W32 Tight junction (TJ) protein expression during engorgement of rat and bovine mammary glands. C. V. Cooper*[1,2,3], K. Stelwagen[2], C. D. McMahon[2], K. Singh[2], V. C. Farr[2], and S. R. Davis[2], [1]*Dexcel Ltd., Hamilton, New Zealand,* [2]*AgResearch, Hamilton, New Zealand,* [3]*Massey University, Palmerston North, New Zealand.*

The pattern of expression of TJ proteins was investigated during engorgement of rat and bovine mammary glands. An increase in mammary TJ permeability was previously shown to occur within 24 h of milk accumulation. The expression of occludin and claudin-1, the major integral transmembrane components of TJ, was determined in two experiments. In experiment 1, Sprague-Dawley rats at peak lactation (d 16) had three abdominal inguinal glands on one side sealed to induce mammary engorgement, the remaining glands were not sealed and acted as suckled controls. Mammary tissue was collected post-mortem at 0, 6, 12, 18, 24 and 36 h after teat sealing (n = 6 rats per time point). In experiment 2, alveolar mammary tissue was collected post-mortem from 42 mid-lactation Holstein Friesian dairy cows at 0, 6, 12, 18, 24, 36 and 72 h following the last milking (n = 6 cows per time point). Immunoblotting showed a characteristic multiple banding pattern for occludin between 60 and 80 kDa. The higher molecular weight (MW) bands were highly phosphorylated and resistant to NP-40 detergent extraction, suggesting they predominantly derive from the tight junction complex. Occludin expression declined during mammary engorgement in rat and bovine glands ($P<0.05$). Claudin-1 migrated in SDS-PAGE as two bands at 22 and 28 kDa. In rats, expression of the 28 kDa band declined within 12 h of mammary engorgement ($P<0.05$), while that of the 22 kDa band, along with lower MW degradation products, increased ($P<0.05$). Both bands were expressed at low levels by 36 h of mammary engorgement. In contrast, claudin-1 protein expression did not alter with engorgement in bovine mammary glands ($P>0.05$). Occludin and claudin-1 expression showed large individual animal to animal variation. Furthermore, the response to mammary engorgement was locally regulated as no changes were detected in suckled control rat mammary glands. Between species variation in the pattern of TJ protein expression suggest that the increase in TJ permeability during milk accumulation is regulated differently between rats and dairy cows.

Key Words: Tight junction, Lactation, Mammary engorgement

W33 Developmental regulation of glucosidase II in mouse mammary gland. J. Feng* and I. K. Vijay, *University of Maryland, College Park.*

The mammary gland synthesizes and secretes large amounts of well-characterized glycoproteins of the milk fat globule membrane and α-lactalbumin during lactation. Previous studies from our laboratory have shown that several glycosyltransferases of the dolichol cycle are coordinately regulated during the growth and differentiation of the mammary gland as it cycles between dormancy and lactation. We have hypothesized that the processing glucosidases I and II would follow a similar pattern of expression in coordination with the glycosyltransferases. The developmental regulation of glucosidase II was investigated in mouse mammary gland. Glucosidase II is a heterodimer of a catalytically active subunit (α subunit) and a smaller subunit (β) that contains the signal for endoplastic reticulum (ER) retention. Mouse mammary glands at different stages of development (n=30 for virgin and post lactating glands; 20 for all the other stages) were examined for glucosidase II mRNA by RT-PCR (both α and β subunits), immunoreactive α and β subunits, and enzyme activity. All three parameters showed a similar pattern, i.e., they were low in tissues from virgin animals, increased steadily during pregnancy and lactation, reaching a peak around mid-lactation, and declined sharply in glands from post-lactating animals. At mid-lactation, glucosidase II α and β subunits mRNA level increased 4-fold relative to the virgin stage. The immunoreactive protein of the two subunits also had 5 and 7-fold increases, respectively. The glucosidase II activity increased nearly 5-fold in mid-lactation compared to virgin stage. These data suggest possible transcriptional and post-transcriptional modulation of glucosidase II during development of the mouse mammary gland. Further, the striking similarity in the regulation of this enzyme and the previously studied glycosyltransferases, when combined with the data on the developmental profile of glucosidase I, indicates that common regulatory signaling cascades may control the enzymes of the glycosylation machinery in the mammary gland. (Supported by N.I.H. grant GM59943.)

Key Words: Glycosylation, Glucosidase II, Mammary gland

Growth & Development

W34 Impact of 5α-dihydrotestosterone on musculoskeletal status of mature laying hens. T. D. Faidley*, S. E. Nicolich, and D. R. Thompson, *Merck Research Laboratories, Somerville, NJ.*

Genetic selection for improved egg production has resulted in aged laying hens that are fragile and depleted of muscle. Selective androgen receptor modulation may offer potential to improve musculature and skeletal structure of these birds. "Spent hens" have become more of a liability than an asset to the industry. We hypothesized that compounds such as 5α- dihydrotestosterone (DHT) may result in muscle and bone gain, thus improving the health and value of aged layers. Subcutaneous injections of 3 mg/kg DHT (5X weekly) were compared to saline injections in mature laying hens (n=10). Hens were housed individually in cages and allowed unlimited access to feed and water. After 3 weeks, DHT treatment decreased ($P<0.05$) egg production (0% vs. 60%), feed consumption (72 g/d vs. 126 g/d), weight gain (-13 g vs. 58 g), and breast fillet as a % of carcass weight (7.2% vs. 8.1%). DHT treatment increased ($P<0.05$) comb redness (a*, 20 vs. 13); and weights of comb (31.2 g vs. 2.2 g), heart (10.6 g vs. 8.1 g), thigh muscle (72.9 g vs. 64.9 g), and metatarsus (23.1 g vs. 21.6 g). DHT treatment had no significant effect on weight of carcass (1337 g vs. 1227 g), whole breast (302 vs. 325), or femur (9.8 g vs. 9.3 g). Breast fillet weight tended to decrease ($P<0.1$) with DHT treatment (97 g vs. 103 g). In summary, DHT treatment was successful in halting egg production and in decreasing feed consumption, however, musculoskeletal effects were inconclusive. Further research is needed to determine if anabolic treatment of aged laying hens can improve welfare and/or economics of egg production.

Key Words: Androgen, Anabolic, Laying hens

W35 Fetus growth at day 78 of gestation in nutrient restricted ewes. M. M. Schwope*, W. J. Means, A. W. Wolf, B. W. Hess, and S. P. Ford, *University of Wyoming, Laramie WY/USA.*

ABSTRACT: Under-nutrition during early gestation can affect muscle development. Our purpose was to determine if fetal growth was affected by nutrient restriction of the gestating ewe. Control (C) ewes were fed 100% of the National Research Council (NRC) recommended diet for gestating ewes. Nutrient restricted (NR) ewes were fed 50% of NRC recommendations during days 28 to 78 of gestation. Control and NR ewes were euthanized (d 78 gestation) prior to removal of gravid uteri. The head and internal organs were removed after the fetus(s) were taken from the uterus. Eviscerated ewes and fetuses were hung by the *Achilles* tendon for 24 to 34 h at 4°C or 15°C, respectively. Subsequently, ewe and fetus *Longissimus dorsi* (Ld) and *Semitendinosus* (St) were removed. Whole body, eviscerated body, Ld, and St weights were recorded. Whole body weight tended ($P = 0.07$)to be lower in NR ewes, although ewe eviscerated weight was not different($P = 0.13$). Fetal whole body ($P = 0.49$) and eviscerated weights ($P = 0.58$) were not different. However, fetal Ld weight as percentage of fetal whole body weight and as percentage of eviscerated fetal weight were different because Ld weights of NR fetuses tended to be heavier ($P = 0.10$) than C fetuses, 3.34 and 2.92 g, respectively. This relationship was not found for fetal St ($P = 0.51$). Ewe Ld and St weights were not different ($P > 0.10$) as percentage of ewe whole body and eviscerated weight. Nutrient restriction of ewes during 28 to 78 d of gestation causes differential changes in muscle development.

Key Words: Fetus, Nutrient restriction, Muscle growth

W36 Dietary supplementation of nucleosides in late pregnant and lactating rats. C. M. De Jesus Arias*, C. E. Oliver, W. L. Keller, and C. S. Park, *North Dakota State University, Fargo ND/USA.*

The objective of this study was to evaluate if the inclusion of nucleosides in the diet of pregnant and lactating rats increases pup performance and immune status. Thirty-two female Sprague-Dawley rats, approximately 14 wk of age and 14 d of gestation, were randomly assigned to either control (nucleotide-free semi-purified diet; Purina Basal Diet # 5755, Ralston Purina, Richmond, IN) or nucleoside (control diet with nucleosides) treatments. The nucleosides were suspended in water by weight in the following proportions: adenosine (1.11), guanosine (1.17), uridine (1.01), cytidine (1.01), and thymidine (1.00). Rats were dosed by gavage daily beginning on d 14 of gestation through d 19 of lactation. Control rats received water, and treatment rats received nucleoside suspension at 0.64 mg/g body weight per d. Dams were weighed every 3 d during gestation and lactation. Feed intake was recorded every other day. On d 3 of lactation, litters were adjusted to 8 pups per dam. Litters were weighed every 3 days through d 19 of lactation. Upon sacrifice, blood was collected from dams (control, 1 dam; treatment, 2 dams) and pups on d 3, 10, 15, and 19 of lactation. Milk yield was estimated on d 15 of lactation. Milk was collected by miniaturized suction apparatus on d 3, 10, 15, and 19 of lactation. Serum and milk immunoglobulin G_{2a} (IgG_{2a}) concentrations were measured by ELISA. The average daily feed intake of the dams was not different between groups (control, 25.7 $\pm$ 9.4 g; treatment, 25.6 $\pm$ 10.4 g). The average daily gain from d 3 to d 15 of treatment pups was approximately 20% higher than that of control pups. Milk yield and IgG_{2a} in serum of the dams (averaged over the trial) were not different between groups. In the treatment group, a significant ($P = 0.02$) correlation (r = 0.61) existed between pup serum concentrations of IgG_{2a} (averaged over the trial) and milk IgG_{2a} (averaged over the trial). Our results indicate that nucleoside supplementation affects pup performance and serum IgG_{2a} concentration.

