M65 Effect of polyethylene glycol on *in vitro* gas production and substrate degradation of diets selected by grazing goats. M. A. Cerrillo-Soto\*, M. Guerrero-Cervantes, G. Nevárez-Carrasco, R. Montoya-Escalante, E. Herrera-Torres, M. Murillo-Ortíz, and A. S. Juárez-Reyes, *Universidad Juárez del Estado de Durango, Durango, Dgo. Mexico.* 

The study was conducted to evaluate the effect of polyethylene gylcol (PEG-6000) on in vitro gas production and substrate degradation of diets selected by goats grazing a shrub and oakland range in the semiarid region of Durango, Mexico. Six Spanish criollo goats (35 to 40 kg BW) were used to obtain diet samples utilizing the handplucking method during Spring (Apr-Jun), Summer (Jul-Sep), Autumn (Oct-Dec) and Winter (Jan-Mar). A total of three days each month from 0900 to 1200 were used to collect samples. Operators followed and observed the animals and manually mimicked animal forage preferences. Collections from six goats were composited to obtain a representative monthly sample. Five hundred mg (DM) samples were incubated in calibrated glass syringes in triplicate with or without PEG-6000 (1 g). The in vitro gas production was recorded at 0, 3, 6, 9, 12 and 24h after inoculation. Moreover, after 24 h of incubation the content of the syringes was completely transferred into a pre-weighed nylon bag (5 cm x 10 cm: pore size 40-60 µm) and thoroughly washed for estimation of the in vitro substrate degradation. Data were analyzed using ANOVA for a completely randomized block design. The addition of PEG-6000 increased the *in vitro* gas production (P < 0.05) at 24h of incubation by 13, 13, 16 and 10% during Winter, Spring, Summer and Autumn, respectively. The in vitro degradation of the substrate was also affected (P < 0.05) by the addition of PEG-6000. Increments of 46, 21, 22 and 36% were recorded during Winter, Spring, Summer and Autumn, respectively. Increases in gas production and substrate degradation by addition of PEG-6000 evidence the in vitro detrimental effects of phenolic compounds contained in the forage selected by grazing goats.

 Table 1. Effect of PEG-6000 on *in vitro* gas production and substrate degradation in goat diets

	Seasons					
	Winter	Spring	Summer	Autumn		
Degraded substrate (% DM)						
With PEG	43.0 <sup>a</sup>	40.0 <sup>a</sup>	37.7 <sup>a</sup>	42.8 <sup>a</sup>		
Without PEG	29.4 <sup>b</sup>	33.0 <sup>b</sup>	30.9 <sup>b</sup>	31.4 <sup>b</sup>		
Mean	26.2	36.4	34.3	37.1		
sem	4.79	5.15	5.08	4.50		
Cumulative gas production at 24h (ml/500 mg DM)						
With PEG	71.4 <sup>a</sup>	81.4 <sup>a</sup>	71.0 <sup>a</sup>	78.5 <sup>a</sup>		
Without PEG	62.9 <sup>b</sup>	72.2 <sup>b</sup>	61.2 <sup>b</sup>	71.2 <sup>b</sup>		
Mean	67.1	76.8	66.1	74.8		
sem	3.52	7.50	7.60	6.50		

Columns with different superscript differ (P<0.05)

Key Words: Grazing goats, *In vitro* gas production, Polyethylene glycol

**M66 Evaluation of cultivated summer pastures for meat goats in Tennessee.** M. Lema\*, K. Souleymane, R. Opio, and C. Fenderson, *Tennessee State University, Nashville.* 

A grazing trial was conducted to evaluate Puna forage chicory Cichorium intybus L., Hybrid Penleaf pearl millet Pennisetum glaucum and Sahara bermudagrass Cynodon dactylon as summer pasture for meat goats. Thirty weaned F1 does (18 Spanish x Kiko, 6 Spanish x Boer and 6 Boer x Kiko) were blocked by body weight and genotype and randomly assigned to the three pasture types. Each pasture type was stocked with 10 does (5 does per replicate) at a stocking rate of 5 does per acre. Puna chicory was 28.3 and 67.7 % higher (P < 0.05) in crude protein (CP). 28.1 and 35.4 % lower (P < 0.05) in acid-detergent fiber (ADF) and 40.0 and 46.0 % lower (P < 0.05) in neutral detergent fiber (NDF) than Penleaf pearl millet and Sahara bermudagrass, respectively. Penleaf pearl millet was 37.7 % higher (P < 0.05) in CP, 10.2 and 10.0 % lower (P < 0.05) in ADF and NDF than Sahara bermudagrass, respectively. Relative Feed Value (RFV), Ca, P, Mg and K contents were significantly higher (P < 0.01) for Puna chicory than for Peanleaf pearl millet and Sahara bermudagrass. Pennleaf pearl millet was higher (P < 0.01) than Sahara bermudagrass in P, K and Mg content. Puna chicory and Pearl millet produced 73 and 70 % higher (P < 0.05) forage CP per ha, respectively than Sahara bermudagrass. Average daily gain and live weight gain per acre of does grazing Puna chicory were significantly higher (P < 0.05) than does grazing Sahara bermudagrass and Penleaf pearl millet.

