

was close to the expected theoretical value of 96% serum protein for the process.

Key Words: Microfiltration, Diafiltration, Milk

764 Isolation and identification of *Micrococcus* spp from Egyptian soft cheese. A. S. Zahran*, *Minia University.*

Micrococci are the predominant bacteria of milk drawn aseptically from the udder. They were also isolated from different cheese varieties. These bacteria constitute a major portion of the secondary cheese microflora and contribute to its flavour through their proteolytic, lipolytic and esterolytic enzymes activities. Fifty strains of *Micrococcus* spp were isolated from Egyptian soft cheese (Domiat). They were identified into four different species as follows *M. varians*, *M. roseus*, *M. sedentarius* and *M. luteus*. The species most often isolated was *M. varians* as it represents 55% of the total isolates, strain *M. varians* DC6 was very active producer of extracellular proteinase and lipase. Immobilized cell culture is a widely used technique for achieving high volumetric efficiency and sustained productivity from microorganisms. The entrapment of bacterial cells in 2% agar substantially improved their enzyme activity. Immobilized cells produced from 40-50% more enzymes than free cells. The agar entrapment method appeared to have no adverse effects on the activity of the cells. The agar beads were structurally stable over five usage cycles. Synthesis of the extracellular enzymes by *M. varians* DC6 appeared to be inducible as no enzymes were detected in the absence of organic nitrogen.

Key Words: *Micrococcus*, Egyptian Soft Cheese, Immobilized Cells

765 Use of exopolysaccharides producing lactic acid bacteria for the production of buffalo milk dahi (yogurt). N. Pandya*¹, S. Kanawjia¹, and R. Dave², ¹*Dairy Technology Department, National Dairy Research Institute, Karnal, India,* ²*Dairy Science Department, South Dakota State University, Brookings.*

Exopolysaccharides (EPS) produced by lactic acid bacteria (LAB) have generated increasing attention among researchers and considered novel and safe food additives. In this experiment, two EPS⁺ cultures (*Lactococcus lactis* subsp. *lactis* NCDC 191 & mixed thermophilic strains NCDC 260) in combination with standard dahi culture (mixed mesophilic strains NCDC 167, EPS⁻) were studied for their effects on incubation pattern, rheology and sensory properties of the dahi made from buffalo milk standardized at 4.5% fat and 9.5% SNF. Both cultures were inoculated at 2% (v/v) rate in three different ratios (1:1, 2:1 and 3:1 of EPS⁻ and EPS⁺ cultures respectively) and incubated at 27 and 32°C for NCDC 191 and NCDC 260, respectively. It was observed that the rate of drop in pH and rate of increase in titratable acidity declined with increasing proportion of EPS⁺ cultures. Rheology and overall sensory properties were improved with increasing ratio of EPS⁺ culture. However, the improvement was significant ($P < 0.05$) only with NCDC 260, but not with NCDC 191. With the increasing proportion of NCDC 260, the viscosity increased from 0.384 to 0.596 Pa.s., the curd tension increased from 35.52 to 46.60 g, and the syneresis reduced from 1.48 to 0.33 ml per 10 g of dahi. The flavor scores decreased significantly ($P < 0.05$) with increasing ratio of NCDC 191 culture whereas, effect of NCDC 260 culture on flavor scores was not significant ($P < 0.05$). The body and texture scores improved up to 2:1 ratio (NCDC 167: NCDC 260) but at a higher ratio of EPS⁺ culture the dahi developedropy consistency and resulted into decline in sensory scores. The appearance scores were improved for both EPS⁺ cultures. A combination of NCDC 167 and NCDC 260 with (2:1) ratio was suggested for commercial production of dahi.

