d 3, 5 and 7 of lactation would increase hourly milk yield (HMY) in gilts with intramammary inflammation. The goal is to improve baby pig performance. Gilts (176.4 kg BW; $n = 15$ in 5 groups of 3) farrowed, and pigs were cross-fostered to equalize litter size to 9.4. The experimental model was lactating Yorkshire crossbred gilts infused with 3.0 ug endotoxin/kg BW into 2 mammary glands at 0715 on each experimental day. The design was a 3X3 Latin Square Design. Litters were separated from dams between hourly weigh-suckle-weigh measurements (from 0800–1700) in order to assess HMY. Gilts were assigned to receive 0, 3, or 5 OT injections (0X, 3X, or 5X) at 0, 4, or 2 h intervals, respectively. Data were analyzed using the GLM option of SAS according to a mixed model design. Rectal temperature (RT) averaged 39.7 °C at 0700, increased to 40.8 °C at 1200 ($P < .01$), and returned to 39.9 °C at 1700 h. Gilts on 3X and 5X had significantly increased RT compared to 0X ($P < .01$). Mean HMY was 100.7, 111.4 and 124.3 g on d 3, 5 and 7 of lactation, respectively ($P < .02$). Mean HMY by 0X gilts was 112.3 g. Mean HMY by 3X and 5X gilts averaged 80.9 g during nursings with no OT injection, and 160.1 g for nursings with OT treatment ($P < .01$). Response to OT treatment attenuated over time in the 5X group, as HMY averaged 173.5 g with first two injections, 137.6 g at third injection, but only 108.9 g at last two injections. The 3X group averaged 150.6, 233.6 and 182.5 g at 0800, 1200 and 1600, respectively. A treatment by time interaction was detected ($P < .01$). Mean HMY (for all nursings) for 0X, 3X and 5X treatment groups averaged 112.3, 117.6 and 106.5 g, respectively ($P > .10$); and there was no difference in 24-h litter weight gains. Results show OT injections improve milk yield at that nursing; but suggest that stress of injections may interfere with response. Data indicate a need for better methods of OT delivery.

Key Words: Porcine lactation, Oxytocin, Endotoxin

1777 Influence of long-term maternal nutrition on ovine fetal growth and development. SP Quigley¹, DO Kleemann¹, SK Walker¹, JA Owens², PI Hynd², G Natrass¹, SR Barritt¹, and PA Speck¹, ¹South Australian Research and Development Institute, South Australia, ²University of Adelaide, South Australia.

Genetic and environmental factors during the pre-natal period affect the growth of the fetus and may set the potential for postnatal growth. The objective of this experiment was to investigate the effect of long-term nutritional manipulation of the pre-natal environment on fetal growth and muscle development in sheep. Mature age Merino ewes ($n=119$) were stratified on liveweight and randomly allocated to 2 nutritional treatments of 1.8 and 0.6 times maintenance requirements (High (H) $n=60$ and Low (L) $n=59$, respectively). Ewes were fed a pelleted diet (60% roughage: 40% grain plus additives) for 12 weeks prior to, and throughout pregnancy. Pregnant ewes from each treatment group were stratified on litter size, determined by ultrasonography on day 44, and liveweight and were randomly allocated to one of three sample points (day 50, 92 and 133 of gestation). Ewes were euthanased and the fetuses recovered, weighed and dissected and internal organ weights recorded. Samples of internal organs and longissimus dorsi, semitendinosus and supraspinatus muscles, were taken for gene expression analysis. H ewes were significantly heavier than L ewes ($p<0.01$) at slaughter on day 50 (73.9 ± 2.1 vs 54.8 ± 2.3 kg), day 92 (80.1 ± 1.8 vs 57 ± 3.4 kg) and day 133 (86.1 ± 2.5 vs 59.3 ± 3.6 kg) respectively. Nutrition had no significant effect on fetal weight and organ development during the first 50 and 92 days of pregnancy ($p=0.44$ and $p=0.26$ respectively). By day 133 fetal weight was significantly different between H (4.08 ± 0.11 kg) and L (3.57 ± 0.15 kg) fetuses ($p<0.01$). Singleton fetuses were heavier (4.20 ± 0.11 kg) than twins (3.57 ± 0.14 kg) ($p<0.01$), while males (4.0 ± 0.13 kg) tended to be heavier than females (3.78 ± 0.11 kg) ($p=0.18$). 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

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. Kissel^{*1}, D.J. Smith², and S.E. Mills¹, ¹Purdue University, ²USDA-ARS Fargo, ND.