Key Words: Meat goat, Penleaf pearl millet, Puna chicory

## **Growth and Development**

M67 Differences in adipogenesis between bovine intramuscular and subcutaneous preadipocytes are not related to expression of PPAR $\gamma_2$  or secretion of PGI<sub>2</sub>. G. Ortiz-Colón\*, A. C. Grant, M. E. Doumit, and D. D. Buskirk, *Michigan State University, East Lansing.* 

The objectives of this study were to determine if intramuscular (IM) and subcutaneous (SC) bovine preadipocytes differ in their expression of peroxisome proliferator-activated receptor  $\gamma_2$  (PPAR) or in their

secretion of prostacyclin (PGI<sub>2</sub>). Preadipocytes isolated from IM and SC adipose tissue of three steers were propagated in culture and upon confluence were exposed to 0 or 25 nM dexamethasone (DEX) for 48 h. After exposure to differentiation media for 12 d, cell lysates were subjected to PPAR immunoblot analysis, which detected an immunoreactive band of  $\approx$ 53 kDa. The relative expression of PPAR was equivalent between IM and SC cells (P = 0.39), and DEX did not affect PPAR abundance (P = 0.98). Heterogeneous preadipocytes

isolated from one steer and clonal preadipocytes derived from a second steer were grown to confluence and exposed to 0 or 25 nM DEX for 48 h. Media were collected every 12 h for 48 h and were assayed for the stable PGI<sub>2</sub> derivatives 6-keto-prostaglandin  $F_{1\alpha}$  and 2,3-dinor-6-keto-prostaglandin F1a. After 12 d in differentiation media, glycerol-3-phosphate dehydrogenase (GPDH) analysis was performed. Intramuscular cells secreted more  $PGI_2$  derivatives than SC cells (P =0.046) and DEX decreased secretion of PGI2 equally in cells from both depots (P < 0.001). The concentration of PGI<sub>2</sub> increased with time (P< 0.001), until 36 h. Although 25 nM DEX increased GPDH activity in both cell populations (P < 0.001), IM cells were less adipogenic than SC cells (P < 0.001). In clonal SC cells, 48 h exposure to 10  $\mu$ M ibuprofen (an inhibitor of prostaglandin synthesis) had no affect (P =0.99) on DEX (25 nM) induction of GPDH activity. Supplementation of differentiation media for 48 h with 123 pg/mL cPGI<sub>2</sub> did not affect adipogenesis of clonal SC cells, either alone, or in combination with DEX, or DEX and ibuprofen (P = 0.99). We conclude that adipogenic differences between IM and SC bovine preadipocytes are not explained by differences in PPAR expression or PGI<sub>2</sub> secretion.

Key Words: Bovine, Preadipocyte, Peroxisome proliferator-activated receptor gamma two

M68 Effect of retinoic acid on sheep preadipocyte gene expression during terminal differentiation. P. Martinez, A. Arana, I. Encio, L. Alfonso, and B. Soret\*, *Universidad Publica de Navarra, Pamplona, Navarra, Spain.* 

Adipogenesis program and its regulation have been extensively studied, mainly using cell lines and primary culture from human, rodents and pigs. Retinoic acid (RA) has been shown as inhibitor of preadipocyte differentiation using those models but its effects on ruminant adipose cells are not well described. We have used sheep primary preadipocyte cells from two anatomical depots (omental and subcutaneous) as a ruminant model to study at a molecular level the effect of RA on preadipocyte differentiation. In brief, stromal-vascular cells were obtained by collagenase digestion and after a period of proliferation were allowed to differentiate by adding serum-free differentiation induction media containing 1.6 µg/ml insulin, 2nM tri-iodothyronine, 10 nM dexamethasone and 10 µM rosiglitazone. To analyze the effect of RA treatment, preadipocytes were treated with 10 µM all-trans retinoic acid through the differentiation period (10 days) and mRNA expression levels of C/EBPB, PPARy, C/EBPa, ADD1, lipoprotein lipase (LPL) and Acetyl CoA carboxylase (ACC) were estimated by quantitative real time PCR. Relative gene expression was performed by normalizing samples (three per treatment) against GAPDH or cyclophilin housekeeping genes, following the Ct method, and RA effect tested computing 95% confidence intervals. Number of differentiated cells was assessed by flow cytometry.

Addition of RA decreased differentiated cells number by 50% in the subcutaneous depot (p<0.001). The expression of adipocyte genes PPAR $\gamma$  and C/EBP $\alpha$  also decreased (p<0.05, day 10) but there was not effect on C/EBP $\beta$  and ADD1 level in subcutaneous cells. This effect was similar but less apparent in omental cells, which showed less differentiated cell numbers than subcutaneous cells (p<0.05) and a low level of gene expression. These results suggest that the inhibitory action of RA on sheep subcutaneous preadipocyte differentiation is mediated by PPAR $\gamma$  and C/EBP $\alpha$  expression and that sheep omental preadipocytes respond less to *in vitro* differentiation.

Key Words: Sheep preadipocytes, Retinoic acid, Transcription factors

**M69** Localization of IGFBP-3 and IGFBP-5 in cultured porcine embryonic myogenic cells. X Gang, E. I. Kamanga-Sollo, M. R. Hathaway, M. E. White\*, M. S. Pampusch, and W. R. Dayton, *University of Minnesota, St. Paul.* 