Key Words: Nucleosides, Pup performance, IgG_{2a}

W37 Effects of specific conjugated linoleic acid (CLA) isomers on growth characteristics in obese Zucker (fa/fa) rats. S. R. Sanders*[1], M. K. Teachey[1], A. Ptock[2], K. Kraemer[2], O. Hasselwander[2], E. J. Henriksen[1], and L. H. Baumgard[1], [1]*University of Arizona, Tucson AZ,* [2]*BASF AG, Ludwigshafen, Germany.*

Growing female obese Zucker (fa/fa) rats were treated (via intra-gastric gavage) for 21 d with either 1) vehicle [corn oil; 2 ml/kg body weight (BW)], 2) CLA mixture [50:50; *trans*-10, *cis*-12 and *cis*-9, *trans*-11 CLA], 3) *cis*-9, *trans*-11 CLA, or 4) *trans*-10, *cis*-12 CLA [all at 1.5 g CLA/kg BW]. Average daily gain (g/d) was significantly (P<0.05) reduced by *trans*-10, *cis*-12 CLA and the CLA mixture (2.50, 1.95, 2.69 and 1.39, for treatments 1, 2, 3 and 4, respectively). There was no treatment effect on average whole-body (minus heart and liver) composition (dry matter basis); fat (70.2%), protein (21.0%) and ash (4.3%). Compared to *cis*-9, *trans*-11 CLA, Zucker rats treated with *trans*-10, *cis*-12 and the CLA mixture had more carcass water (38.0, 40.8, 37.0 and 39.0% for treatments 1, 2, 3 and 4). There was no treatment effect on soleus and plantaris muscle weights. Treatment had no effect on heart or liver weight, nor heart or liver weight as a percentage of body weight, but *trans*-10, *cis*-12 CLA significantly increased liver lipid content (23.6, 22.0, 24.4 and 31.0% for treatments 1, 2, 3 and 4). Carcass fatty acid analysis indicated *cis*-9, *trans*-11 CLA content averaged 0.2, 2.0, 1.9 and 0.5 and *trans*-10, *cis*-12 CLA averaged <0.1, 1.2, 0.5 and 1.7 g/g of fat for treatments 1, 2, 3 and 4. Liver fatty acid analysis indicated *cis*-9, *trans*-11 CLA averaged 0.2, 1.3, 1.6 and 0.7 and *trans*-10, *cis*-12 CLA averaged <0.1, 0.7, 0.5 and 1.4 g/g of fat for treatments 1, 2, 3 and 4. Ratios of $C_{16:0}/C_{16:1}$ and $C_{18:0}/C_{18:1}$ (a proxy of Δ^9-desaturase capability) were not affected in hepatic lipids (9.8 and 1.8, respectively). The palmitate ratio was unaffected in carcass fats (4.3) but *trans*-10, *cis*-12 increased the ratio of $C_{18:0}/C_{18:1}$ (0.14, 0.12, 0.13 and 0.17 for treatments 1, 2, 3 and 4). Similar to previous reports, CLA increased hepatic lipid content, but the ability of CLA to alter body composition in obese Zucker rats remains questionable.

Key Words: CLA, Zucker Rat, Body Composition

W38 Body composition and carcass fatty acid profiles in hybrid striped bass treated with recombinant bovine somatotropin (rbST). S. R. Sanders*[1], J. L. Collier[2], L. H. Baumgard[1], and R. J. Collier[1,2], [1]*University of Arizona,* [2]*AquaTrophics Inc., Tucson, AZ.*

Eleven hybrid striped bass initially weighing 10 g were injected with passive integrated transponder tags and sorted into two 160 L tanks housed in an unheated greenhouse for 196d. Fish were randomly assigned to either IP injections of 0.5 ml rbST (100 mg/g body weight [BW]) or normal saline 4x during the first 33d. After d33, both groups were untreated, managed identically and harvested on d196. Fish were fed floating steelhead pellets 2x/d to achieve a total daily intake of 3-5% BW. Immediately after the 4th IP injection, due to ambient temperature changes, water temperature decreased from 22 to 17C, resulting in all fish consuming little if any feed. Feed intake resumed as temperatures returned to 22C (54d post 4th IP injection). Overall (d1-196) average daily gain (ADG) was not affected by treatment (557 mg/d), but ADG during IP injections (d1-56) was increased 42% by rbST, although not significant (P>0.1). Overall fish length gain was not altered by treatment (5.1 mm/d) however, during the IP injection phase, rbST increased (P<0.001) fish length gain (9.1 vs. 6.8 mm/d). Whole body composition analysis 140d post rbST administration indicated no difference in carcass dry matter (30.6%), fat (35%), protein (47.6%) or ash (16.6% [fat, protein and ash reported on a dry matter basis]). Fatty acid analysis indicated fish treated with rbST tended (P=0.11) to have a higher unsaturated fatty acid content (71.5 vs. 68.5%) and reduced *de novo* fatty acid contribution (C_{12}-$C_{14:1}$; 62 vs. 70 mg/g of fat). Fish treated with rbST had increased (P=0.05) proportions of long chain PUFA ($\geq C_{20:1}$; 177 vs. 129 mg/g fat), but no difference in the Δ^9-desaturase index ($C_{14:0}/C_{14:1}$, $C_{16:0}/C_{16:1}$, $C_{18:0}/C_{18:1}$). Body composition analysis indicates the beneficial effects of rbST on nutrient partitioning in young hybrid striped bass are lost or diluted over time (140d) following rbST treatment cessation; preliminary data suggest rbST enhances ADG and has beneficial effects on carcass fatty acid profile.

Key Words: rbST, Fish, Body composition

W39 Effect of restricted post-weaning growth resulting from reduced floor and feeder space on pig growth performance in a wean-to-finish system. B. F. Wolter[1], M. Ellis[2], J. M. DeDecker*[2], B. P. Corrigan[2], S. E. Curtis[2], E. N. Parr[3], and D. M. Webel[3], [1]*The Maschhoffs LLC, Carlyle, IL/USA,* [2]*University of Illinois, Urbana, IL/USA,* [3]*United Feeds, Inc., Sheridan, IN/USA.*

The effect of reduced post-weaning growth resulting from restricted floor and feeder-trough space on subsequent growth to slaughter was investigated in a wean-to-finish system. The study was carried out from weaning (5.5 0.01 kg BW; 17 d of age) to end of wk 25 post-weaning. Pigs (n =1,728) were used in a randomized block design with a 2 x 2 x 2 factorial arrangement of treatments: 1) floor space (High [0.630 m^2/pig] vs Low [0.315 m^2/pig]), 2) feeder-trough space (Unrestricted [4 cm/pig] vs Restricted [2 cm/pig]), and 3) duration of floor- and feeder-trough-space treatment (12 vs 14 wk post-weaning). The study was carried out in two periods; Period 1 was from weaning to the end of the treatment period (i.e. wk 12 or wk 14 post-weaning); Period 2 was from the end of the treatment period to wk 25, during which pigs on all treatments had the same floor and feeder space. During Period 1 both Low floor space and Restricted feeder space reduced ADFI (P < 0.05), but ADG was only reduced by Low floor space (P < 0.01). Pigs on treatment for 14 compared to 12 wk had higher (P < 0.01) ADG and ADFI. Neither feeder space nor treatment duration affected growth performance during Period 2. However, during Period 2 pigs on the Low compared to High floor space had increased ADG and G:F with the difference being greater for pigs on treatment for 14 than 12 wk (floor space x treatment duration interaction; P < 0.05). However, Low floor-space pigs tended (P = 0.06) to be lighter than High floor-space pigs at the end of Period 2. Carcass measures at end of Period 2 were not influenced (P > 0.05) by any treatment. In summary, pigs with restricted growth due to Low floor space until 12 or 14 wk post-weaning had increased growth and feed efficiency in the subsequent period to wk 25 post-weaning.

Key Words: Feed trough, Floor space, Pigs

W40 Refolding and purification of unprocessed porcine myostatin expressed in *E. coli*. H. J. Jin, Y. S. Kim*, and M. A. Dunn, *University of Hawaii, Honolulu HI*.

Myostatin is a growth and differentiation factor that suppresses skeletal muscle growth. Like many other TGF-β family member proteins, it is expressed as a prepropeptide that yields a mature form of myostatin after proteolytic processing at the paired basic residues (Arg-Lys-Arg-Arg). Since unprocessed pure myostatin is not currently available, the objective of this study was to purify unprocessed, refolded, porcine myostatin expressed in *E. coli*. Recombinant myostatin inclusion bodies harvested from *E. coli* were solubilized (1 mg/ml) in a buffer solution (50 mM CAPS, pH 11.0 containing 0.3% N-lauroylsarcosine and 1 mM DTT). Then, the inclusion body solution was diluted 100 times with refolding buffer (10 mM Tris buffer containing reduced and oxidized glutathionine, pH 8.5) and incubated at 4°C for 7 days. After dialysis in 20 mM Tris buffer (pH 8.5), the myostatin containing solution was subjected to anion exchange chromatography, and fractions containing the recombinant refolded myostatin were collected and combined. The combined solution was subjected to size exclusion chromatography to further purify the refolded myostatin. The purified myostatin formed a monomer under reduced conditions, and a dimmer under non-reduced conditions in SDS-PAGE analysis. Upon incubation with furin, an endopeptidase cleaving the paired basic residues, the unprocessed recombinant myostatin (50 kD) yielded 37 kD and 15 kD proteins, corresponding respectively to the prodomain and mature form of myostatin. Based on the current biochemical results, it is concluded that the refolded native form of unprocessed myostatin could be obtained from *E. coli* expressed inclusion bodies with high efficiency (15% yield) and that the unprocessed myostatin is a substrate for furin.

Key Words: Protein purification, Myostatin, Furin

W41 Effect of flax supplementation and a combined trenbolone acetate and estradiol implant on muscle satellite cell activity in beef cattle. J. D. Dunn*, A. T. Waylan, J. P. Kayser, E. K. Sissom, and B. J. Johnson, *Kansas State University, Manhattan*.

Objectives of this study were to evaluate the effects of 5% ground flaxseed (FLAX) and a combined TBA/E_2 growth promotant, Revalor-S, (IMP) on muscle satellite cell proliferation and differentiation. Sixteen yearling crossbred steers (initial BW = 397 kg) were randomly assigned to one of four treatments: 1) FLAX/IMP, 2) No FLAX/IMP, 3) FLAX/No IMP, 4) No FLAX/No IMP. Steers were allowed ad libitum access to a 93% concentrate diet for the entire study. Biopsy samples (3.5 g) were obtained from the longissimus muscle on d 0, 14, and 28. Satellite cells were isolated from the biopsy samples by enzymatic digestion and differential centrifugation. Satellite cells from each steer were resuspended in Dulbecco's Modified Eagle Medium (DMEM) with 10% fetal bovine serum and plated on two wells of two four-well tissue culture plates coated with reduced growth factor matrigel. Cultures from each steer were stained 24 h post-plating with Hoechst 33342 and nuclei were counted. At 96 h post-plating, cells from each steer were put into a fusion-promoting media of DMEM with 3% horse serum and 1.5 μg/mL BSA-linoleic acid. At 192 h, cultures were stained and counted for total and myotube nuclei. FLAX or IMP had no effect on satellite cell activity. However, nuclei at 24 h post-plating increased ($P<0.001$) from d 0 to 28 whereas total nuclei 192 h post-plating were unchanged ($P>0.10$). Myotube nuclei increased ($P<0.05$) from d 0 to 28 and thus fusion percentage also increased ($P<0.05$) from d 0 to 28. Cell yield per g of muscle tissue also increased ($P<0.05$) from d 0 to 28 while number of doublings decreased ($P<0.001$). These data suggest satellite cells were activated *in vivo* over the 28 d period and that the cells lost proliferative capacity when placed in culture. Also, the increases in myotube nuclei and fusion percentage over time indicate that isolated satellite cells became more inclined to differentiate into muscle over the feeding period regardless of FLAX or IMP.

Key Words: Satellite cell, Beef cattle, Trenbolone acetate

W42 Walking temporal variables of the sound and lame dairy cow. M. C. Nicodemus* and A. M. Chapa, *Mississippi State University, Mississippi State, MS*.