Ractopamine is a β -adrenergic receptor (β AR) ligand that enhances protein gain in animals. The commercial product is a mixture of four stereoisomers. In rats, the levorotatory RR isomer seemed to be responsible for the growth response of ractopamine, consistent with known requirements for biological AR-ligand activation. In order to determine the AR stereoselectivity for ractopamine stereoisomers, binding affinities and adenyl cyclase (AC) activation were determined using cloned porcine β 1- and β 2-AR. Chinese hamster ovary cells stably expressing the porcine β 1AR or β 2AR were grown to confluence and cell membranes collected by 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 β 1- and β 2AR (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 β 1- and β 2-AR, respectively). Isoproterenol stimulated AC activity 600% relative to basal rates with both β AR subtypes. Ractopamine stereoisomers did not significantly stimulate AC through the β 1AR at moderate (near Kd) or high (10^{-4} M) concentrations. In contrast, the RR isomer increased AC activity 200 to 300% through the β 2AR 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 β 2AR. The porcine β AR exhibited stereoselectivity towards ractopamine stereoisomers with the RR isomer exhibiting the highest affinity for β 1- and β 2-AR. In contrast, ractopamine stereoisomers seemed to be more effective at eliciting cAMP responses from β 2AR than β 1AR.

Key Words: Pig, Receptor, Adrenergic

1779 Leptin in neonatal pigs: effects of oral versus intramuscular administration. N.C. Whitley¹, E.L. McFadin-Buff^{*2}, P.R. Buff², and D.H. Keisler², 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 greatest during the first hours after parturition, then decline and remain low throughout the remainder of lactation. This observation indicates that leptin may influence the neonate if leptin absorption occurs with oral consumption of leptin. The objective of this study was to compare peripheral serum concentrations of leptin in neonatal pigs after oral or intramuscular administration of leptin. Thirty-three pigs from 6 sows were used. Between 5 and 20 hours after birth, pigs were weighed, numbered, and treated with either a single intramuscular injection of sterile saline (C), or 2.5 mg/g BW ovine leptin, administered either orally (O) or intramuscularly (I). Pigs from each treatment were chosen for sacrifice at 2, 4, 6, or 8 h after treatment. At euthanasia, blood samples were collected for analysis of serum leptin levels. Liver tissue samples were collected and total liver RNA was used for determination of leptin receptor mRNA expression using porcine 28S and leptin receptor probes in a slot blot technique. There was no effect of time after treatment on either variable. Both oral ($P < .07$; tendency) and injected ($P < .001$) leptin increased serum concentrations of leptin over that of control pigs, with injection producing the highest serum leptin concentrations ($P < .001$). Serum leptin averaged 6.2 ± 4.9 , 19.5 ± 4.5 and 101.7 ± 5.4 ng/ml for controls, O and I pigs, respectively. Leptin receptor mRNA (leptin receptor mRNA:28S mRNA) 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 sows soon after parturition were also 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