The proliferation-suppressing actions of myostatin and TGF-beta in porcine embryonic myogenic cell (PEMC) cultures are mediated, at least in part, by insulin-like growth factor binding protein (IGFBP)-3 and IGFBP-5. Consequently, understanding the mechanism of action of these IGFBPs in myogenic cells is important to understanding how TGF-beta and myostatin regulate growth of muscle. We have used anti-rpIGFBP-3 (anti-BP3), anti-rpIGFBP-5 (anti-BP5) and anti-desmin antibodies to localize IGFBP-3, IGFBP-5 and desmin, respectively, in PEMC. IGFBP-3 was detected in the cytoplasm and nuclei of desmin-positive, mononucleated cells in proliferating PEMC cultures; thereby, establishing that IGFBP-3 is present in PEMC (controls using non-specific IgG show no staining). Similarly, IGFBP-3 was detected in cultured PEMC myotubes. In proliferating PEMC cultures, treatment for 24 h with 20 ng TGF-beta/ml medium resulted in an 80% increase (p<0.01) in the number of nuclei containing IGFBP-3. Myogenic cells pre-treated with anti-BP3 for 24 h prior to immunohistochemical localization showed dramatically reduced intracellular levels of IGFBP-3 as compared to control cells that received no pre-treatment. This confirms reports in other cell types that a significant portion, if not all, of the intracellular IGFBP-3 represents uptake of secreted IGFBP-3. Additionally, these results establish that anti-BP-3 interferes with the transport of IGFBP-3 into myogenic cells. IGFBP-5 was detected in the cytoplasm and nuclei in proliferating and fused PEMC cultures (controls with non-specific IgY show no staining). Localization of IGFBP-3 and IGFBP-5 in PEMC at different stages of differentiation or after treatment with specific growth factors should lead to a greater understanding of the roles of these IGFBPs in muscle growth and differentiation.

Key Words: IGFBP-3, IGFBP-5, Muscle

**M70** Use of RNA interference (RNAi) to silence IGFBP-3 and IGFBP-5 expression in porcine embryonic myogenic cell cultures. X. Gang, M. R. Hathaway, M. E. White, E. I. Kamanga-Sollo, M. S. Pampusch, and W. R. Dayton\*, *University of Minnesota, St. Paul.* 

Insulin-like growth factor binding proteins (IGFBP)-3 and -5 play a significant role in the mechanism by which TGF-beta and myostatin suppress proliferation of porcine embryonic myogenic cells (PEMC) and porcine muscle satellite cells (PMSC). RNA interference (RNAi) utilizing small inhibitory RNA (siRNA) is currently extensively used to silence specific genes in mammalian cells. Consequently, we have cloned a small hairpin (sh) IGFBP-3 RNA sequence (complementary 19mer siRNA sequences separated by a hairpin loop) into the pSilencer 2.1 (Ambion) siRNA expression vector. Electroporation was used to transiently transfect this construct into PEMC cells (IGFBP-3-silenced PEMC). As a control, the same electroporation procedure was used to transfect the vector containing a nonsense sequence supplied by the manufacturer into PEMC (mock-silenced cells). As compared to mock-silenced or non-transfected control cells, IGFBP-3-silenced PEMC showed a 90% reduction (p<0.01) in IGFBP-3 protein and mRNA levels. Neither IGFBP-2 nor IGFBP-5 mRNA or protein levels were significantly affected in the IGFBP-3-silenced cell population, indicating that the suppression of IGFBP-3 production was specific. Suppression of IGFBP-3 production in IGFBP-3-silenced PEMC persisted for at least 120 h after transfection, providing ample time to accomplish experiments assessing the effects of IGFBP-3 knock-down

on proliferation and cell signaling. Additionally, immunohistochemical studies show that intracellular IGFBP-3 levels were dramatically reduced in IGFBP-3-silenced cells as compared to mock-silenced cells. We also have identified an shRNA that reduces IGFBP-5 mRNA in PEMC by 95% (as compared to mock-silenced control PEMC) (p<0.01) without significantly altering the levels of IGFBP-2 or -3 mRNA or protein. Silencing IGFBP-3 and IGFBP-5 production in PEMC cells will provide a valuable research tool for use in assessing the role of these IGFBPs in mediating the anti-proliferative effects of myostatin and TGF-beta on these cells.

Key Words: IGFBP-3, IGFBP-5, Muscle

## **M71** Effects of clenbuterol and serum on the activation of mitogenactivated protein kinase in cultured bovine satellite cells. J. M. Scheffler\* and S. J. Jones, *University of Nebraska, Lincoln.*

The direct effects of β-adrenergic agonists such as clenbuterol on satellite cells is poorly understood. The mitogen-activated protein (MAP) kinase cascade has been implicated in the regulation of skeletal muscle growth. Therefore, the objective of this study was to examine the effects of clenbuterol and serum on MAP kinase activity of cultured bovine satellite cells (BSC). BSC were seeded onto 6 well plates and grown to 80% confluence and pretreated with either serum-free media or media containing 10% fetal bovine serum (FBS) for 16 h. At the end of pretreatment, cells were fed with serum-free media, media with 10% FBS or pretreatment media with the addition of  $2 \times 10^{-11}$  M clenbuterol. Serum-starved cells were solubilized at 0, 0.5, 1, 2, 4 or 8 h post treatment then frozen until analysis. Cells pretreated with 10% FBS were solubilized at 0, 1 and 4 h post treatment. Activation of MAP kinase was determined by western blot analysis using antiphosphorylated MAP kinase antibodies. Administration of 10% FBS media to serum-starved cells resulted in a 7.5-fold increase (P<0.0001) in phosphorylated MAP kinase at 0.5 h compared to cells fed serumfree media. The level of MAP kinase phosphorylation declined over the 8 h study and dropped to levels similar to that observed in cells fed serum-free media at 8 h (P>0.05). Clenbuterol had no effect (P>0.05) relative to cells fed serum-free media. Removal of serum from BSC resulted in a dramatic reduction (P<0.001) in MAP kinase phosphorylation compared to cells fed 10% FBS at 1 and 4 h. MAP kinase phosphorylation in BSC fed a combination of 10% FBS and clenbuterol were intermediate (P<0.01) to those fed 10% FBS media and cells fed serum-free media at 1 hr. At 4 hr there was no difference in MAP kinase phosphorylation in serum-fed cells with or without clenbuterol. These results indicate that factors in serum play an important role in MAP kinase activation while clenbuterol has little effect on MAP kinase phosphorylation under the conditions used in this study.