Key Words: Exopolysaccharides, Buffalo Milk, Dahi

ASAS - Growth and Development II

766 Influence of dietary protein and lactose levels on protein synthesis and translation initiation factor activation in neonatal pigs. J. W. Frank*¹, J. Escobar¹, A. Suryawan¹, H. V. Nguyen¹, C. W. Liu¹, S. R. Kimball², L. S. Jefferson², and T. A. Davis¹, ¹*USDA/ARS Children's Nutrition Research Center, Baylor College of Medicine, Houston, TX,* ²*College of Medicine, The Pennsylvania State University, Hershey.*

Parenteral infusion of insulin (INS) and amino acids increases protein synthesis (PS) and eukaryotic translation initiation factor (eIF) activation in skeletal muscle and liver. Pigs (N = 25; BW = 1.6 kg) were enterally fed isocaloric milk diets with three levels of protein (5, 10, and 15 g/kg/d) and two levels of lactose (11 and 23 g/kg/d) from 1 to 7 d of age. On d 7, pigs were gavage fed after a 4 h fast and blood samples were collected every 30 min for 1.5 h. Pigs were then euthanized and tissues harvested. Daily gain and PS in the longissimus dorsi and gastrocnemius muscles and liver were not influenced by lactose level, but increased with dietary protein and plateaued at the 10 g/kg/d level ($P < 0.01$). Plasma INS was greater in the high lactose fed pigs ($P < 0.01$) and lower in pigs fed the lowest protein diet ($P < 0.001$). Plasma branched-chain amino acids were influenced by dietary protein level ($P < 0.001$). Liver and muscle protein kinase B phosphorylation was greater in the high lactose fed pigs ($P < 0.05$). Liver and muscle ribosomal protein S6 kinase and liver 4E-BP1 phosphorylation increased with dietary protein and plateaued at the 10 g/kg/d level ($P < 0.01$), while muscle 4E-BP1 phosphorylation continued to increase to the highest protein level ($P < 0.001$). The association of eIF4G to eIF4E increased with dietary protein level ($P < 0.05$) and was not influenced by lactose level. The results suggest that growth and PS in neonatal pigs are influenced by dietary protein intake. These changes involve modulation of the availability of eIF4E for eIF4F complex assembly and may be mediated by plasma insulin and amino acid levels. (NIAMS AR 44474, USDA 58-6250-6-001)

Key Words: Pigs, Protein Synthesis, Translation Initiation

767 Infusion of a physiological dose of leucine stimulates muscle protein synthesis in neonatal pigs by enhancing the activity of translation initiation factors. J. Escobar*¹, J. W. Frank¹, S. R. Kimball², A. Suryawan¹, H. V. Nguyen¹, C. W. Liu¹, L. S. Jefferson², and T. A. Davis¹, ¹*USDA/ARS, Children's Nutrition Research Center, Baylor College of Medicine, Houston, TX,* ²*Cellular and Molecular Physiology, College of Medicine, Pennsylvania State University, Hershey.*

In adult rats, skeletal muscle protein synthesis increases in response to pharmacological doses of leucine (Leu) administered orally. The effect of a physiological rise in plasma Leu on skeletal muscle protein synthesis has not been investigated in neonatal pigs, which are highly sensitive to amino acid and insulin stimulation. Thus, 24 crossbred pigs were food-deprived for 12 h and intra-arterially infused with Leu (0 or 400 mmolkg⁻¹h⁻¹). Protein synthesis was measured after 60 or 120 min in liver, and longissimus dorsi and gastrocnemius muscles. Infusion of Leu increased ($P < 0.01$) plasma Leu 2.5- to 3.4-fold while plasma insulin and glucose were unchanged. Infusing Leu for 120 min, but not for 60 min, reduced ($P < 0.05$) plasma essential amino acids levels. Infusing Leu for 60 and 120 min increased ($P < 0.05$) phosphorylation of eukaryotic initiation factor (eIF) 4E binding protein-1 (4E-BP1), ribosomal protein (rp) S6 kinase (S6K1), and rpS6, and decreased the amount of eIF4E associated with its repressor, 4E-BP1, in longissimus dorsi muscle. In liver, phosphorylation of 4E-BP1, S6K1 and rpS6, as well as eIF4E associated with 4E-BP1 were not affected by Leu infusion. Leucine infusion for 60 min increased protein synthesis in longissimus dorsi (38%, $P = 0.04$) and gastrocnemius (67%, $P = 0.005$) muscles, but not in liver ($P = 0.11$). Leucine infusion for 120 min did not increase protein synthesis in skeletal muscle and reduced protein synthesis in liver (25%, $P < 0.01$). Thus, a physiological increase in plasma Leu stimulates protein synthesis in skeletal muscle of neonatal pigs by increasing eIF4E availability for eIF4F assembly. Moreover, this response appears to be insulin-independent, substrate-dependent, and tissue-specific (NIAMS AR 44474, USDA58-6250-6-001).