1780 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 in diet on several growth related hormones and fat metabolism in finishing pigs. A total of 84 cross bred finishing pigs (average initial BW of 56 ± 0.37 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 ± 0.71 kg for control group, 90.55 ± 1.51 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 allowed to clot overnight at 4 °C. Serum was then harvested after centrifugation and stored at -20 °C until assayed. The addition of NMA in diet increased the IGF-I, Insulin, T₃, T₄ and TSH levels in serum with 50.68% ($p < .05$), 62.61% ($p < .02$), 123.33% ($p < .01$), 60.58% ($p < .03$) and 78.40% ($p < .02$), respectively. Meanwhile, cAMP level and IGF-I level in the liver were increased with 26.93% ($p < .01$) and 26% ($p < .03$) with addition of NMA. In contrast, cAMP level in the hypothalamus was decreased 21.89% ($p < .01$). The data from subcutaneous fat (10th rib) analysis showed that supplement of 50 mg/kg NMA decreased the total activities of malic dehydrogenase (MDH) by 20.54% ($p < .04$), glucose-6-phosphate dehydrogenase (G-6-DPH) by 16.97% ($p < .04$), and decreased the specific activities of MDH and G-6-DPH by 37.46% ($p < .001$) and 35.06% ($p < .01$), respectively. The hormone sensitive lipase (HSL) total activity was increased by 25.00% ($p < .02$) in NMA treated pigs. These 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 Studies on lipid metabolism in hepatocytes from growing pigs. T.J. Caperna^{*1}, 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 interactions 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 dexamethasone (10^{-6} M) 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 20 to 500ng/ml. On day four, medium containing 0.23mM 1-¹³C-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% ($P < 0.02$, $n=4$); chronic rh-leptin exposure (200ng/ml, 2.5d) had no apparent effect on palmitate OX under these conditions. For determination of ketogenesis, cells were maintained in DMEM/M199 containing both I and G between 1 and 100ng/ml with or without rh-leptin. Medium was collected on the fourth day, following a 24h incubation period and β -hydroxybutyrate (BHB) and acetoacetate (ACAC) were determined fluorometrically using a BHB-dehydrogenase-NADH coupled system. Ketone production was stimulated by G and inhibited by I; depending on the G:I ratio (0.01:1 to 100:1), total ketone production ranged between 100 and 400nmol/mg protein/24h. Addition of rh-leptin had no consistent effect on ketone production or BHB:ACAC ratio (0.98 control, 0.97 rh-leptin). These data suggest that while pig hepatocytes are indeed

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. Poulos, *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, or stained for lipid. 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 + dexamethasone (DEX) for 5 days after confluence did not increase preadipocyte number ($P > .05$), 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 ($75 \pm 12\%$ and $75 \pm 9\%$; respectively) but was reduced ($P < .05$) to $46 \pm 6\%$ 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 LXR α ligands on adipocyte differentiation. T.D. Brandebourg* and C.Y. Hu, *Oregon State University, Corvallis*.

We recently demonstrated that ligand activation of LXR α , an orphan receptor expressed in adipose tissue, negatively regulated adipocyte differentiation. Administration of an LXR α agonist inhibited differentiation of 3T3-L1 cells while geranylgeraniol (GG), a metabolite of mevalonic acid that antagonizes LXR α , blunted the inhibitory effect of the LXR α agonist. The objective of this study was to extend those results by evaluating the effect of several LXR α 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 LXR α ligands or cholesterol (Chol), which neither activates nor binds LXR α , 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 ($p < .001$), 53% at 5 mM ($p < .01$), 61% at 10 mM ($p < .001$) and 66% at 20 mM ($p < .001$). Administration of 22R decreased GPDH activity by 9% at 2.5 mM ($p < .05$), 17% at 5 mM ($p < .001$), 35% at 10 mM ($p < .001$) and 41% at 20 mM. Administration of 20S decreased GPDH activity by 12% at 2.5 mM ($p < .001$), 20% at 5 mM ($p < .01$), 27% at 10 mM ($p < .01$) and 57% at 20 mM ($p < .001$). Conversely, administration of 22S, which is known to bind LXR α 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 LXR α 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 LXR α antagonist that may prove useful in subsequent studies of the role of LXR α 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 confluency in 10% fetal bovine serum in DMEM/F12 (50:50) medium. At confluency, 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 μ M 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 μ M) 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. Helman*¹, E. H. Lonergan¹, S. M. Lonergan¹, and G. M. Davenport², ¹Iowa State University, Ames, IA, ²The 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 analyses. Western blots were completed using an anti-calpastatin antibody (MA3945, Affinity Bioreagents). p94 blots were examined with an anti-p94 antibody (NCL-CALP-12A2, Novocastrol Labs). 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