Key Words: Clenbuterol, Mitogen-activated protein kinase, Bovine satellite cells

M72 Production of a polyclonal antibody against unprocessed chicken myostatin and the effects of in-ovo administration of the antibody on post-hatch broiler growth and muscle mass. N. K. Bobbili\*, Y. K. Lee, and Y. S. Kim, *University of Hawaii*, *Honolulu*.

Myostatin, a member of the TGF- $\beta$  superfamily, is a potent negative regulator of skeletal muscle growth. The objective of this study was to produce a polyclonal antibody against unprocessed chicken myostatin and to examine the effects of in-ovo administration of the antibody

on post-hatch broiler growth and muscle mass. Unprocessed form of chicken myostatin, which had been expressed in E. coli and purified by electro-elution of myostatin bands after fractionation by SDS-PAGE, was used as an immunogen in producing a polyclonal antibody against unprocessed myostatin in rabbit. In Western blot analysis, the antibody showed a strong binding affinity to commercially available myostatin prodomain with little binding affinity to mature myostatin. The antibody also demonstrated a certain level of cross-reactivity with pTGF-ß1 and rhBMP2, but not with pTGF-ß2, rhTGF-ß3, rhBMP3 and rhBMP5 in Western blot analysis. To examine the effects of in-ovo administration of the antibody, eggs were injected once with 35 µg antibody in 50 µL PBS per egg either into the albumen (Alb) or yolk (Yolk) on day 3 of incubation. Controls (Con) received no injection. After hatch, chicks were raised for 28 d. The broilers of the Yolk group had significantly (P<0.05) lower body wt (8.5%) at 7 d than the Con group. At 14, 21 and 28 d, the mean body wt of the Yolk group was lower (5%) than that of the Con, but the difference was not statistically significant. Thigh and leg muscle wt of the Yolk group was significantly (P<0.05) lower (10%) than that of the Con at 28 d. In contrast, no significant effects on body and muscle mass were observed when the antibody was injected into the albumen. In summary, the polyclonal antibody raised against the unprocessed chicken myostatin binds to myostatin prodomain, and injection of this antibody to the yolk of eggs appeared to decrease muscle mass in chicks hatched from these eggs.

Key Words: Polyclonal anti-myostatin antibody, Myostatin, In-ovo administration

**M73** Maternal immunization against myostatin enhances posthatch broiler growth and muscle mass. Y. S. Kim<sup>1</sup>, Y. C. Huh<sup>2</sup>, and C. J. Kim\*<sup>2</sup>, <sup>1</sup>University of Hawaii, Honolulu, <sup>2</sup>Chungnam National University, Daejeon, Korea.

Myostatin, a member of the TGF- $\beta$  superfamily, is a potent negative regulator of skeletal muscle growth. The objective of this study was to examine the effect of maternal immunization against myostatin in broiler hens on post-hatch broiler growth and skeletal muscle mass. Twelve 5 month-old Cobb broiler hens were divided into four groups: CON, rMYO, MYO1 and MYO2. The CON group was immunized with 1 mg of keyhole lymphet hemocyanin (KLH), rMyo with 1 mg of recombinant active form of myostatin, MYO1 with 1 mg of 24-mer myostatin peptide-KLH conjugate, and MYO2 with 1 mg of 15-mer myostatin peptide-KLH conjugate. Hens in each group were housed together with one 5 month-old Cobb rooster, and the roosters in each group were rotated weekly. Antibody titers were detected in hens' sera, yolk IgY and post-hatch chicks' sera of the rMYO, MYO1 and MYO2 groups, indicating a transfer of antibodies into fertilized eggs and post-hatch chick's circulation. Post-hatch broilers from the rMYO and MYO2 groups showed significantly enhanced growth as compared to the CON group, resulting in 7.6% (rMYO) and 9.1% (MYO2) increase in body weight at 28 d after hatch. Similar to the body weight response, the carcass weight of the MYO2 was significantly heavier than that of the CON. The weights of breast muscle and thigh-leg of the rMYO and MYO2 groups were significantly heavier than those of the Con group. The percentages of breast muscle mass to body mass of the rMYO and MYO2 groups were significantly higher than that of the CON group, indicating that the growth-enhancing response was more selective in skeletal muscles than in other tissues of the body. In contrast, the growth of the MYO1 group was not significantly different from that of the CON group. No significant difference was also observed between the CON and MYO1 in carcass and muscle

weight. The results of this study indicate that maternal immunization against myostatin is a potential means to improve skeletal muscle growth of broilers.

Key Words: Myostatin, Maternal immunization, Broiler

M74 Effects of colostrum (C) feeding and dexamethasone (Dexa) treatment on sodium-dependent glucose co-transporter-1 (SGLT1) in the small intestine of neonatal calves. H. M. Hammon\* and U. Schoenhusen, *Research Institute for the Biology of Farm Animals (FBN)*, *Dummerstorf, Germany*.