Lameness in dairy cattle is associated with milk production loss emphasizing the importance of early lameness detection. The human eye is limited in detecting subtle lameness so that additional detection methods are needed. Kinematic analysis has been effective in measuring subtle equine lameness, but research in dairy cattle is lacking. The objectives of this study were to determine sound and lame walking temporal variables of dairy cows. 5 sound lactating dairy cows and 5 lame cows with a lameness score between 3-5 were freely walked at a consistent velocity (1.2-1.4 m/s) through an enclosed, calibrated runway. 4 consistent, straight strides with easily detected hoof impacts were used from each cow. 60 Hz frame-by-frame analysis determined stride duration and the following temporal variables, which were calculated as percent of stride: stance durations, limb supports, and advanced placements and lift-offs. Means (SD) were calculated and paired t-tests performed to determine significant differences between left and right variables ($P<0.05$) in which variables were collapsed if no significant differences were found. The sound and lame walks were 4-beat stepping gaits with a lateral footfall sequence and alternating periods of bipedal and tripedal support. The sound walk was symmetrical as the left and right variables were insignificantly different while the lame walk demonstrated asymmetry. Stride duration was similar for both walks (sound: 1302+66 ms; lame: 1332+157 ms). In both walks the forelimbs spent 69+2% of the stride in stance and 67+3% in the hind with a 4+2% increase in stance in the lame limb. The majority of the stride in both gaits was spent in tripedal support (sound: 71+2%; lame: 78+4%) with equal bipedal support in the sound walk (diagonal: 13+2%; lateral: 16+3%) and unequal in the lame (diagonal: 7+4%; lateral: 15+5%). Advanced placements and lift-offs were regular in the sound walk, but the lateral and diagonal advanced lift-offs were irregular for the lame (lateral: RH-RF=23+1%, LH-LF=25+4%; diagonal: RF-LH= 31+1%, LF-RH=33+13%). Understanding the sound and lame walking temporal variables of the dairy cow will assist in the early detection of lameness.

Key Words: Dairy cow, Temporal variables, Lameness

W43 Effect of melengestrol acetate (MGA) on bovine muscle satellite cell proliferation and differentiation. E. K. Sissom*, J. P. Kayser, A. T. Waylan, J. D. Dunn, and B. J. Johnson, *Kansas State University, Manhattan*.

Melengestrol acetate (MGA) increases growth rate and inhibits estrus in feedlot heifers, but the effect of MGA on skeletal muscle growth and differentiation has not been studied. The purpose of these experiments was to investigate the effects of MGA on cultured bovine muscle satellite cell proliferation and differentiation. Satellite cells were used to assess the effects of MGA in a dose titration (0, 1 nM, 10 nM, 100 nM, 1 μM, 10 μM, and 100 μM) study on [^{3}H]-thymidine incorporation (TI). Cells were plated in Dulbecco's Modified Eagles Medium containing 10% fetal bovine serum. MGA was added at 0 or 48 h after plating. At 72 h, [^{3}H]-thymidine was added and incubated for 3 h. Cultures were allowed to differentiate, and nuclei were stained at 168 h with Hoechst 33342 to determine the effect of MGA (10 nM and 100 μM) addition the first 48 h on extent of differentiation and absolute myotube nuclei number. MGA addition resulted in a dose-dependent decrease ($P<0.05$) in DNA synthesis as measured by TI. The addition of 1 nM MGA did not affect ($P>0.05$) TI in bovine satellite cells as compared to a control medium (no MGA). The addition of 10 nM, 100 nM, and 1 μM MGA to cultures of proliferating bovine satellite cells reduced TI approximately 27, 25, and 28%, respectively, as compared to a control medium. MGA doses of 10 and 100 μM further reduced TI approximately, 50 and 57%, respectively, as compared to control cultures. MGA addition (10 nM) did not alter ($P>0.10$) the extent of differentiation or myotube nuclei number at 168 h. However, 100 μM MGA addition reduced ($P<0.05$) both fusion percentage and myotube nuclei number as compared to control cultures. These data obtained with concentrations $\geq$ 10 nM, a concentration several orders of magnitude greater than observed *in vivo*, suggest that MGA has pharmacological effects on bovine muscle cell proliferation and differentiation. This *in vitro* test system may be useful for evaluation of mechanism for anabolic compounds.

Key Words: Melengestrol acetate, Bovine, Muscle

W44 Ontogenetic changes in fatty acid profiles from different tissues in growing Holstein bull calves. H. C. Hafliger, III*, P. C. Gentry, S. R. Sanders, L. H. Baumgard, and R. J. Collier, *University of Arizona, Tucson, AZ*.

Holstein bull calves were euthanized at 4 wk (n=6) or 12 wk (n=5) of age. Calves were fed milk replacer until 12 d of age, then a corn-based starter feed was offered ad libitum. At slaughter, abdominal (kidney) fat, skeletal muscle and hepatic tissue were snap frozen in liquid nitrogen, then stored at -80°C until assayed. In skeletal muscle and liver tissue, few differences between specific fatty acids, besides the trans profile, or Δ^9-desaturase ratios due to age were detected. In skeletal muscle, age significantly increased the content of *trans*-10, *trans*-11, and *trans*-12 from 5.0, 1.7 and 1.1 to 26.3, 3.2 and 2.1 mg/g of fat, respectively. Furthermore, skeletal muscle *trans*-10, *cis*-12 CLA content was increased ($P<0.09$) from <0.1 to 0.5 mg/g at wk 4 and 12, but *cis*-9 *trans*-11 CLA was unaffected by age and averaged 1.2 mg/g of fat. In hepatic tissue the trans profile remained stable with increasing age averaging 1.3, 2.3, 35.1 and 5.3 mg/g for *trans*-6-8, *trans*-9, *trans*-10 and *trans*-11 $C_{18:1}$ respectively, but *trans*-12 $C_{18:1}$ increased ($P<0.05$) from 1.9 to 3.0 mg/g from wk 4 to 12. Hepatic *trans*-10, *cis*-12 and *cis*-9, *trans*-11 CLA content did not change with age and averaged 2.8 and 2.3 mg/g of fat. In abdominal fat, *cis*-9, *trans*-11 and *trans*-10, *cis*-12 CLA increased ($P<0.05$) from wk 4 to 12 (1.4 to 2.1 and <0.1 to 1.0 mg/g of fat). Similar to hepatic tissue and skeletal muscle, the trans profile markedly increased with age and this was especially true for *trans*-10 $C_{18:1}$ which increased from 18 to 47 mg/g of fat respectively. Adipose ratios of $C_{14:0}/C_{14:1}$, $C_{16:0}/C_{16:1}$, and $C_{18:0}/C_{18:1}$ (proxy for Δ^9-desaturase) increased with age ($P<0.05$) suggesting an increase in rumen biohydrogenation and/or a decrease in the Δ^9-desaturase system. Concentrations of $C_{12:0}$, $C_{14:0}$, and $C_{14:1}$ decreased ($P>0.05$) symptomatic of a decrease in *de novo* synthesis and/or an increase in long chain fatty acid ($>C_{16:1}$) incorporation, which was observed. Overall as calves aged, products of rumen biohydrogenation tended to accumulate in tissues while *de novo* synthesized fatty acids decreased in content.

Key Words: Fatty acid, CLA

W45 Tissue deposition rates and empty body composition of purebred and crossbred Nellore bulls. A. Berndt[1], G. M. da Cruz[2], G. F. Alleoni[2], M. Alencar[3], and D.P.D. Lanna*[1], [1]*ESALQ/USP, Piracicaba, SP, Brazil,* [2]*CPPSe, EMBRAPA, Sao Carlos, SP, Brazil,* [3]*IZ, Nova Odessa, SP, Brazil*.

Nellore (NE) and crossbred Canchim x Nellore (CN), Angus x Nellore (AN) and Simental x Nellore (SN) young bulls with initial empty body weight of 294.3 kg were fed for 92-161 days. The diet had 60% corn silage and 40% concentrate, 13.8% CP and 71.5% TDN on a dry matter basis. Daily empty body gains (kg/day) were 1.34 (AN), 1.12 (CN), 1.39 (SN) and 1.03 (NE). To obtain baseline body composition 14 animals of the same group were slaughtered before feedlot. Animals were slaughtered when estimated hot carcass weight was greater than 225 kg and ultrasound backfat thickness over 4 mm. Results are presented on table 1. Data were analysed by GLM proceeding of SAS (SAS, 2001). Crossbreeding greatly improved growth rates and protein deposition rates, particularly for Angus and Simental. Nellore purebred and Canchin crossbred had the fattest gain. Angus and Simental were leaner at the same empty body weight. Crossbreeding improves the potential for carcass production from Nellore cows, however calves have increased net protein and energy requirements.

Table 1:	AN	CN	SN	NE
Empty Body Composition(%)				
Water	55.69^b	54.21^b	57.64^a	52.00^c
Ether Extract	20.70^b	22.30^b	18.59^c	24.68^a
Protein	18.94^b	18.84^b	19.06^a	18.70^c
Ash	4.67^b	4.65^b	4.71^a	4.62^c
Energy (Mcal/kg)	3.02^b	3.16^b	2.82^c	3.38^a
Period gain rates (kg/day)				
Water	0.63^a	0.44^b	0.70^a	0.36^b
Ether Extract	0.41^a	0.44^a	0.37^a	0.45^a
Protein	0.25^a	0.20^b	0.26^a	0.18^c
Ash	0.066^a	0.055^b	0.068^a	0.050^b
Energy (Mcal/day)	5.25^a	5.22^a	4.98^a	5.24^a
Empty Body Gain Composition (%)				
Water	46.59^a	38.18^b	49.83^a	33.38^b
Ether Extract	30.21^b	39.24^a	26.78^b	44.39^a
Protein	18.29^a	17.73^b	18.51^a	17.41^b
Ash	4.91^a	4.85^{ab}	4.88^a	4.81^b
Energy (Mcal/day)	3.87^b	4.69^a	3.56^b	5.16^a

Key Words: Body composition, Tissue deposition rates, Nellore crossbred

W46 Morphological, behavioral and physiological measurements and their relationships with growth in beef cattle. K. Uetake*[1], T. Ishiwata[1], N. Abe[2], and T. Tanaka[1], [1]*School of Veterinary Medicine, Azabu University,* [2]*Faculty of Agriculture, Tamagawa University*.