1786 Growth of myoblasts derived from genetically different mice, pigs, and cattle. C. Rehfeldt^{*1}, G. Nürnberg¹, U.K. Zettl², E. Mix², M. Wittstock², U. Renne¹, H.J. Papstein¹, and K. Ender¹, ¹Research Institute for the Biology of Farm Animals, Dummerstorf, 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 three lines of mice selected for more than 70 generations for 6-weeks body weight (DU-6), protein content (DU-6P), or an index from body weight and physical endurance fitness (DU-6+LB), and from a control line (DU-Ks); from wild-type (WP) or German Landrace domestic pigs (DP); and from Holstein Friesian (HF) or White Blue Belgium (WBB) cattle. During ten days of cultivation in DMEM with 8% FBS the cells of each selected mouse line accumulated more DNA and protein as compared to the control line ($P < 0.01$) with DU-6+LB > DU-6 > DU-6P ($P < 0.05$). Porcine myoblasts from *Semiteindinosus* muscle of neonatal WP or DP were grown for six days in MEM α with 10% FBS. The cells from the highly selected DP accumulated less DNA than WP cells (1.06 vs 1.71 g/well, $P < 0.0001$). There were only small differences in the growth kinetics between HF and WBB myoblasts from *Biceps brachii* muscle of three month old fetuses grown for six days in DMEM with 10% FBS. The WBB myoblasts synthesized more protein than HF myoblasts from d 4 to 6 of cultivation (17 to 30%, $P < 0.0001$) whereas DNA levels did not clearly differ. Serum deprivation by changing to 1% FBS during exponential growth as examined in the murine cell lines caused significant decreases in DNA and protein levels ($P < 0.01$) with 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 percentages of apoptotic cells were found in cultures of DU-Ks ($P < 0.01$) and DU-6 ($P < 0.001$) 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

1787 Solubilization and purification of a recombinant chicken myostatin expressed as inclusion bodies in *E. coli*. Y. S. Kim^{*1}, K. S. Baek², 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 myostatin 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 conditions, but not at pH 8 and 9. A significant proportion of recombinant myostatin stayed as heterogeneous multimer forms in the solubilized inclusion bodies, and increasing pH favored monomer formation. Thus, 30 min boiling or 25°C overnight incubation at pH 9 provided the optimum condition for inclusion body solubilization, and the proportion of monomer was approximately 50 % in these conditions. Solubilized inclusion 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 heterogeneous 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

1788 Stair-step compensatory growth regimen in dairy heifers and its effects on transition health. M.S. Laubach^{*1}, D.E. Schimek¹, D.B. Carlson¹, A.M. Encinas¹, J.L. Burton², J.W. Schroeder¹, W.L. Keller¹, and C.S. Park¹, ¹North Dakota State University, ²Michigan State University.

The objective of this study was to determine if a one stair-step gestational nutrition regime affects metabolic status and lactation potential of dairy heifers during the transition period. Nineteen Holstein heifers averaging 511 kg of body weight were randomly divided into two treatments. 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-implantation 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 hours of calving), 0.5, 1, 2, 3, 5, 7, 10, 13 to monitor various metabolites, white blood cell counts, and lymphocyte populations (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 (before $P = 0.06$, after $P = 0.01$). There were no differences 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. Moallem^{*1}, G. E. Dahl¹, E. K. Duffey-Tower¹, A. V. Capuco², 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 undegraded 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 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. Weekly blood progesterone 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, WH and HH were increased by both RUP and bST alone while growth responses to the combination of RUP and bST (RUPbST) 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. Skeletal 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 protein limited growth during the early post-weaning period, whereas circulating bST was more important during the time just prior to puberty.

Treatments						Effects <i>P</i> <		
Item	Control	bST	RUP	RUP bST	SEM	bST	RUP	RUP* bST
Body weight								
315d, kg	290.4	302.3	306.6	333.2	5.8	0.153	0.011	0.931
Body weight								
gain, kg/d	0.935	0.974	0.983	1.091	0.04	0.013	0.006	0.228
Wither height								
315d, cm	100.6	101.7	102.8	103.5	0.85	0.317	0.024	0.846
Wither height								
gain, cm/d	0.134	0.137	0.144	0.150	0.004	0.259	0.005	0.745
Hip height								
315d, cm	106.4	106.9	108.2	109.3	0.89	0.360	0.021	0.757
Hip height								
gain, cm/d	0.139	0.146	0.151	0.159	0.003	0.018	0.001	0.853

Key Words: bST, RUP, Skeletal growth

1790 Effects of added rumen undegraded protein and bovine somatotropin administration on organ and tissue weights in prepubertal dairy heifers. U. Moallem¹, G. E. Dahl^{*1}, A. V. Capuco², R. L. Baldwin², and R. A. Erdman¹, ¹University of Maryland, College Park, ²USDA-ARS, Beltsville, MD.