Key Words: Leucine, Translation Initiation Factors, Protein Synthesis

768 Body protein deposition response following sudden changes in ideal protein intake differs between pig types. H. R. Martínez* and C. F. M. de Lange, *Department of Animal and Poultry Science, University of Guelph, ON, Canada.*

The objective of this study was to evaluate the extent and dynamics of compensatory growth following a period of lysine intake restriction in two pig types. In Exp. 1, 40 Yorkshire barrows (14.4 ± 1.6 BW) were assigned to one of two diets (control, - 50% lysine) and fed restricted at 75% of voluntary daily DE intake according to NRC (1998; % of NRC) from 15 to 35 kg BW. Thereafter, pigs were fed restricted (75% of NRC) or ad libitum diets that were not limiting in ideal protein. In Exp 2, 57 Yorkshire entire male pigs (15.8 ± 0.9 BW) were assigned to one of three diets (control, -30% or - 45% lysine) and fed restricted (90% of NRC) from 15 to 38 kg BW; thereafter they were fed at 90% of NRC diets not limiting in ideal protein. BW gain and body composition (serial slaughter) was monitored for at least 30 days after lysine intake restriction was removed. Lysine intake restrictions reduced growth rate ($P < 0.01$; 533 vs 410 g/d in Exp. 1; 794 vs 672 and 648 g/d in Exp. 2). Based on BW gain, no compensatory growth (CG) occurred in barrows ($P = 0.95$) due to previous lysine intake restriction; there were no interactive effects of feeding regime and previous diet lysine level on BW gain ($P = 0.74$). In entire males at 38 kg BW, lysine intake restriction tended to increase body lipid to protein ratios (L/Pr; 0.83, 0.95, 1.06; SE 0.08, $n = 2$). Entire males showed full CG. The BW gain was inversely related to previous diet lysine levels between 38 and 53 kg (1104, 1152, 1210 g/d; $P < 0.03$) and between 38 and 110 kg BW (1100, 1166, 1180 g/d; $P < 0.01$). Body composition (L/Pr; 1.07, 1.12, 1.12; SE 0.05) and carcass characteristics (backfat 20.0, 20.4, 20.7 mm, SE 1.79; loin area, 4596, 4579, 4557 mm², SE 136; colour; carcass protein/body protein mass, 1.16, 1.19, 1.19, SE 0.06) in entire males at 110 kg BW were not influenced ($P > 0.10$) by diet lysine level between 15 and 38 kg BW. CG is more likely to occur in growing pigs with higher lean tissue growth potentials.

Key Words: Compensatory Growth, Body Composition, Pig Type

769 Human somatotropin is more efficacious than porcine somatotropin in growing pigs. F. R. Dunshie*, *Department of Primary Industries, Werribee, Victoria, Australia.*