Glucocorticoids and C feeding affect glucose metabolism in neonatal calves, but less is known concerning effects on intestinal glucose transport. We have studied the effects of Dexa and C supply on gene expression and protein content of SGLT1 in the duodenum and jejunum of neonatal calves to test the hypothesis that glucocorticoids and C feeding differently affect SGLT1 in neonatal calves. Twenty-eight male calves were randomly divided into four groups (FD<sup>-</sup>, FD<sup>+</sup>, CD<sup>-</sup>, CD<sup>+</sup>). Calves of FD - and FD+ were fed milk-based formulas (same protein and energy content than C, but only marginal amounts of bioactive substances and immunoglobulins), whereas calves of CD<sup>-</sup> and CD<sup>+</sup> received C for 3 d. On d 4 all calves received a milk replacer twice daily. Calves of FD<sup>+</sup> and CD<sup>+</sup> were injected Dexa [30 µg/kg BW per d] twice daily at feeding times. Calves were euthanized on d 5 of life after 16 h without food, mid-duodenum and mid-jejunum were removed quickly, frozen in liquid nitrogen and stored at -80°C until analyzed. Total RNA was extracted from duodenal and jejunal mucosa and mRNA abundances for SGLT1 were quantified by real-time RT-PCR relative to housekeeping genes. Protein expression of SGLT1 in brush border membrane vesicles (BBMV) was quantified by SDS PAGE and immunoblot. The General Linear Model of SAS was used to examine feeding and Dexa effects on gene and protein expression of SGLT1. Abundance of SGLT1 mRNA and protein were higher (P < 0.05) in duodenal than jejunal mucosa, but mRNA levels showed no differences with regard to feeding or Dexa treatment. Protein content of SGLT1 in jejunal BBMV showed a significant feeding × treatment interaction with highest protein content of SGLT1 in FD<sup>+</sup>. Mucosal SGLT1 gene and protein expression depend on intestinal site and Dexa effects on SGLT1 protein expression in jejunal BBMV depend on diet. Lack of feeding effects on intestinal SGLT1 expression is probably due to measuring intestinal SGLT1 in the fasting state.

Key Words: Neonatal calf, SGLT1, Colostrum

**M75 Oral nucleotides enhance immune status of neonatal dairy calves.** K. M. Ballou\*, D. E. Schimek, W. L. Keller, M. L. Bauer, and C. S. Park, *North Dakota State University, Fargo.* 

The aim of this study was to evaluate the effect of a one-time oral dose of nucleotides at birth on subsequent calf health and immune status. Eighteen colostrum-deprived, newborn Holstein calves ( $46.4 \pm 4.0$  kg initial body weight) were assigned randomly to purified milk replacer or purified milk replacer plus nucleotides (one-time dose, at birth) at twenty times the level found in cow milk (adenosine monophosphate = 1.60, cytidine monophosphate = 3.20, guanosine monophosphate = 1.33, inosine monophosphate = 1.74, and uridine monophosphate = 27.55 µmol/kg body weight). Milk replacer was fed by dry powder weight at 0.7% of the body weight and reconstituted with 1.9 L of warm water twice daily. Calves were housed in hutches at the North Dakota State University Dairy Research Unit. Signs of morbidity (ocular and nasal discharge, depression/lethargy, respiratory abnormalities, and rectal temperature) were noted daily. Scours scores [4 point scale (1

J. Anim. Sci. Vol. 84, Suppl. 1/J. Dairy Sci. Vol. 89, Suppl. 1

= normal; 4 = watery)] were assessed daily for 4 wk. Calves were weighed weekly for 8 wk. Blood was taken by jugular venipuncture at h 0 and 24, and d 7, 14, 21, and 28. Serum was analyzed for glucose, nonesterified fatty acids, immunoglobulins G (IgG) and M (IgM). Nucleotide supplementation did not affect body weight on d 28 (49.0  $\pm$  5.5 vs. 50.7  $\pm$  5.2 kg) or on d 56 (68.0  $\pm$  6.4 vs. 66.1  $\pm$  6.9 kg); morbidity; mortality; or serum nonesterified fatty acids (359.8  $\pm$  32.7  $\mu$ Eq/L), glucose (97.9  $\pm$  4.57 mg/dL), IgM on d 28 (113.9  $\pm$  19.9 mg/dL), or scours scores (1.23  $\pm$  0.08) for the first 2 wk. Nucleotide–fed calves had higher (*P* = 0.01) serum IgG on d 28 than control–fed calves (647.7  $\pm$  87.8 vs. 1006.6  $\pm$  93.8 mg/dL). Results suggest that a one–time oral dose of nucleotides at birth may enhance immune status of neonatal calves by increasing serum IgG.

Key Words: Nucleotide, Immune status, Dairy calf

M76 Effects of nutrition and weaning age on performance of ewes and lambs and incidence of subclinical mastitis in santa inês breed. S. Fenandes, E. R. Siqueira, P. F. Domingues, E. V. Z. Estasieniuk, L. S. Serrão, and R. M. S. Emediato\*, *São Paulo State University*, *Botucatu, São Paulo, Brazil.* 

Twenty nine multiparous Santa Inês ewes, all of them at the same reproductive age, were submitted to two nutrition levels over the last gestation month and lactation (corn silage diet and corn silage plus concentrate diet) and two weaning ages (45 and 70 days). The aim of this study was to evaluate the effects of these treatments on ewe and lamb performance and on the incidence of subclinical mastitis. Treatments 1 (corn silage and weaning age 45 days) and 3 (corn silage and weaning age 70 days) diets were composed of corn silage (7.43 % CP; 69.65 % TDN; 28.97 % DM; 22.45% CF; 4.37% FC and 3.73% ash) which simulated pasture condition. The animals in treatment 2 and 4 were fed corn silage plus concentrate, in a 65:35 ratio. The whole diet composition was 13.4% CP and 65% TDN, according to NRC (1985) requirements for lactation. Milk production was determined weekly, starting in the second week after parturition, according to Susin et al. (1995). Subclinical mastitis diagnostic was performed by California Mastitis Test (CMT), microbiological culture and somatic cell count. Corn silage plus concentrate diet treatments presented the best parturition weight, milk production and lamb performance from birth to weaning (P<.05). Regardless nutrional levels and weaning age, subclinical mastitis was reported in 37.93% of the ewes.