Twenty-four dairy heifers were used to determine the effects of added rumen undegraded 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 0.1mg/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 empty body weight (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 which was observed in the RUP group. At 10 month of age, bST increased the average weights of heart (28%, *P* < 0.01), kidney (22.3%, *P* < 0.01) and liver (17%, NS), and as the relative part of EBW by 15% (*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

1791 Effects of added rumen undegraded protein and bovine somatotropin administration on body composition in prepubertal dairy heifers. U. Moallem¹, K. R. McLeod², A. V. Capuco², K. E. Duffey-Tower¹, G. E. Dahl¹, and R. A. Erdman^{*1}, ¹University of Maryland, College Park, ²USDA-ARS, Beltsville, MD.

The objective of this study was to test effects of added rumen undegraded 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 mcal/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* < .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.

Mean (5 and 10 mo)	Treatments					Treatments effects (P<)		
	Control	bST	RUP	RUP bST	SEM	bST	RUP	RUP*
Ash, %	4.88	5.29	4.78	5.42	0.25	0.06	0.96	0.65
Crude protein, %	18.83	19.83	18.61	19.97	0.72	0.13	0.95	0.80
Fat, %	12.11	11.86	12.03	9.98	0.54	0.05	0.09	0.12
Energy, mcal/kg	2.11	2.17	2.11	1.97	0.05	0.62	0.12	0.1
Empty body (10 mo of age)								
Weight, kg	217.4	241.0	228.7	238.2	7.8	0.07	0.60	0.40
Ash, kg	10.7	12.4	10.5	13.3	0.95	0.04	0.69	0.55
Crude protein, kg	40.9	47.6	40.4	48.0	3.1	0.06	0.98	0.88
Fat, kg	31.5	34.6	33.5	28.7	3.1	0.47	0.75	0.16
Energy, mcal	509.0	576.3	528.6	522.6	34	0.34	0.58	0.26

Key Words: bST, RUP, Body composition

1792 Relationships between concentration of serum immunoglobulins and growth rate of dairy heifers. W. Jarmuz¹, I. Szelag², and R. Skrzypek^{*2}, ¹IGiHZ PAN Jastrzebiec, ²Agricultural University of Poznan, Poland.

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 immunoglobulin 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 age of 90 days 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-colostral period of life.

Key Words: Heifers, Serum immunoglobulins, Growth rate

1793 Growth Performance, Metabolic and Endocrine Traits in Calves Pair-fed by Automate or by Bucket During the First Month of Life. H.M. Hammon^{*}, A. Nussbaum, G. Schiessler, and J.W. Blum, University of Berne, Switzerland.

Amounts of colostrum (C) and milk intake greatly influence postnatal development of calves. However, as indicated by suckling calves in

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 thyroxine 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 GrB. Preprandial plasma TG (on d 7) were higher and mean postprandial concentrations (d 7) tended to be higher in GrA than GrB. Insulin concentrations (d 7) tended to be higher in GrA than 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 GrB. Plasma IGF-I was higher in GrA than 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

1794 Age-related changes of the somatotrophic axis in cloned Holstein calves. K.E. Govoni*, X.C. Tian, G.W. Kazmer, M. Taneja, B. Enright, A.L. Rivard, X. Yang, and S.A. Zinn, *University of Connecticut, Storrs, CT.*

Developmental anomalies, as well as high rates of fetal and post-natal mortality have been reported in somatic-cell cloned cattle. Our objective was to determine if the development of the somatotrophic 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 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 somatotrophic 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 somatotrophic 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-I