While the improvements in productive efficiency in response to daily porcine somatotropin (pST) injection are well documented, there is some evidence that human ST (hST) may be even more efficacious than pST. If the relative differences in efficacy between the two ST's is great enough, then variants of human somatotropin may be worth pursuing as a daily injectable for pigs. Forty two individually-penned crossbred gilts (60.4 kg) were stratified on live weight into 14 blocks and then one pig from each block was then randomly assigned to a treatment group consisting of either 7 doses of pST (0, 1.0, 2.0, 3.0, 4.0, 5.0 and 6.0 mg/day) and 7 doses of hST (0, .2, .4, .6, .8, 1.0 and 1.2 mg/day) for 7 days. The regression equation relating feed conversion ratio (FCR) to dose of pST and hST was described by the following equation (with standard errors in brackets): $FCR = 2.4976 (.0817) - .1279 (.259) \times pST \text{ dose} - .409 (.130) \times hST \text{ dose}$. Thus, the FCR for control animals was 2.50, and decreased by .128 and .409 for every mg/d increase in dose of pST and hST, respectively. Therefore, hST was approximately 3.2x more effective in decreasing FCR than pST. While the effects of ST on plasma urea nitrogen (PUN) could not be as easily explained by regression techniques, both pST and hST significantly ($P < .001$) reduced PUN. Application of pST at 5x the dose of hST was more effective in reducing PUN, indicating that the relative difference in potency was less than 5x. Therefore, it appears that hST is approximately 3x more effective in pigs than pST, at least in the short term.

Key Words: Growth, Swine, Growth Hormone

770 Concentrations of insulin-like growth factors I and II in embryonic fluids of chickens, ducks, and turkeys. D. M. Karcher*¹, J. P. McMurtry², and T. J. Applegate¹, ¹*Animal Sciences, Purdue University, West Lafayette, IN,* ²*USDA/ARS/GBL, Beltsville, MD.*

Insulin-like growth factors (IGF-I, IGF-II) are present in the amniotic fluid of developing mammals and ingestion of the amniotic fluid has been

shown to potentially impact the proper development of the gastrointestinal tract. To investigate IGF-I and II changes in avian embryo fluids, 250 duck eggs, 200 turkey eggs, and 222 chicken eggs were incubated, and the embryos were staged according to Hamburger and Hamiltons classification (1951). Amniotic and allantoic fluid IGF-I and -II concentrations were determined by a chicken radioimmunoassay which has 100% reactivity between all species. When the beak was prominent on the embryo, stage 29, the mean IGF-I concentration (conc.) of the chicken amniotic fluid was 97.9% greater than the mean turkey conc. and 81.5% greater than the mean duck conc. ($P \leq 0.05$). At stage 40, when the external features are complete, the chicken amniotic fluid IGF-I conc. was 34.6% greater in the duck amniotic fluid and 17.5% greater than in turkey amniotic fluid ($P \leq 0.05$). However, at stage 37 and 38 (eye-lids narrow and leg scales appear) the IGF-I conc. in turkey amniotic fluid was 70-80% higher compared to the chicken or duck conc. ($P \leq 0.05$). Within species, amniotic IGF-I conc. were low at stage 29 with maximum conc. occurring at the time of imbibing (stage 42 duck and turkey; stage 44 for chicken). The amniotic IGF-I conc. in the chicken had a cubic relationship ($r^2 = 0.41$; $P = 0.0003$) with a 95% difference ($P \leq 0.05$) between the lowest and highest conc. Duck amniotic IGF-I had a linear relationship ($r^2 = 0.73$; $P < 0.0001$) with a 94% ($P \leq 0.05$) difference observed between highest and lowest conc. The IGF-I conc. in the turkey amniotic fluid followed a similar trend with a quadratic relationship ($r^2 = 0.43$; $P = 0.0006$) and an observed difference in minimum and maximum conc. of 98% ($P \leq 0.05$). Amniotic fluid IGF-II was not significantly ($P > 0.05$) different across species. Overall, the conc. of IGF-I and II varies across and within species.

Key Words: Insulin-Like Growth Factors, Embryonic Fluids, Duck

771 Selection for growth does not alter jejunal glucose absorption and energy metabolism in mice. Y. K. Fan¹, W. J. Croom, Jr.*², I. L. Taylor³, L. R. Daniel², A. R. Bird⁴, B. W. McBride⁵, V. L. Christensen², and E. J. Eisen², ¹*National Chung Hsing University,* ²*North Carolina State University, Raleigh,* ³*Tulane University, New Orleans, LA,* ⁴*CSIRO Health Sciences and Nutrition, Australia,* ⁵*University of Guelph, ON, Canada.*