The objective of this study was to determine the important parameters that regulate skeletal and longissimus muscle growth of beef cattle. Thirty-five crossbred (Japanese Black X Holstein) steers transported to a farm at 6-10 mo of age were managed under pen conditions. Each of the three pens (6.0 m X 9.5 m each) consisted of 11-12 steers. Serum and plasma samples from the jugular vein (concentrations of 7 hormones and 5 nutrients), ultrasonic images between the 6th and 7th rib (longissimus muscle area (LMA) and beef marbling score (BMS)), physical measurements (body weight and 10 parts of measurements), temperament scores at 5 different handling conditions, and behavioral observations using the instantaneous sampling with 10-min intervals for 2 h after morning and evening feedings (17 behavioral categories) were collected 1, 3, 5 mo after their entry into the farm. The average daily gain (ADG) and increase in LMA (ILMA) were also determined. A factor analysis with principal components and orthogonal varimax rotation determined 8 common clusters of measurements. As for growth-related measurements, ADG, the body weight 1 mo later, chest width, and the frequency of investigative behavior constituted a cluster. ILMA clustered with triglyceride and total cholesterol concentrations, LMA 1 mo later, and temperament scores at blood sampling and ultrasonic recording. ADG was not correlated with ILMA. BMS, leptin concentrations, thurl width, and the frequencies of lying and eating hay clustered together. Vitamin A concentrations entered a cluster of cathecholamine and cortisol concentrations, the frequency of grooming with pen facilities, entry order into the crush, and a temperament score on the scales. Vitamin A concentrations also tended to be correlated with insulin ($r = 0.31$, $P = 0.07$) and leptin ($r = 0.27$, $P = 0.12$) concentrations. Vitamin A may play an important role in the hormonal system(s) that regulate stress responses and longissimus muscle growth in the cattle.

Key Words: Beef cattle, Growth, Hormonal system

W47 Parameters for a refined model of ruminant growth and composition. J. W. Oltjen*[1], A. B. Pleasants[2], T. K. Soboleva[2], and V. H. Oddy[3], [1]*University of California, Davis, California,* [2]*Ag Research, Hamilton, New Zealand,* [3]*Meat and Livestock Australia, Sydney, Australia*.

We have refined the prediction system for ruminant animal growth and composition developed previously (Oltjen et al., 2000, Modelling Nutrient Utilization in Farm Animals, pp. 197-209, CABI Publishing, New York). The model represents body protein in two pools, viscera (v) and non-viscera (m). Using sheep datasets (Ferrell et al., 1986, Brit.

J. Nutr. 56:595) and New South Wales (unpublished), we have simplified the adjustments in the model for protein gain and loss of body fat (f) at near maintenance feeding, and more precisely estimated variable maintenance parameters. In the model muscle and viscera each have an upper bound (m* and v*, respectively). For muscle m* is genetically fixed; however, v* is affected by energy intake and muscle (protein) mass. Net energy intake above maintenance (NEG) is used for m and v gain before its use for fat accretion. The model is expressed in terms of energy (kJoules), with parameters k_m (0.353), c_m (1340 kJ d^{-1}), k_v (0.050 d^{-1}), cs_1 (0.314 d) and cs_2 (0.0416). Our new work has allowed simplification of previous equations so that the adjustment allowing gain of muscle or viscera at zero retained energy is f_a with e2 (3.4), and the previous equations for visceral growth have been simplified: dm/dt = k_m (NEG + $c_m f_a$) (1 - m/m*); dv/dt = k_v (v* - v); df/dt = NEG - dm/dt - dv/dt; f_a = (1-m/m*)^{e2}; v* = cs_1 MEI + cs_2 m. Maintenance energy (HP_{maint}) includes a variable coefficient on body weight: $HP_{maint} = \alpha_t$ $EBW^{0.75}$; $\alpha_t = \alpha_0$ (1 + b (MEI_t/MEI_0 - 1)(1-$e^{-t/\tau}$)) which results in a lag in change of maintenance requirements after intake changes from MEI_0 to MEI_t. Here EBW is empty body weight (kg), t is time (days), b (0.116) and τ (20.0 d) are constants; MEI_0 and α_0 are original values of intake and the maintenance coefficient. Sheep growth and composition is more accurately predicted with the revised model, and the model predicts sheep (Ferrell et al., 1986) empty body weight and fat content ($\pm$ 2.1 kg and 2.3%-units, respectively) more accurately than the current Australian feeding system. New additions refine predictions at levels of energy intake at or below maintenance.

Key Words: Growth, Composition, Mathematical modeling

W48 A dynamic model to predict the composition of fat-free matter gains in cattle.

C. B. Williams*, *USDA, ARS, U.S. Meat Animal Research Center, Clay Center, NE.*

Composition of empty BW (EBW) was described in terms of ether-extractable lipid (FAT) and fat-free matter(FFM) and the terms dEBW, dFAT, and dFFM were used to represent daily gains in these components. The dFFM is composed of protein, water, and ash, and a model was developed to predict the composition of dFFM. The conceptual approach used in model development was based on experimental data that showed as cattle grew from birth to maturity: a) the water content of FFM decreased and the protein and ash content increased, b) the protein content of FFM increased at a decreasing rate, and c) the protein:ash ratio in the FFM dry matter was fixed. These results suggest that as cattle grow and mature, gains in FFM would contain increasing amounts of protein, and the protein content of dFFM would increase at a decreasing rate as the dFFM content of dEBW decreased. Mathematical functions were formulated to represent these concepts, and a set of equations were derived to predict composition of dFFM. The protein content of dFFM was predicted as a function of the fraction of dEBW that was dFFM, FAT content of EBW, and dFFM. A fixed protein to ash ratio of 4.26:1 was used to calculate the amount of ash, and water was obtained as a residual. Gain in EBW, dFAT, and dFFM of Hereford x Angus steers from birth to 500 kg BW was simulated with a previously published model, and the above model was used to predict composition of dFFM. Predicted response curves of the EBW components over the growing period were similar in shape to observed data. Regression analysis was used to investigate the relationship between protein weight and FFM weight. Results showed a linear relationship with no evidence for curvilinearity in the predicted data and two experimental data sets. The coefficient on FFM in the predicted data was 0.249 (SE=0.0008), and in the two sets of experimental data, the coefficients on FFM were 0.247 (SE=0.003) and 0.25 (SE=0.004). These results support the conclusion that the model is capable of accurately representing the real system.

Key Words: Model, Body Composition, Protein

W49 The effect of nitrogen and forage source on feed efficiency and structural growth of prepubertal Holstein heifers. .

P. J. Kononoff*[1], A. J. Heinrichs[1], and M. T. Gabler[1], [1]*Department of Dairy and Animal Science, The Pennsylvania State University.*

Eighty Holstein heifers averaging 189.6 + 6.8 kg of BW were used to evaluate the effects of forage level and rumen degradable nitrogen source on feed efficiency, structural growth, and body condition score (BCS). A randomized complete block design was used with heifers blocked according to weight (> 136.1 kg and < 136.1 kg). Heifers were assigned one of four treatment diets that were arranged in a 2 X 2 factorial. Treatments were constructed with two levels of forage (65 or 75%) and two protein sources. Forage sources were a mixture of corn silage and chopped timothy hay. Protein sources were either soybean meal (SBM) or a slow release urea product (Optigen 1200 , CPG Nutrients, Syracuse, NY), which was fed at 1.8 % of diet DM on low forage diets and 1.3 % of the diet DM on high forage diets. Body weight was measured weekly on two consecutive days and used to adjust intake to be approximately 2.2% of BW on a DM basis. Diets were fed as a TMR using the Calan Door System for measuring individual DMI. To determine change in structural growth, wither/hip height, hip width, and heart girth were measured weekly. In addition, blood samples were collected weekly 4 hr post feeding, to determine plasma urea nitrogen concentration. Average daily gain and feed efficiency did not differ between rations of different forage level or nitrogen source, averaging 0.87 + 0.05 kg and 7.4 + 0.5 respectively across treatments. Similarly, no differences were observed in change of wither/hip height, hip width, or heart girth. No differences were observed in plasma urea nitrogen, which averaged 12.3 + 0.4 mg/dl across treatments. Results of this experiment suggest that feeding moderately different levels of forage along with either SBM or Optigen 1200, does not result in any significant differences in main or interactive effects in feed efficiency or structural growth. Optigen 1200 can be used in heifer diets to effectively replace SBM as a nitrogen source in either high or low forage rations.

Key Words: Heifer growth, Feed efficiency, Slow release urea

W50 Effects of Prepubertal Growth Rate and POSILAC® Treatment of Replacement Dairy Heifers on Subsequent Milk Production and Economics.

J. L. Vicini*[1], D. T. Galligan[2], S. E. Bettis[1], C. R. Bilby[1], S. C. Denham[1], R. L. Hintz[1], J. L. Holst[1], T. H. Klusmeyer[1], E. D. Plunkett[1], B. A. Crooker[3], W. J. Weber[3], and M. E. Van Amburgh[4], [1]*Monsanto Co, St. Louis, MO,* [2]*University of Pennsylvania, Kennett Square, PA,* [3]*University of Minnesota, St. Paul, MN,* [4]*Cornell University, Ithaca, NY.*

Holstein heifers (N = 715) at AZ, ID, MN and NY were used to determine if prepubertal growth rate and bST affect subsequent milk yield. Heifers were assigned randomly to pens and each site had 2 pens/treatment. Treatments were in a 2X3 factorial arrangement of three feed management programs with or without POSILAC (bST; 500 mg/14 d). Feed management programs were restricted intake of a control diet (CECP) to achieve an ADG of 0.6 to 0.8 kg/d, ad lib intake of a high energy diet (HECP), and ad lib intake of a high energy and high metabolizable protein diet (HEHP). Intake of CECP-bST heifers was matched to their CECP counterparts. The HECP and HEHP diets contained more energy than CECP and HEHP had more MP than CECP or HECP. At treatment initiation, BW averaged 162 (range: 135 to 189) kg and age averaged 172 (range: 128 to 211) d. After the 140-d treatment period, all heifers within a site were fed CECP for an ADG of 0.6 to 0.8 kg until 4 to 6 wks prior to calving. Lactation management was by normal site procedures and 634 heifers calved. Mean milk yield during the 252-d lactation was 28.4, 27.0, 27.5, 28.5, 27.9 and 26.1 kg/d for CECP, HECP, HEHP, CECP-bST, HECP-bST and HEHP-bST cows. Milk yield of HECP cows was less than CECP, as has been reported previously. Yields of CECP and HECP-bST cows did not differ. The reason HEHP-bST cows produced less milk is not known. Annual cost of producing 35 heifers per treatment was compared to CECP after correcting for change in number of replacements and accounting for feed costs and difference in milk yield. Costs were -2548, -3973, -443, -2692 and -2272 $US for HECP, HEHP, CECP-bST, HECP-bST and HEHP-bST. BST increased production of HECP cows to values similar to that of CECP cows, but the magnitude of change did not economically justify the use of bST.