1795 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 divided into several small portions per d. In this study growth performance and metabolic and endocrine traits were compared in calves fed by a computer-programmed automate either restricted amounts of C (GrR; n = 7) and milk or C and milk ad libitum (GrL; n = 7). Feed intake and meal frequencies were measured by the automate. Blood samples were taken after birth on d 1, 2, 14, 21, and 28 before feed intake and on d 3 and 7 before and 1, 2, 4, 6 and 8 h after feed intake. Plasma concentrations of total protein (TP), albumin, glucose, non-esterified fatty acids (NEFA), triglycerides and cholesterol were measured photometrically. Plasma concentrations of insulin, glucagon and insulin-like growth factor (IGF-I) 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. There were group differences of dry matter intakes and meal sizes per visit (GrL > GrR) and total visits (with and without milk intake) at the automate (GrR > GrL). Feed intake in GrL increased from 1 to 4 d, then remained stable and was always higher than in GrR. Weight gain was greater in GrL than GrR in wk 1. There were significant group differences of plasma concentrations of TP (GrL > GrR; d 28), albumin (GrR > GrL; d 1 - 2), cholesterol (GrR > GrL; d 28), NEFA (GrR > GrL; d 1 - 2), insulin (GrL > GrR; d 1 - 2), and IGF-I (GrR > GrL; d 1 and 28). In conclusion, calves fed by automate ad libitum were capable of ingesting 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

1796 Glucose metabolism in Holstein and Jersey calves fed milk replacer once versus twice daily. C. M. Cheatham*¹, C. C. Williams¹, J. M. Fernandez¹, W. A. Nipper¹, H. G. Bateman, II¹, J. C. Lovejoy², D. T. Gantt¹, L. R. Gentry¹, and G.E. Goodier¹, ¹*Louisiana State University Agricultural Center, Baton Rouge, LA,* ²*Pennington 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 calf starter and water throughout the study. At weeks 3 and 6, Minimal Model intravenous glucose tolerance tests were performed to assess glucose effectiveness (S_G), an estimate of insulin-independent glucose disappearance; insulin sensitivity (S_I), an estimate of insulin-dependent glucose disappearance; and acute insulin response ($AIR_{glucose}$), insulin secretion relative to glucose administration. Of the three parameters measured, Jersey calves differed from Holsteins only by having greater S_I ($P < 0.05$). The S_I and S_G were similar for calves regardless of MR feeding frequency ($P > 0.10$). However, S_I decreased with age for all calves ($P < 0.05$) while $AIR_{glucose}$ increased with age ($P < 0.05$). In calves fed MR 2x, $AIR_{glucose}$ 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

1797 Evaluation of bovine or porcine plasma in calf milk replacers on mortality, morbidity, intake and growth of young dairy calves. J. D. Quigley, C. J. Kost, and T. M. Anspach*, *APC Company, Inc., Ames, IA.*

Replacement of a portion of the whey protein concentrate with spray-dried animal plasma in calf milk replacer (CMR) formulations can provide a source of immunologically active IgG without markedly affecting digestibility or intake. However, data indicating the importance of the species origin of animal plasma are lacking. In this study, Holstein bull calves (n = 120) at the APC Company Research Facility in Ames, IA were fed one of three CMR for 42 d. Experimental CMR were formulated to contain whey protein concentrate (WPC) as the primary protein source or WPC plus 5% spray-dried bovine plasma (SDBP) or WPC plus 5% heat stable porcine plasma (SDPP). Calves were also offered commercial calf starter and water for ad libitum consumption. Mortality was reduced from 25% to 7.5% and 5.0% when calves were fed CMR containing WPC, SDBP, or SDPP, respectively. Morbidity, measured as number of days that calves had diarrhea (scours) was reduced by 30% (P < 0.01) when SDBP or SDPP were fed. Mean number of days with scours were 6.1, 3.9 and 4.7 d for calves fed WPC, SDBP and SDPP, respectively. Fecal scores during the 42-d study tended to be reduced (P < 0.10) and feed efficiency tended (P < 0.10) to be improved when SDBP or SDPP was fed. Mean fecal scores (1 = normal feces to 4 = severe scours) were 1.66, 1.59 and 1.61 for calves fed CMR containing WPC, SDBP and SDPP, respectively. Mean feed efficiency was 170, 265 and 211 g of BW gain/kg DM intake, respectively. Calves fed SDPP tended (P < 0.10) to consume less starter, total DM and protein compared to calves fed SDBP and tended (P < 0.10) to have lower BW gain during the first 28-d of the study. Mean BW gain from d 0 to d 28 was 83, 119 and 80 g/d for calves fed WPC, SDBP and SDPP, respectively. There were no differences in BW gain after 28 d, and calves ended the study weighing 56.4, 58.2, and 56.8 kg, respectively. Both SDBP and SDPP were effective in reducing morbidity and mortality during the trial and may be effective adjuncts to a calf management program.