Key Words: Lamb, Sheep lactation, Sheep milk

**M77 Opioid agonist modulation of long term food intake in sheep.** F. Y. Obese<sup>1</sup>, B. K. Whitlock<sup>1</sup>, F. C. Buonomo<sup>2</sup>, and J. L. Sartin<sup>\*1</sup>, <sup>1</sup>*Auburn University, Auburn, AL*, <sup>2</sup>*Monsanto Co, St Louis, MO*.

Opioid receptors mu and kappa have been suggested as regulators of food intake and have further been suggested as downstream regulators of agouti related peptide function. Syndyphalin-33 (SD33), Tyr-Dmet (o) –Gly-methylphenethylamide, is a mu opioid receptor ligand suggested to activate central receptors after intravenous administration. Experiments were conducted to determine the effect of SD33 on food intake and to assess its ability to alter food intake in an endotoxin disease model. Five mixed-breed, castrate male sheep were housed indoors in individual pens in a temperature-and light-controlled facility. Animals had ad libitum access to water and concentrate feed, which contained 12% crude protein and was calculated to meet 100% of daily requirements. Saline (0.9%) or SD33 (0.05 or 0.1  $\mu$ mol/kg BW) were injected iv into sheep. Each treatment was administered to each sheep with at least 1-wk interval between treatments. The order of treatments

was randomized. The 0.1µmol/kg BW dose of SD 33 increased feed intake at 24 h (P = 0.006) and 48h (P = 0.019) relative to saline. Exp 2 determined whether SD33 effects on food intake were mediated by actions on opioid receptors and whether its activity can counteract the reduction in feed intake associated with administration of bacterial endotoxin (lipopolysaccharide; LPS). Saline (0.9%), SD 33 (0.1 µmol/kg BW), Naloxone the opioid receptor antagonist (NAL; 1mg/kg BW), LPS (1µg/kg BW), NAL plus SD 33 and LPS plus SD33, were injected i.v. The naloxone or LPS were injected to the sheep 5 min before SD33. Cumulative feed intake at 24 and 48h following injections were determined. The administration of naloxone reduced food intake at 24h and 48h (P<0.05) while the SD 33 combined with NAL did not, suggesting SD33 acted on food intake through the opioid receptor. LPS alone decreased food intake at 24 (P<0.05) and 48 h (P<0.05) relative to saline controls and SD33 failed to reverse the reduction in feed intake induced by LPS. These data suggest that SD33 activates food intake after iv injection and its effects are mediated via the opioid receptors in sheep. Supported by USDA Grant no. 2004-35206-14136.

Key Words: Opioid, Sheep, Appetite

M78 Effects of feeding *ad-lib* fresh milk or milk replacer during nursing and added protein at pre-puberty period to Holstein heifers on growth rates and production during first lactation. U. Moallem\*<sup>1</sup>, D. Werner<sup>2</sup>, H. Lehrer<sup>1</sup>, M. Katz<sup>1</sup>, L. Livshitz<sup>1</sup>, I. Bruckental<sup>1</sup>, and A. Shamay<sup>1</sup>, <sup>1</sup>Institute of Animal Science, ARO, Israel, <sup>2</sup>Extension Service, Ministry of Agriculture, Israel.

The objective of this study was to test the effects of feeding ad-lib fresh milk vs. milk replacer and 2% added protein at pre-puberty period on growth rates and production during first lactation. Forty-six 3 d old Israeli-Holstein calves were individually housed and randomly assigned to one of two treatments: 1) Milk replacer (MR- 12% fat and 23% protein; DM basis) - free access to milk replacer in two 60-min meals per day until weaning (60 d of age) and; 2) Milk (M) - free access to fresh milk as in treatment 1. Water and starter mix (18% protein; DM basis) were offered freely and daily individual feed intakes were recorded until 90 d of age. From 60 d of age, all heifers were fed the same diet and from 90 d of age they were housed together. During 150 to 300 d of age, M and MR calves were divided into two subgroups: each control (C) subgroup was fed a regular growing diet (13.2% protein; DM basis) and each treatment subgroup was supplemented with 2% of protein (P), creating 4 groups: MC, MP MRC and MRP. From 300 d of age all heifers were fed the same diet until calving. Weekly measurements of live body weight (LBW), hip height (HH), withers height (WH), hip width (HW) and heart girth (HG) were taken until 90 d of age, and then every two wks. Average daily total DMI was higher in MR than in M group (1.35 vs. 1.26 kg/d; P < 0.0001). At weaning, LBW of M calves was significantly higher than of MR calves (P < 0.01), while HG and HW tended to be higher (P < 0.09). Neither nursing management nor protein supplementation affected skeletal measurements at first calving; however, the LBW and HW of MP group were significantly higher than in MRP group. Milk and FCM (3.5%) production during the first lactation were significantly higher in MP group than in all other groups (P < 0.007). In conclusion, nursing ad-lib milk compared to ad-lib milk replacer tended to increase LBW but not skeletal size, and increased milk production during the first lactation. These results are in agreement with our previous findings.