Key Words: Heifers, Economics, bST

W51 Effects of feed management program and POSILAC® on prepubertal growth rate of replacement dairy heifers. J. L. Vicini*[1], S. E. Bettis[1], C. R. Bilby[1], S. C. Denham[1], R. L. Hintz[1], J. L. Holst[1], E. D. Plunkett[1], B. A. Crooker[2], W. J. Weber[2], H. Chester-Jones[2], and M. E. Van Amburgh[3], [1]*Monsanto Co., St. Louis, MO,* [2]*University of Minnesota, St. Paul, MN,* [3]*Cornell University, Ithaca, NY.*

Holstein heifers (N = 715) at AZ, ID, MN and NY were used to determine effects of bST and feeding additional energy and metabolizable protein (MP) on growth of replacement dairy heifers. Heifers were assigned randomly to pens and each site had two pens per treatment. Treatments were in a 2X3 factorial arrangement of three feed management programs with or without POSILAC (bST; 500 mg/14 d). Feed management programs were restricted intake of a control diet (CECP) to achieve an ADG of 0.6 to 0.8 kg, ad libitum intake of a high-energy diet (HECP), and ad libitum intake of a high energy and high MP diet (HEHP). Intake of CECP-bST heifers was matched to that of their CECP counterparts. The HECP and HEHP diets contained more energy than CECP. Diet CP content varied but HEHP had more MP than CECP or HECP. At treatment initiation, BW averaged 162 (range: 135 to 189) kg and age averaged 172 (range: 128 to 211) d. After the 140-d treatment period, all heifers within a site were fed CECP for an ADG of 0.6 to 0.8 kg until 4 to 6 wks prior to calving. During the 140-d treatment period, ADG for CECP, HECP, HEHP, CECP-bST, HECP-bST and HEHP-bST heifers was 0.77, 1.13, 1.21, 0.81, 1.13 and 1.29 kg, respectively. From the end of the treatment period to the first post-calving body weight, ADG was 0.52, 0.61, 0.63, 0.56, 0.64 and 0.64 kg, respectively. The percentage of heifers that became pregnant was 90.5, 89.1, 88.4, 95.9, 90.8 and 94.9% for CECP, HECP, HEHP, CECP-bST, HECP-bST and HEHP-bST heifers, respectively. Age at calving was 25.2, 22.4, 21.9, 24.9, 22.6 and 21.7 mo for CECP, HECP, HEHP, CECP-bST, HECP-bST and HEHP-bST heifers, respectively. HECP and HEHP increased ADG and decreased age at calving. BST did not significantly affect ADG. High-energy diets and bST did not adversely affect percent of heifers that became pregnant.

Key Words: Heifer, Growth, bST

W52 Associations between first lactation milk yields and pepubertal and peripubertal growth rates of Holstein heifers fed diets with different concentrations of protein and energy, protein:energy ratios and injected with bST. T.I. Belloso*, M. Liboni, M.S. Gulay, M.J. Hayen, K.C. Bachman, and H.H. Head, *University of Florida.*

Holstein heifers that completed 150 DIM were used to evaluate effects of diet, season of calving (SEA), and bST on milk yield (MY), body weight (BW) and body condition score (BCS). They were from a group of 121 heifers raised on four diets and bST in a 2x2x2 factorial arrangement of treatments. Diets fed from 100-120 d of age to 341 Kg BW contained 14%(L) or 19% (H) CP with energy at 100% (L) or 110% (H) of NRC (1989) to give four protein-energy diet groups LL (n=27), LH (n= 21), HL (n=24) and HH (n=20) that contained 50, 55, 65, and 73 g of CP/Mcal ME, respectively. Half the heifers in each group were injected biweekly with bST (POSILAC®, 500 mg bST/1.4 mL): 0.2 mL to 181 kg BW, 0.3 mL from 182-273 Kg BW, and 0.4 mL above 273 Kg BW to provide about 5.1, 7.6 and 10.2 mg bST/d, respectively. At 341 Kg BW all heifers were fed the same diet and bST was discontinued. Average daily BW gains (ADGs) to 341 kg BW were 0.894, 0.972, 1.007 and 0.989 kg/d. Diet affected days to reach final BW, ADG and height at withers ($P < 0.01$). ADG of bST injected heifers was about 4.1% greater but no effects of bST were detected for other growth measures. No differences in number of inseminations, calving age, or BW and BCS at calving were detected due to diet, bST or SEA. Mean MY through 150 DIM were 29.8 (LL), 30.3 (LH), 29.9 (HL) and 29.5 (HH) kg/d; means did not differ (P=0.9475) and no effects of bST (29.7 vs. 30.0 kg/d; P= 0.6731) or bST*SEA interaction (P=0.4545) were detected. There was a significant effect of SEA on mean MY ($P< 0.0112$); heifers that calved in cooler months produced more milk (2.8 kg/d) but the two- and three-factor interactions among diet, SEA and bST were not significant. During lactation BW and BCS did not differ due to diet, ADG, or bST. Overall, no positive or negative effects of feeding diets with different protein to energy concentrations and ratios or injecting bST were detected on breeding or calving traits or on MY.$\leq leq$

Key Words: Heifers, Milk yield, Growth rates

W53 IGF binding protein-2 reduces the mitogenic effect of IGF-I, but not des-IGF-I, in MAC-T bovine mammary epithelial cells. B. E. Etchebarne* and M. J. VandeHaar, *Michigan State University.*

Insulin-like growth factor-I is a potent mitogen of mammary epithelial cells. The IGF binding proteins (IGFBP) alter IGF bioactivity and can affect mammary cells directly. Very little work has been done with IGFBP-2, although it is the major IGFBP synthesized by mammary epithelial cells in vitro. Our objective was to determine the effect of IGFBP-2 on proliferation of bovine mammary epithelial cells. The MAC-T bovine mammary cell line was used in this study with cells plated at 5000 cells/well in 24-well plates coated with collagen. Cells were first incubated with 10% FBS for 24 h, then serum-free media for 48 h, and then with treatments for 72 h with one change of media at 48 h. In one study, treatments were 0, 1, 10, or 100 ng of IGF-I/ml along with 0, 10, or 100 ng of IGFBP-2. In a second study, treatments were no IGF-I, 1 ng/ml IGF-I, 1 ng/ml des-IGF-I, or 0.5 ng/ml betacellulin along with 0, 10, or 100 ng IGFBP-2. Cell proliferation was assessed by measuring incorporation of 3H-thymidine included in the media for the final 2 h of treatment. Both IGF-I and des-IGF-I at 1 ng/ml increased 3H-thymidine incorporation to 300% that of cells in control media ($P < 0.01$) and higher doses caused no further stimulation. In cells treated with 1 ng of IGF-I, IGFBP-2 at 100 ng (25 molar ratio excess) abolished 75% of the mitogenic effect of IGF-I ($P < 0.01$), and IGFBP-2 at 10 ng decreased proliferation slightly. Neither 10 nor 100 ng of IGFBP-2 had any effect in cells treated with 10 or 100 ng of IGF-I. IGFBP-2 also did not alter cell proliferation in cells treated with 1 ng of des-IGF-I, indicating that binding of IGFBP-2 to IGF-I was critical for its inhibitory effect. IGFBP-2 caused only a small decrease in basal cell proliferation. Betacellulin increased cell proliferation 25% and was not inhibited by IGFBP-2. In conclusion, IGFBP-2 decreases the mitogenic effect of IGF-I in cultured mammary epithelial cells, and the likely explanation for this effect is that it binds IGF-I, thereby preventing its interaction with the IGF-I receptor.

Key Words: Mammary epithelial cells, IGF-I, IGFBP-2

W54 Changes in plasma leptin from birth to puberty in dairy cattle. S. S. Block*, J. M. Smith, R. A. Ehrhardt, M. C. Diaz, R. P. Rhoads, M. E. Van Amburgh, and Y. R. Boisclair, *Cornell University.*

Leptin is thought to play a critical role in regulating energy metabolism throughout mammalian life. In growing dairy cattle, plasma leptin has been proposed as a partial mediator of the effects of nutrition on reproductive and mammary development. However, the developmental stage at which the plane of nutrition increases plasma leptin has not been well defined. Further, it is unknown whether the onset of puberty is affected by plasma leptin concentration in dairy cattle. To investigate these questions, two studies were performed. In the first study, neonatal calves were fed a milk replacer at levels supporting an average daily gain of 570 g/d (LOW) or 1210 g/d (HIGH). Weekly blood samples were obtained until slaughter at 105 kg body weight. Plasma leptin and adiposity remained constant in the LOW calves, but started to increase by the third wk of age in the HIGH calves. In the second study, 3-5 mo old heifers were fed a TMR supplemented with either calcium salts of palm fat or conjugated linoleic acids at levels sustaining an average daily gain of approximately 1.0 kg/d. Blood samples were obtained until the third post-pubertal luteal phase. The fat source had no effects on growth parameters, body composition, age at puberty or plasma leptin. Therefore, plasma leptin was reanalyzed as a function of age from start of treatment until slaughter. The plasma concentration of leptin remained nearly constant at 2.3 ng/ml until 1 yr of age when a rise in plasma leptin became obvious. Puberty occurred with equal frequency either around 1 yr of age when plasma leptin was nearly constant or later when leptin was rising rapidly. We conclude that plasma leptin is regulated by nutrition in early postnatal life, but that a sudden increase in plasma leptin is not required for the onset of puberty in dairy cattle.

Key Words: Leptin, Cattle, Puberty

W55 Calf socialization, non-forage fiber supplementation and rumen development in white and pink veal production systems. C. W. Cruywagen*[1] and L. C. Hoffman, [1]*University of Stellenbosch, South Africa*.

Thirty Holstein calves were used to compare pink (early weaning) and white (all-milk) veal production systems. Treatments were included where calves were able to socialize, while the effect of a non-forage fiber supplement (malt pellets) on abnormal oral behavior and rumen development was also investigated in the white veal system. Treatments were (i) white veal, milk replacer only, individual housing (WMI), (ii) white veal, milk replacer plus malt pellets (200 g/d), individual housing (WSI), (iii) white veal, milk replacer plus malt pellets, group housing (WSG), (iv) pink veal, early weaning (5 weeks), individual housing (PEI) and (v) pink veal, early weaning, group housing (PEG). All the white veal calves received milk replacer at an increasing level (3 L per day at 4 d, 18 L per day at 18 weeks). All calves were slaughtered at 18 weeks of age. Treatment had no effect on final body weight (P=.56) or total body weight gain (P=.45). Final body weights were 159.7, 169.2, 157.5, 166.2 and 161.2 kg and total body weight gains were 116.3, 128.9, 114.9, 125.0 and 120.2 kg for WMI, WSI, WSG, PEI and PEG, respectively. Carcass weight (P<.01) and dressing percentage (P<.001) were significantly affected by treatment, favoring the white veal treatments. Carcass weights were 90.3, 97.0, 86.6, 80.3 and 82.0 kg and dressing percentages were 56.6, 57.4, 55.0, 48.3 and 50.9 % for WMI, WSI, WSG, PEI and PEG, respectively. No rumen papillae development was observed in the all-milk treatments. Papillae development was negligible in white veal calves receiving supplemental malt pellets, but supplementation appeared to decrease tongue rolling. In the pink veal treatments, calf starter and finishing rations had a significant effect on the development of rumen papillae. Papillae length in these treatments varied between 4.5 and 11 mm and papillae density between 34 and 82 papillae cm^{-2}. It was concluded that the ability to socialize had no beneficial effect on any of the measured parameters and that supplemental malt pellets to prevent abnormal oral behavior had no effect on rumen development.

Key Words: Calves, Veal production, Rumen development

W56 Glucose metabolism in neonatal calves: effects of glucocorticoids and dependence on colostrum feeding. S. N. Sauter, J. W. Blum, and H. M. Hammon*, *University of Berne, Berne, Switzerland*.