Key Words: calves, milk replacer, immunity

1798 Body composition of Piedmontese x Hereford and Wagyu x Hereford newborn calves. P.L. Greenwood^{1,2}, H. Hearnshaw^{1,3}, D.W. Hennessy^{1,3}, J.M. Thompson^{1,4}, and G.S. Harper^{1,5}, ¹*Cooperative Research Centre for Cattle and Beef Quality, Armidale, Australia*, ²*NSW Agriculture Beef Industry Centre, Armidale, Australia*, ³*NSW Agriculture Research and Advisory Station, Grafton, Australia*, ⁴*University of New England, Armidale, Australia*, ⁵*CSIRO Livestock Industries, Brisbane, Australia.*

Newborn calves of extreme genotypes for marbling and muscle growth potential were studied to provide base-line data for studies aimed at elucidating the cellular basis and regulation of growth and development of carcass tissues. Female calves born to Hereford cows and sired by Piedmontese (PxH calves, LW range 31.4 to 42.0 kg, n = 10) or Wagyu (WxH calves, 27.6 to 35.4 kg, n = 8) bulls were slaughtered within 24 h of birth. Organs and selected muscles and body tissues were dissected out and weighed. Empty body weights were determined, and carcasses prepared to a standard before being Computer Tomography (CT)-Scanned. Carcass composition was determined by integration of cross-sectional tissue areas from serial CT body scans. As a proportion of empty body weight, PxH calves had (P<.05) higher dressing percentage than WxH calves (mean \pm SEM, 57.6 \pm .6% vs 54.9 \pm .5%, respectively), more carcass lean (75.1 \pm .4% vs 72.9 \pm .6%), less carcass fat (4.8 \pm .1% vs 5.2 \pm .1%) and bone (20.1 \pm .4% vs 21.9 \pm .5%), larger *M. semitendinosus* (.45 \pm .01% vs .38 \pm .01%) and *M. longissimus et lumborum* (.46 \pm .02% vs .39 \pm .02%), and lower combined weight of organs (9.0 \pm .1% vs 10.2 \pm .2%). PxH calves also tended to have less skin (11.5 \pm .3% vs 12.3 \pm .3%, P<.10) than their WxH counterparts, but there was no difference in the proportion of abdominal plus kidney fat (.55 \pm .04% vs .61 \pm .04%, P>.10). When adjusted for differences in empty body weight, significant effects (P<.05) remained for weight of fat in the carcass, weight of *M. semitendinosus* and combined weight of organs. The results demonstrate that differences in the distribution of body tissues between Wagyu x Hereford and Piedmontese x Hereford cattle are apparent at birth.

Key Words: Cattle, newborn, body composition

1799 Post-weaning growth of cattle destined for Japanese and Korean markets: Relationships between growth during backgrounding and intramuscular fat percentage (IMF%) at slaughter. M.J. McPhee^{1,2}, S. Harden^{1,3}, P.L. Greenwood^{1,2}, and V.H. Oddy^{1,4}, ¹*Cooperative Research Centre for Cattle and Beef Quality, Armidale, Australia*, ²*NSW Agriculture, Beef Industry Centre, University of New England, Armidale, NSW 2351, Australia*, ³*NSW Agriculture, Tamworth Centre for Crop Improvement, Tamworth, NSW 2340, Australia*, ⁴*Meat and Livestock Australia, 165 Walker Street, North Sydney, NSW 2060, 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 linear 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<.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 \pm 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 μ g/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 x 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 x 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. L. A. L. Chardulo^{*1}, J. A. Ferro², A. C. Silveira¹, L. R. Furlan¹, M. D. B. Arrigoni¹, H. N. Oliveira¹, M. I. T. Ferro², and M. Macari², 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 males of five breeds (Charolais, Gelbvieh, Aberdeen Angus, Hereford and Simmental) and Simmental × Nelore 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 ($p < .01$). Males had better performance than females for all characteristics evaluated ($p < .01$). 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 ($p < .01$). Aberdeen Angus animals had higher triiodothyronine (T_3) levels and higher IGF-I levels during finishing phase as compared to the other breeds ($p < .01$). Correlations between T_3 and subcutaneous fat ($r = 0.76$, $p < .01$) and marbling ($r = 0.79$, $p < .01$) were significant as well as between IGF-I and subcutaneous fat ($r = 0.67$, $p < .01$) and marbling ($r = 0.59$, $p < .01$). 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 Beermann^{1,2}, 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 × 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 ($P = 0.07$), and circulating IGF-1 concentration was not altered. Estradiol administration increased plasma estradiol 18-fold (2.4 to 48.5 pg/ml) but did not alter plasma PUN or IGF-1 gene expression in skeletal muscle (all $P > 0.1$). An interaction between protein intake and estradiol treatment was observed for IGF-1 mRNA abundance in skeletal muscle ($P = 0.11$). When the higher protein diet was fed estradiol treatment increased IGF-1 mRNA abundance in skeletal muscle 42% compared to excipient treatment. Results from this study indicate that 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 Beermann^{1,2}, 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 estradiol administration (175 micrograms at 12-h intervals) 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 euthanized 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 concentration (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/d), 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