Key Words: Nursing management, Skeletal growth, Milk production

**M79** Performance of calves fed whole milk and milk replacer in different sequences. M. C. Scott\*, R. E. James, and M. L. McGilliard, *Virginia Polytechnic Institute and State University, Blacksburg.* 

Previous studies have compared performance of calves receiving either pasteurized waste milk or whole milk to that of calves receiving 20% protein and 20% fat milk replacer on an equal volume basis. The objective of this study was to compare performance of calves fed different sequences of whole milk (M), 25% protein and 30% fat, to a 28% protein and 20% fat milk replacer (MR). Milk replacer was reconstituted to 14% DM to be isocaloric to M, both providing 5 mcal/kg of metabolizable energy on an as fed basis. Holstein, Jersey, and reciprocal crosses, both bull (n=17) and heifer calves (n=46) were fed for 8wk. Treatment (TRT) 1 calves received M for 28 d and then MR until weaning at 56 d. Treatment 2 calves represented the reverse of TRT 1, whereby calves received MR for 28 d and switched to M until weaning. Calves on TRT 3 were fed MR for the entire 8 wk. Feeding rates were determined by calf birth weight. Liquid diets were fed twice daily at the rate of 5.5, 7.3 and 8.2 kg/d as-fed for birth weights of less than 27, 27-36 and greater than 36 kg. Calf body weight (BW), wither height (WH), hip height (HH), hip width (HW), body length (BL), and heart girth (HG) were recorded at 1 d, 28 d  $\pm$ 3 d, 35 d  $\pm$ 3 d, and 56 d of age. Starter grain intake (GI) was measured twice weekly. Starter grain and water were offered ad libitum after 1 w of age. Four periods of time were evaluated; birth to weaning, the first four weeks (P1), the transition period (TP) of wk 4 to wk 5, after calves switched diets, and until 8 wk (P2). Measures of BW, WH, HH, HW, BL and HG gain were not different among treatments over the entire period. Similarly, GI through 8 wk was not different. Calves on TRT 1 had a more rapid ADG, 0.67 kg/d  $\pm$  .07 SE during TP than TRT 2.  $0.57 \text{ kg/d} \pm .07 \text{ SE}$  (P $\leq 0.05$ ). This was probably caused by increased water retention due to greater mineral content of MR relative to the M. The trial demonstrated similar growth through 8 wk with either sequence of M and MR or only MR when offered on an isocaloric basis.

Key Words: Calf, Milk replacer

**M80 Development of specific breeds equations to estimate chemical empty body composition using the 9-10-11<sup>th</sup> rib cut composition.** A. Berndt<sup>1</sup>, G. M. da Cruz<sup>3</sup>, G. F. Alleoni<sup>4</sup>, M. M. Alencar<sup>3</sup>, and D. P. D. Lanna<sup>\*2</sup>, <sup>1</sup>*APTA/SP*, *Andradina, São Paulo, Brazil*, <sup>2</sup>*ESALQ/USP*, *Piracicaba, São Paulo, Brazil*, <sup>3</sup>*EMBRAPA/CPPSe, São Carlos, São Paulo, Brazil*, <sup>4</sup>*IZ/SP*, *Nova Odessa, São Paulo, Brazil*.

Simple linear regressions were obtained from empty body chemical composition and Hankins & Howe 9-10-11th rib cuts chemical composition for purebred and crossbred Nellore bulls. One hundred and eighty eight (188) Nellore (NE) and crossbred Canchim x Nellore (CN), Angus x Nellore (AN) and Simental x Nellore (SN) young bulls were used. Bulls came from a 300 Nellore dams randomly mated to several representative bulls for each breed. Bulls initial empty body weight (EBW) was 288 kg and they were fed for 57-186 days. Animals were slaughtered when estimated hot carcass weight was greater than 225 kg and ultrasound back fat thickness reached 4-5 mm. Diet had 60% corn silage and 40% concentrate, 13.8% CP and 71.5% TDN on a DM basis. Direct empty body composition was collected on 115 animals, for which all tissues including blood, head, hide, feet, viscera and carcass were frozen, grounded, homogenized and sampled for chemical analysis. Of these 115 bulls, 48 animals were slaughtered before feeding to obtain baseline body composition and 67 were slaughtered after the feeding period. The individual predictive equations for percent empty body water and ether extract using the percentage water in the 9-10-11<sup>th</sup> rib cut are presented on table 1.

Table 1: Regression equations using %water of 9-10-11<sup>th</sup> rib cut as independent variable to predict empty body composition.

Genetic group	Dependent variable (% EBW)	Regression equation	R <sup>2</sup>
AN	EE	y = -1.2124x + 111.1	0.8051
AN	Water	y = 0.5294x + 31.96	0.8950
CN	EE	y = -1.2744x + 114.22	0.8196
CN	Water	y = 0.581x + 28.61	0.8870
NE	EE	y = -1.1648x + 107.09	0.8716
NE	Water	y = 0.5348x + 31.70	0.8986
SN	EE	y = -1.4296x + 124.54	0.9151
SN	Water	y = 0.6447x + 23.63	0.9129

Key Words: Empty body composition, Nellore crossbred, 9-10-11<sup>th</sup> rib cuts

**M81** Phenotypical characterization of genetically different cattle in segregating family structures, growth and carcass characteristics. R. Pfuhl\*, O. Bellmann, J. Wegner, K. Ender, and C. Kühn, *Research Institute for the Biology of Farm Animals, Dummerstorf, Germany.* 