Plasma glucose concentrations in new-born calves are influenced by colostrum (C) feeding and by glucocorticoids. We have tested the hypothesis that a high glucocorticoid status after birth as well as C feeding influence glucose metabolism in association with an increase of hepatic expression and activities of phosphoenolpyruvate carboxykinase (PEPCK; EC 4.1.1.32) and pyruvate carboxylase (PC; EC 6.4.1.1), two key enzymes of the hepatic gluconeogenesis. Calves (n=7, each group) of $GrFD^-$ and $GrFD^+$ were fed a milk-based formula (F), whereas calves of $GrCD^-$ and $GrCD^+$ were fed C. Dexamethasone (DEXA; 30 μg/[kg body weight × d]) was injected to calves of $GrFD^+$ and $GrCD^+$. Calves were fed C or F for the first 3 d, milk replacer on d 4, and were euthanized on d 5 of life. On d 1, 2, 4 and 5, plasma concentrations of glucose and insulin were measured, and on d 5 mRNA concentrations and activities of PEPCK and PC were measured in liver. Plasma glucose concentrations were higher ($P < 0.01$) in DEXA-treated calves than in controls on d 1, 2, 4, and 5 and were higher ($P < 0.05$) in C-fed than in F-fed calves on d 4. Plasma insulin concentrations were higher ($P < 0.001$) in DEXA-treated than in non-treated calves with a greater DEXA effect in C-fed calves on d 4 and 5. Mitochondrial PEPCK mRNA was higher ($P < 0.05$) in C-fed than in F-fed calves, but cytosolic PEPCK mRNA showed no group differences. Expression of PC was lower ($P < 0.001$) in DEXA-treated than in non-treated calves and tended to be lower ($P < 0.1$) in C-fed than in F-fed calves. Activities of PEPCK on d 5 decreased ($P < 0.001$) after DEXA treatment. PEPCK activities were higher ($P < 0.1$) in $GrCD^-$ than in $GrFD^-$. Activities of hepatic PC were lower ($P < 0.1$) in DEXA treated than in non-treated calves. Elevated plasma glucose concentrations after DEXA treatment did not result from increased hepatic gluconeogenic activities, because DEXA did not stimulate hepatic gluconeogenic enzymes. However, C feeding increased glucose concentrations possibly in part due to elevated hepatic gluconeogenesis.

Key Words: Neonatal calf, Gluconeogenesis, Dexamethasone

W57 Effects of age and accelerated growth on circulating concentrations of β-carotene and vitamins A, E, and D in milk replacer-fed calves. M. R. Foote*[1], B. J. Nonnecke[2], M. A. Fowler[3], B. L. Miller[3], T. E. Johnson[3], D. C. Beitz[1], and R. L. Horst[2], [1]*Iowa State University, Ames, IA,* [2]*National Animal Disease Center, ARS, USDA, Ames, IA,* [3]*Land O'Lakes Inc., Webster City, IA*.

Effects of feeding intensified diets to neonatal calves on growth performance and protein utilization have been described. However, effects of accelerated growth on other nutritional parameters, including vitamin utilization have not been described. The current study evaluated effects of age and plane of nutrition on the plasma concentrations of β-carotene and vitamins D (25-hydroxyvitamin D_3), A (retinol), and E (RRR-α-tocopherol) in milk replacer-fed calves. Twenty-two Holstein bull calves were fed a standard (0.57 kg/d of a 22% CP, 20% fat milk replacer, n=11) or an intensified (1.14 kg/d of a 28% CP, 20% fat milk replacer, n=11) diet from 1 through 7 wk of age. Texturized calf starter was fed ad libitum to calves fed the intensified diet, but limit-fed to calves on the standard diet to target an average daily weight gain of 0.36 kg. Average daily weight gain of the intensified calves (0.58 kg) was greater ($P < 0.05$) than that of the standard calves (0.26 kg). For all calves, β-carotene, retinol, and RRR-α-tocopherol concentrations in plasma decreased markedly ($P < 0.05$) from wk 1 to wk 2 of the study. 25-Hydroxyvitamin D_3 concentrations increased ($P < 0.05$) from wk 1 to wk 2. Concentrations of 25-hydroxyvitamin D_3 in calves fed the standard diet, however, decreased after wk 6, and were lower ($P < 0.05$) than intensified calves by wk 8. Unlike calves on the standard diet, calves fed an intensified diet had decreased ($P < 0.05$) concentrations of retinol and RRR-α-tocopherol by wk 8. These results suggest that feeding an intensified diet during the neonatal period may increase the demand for retinol and RRR-α-tocopherol. These demands are likely associated with increased growth. These age and dietary related changes in vitamin status may impact maturation of neonatal immune function ultimately affecting the neonatal calf's susceptibility to infectious disease.

Key Words: Calves, Vitamins, Growth

W58 Cell proliferation, apoptosis and B- and T-lymphocyte numbers in gut-associated lymphoid tissue and thymus of neonatal calves: Effets of dexamethasone (DEXA) and colostrum feeding. J. Norrman*, C. W. David, S. N. Sauter, H. M. Hammon, and J. W. Blum, *University of Berne, Berne, Switzerland*.

Glucocorticoids influence immune reactions. We have tested whether an enhanced glucocorticoid status induced by DEXA influences proliferation, apoptosis and B- and T-lymphocyte numbers in Peyer's patches (PP) of ileum and thymus. Calves fed colostrum (C) or a formula (F) that contained no immunoglobulin G, hormones and growth factors. DEXA (30 micrograms/kg body weight × d for 4 d) was i.m. injected to $GrFD^+$ and $GrCD^+$. On first 3 d calves of $GrCD^{--}$ and $GrCD^+$ were fed C and of $GrFD^{--}$ and $GrFD^+$ were fed F. On d 4 all calves received a milk replacer. There were significant effects ($P < 0.05$) of DEXA treatment (decrease of cell proliferation rates in follicles of PP and thymus, increase of apoptotic rate in follicles of PP and thymus, decrease of B-lymphocyte numbers in follicles of PP, increase of B-lymphocyte numbers in domes of PP, increase of T-lymphocyte numbers in follicles of PP and decrease of intraepithelial T-lymphocyte numbers). There were significant effects ($P < 0.05$) of C feeding (decrease of cell proliferation rates in follicles of PP, of B-lymphocyte numbers in interfollicular areas, domes and follicular-associated epithelium of PP; increase of cell proliferation rate in thymus). A DEXA × feeding interaction ($P < 0.001$) was found on cell proliferation rate of the thymus. In conclusion, DEXA treatment decreased cell proliferation rates in follicles of PP and thymus and enhanced apoptotic rates in follicles of PP, but DEXA effects on B- and T-lymphocyte numbers in PP compartments were not uniform. C feeding decreased cell proliferation rates in follicles of PP and numbers of B-lymphocytes in domes, in follicular-associated epithelium and in interfollicular areas of PP, but enhanced cell proliferation rates in thymus. Furthermore, C feeding selectively modulated DEXA effects in the thymus.

Key Words: Immune system, Lymphocyte, Neonatal calf

W59 Growth hormone, insulin, and glucose responses to infusion of amino acids in developing dairy calves. C. C. Williams*, I. A. Norris, C. C. Stanley, L. R. Gentry, D. L. Thompson, Jr., H. G. Bateman, and D. T. Gantt, *Louisiana State University Agricultural Center, Baton Rouge, LA*.

Twenty-four female Holstein calves were randomly assigned to one of 4 treatments to evaluate the efficacy of amino acids as growth hormone (GH) or insulin secretagogues in neonatal dairy calves and to monitor the changes in these responses as calves undergo the transition to becoming functional ruminant animals. Treatments consisted of physiological saline (SAL); arginine (ARG, 0.5 g/kg BW); aspartic acid (ASP, 0.5 g/kg BW); and ornithine (ORN, 0.5 g/kg BW). Challenges were conducted at 1 month of age (prior to weaning) and again at 3 months of age. After an overnight period of feed deprivation, calves were fitted with indwelling jugular catheters, and approximately 1 hour later treatment solutions were infused. Samples of blood were collected via catheters at -30, -20, -10, 0, 10, 20, 30, 40, 50, 60, 75, 90, 105, and 120 min relative to onset of infusions for measurement of plasma GH. Samples collected at minutes -10 through 60 were analyzed for plasma glucose and insulin concentrations. In addition, baseline plasma samples obtained at 0 min were analyzed for thyroxine (T4), albumin, and urea nitrogen (PUN). An acute release of GH was induced ($P < 0.05$) by ASP in calves at 1 and 3 months of age. Peak concentrations of GH in response to ASP were greater ($P < 0.05$) in calves at 1 month of age. There was a treatment by time interaction ($P < 0.05$) in response to ARG and ORN for insulin concentrations, with increases observed 10 to 20 min post infusion. Consequently, glucose concentrations were decreased ($P < 0.05$) 30 min after infusion in calves infused with ARG and ORN. Baseline concentrations of PUN and albumin were similar for all calves ($P > 0.05$). Concentrations of T4 were lower ($P < 0.05$) in SAL treated calves, but no biological significance of this effect could be determined. These data indicate that ASP is effective in eliciting a GH response in young dairy calves, while ARG and ORN stimulate insulin release.

Key Words: Growth hormone, Insulin, Secretagogue

W60 Cell proliferation and apoptosis rates and B- and T-lymphocytes numbers in gut-associated lymphoid tissues, thymus, and lymphnodes of pre-term and full-term calves. C. W. David, J. Norrman, H. M. Hammon, and J. W. Blum*, *University of Berne, Berne, Switzerland*.

Morbidity and mortality due to insufficient immune functions are high in neonatal calves, especially if born pre-term. We have studied cell proliferation and apoptosis rates and B- and T-lymphocyte numbers in Peyer's patches (PP) of ileum, thymus, and mesenterial and prescapular lymphnodes (LM and LP) in unfed pre-term calves (GrP; born 13 d before normal term) on d 1 and in unfed full-term calves on d 1 (GrF) and on d 5 of life after feeding colostrum (C) for 3 d (GrC). In GrF compared with GrP there were higher ($P<0.05$) numbers of proliferating and apoptotic cells in interfollicular areas of PP, of T-lymphocytes in follicles and interfollicular areas of PP and within villus epithelia, of proliferating and apoptotic cells in LM and LP, of B-lymphocytes in paracortex and follicles of LM and LP, and of proliferating cells in cortex and medulla of thymus. In GrF compared with GrC there were higher ($P<0.05$) numbers of proliferating cells in follicles, interfollicular areas and domes of PP, but lower ($P<0.05$) numbers of apoptotic cells in follicles, interfollicular areas and domes of PP, and lower numbers of T-lymphocytes in follicles and interfollicular areas of PP and within villus epithelia. In thymus cortex and medulla numbers of proliferating cells were higher ($P<0.05$) in GrC than in GrF. In conclusion, studied lymphoid sites differed with respect to ontogenetic changes. Apoptotic rates were generally smaller at all sites of PP in GrC than in GrF and proliferation rates increased from GrP to GrF and from GrF to GrC in all tissues. Numbers of T-lymphocytes in PP were higher in GrF than in GrP, but lower in PP in GrC than in GrF, except in the domes. Numbers of B-lymphocytes did not change in PP despite of high proliferation and low apoptotic rates, suggesting that they leave PP during the first days of life. Interestingly, C feeding decreased T-lymphocyte numbers and increased apoptotic rates in PP.