1804 Effects of immunization against LHRH on growth performance, sex characteristics, and meat quality of intact male pigs. C. Y. Liu^{*1}, L. C. Cheng¹, P. C. Yang¹, T. Y. Chang^{2,3}, M. Shen³, C. L. Finstad³, and C. Y. Wang^{2,3}, ¹Pig Research Institute Taiwan, ROC, ²United Biochemical, Inc., Asia, ROC, ³United Biochemical, Inc., USA.

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) μ g 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 μ g level ($P < .05$). Tenderness and juiciness of longissimus muscle chops did not differ among groups by sensory panel evaluations, but chops from immunized animals had less off-flavor ($P < .01$) than intact controls. Genital tract weights (testes, epididymes, seminal vesicles, and prostate glands), measured at slaughter, were significantly ($P < .001$) decreased by immunization, and both doses of anti-LHRH vaccine had similar efficacy. Consistently, plasma testosterone concentrations were completely inhibited ($P < .001$) 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

1805 The effects of zinc and thyroid hormone on the expression of growth hormone and thyroid stimulating hormone in primary rat anterior pituitary cells. A.L. Rivard^{*}, M.A. Shaller, H.C. Freake, and S.A. Zinn, University of Connecticut.

Chelation of zinc with DTPA (diethyltriaminepentaacetic acid) stimulates expression of growth hormone (GH) in tri-iodothyronine (T_3)-treated rat pituitary tumor (GH3) cells. To determine whether zinc chelation stimulates GH expression in rat anterior pituitary cells, 50 to

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 levels of expression for GH and TSH, respectively. To evaluate RT-PCR, preliminary experiments were performed with GH3 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 control cells. Expression of TSH mRNA was greater in DTPA treated cells compared with controls ($p < .01$), 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% ($p = .03$). However, in contrast to GH3 cells, DTPA did not stimulate GH mRNA levels in T₃-treated pituitary cells ($p = .18$). In conclusion, while zinc chelation increases TSH mRNA, in the presence 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

1806 Effects of dietary conjugated linoleic acid (CLA) on the composition and function of peripheral blood mononuclear leukocyte populations in heifer calves. J.M. Smith^{*1}, B.J. Nonnecke², M.E. Van Amburgh¹, B.A. Pesch², and J.A. Harp², ¹Cornell University, Ithaca, NY, ²National 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 CD3+ T cells (and CD4+, CD8+, and $\gamma\delta$ 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 chicks and rodents 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 \pm 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 \pm 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

AMSA/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^{*1}, A. M. Scholz², and V. G. Pursel¹, ¹USDA, Agricultural Research Service, Beltsville, MD, ²Ludwig Maximilians University-Munich, Oberschleissheim, 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 \pm 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 \pm 7.0%; shoulder region, 23.8 \pm 6.7%; ham region 22.9 \pm 6.7%; loin region, 23.7 \pm 7.6%; and the side region 27.9 \pm 7.6%. The correlation (R^2) 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 R^2 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