The physiological mechanisms which affect the transformation of nutrients into body fat in bulls of secretion type or into muscle tissue in bulls of accretion type is still not fully revealed. Hence, we designed a study of segregating family structures using Charolais (Ch) cattle as a representative for the accretion type and German Holstein (GH) cattle as a representative for the secretion type of cattle. In further experiments, the P<sub>0</sub> generation was characterized phenotypically. This study compares selected phenotypical results of the F2 bull generation (n=65) with the P<sub>0</sub> data to get first insights in potential segregating of growth and carcass traits. The F<sub>2</sub> bulls of five segregating families showed an average final weight at 18 mo between 676.69 kg and 739.18 kg. These data were intermediate between the P<sub>0</sub> Charolais bulls, which gained a final weight of 750.6 kg and were 84.7 kg heavier than the GH bulls with 665.9 kg (P<0.001). The Charolais bulls exhibit a hot carcass weight (HCW) of 450.26 kg, the GH bulls 356.74 kg (P< 0.001). The average HCW of the  $F_2$  bull families extends from 387.85 kg to 414.46 kg and is within the  $P_0$  data (P < 0.001). The Rib eye area of the F<sub>2</sub> bulls (103.18 cm<sup>2</sup> – 108.17 cm<sup>2</sup>) varies (P < 0.001) within the  $P_0$  bulls (Ch = 125.82 cm<sup>2</sup> and GH = 82.14 cm<sup>2</sup>). The average dressing percent in the  $F_2$  bull families (56.46 %– 58.01 %), was between the  $P_0$  bulls (Ch = 60.31, GH = 53.96) with (P< 0.001). The inner fat content of the F<sub>2</sub> bulls varies from 39.6 kg to 43.3 kg and is within the P<sub>0</sub> range (Ch = 35.4 kg, GH = 51.06 kg) with (P< 0.001). In conclusion, the F<sub>2</sub> animals showed intermediate data with high variation between the values of the P<sub>0</sub> animals in the observed traits, which confirms the expected results for the F<sub>2</sub> generation. No atypical effects of these traits were recorded. Further experimentation will be conducted on the F<sub>2</sub> bulls.

Key Words: Cattle, Carcass, Growth

**M82** Residual feed intake (RFI), behavioral, and physiological measures in Angus Bulls. J. P. Cassady\*, C. S. Whisnant, M. H. Poore, and G. B. Huntington, *North Carolina State University, Raleigh*.

The objective was to measure RFI in 56 registered Angus bulls (285  $\pm$  34 kg BW, 275  $\pm$  21 d old) from one herd and to relate RFI to physiological and economically important traits. After completing a post-weaning vaccination and parasite elimination program, bulls were adapted to a corn silage-based diet (140 g CP, 1.73 Mcal NEm and 1.22 Mcal NEg per kg DM), and trained to use individual feeding gates. They were blocked based on BW and sire into groups of 12 and fed the same diet for 84 d. They were weighed every 14 d, and measures of temperament, chute escape velocity, hip height, scrotal circumference, blood samples, for determination of circulating concentrations of testosterone, triiodothyronine (T3) and thyroxine (T4), and ultrasound measures of body composition were collected at d 8, d 58, and d 84. At the end of the study rate of eating was measured on the eight bulls with the highest and lowest RFI (n = 16). Mean  $\pm$ SD RFI (predicted minus measured) was  $0.12 \pm 0.73$  kg DM/d. Mean  $\pm$  SD ADG and DMI were 1.42  $\pm$  0.20 and 7.3  $\pm$  1.7 kg. Residual feed intake was positively correlated with eating rate, BW, BW gain, hip height, scrotal circumference, and testosterone on d 8 (P < 0.05) and negatively correlated with T3 on d 8 (P < 0.08). Increased ribeve area (P < 0.07) and calmer temperament (P < 0.13) tended to correlate with RFI. For 6 bulls greater than 1 SD from the mean, RFI was  $1.40 \pm$ 0.37, and for 7 bulls less than 1 SD from the mean RFI was -1.10  $\pm$ 0.56. Compared to the 8 bulls with lowest RFI (-1.03 kg/d), the 8 bulls with the highest RFI (1.24 kg/d) ate faster, were taller and heavier, gained weight faster (P < 0.05), and tended (0.05 < P < 0.20) to be calmer in the weigh box, slower to leave the chute, had larger ribeye area, greater increases in subcutaneous fat over the rib and rump, and less increase in hip height from d 58 to d 84. We conclude that differences in physiological and behavioral traits of bulls likely explain important components of RFI.

Key Words: Beef cattle, Residual feed intake, Efficiency

## Meat Science and Muscle Biology

**M83 Fatty acid profile in selected rodent and fish species from Colombia.** L. L. Betancourt<sup>\*1</sup> and G. J. Díaz<sup>2</sup>, <sup>1</sup>Universidad de La Salle, Facultad de Zootecnia, Bogotá, Distrito Capital, Colombia, <sup>2</sup>Universidad Nacional de Colombia, Facultad de Medicina Veterinaria y Zootecnia, Bogotá, Distrito Capital, Colombia.

The fatty acid profile of muscle tissue of three rodent species and nine fish species was investigated. Rodent species included capybara (Hydrochaeris hydrochaeris), guinea pig (Cavia porcellus), and agouti (Agouti sp.), which are commonly eaten by native Colombian people. The fish species investigated were: trout (Oncorhynchus mykiss), tilapia (Oreochromis sp.), common two-banded seabream (Diplodus vulgaris), black capu (Colossoma macropomum), tiger shovelnose catfish (Pseudoplatystoma fasciatum), jau catfish (Paulicea luetkeni), matrincha (Brycon sp.), pictus catfish (Pimelodus grosskopfii) and capitán (Eremophilus mutisii). A total of 500 gr of muscle tissue was lyophilized, and its fat content was extracted with organic solvents. Fatty acids extracted from the fat were analyzed as methyl-esthers using gas chromatography with flame-ionization detection. Individual fatty acids were reported as percent of total fatty acids. The capybara, agouti, and guinea pig omega-3 (n-3) fatty acid content in muscle tissue was 21.3, 13.0, and 9.3%, respectively. These levels of n-3 fatty acids are higher that those observed in beef, pork or poultry. Among

J. Anim. Sci. Vol. 84, Suppl. 1/J. Dairy Sci. Vol. 89, Suppl. 1