Key Words: Immunology, Lymphocytes, Neonatal calf

W61 Effects of dexamethasone (DEXA) and growth hormone (ST) on glucose production in calves. H. M. Hammon*[1], J. W. Blum[1], and S. S. Donkin[2], [1]*University of Berne, Berne, Switzerland,* [2]*Purdue University, West Lafayette, IN*.

The hypothesis was tested that DEXA and ST increase glucose production in calves by stimulating hepatic gluconeogenesis and glycogenolysis. Calves (n=24) were randomly divided in 4 groups and were treated from d 3 to d 42 of life. CNTL received saline, DX was daily treated with DEXA (30 μg/kg body weight; Azium, Schering-Plough, Terre Haute, IN), GH was treated with 500 mg recombinant bovine ST (rbST; Posilac, Monsanto, St. Louis, MO) every 14 d, and DXGH was treated with DEXA and rbST; dosages were as in DX and GH. Blood samples (d 3, 7, 14, 28, 42) and liver samples (d 7, 14, 28, 42) were analyzed for glucose and insulin in blood plasma and mRNA and activities of phosphoenolpyruvate carboxykinase (PEPCK; EC 4.1.1.32) and pyruvate carboxylase (PC; EC 6.4.1.1) as well as glycogen content in liver. Glucose concentrations in DXGH were highest ($P < 0.01$) on d 14 and were higher ($P < 0.05$) in DXGH than in DX on d 42. Insulin concentrations in DXGH were higher ($P < 0.05$) than in CNTL on d 7 and were higher ($P < 0.05$) than in all other groups from d 14 to d 42. Insulin concentrations in DX were higher ($P < 0.05$) than in CNTL from d 7 to d 28 and were higher ($P < 0.05$) in DX than in GH on d 28. Expression of PEPCK was lower ($P < 0.05$) on d 7 and 28 in DX and DXGH than in CNTL and GH and on d 14 and 42 was lower or tended to be lower in CNTL than in DX ($P < 0.05$) and DXGH ($P < 0.1$). Expression of PC was lower ($P < 0.05$) on d 7 in DX and DXGH than in CNTL and GH and on d 14 tended to be lower ($P < 0.1$) in DXGH than in CNTL. Activities of PEPCK were higher ($P < 0.05$) on d 14 and tended to be higher on d 28 ($P < 0.1$) in DXGH than in CNTL and DX. PC activities on d 14 and 28 were lower ($P < 0.05$) in DX and DXGH than in CNTL and GH. Glycogen content in liver was reduced by DEXA and ST alone and in combination. The data indicate age-dependent expression of mRNA and activity of gluconeogenic enzymes and an age-dependent response to DEXA and the combination of DEXA and ST, but no response to ST alone.

Key Words: Glucocorticoids, Gluconeogenesis, Growth hormone

W62 The response of the somatotropic axis to growth hormone (ST) and dexamethasone (DEXA) in calves. H. M. Hammon*[1], H. Sauerwein[2], J. W. Blum[1], and S. S. Donkin[3], [1]*University of Berne, Berne, Switzerland,* [2]*Bonn University, Germany,* [3]*Purdue University, West Lafayette, IN*.

Glucocorticoids inhibit postnatal growth, but stimulate the somatotropic axis around birth. We have studied effects of DEXA treatment on the somatotropic axis and on the response of the somatotropic axis to ST. Calves (n=24) were randomly divided in 4 groups and were treated from d 3 to d 42 of life. CNTL received saline, DX was daily injected DEXA (30 μg/kg body weight [BW]; Azium, Schering-Plough, Terre Haute, IN), GH was treated with 500 mg recombinant bovine ST (rbST; Posilac, Monsanto, St. Louis, MO) every 14 d, and DXGH was treated with DEXA and rbST; dosages were as in DX and GH. Blood samples (d 3, 7, 14, 28, and 42) and liver biopsy samples (d 7, 14, 28, and 42) were analyzed for ST, insulin-like growth factor (IGF)-I, and IGF binding protein (IGFBP)-3 in blood (by RIA or EIA) and ST receptor (STR) and IGF-I mRNA in liver (by Northern blot). BW increased ($P < 0.05$) in CNTL and GH up to d 42 and in DX and DXGH up to d 28, but then decreased ($P < 0.05$) up to d 42. Plasma ST concentrations were highest ($P < 0.01$) in GH on d 7 and 14 and were higher ($P < 0.05$) in DXGH than in CNTL and DX on d 7. Plasma IGF-I concentrations in DXGH were higher ($P < 0.05$) on d 7 and 14 than in CNTL and DX and were higher ($P < 0.05$) on d 28 than in all other groups. IGF-I concentrations on d 42 were lowest ($P < 0.05$) in DX. Plasma IGFBP-3 concentrations were higher ($P < 0.05$) on d 7 in DXGH than in CNTL and on d 14 and 28 in DXGH than in all other groups. IGFBP-3 concentrations were higher ($P < 0.05$) on d 28 in GH than in DX and on d 42 were lowest ($P < 0.05$) in DX. STR mRNA increased ($P < 0.05$) on d 14 in DXGH and decreased ($P < 0.05$) on d 42 in DX and DXGH. IGF-I mRNA increased ($P < 0.05$) on d 7 and 14 in GH and DXGH and decreased ($P < 0.05$) on d 42 in DX and DXGH. In conclusion, DEXA depressed postnatal growth, but not before d 42. There was a weak response of the somatotropic axis to ST, but DEXA greatly enhanced the response of the somatotropic axis to ST.

Key Words: Veal calves, Glucocorticoids, Somatotropic axis

W63 Small intestinal and colon morphometry, epithelial cell proliferation, and absorptive capacity in neonatal calves fed milk-derived insulin-like growth factor-I (IGF-I) or a colostrum extract. B. Roffler[1], A. Fäh[1], S. N. Sauter[1], H. M. Hammon[1], P. Gallmann[2], G. Brem[3], and J. W. Blum*[1], [1]*University of Berne, Berne, Switzerland,* [2]*Swiss Federal Dairy Research Station, Liebefeld, Switzerland,* [3]*University of Vienna, Vienna, Austria*.

Concentrations of non-nutritional factors, such as insulin-like growth factor-I (IGF-I), in bovine colostrum (C) are high and can modulate neonatal intestinal development and function. In neonatal calves we have investigated effects on intestinal epithelial cell morphology, proliferation, apoptosis, and absorption of feeding milk-born human IGF-I (hIGF-I) or a bovine C extract. Calves were fed a milk-based formula containing amounts of nutrients comparable as in C for the first 3 d and a milk replacer from d 4 on. Formula and milk replacer contained only traces of non-nutritional factors such as IGF-I and insulin. In experiment 1, supraphysiological amounts of hIGF-I (3.8 mg/L formula; secreted by transgenic rabbits with their milk) were added to the formula. Xylose appearance in blood (after feeding xylose on d 5) and intestinal parameters (after euthanasia on d 8) did not differ between groups. In experiment 2, an extract of first-milked bovine C that provided physiological amounts of IGF-I (0.50, 0.15 and 0.09 mg IGF-I/L formula on d 1, 2, and 3, respectively and 0.09 mg IGF-I/L milk replacer on d 4) was added to formula or milk replacer. Plasma xylose concentration in the control group was transiently higher than in calves fed the C extract. On d 5 (after euthanasia) villus circumferences and heights in small intestine and epithelial cell proliferation rate in intestine were higher in calves fed the C extract than in controls. In conclusion, orally administered hIGF-I from transgenic rabbits had no effect on the intestinal tract. However, feeding a bovine C extract enhanced intestinal villus size, although it appeared to transiently decrease the absorptive capacity.

Key Words: Growth factors, Intestine, Neonatal calves

W64 Effect of a short-term fast on intestinal disaccharidase activity and villus morphology in piglets suckling insulin-like growth factor-I (IGF-I) transgenic sows. J. L. Hartke*, M. H. Monaco, M. B. Wheeler, and S. D. Donovan, *University of Illinois, Urbana, IL*.

We have shown that oral IGF-I increases lactase (LPH) activity in piglets compared to non-IGF-I treated pigs. Differences in LPH activity were greatest when piglets were killed in a post-absorptive state. Further, stable isotope tracer studies suggest that IGF-I up-regulates LPH activity by suppressing proteolytic degradation of LPH and its precursor (proLPHh). The current study was conducted using transgenic sows that over-express IGF-I in milk. We hypothesized that LPH activity would be maintained at a higher level in piglets suckling IGF-I transgenic sows (IGF-I) than piglets suckling non-transgenic sows (CON) following a short-term fast. Following farrowing, litters were normalized to 10 piglets. On d6, 30 piglets suckling IGF sows and 30 piglets CON sows were randomly assigned to 3 treatments: fed piglets (0h) remained with the sow until euthanasia; fasted piglets were removed from the sow 6 (6h) or 12 hours (12h) prior to euthanasia on d7. Serum IGF-I and IGF-I binding proteins (IGFBP) were measured. Intestinal weight, length, protein and DNA content, disaccharidase activity and villus morphology were assessed. Serum IGF-I did not differ between CON and IGF-I, but was lower at 12h compared to 0h ($p<0.05$). Serum IGFBP-4 was lower at 12h compared to 0h and IGFBP-1 was higher at 12h vs. 0h or 6h ($p<0.02$). No effects of IGF-I or fasting were noted for jejunal protein or DNA content. Jejunal villus height and width were greater at 6h and 12h compared to 0h ($p<0.05$). Crypt depth differed between all groups and increased over time ($p<0.05$). Disaccharidase activity was unaffected by fed state, however IGF-I piglets had greater jejunal LPH ($p<0.01$) and sucrase ($p=0.025$) activities. In summary, short-term fasting reduced serum IGF-I, but increased villus surface area. Piglets suckling IGF-I sows exhibited increased disaccharidase activity regardless of fed state. (Funded by the USDA CSREES under project NRICGP 00-35206).

Key Words: IGF-I, Fasting, Disaccharidase

W65 Temporal and spatial expression of MUC1 mRNA along the gastrointestinal tract. C. Liu*, A. K. Erickson, and D. R. Henning, *South Dakota State University, Brookings SD/USA*.

MUC1, a heavily glycosylated membrane-associated mucin, is a major component of milk fat globule membranes (MFGM). The role that MUC1 from MFGM plays in the milieu between mother and offspring is not well understood. One possible role for milk MUC1 may be to mimic intestinal MUC1 on the surface of the epithelial cells lining the gastrointestinal (GI) tract in order to block the binding of bacteria to host epithelial cells. To begin to evaluate this possibility, we needed to know the MUC1 expression pattern along the GI tract, especially its distribution in the neonatal GI tract. Consequently, the current study was designed to evaluate temporal and spatial expression of MUC1 mRNA along the porcine GI tract. We used a reverse-transcription polymerase chain reaction (RT-PCR) approach with primers based on conserved sequences between human and mouse MUC1 to obtain the sequence of a 603 bp segment of porcine MUC1 cDNA from porcine lactating mamary gland. Using these same primers, we developed a quantitative RT-PCR procedure, with normalized beta-actin mRNA expression as an internal control, to assess the level of expression of MUC1 mRNA in different sections of the GI tract (stomach, duodenum, jejunum, ileum, and colon) from pigs of different ages (1-day, 3-weeks, 6-weeks, and 6-months). Our results indicate that MUC1 mRNA was expressed in a tissue-specific manner in porcine GI tracts with high expression in the stomach, moderate expression in the duodenum and colon, and virtually undetectable expression in the jejunum and ileum. No obvious age-related difference in MUC1 mRNA expression was detected.

Key Words: Porcine MUC1, mRNA expression, Gastrointestinal tract

W66 Cloning and characterization of the bovine class 1 and class 2 insulin-like growth factor-I mRNA . Y. Wang*, S. E. Price, D. E. Eversole, and H. Jiang, *Virginia Polytechnic Institute & State University*.

Insulin-like growth factor-I (IGF-I) is an important regulator of growth, development, and metabolism, and is the primary mediator of the growth-promoting activity of growth hormone (GH). The IGF-I polypeptide has been indicated to be generated from two classes of IGF-I mRNA containing either exon 1 (class 1IGF-I mRNA) or exon 2 (class 2 IGF-I mRNA) as the leader exon in several species. The objective of this study was to identify class 1 and class 2 IGF-I mRNA in cattle and compare their expression in different tissues, at different developmental stages, and in response to GH, as well as their translatability. Three class 1 IGF-I complementary DNA (cDNA) corresponding to three different transcription start sites in exon 1 and one class 2 IGF-I cDNA were identified from adult cattle liver using $5'$ rapid amplification of cDNA ends ($5'$ RACE). The expression of these four IGF-I mRNA variants were further confirmed by ribonuclease protection assays (RPAs). The RPAs also revealed the presence of two additional class 1 and one additional class 2 IGF-I mRNA variants in bovine tissues. Both classes of IGF-I mRNA were expressed in all tissues examined, including adipose, brain, adrenal gland, heart, kidney, liver, lung, skeletal muscle, rumen, small intestine, pituitary, and spleen, with the highest level in liver and with class 1 being more abundant than class 2 IGF-I mRNA. The levels of both class 1 and class 2 IGF-I mRNA were higher in adult liver than in fetal liver ($P < 0.05$) and were coordinately increased in the liver of steers in response to GH administration ($P < 0.05$). In vitro translation analyses indicated that the luciferase reporter mRNA fused to a class 1 IGF-I $5'$-untranslated region ($5'$-UTR) was translated approximately four times efficiently as the luciferase reporter mRNA fused to a class 2 IGF-I $5'$-UTR. These results together suggest that as in several other species, IGF-I gene is also expressed as class 1 and class 2 transcripts in cattle, with class 1 IGF-I mRNA contributing more to the IGF-I polypeptide than class 2 IGF-I mRNA and that the expression of both classes of IGF-I mRNA is sensitive to developmental and hormonal (i.e. GH) factors.

Key Words: Cattle, Insulin like growth factor, $5'$ Untranslated region

W67 Effects of fasting on serum insulin-like growth factor I and liver insulin-like growth factor I and growth hormone receptor mRNA in cattle . Y. Wang, S. Eleswarapu, W. E. Beal, W. S. Swecker, R. M. Akers, and H. Jiang*, *Virginia Polytechnic Institute & State University*.

Nutritional deprivation decreases blood insulin-like growth factor I (IGF-I) concentrations in a variety of species. In this study we tried to understand the underlying mechanism by determining the effects of fasting on the levels of total IGF-I and total GHR mRNA, as well as the levels of individual IGF-I and GHR mRNA variants in the liver of young steers. Fasting for nearly three days decreased the levels of serum IGF-I by 63% ($P < 0.01$) and this decrease was associated with a 75% decrease ($P < 0.01$) in total IGF-I mRNA in the liver. Fasting-induced decrease in liver IGF-I mRNA was further found to be caused by an equal decrease in the levels of both class 1 and class 2 IGF-I mRNA. In addition to IGF-I mRNA, fasting also decreased the levels of total GHR mRNA in the liver ($P < 0.05$) and this decrease was associated with a decrease in the levels of GHR mRNA variants 1C3 ($P < 0.05$) and 1A ($P = 0.08$). Fasting did not affect the levels of two other major GHR mRNA variants, 1B and 1C2. These results together suggest the following mechanism for fasting-induced decrease in blood IGF-I: fasting decreases the levels of GHR mRNA variants 1C3 and 1A in the liver, thereby decreasing GHR number, thereby decreasing GH-induced expression of IGF-I mRNA, thereby decreasing IGF-I secretion from the liver, and thereby decreasing blood IGF-I.

Key Words: Cattle, Insulin like growth factor, Liver

W68 The bovine growth hormone receptor promoter 1 is positively regulated by hepatocyte nuclear factor 4γ via the same element for hepatocyte nuclear factor 4α. H. Jiang*[1], M. C. Lucy[2], and Q. Xu[1], [1]*Virginia Polytechnic Institute & State University*, [2]*University of Missouri*.

Transcription of growth hormone receptor (GHR) gene is directed by multiple promoters. One promoter, named GHR P1, is responsible for liver- and postnatal stage-specific expression of the GHR mRNA variant 1A. We previously found that the region between nucleotide -218 and nucleotide -151 (relative to the transcription start site) of GHR P1 plays a role in regulating the promoter activity, through interactions with a transcription factor named hepatocyte nuclear factor 4α (HNF-4α). Deoxyribonuclease I footprint analyses and electrophoretic mobility shift assays indicated that the -218/-151 region might bind additional transcription factors in the liver. The objective of this study was to identify these additional transcription factors. Using the yeast-one hybrid system with the -218/-151 region as bait, we have isolated dozens of putative clones from a bovine liver cDNA library. Nucleotide sequencing identified several of the clones as hepatocyte nuclear factor 4γ (HNF-4γ) in addition to HNF-4α. Sequence analyses indicated that HNF-4γ and HNF-4α were encoded by different genes. Electrophoretic mobility shift assays revealed that HNF-4γ bound to the same element consisting of direct repeats of GGTCA between nucleotide -196 and nucleotide -178, to which HNF-4α had been found to bind. Ribonuclease protection assays indicated that like HNF-4α, HNF-4γ mRNA was highly expressed in liver, absent in most tissues, and more abundant in adult liver than in fetal liver. Co-transfection analyses demonstrated that HNF-4γ was able to enhance the GHR P1 activity in the presence or absence of HNF-4α and that this enhancement was dependent on the GGTCA repeats in the -196/-178 region. These results together suggest that HNF-4γ is another transcription factor for the liver- and postnatal stage-specific GHR P1, which positively regulate the GHR P1 activity via the same element for HNF-4α.

Key Words: Transcription factor, Growth hormone receptor, Liver

W69 Gender differences in serum insulin-like growth factor (IGF)-I and IGF binding proteins in eight exotic species. K.E. Govoni*, D. Goodman, R.M. Maclure, and S.A. Zinn, *University of Connecticut, Storrs, CT*.

The somatotropic axis is important in the regulation of growth. Increased concentrations of IGF-I and IGF binding protein (BP)-3 and decreased concentrations of IGFBP-2 are associated with increased growth rates in cattle and swine, however limited experiments have been done to examine the somatotropic axis in exotic species. The overall objective of this experiment was to determine serum concentrations of IGF-I, IGFBP-2 and IGFBP-3 in eight different exotic species. Serum samples were collected from male (M) and female (F) Java Banteng (5M; 3F), Bongo (5F; 3M), Addra Gazelle (4M; 4F), Giant Eland (6M; 2F), Nile Lechwe (5M; 3F), Roan Antelope (4M; 4F) and White Rhinoceros (4M; 4F). Blood samples were collected at two different time points, from each animal. At each time point, on average, F were older than M for all species except Nile Lechwe and White Rhinoceros. In addition, one sample was collected from eight (5M; 3F) Asian Elephants. Concentrations of IGF-I were determined by RIA and concentrations of IGFBP-3 and -2 were determined by Western Ligand Blot. Concentrations of IGF-I, IGFBP-3 and IGFBP-2 were detectable in all species. Average concentrations of IGF-I, IGFBP-3 and IGFBP-2, for all species, range from 17 to 442 ng/mL, 17 to 178 arbitrary units (AU) and 10 to 61 AU, respectively. In general, average concentrations of IGF-I and IGFBP-3 were greater in M and concentrations of IGFBP-2 were greater in F. Concentrations of IGF-I were greater in M than F ($P < 0.05$) in Java Banteng and in Nile Lechwe. There was a trend for greater concentrations in M than F ($P < 0.10$) in Bongo, Roan Antelope and White Rhinoceros. Concentrations of IGF-I increased with age in Java Banteng ($P = 0.08$) and in M Nile Lechwe ($P < 0.05$) and decreased in White Rhinoceros ($P = 0.07$) and F Nile Lechwe ($P < 0.05$). Concentrations of IGFBP-3 in Java Banteng were greater in M than F ($P < 0.01$) and increased with age ($P < 0.01$). Concentrations of IGFBP-2 were greater in F than M in Elephants ($P < 0.05$) and in Roan Antelope ($P = 0.08$). Although relatively few samples were collected, gender and age differences were observed, in some of the species, which parallel differences observed in domestic species.

Key Words: Insulin-like growth factor binding proteins, Insulin-like growth factor-I, Exotic species

Meat Science & Muscle Biology: Manipulation of Meat Quality

W70 Antioxidant effects of rosemary extract and whey powder on the oxidative stability of wiener sausages during 10 months frozen storage. S. A. Coronado[1], F. R. Dunshea[2], and N. P. Shah[1], [1]*Victoria University, Melbourne, Australia*, [2]*Victorian Institute of Animal Science, Werribee, Australia*.

Lipid oxidation is a major problem encountered in meat processing. Fishmeal is added directly to pig feed in order to provide protein or energy and to increase dietary vitamin A and D. However, high levels of fish oil render the animal fat more prone to oxidation while introducing fishy odors into the meat product. The aim of this study was to investigate the stability of wiener sausages prepared from pork obtained from pigs fed diets containing vitamin E (10 or 200 mg α-tocopheryl acetate per kg feed) and fish-meal (0 or 5%) and manufactured with or without an antioxidant (0.03% rosemary extract or 2.5% sweet whey). Twelve (Large White x Landrace) gilts were randomly allotted to four dietary treatments containing two levels of vitamin E (10 or 200 mg/kg) and two levels of fish meal (0 or 5%) using a 2 x 2 factorial design. Wiener sausages were manufactured from meat obtained from animals after slaughter and stored for 5 days at 4°C with or without antioxidants. The oxidative stability of the wieners was examined over ten months of frozen storage. Lipid oxidation in the product was measured by means of thiobarbituric acid reactive substances (TBARS) and fluorescence shift. Sensory evaluation of the product to detect oxidative changes was also carried out. No lipid oxidation as measured by TBARS, fluorescence shift and sensory analysis was observed in wieners stored at -20°C for ten months. The oxidative stability of wieners was unaffected ($P > 0.05$) by dietary treatments or by the addition of antioxidants. Dietary vitamin E lowered TBARS values and helped retard lipid oxidation.

Key Words: Antioxidant, Oxidation, Wiener