The present study was designed to investigate the effects of genotype and sodium monensin (NaM) on whole body energetics and jejunal function (JF) using 4 different genetic lines of mice. The lines were M16 (selected for rapid growth), randomly bred controls (ICR) and their reciprocal crosses (M16xICR and ICRxM16). Eight-week-old mice from each line were administered either NaM (20 μ M) or excipient via the drinking water for 14d ($n=6$ mice per cell). Whole-body O₂ consumption was measured on day 11. On day 14, mice were euthanized and the jejunum dissected for measurement of jejunal protein and DNA, total, serosal and mucosal O₂ consumption, jejunal uptake rate and whole jejunal glucose uptake. The apparent energetic efficiency (APEE) of glucose uptake was calculated. M16 mice were larger, had greater feed and water intake and greater body fat %. Selection for growth had no effect on intestinal weight adjusted for fasted body weight, increased SI density (mg/cm) and decreased adjusted SI length ($P < .01$). M16 mice had greater intestinal villus width, crypt depth and enterocyte height ($P < .05$) but decreased villus height/crypt depth ($P < .05$). No changes were noted in serosal or mucosal O₂ consumption. M16 mice had lower jejunal glucose uptake rates, total jejunal glucose absorption adjusted for BW ($P < .01$). APEE of glucose uptake decreased 51% in M16 compared to ICR ($P < .05$). Reciprocal crosses (ICRxM16 vs. M16xICR) had few effects on physiological parameters measured as compared to ICR. NaM decreased feed efficiency (mg gain/g feed) and daily water intake across all lines ($P < .01$) but had no effect on adjusted whole-body O₂ consumption. Jejunal protein/DNA decreased as well as jejunal villus width ($P < .05$) with NaM. Selection for growth resulted in less glucose absorption and efficiency of glucose transport. This supports the existing hypothesis that selection for growth does not inevitably result in a concomitant increase in jejunal function or APEE of nutrient absorption.

Key Words: Genotype, Jejunal Absorption, Monensin

772 Effect of microbial colonisation on activated caspase-3 protein and gene expression in the gnotobiotic pig small intestine. B. P. Willing*, R. H. Siggers, T. W. Shirkey, B. G. Goldade, J. K. Marshall, B. Laarveld, and A. G. Van Kessel, *Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, SK, Canada.*

Previous gnotobiotic pig experiments have shown dramatic differences in ileal morphology in germ-free (GF) and mono-associated as compared to conventionalized pigs, most prominently characterized by as much as a 2-fold increase in villus length. In three experiments sixteen piglets were derived by caesarian-section, allocated to one of four treatment groups including GF, mono-associated with non-pathogenic *Escherichia coli* (EC) or *Lactobacillus fermentum* (LF) or conventionalized with sow feces (CV) and reared to 13 (exp. 1) or 14 (exp. 2&3) d of age. In experiment 3 the LF group was contaminated with *Klebsiella pneumoniae* making it di-associated (LFKP). In experiment 1, whole intestinal tissue was collected at 75% of the small intestine (SI) length. In experiments 2 and 3, villus tip epithelial cells were harvested from 80 cm lengths of SI with 1.5mM EDTA using the distended sac method. To characterize the morphological differences in relation to apoptosis, activated caspase-3 protein and caspase-3 gene expression were measured by Western blot and quantitative PCR (qPCR). Activated caspase-3 protein was detectable in whole intestinal tissue in CV animals only. In the same tissue, caspase-3 mRNA abundance was similar among CV, LF and EC but reduced 1.8 fold in GF ($P < 0.05$). In villus tip cells there was a slight trend for reduced caspase-3 expression in GF, EC, LF and LFKP treatment groups relative to CV. Results indicate that analysis of both transcript and activated protein abundance can yield complementary information regarding apoptotic activity. Apoptotic activity appeared lower in GF and mono-associated pigs however, relative differences in gene expression in whole tissues versus villus tip epithelial cells suggest that markedly increased cellular infiltration of lamina propria and/or development of peyers patches in CV pigs may contribute significantly to apoptotic activity observed when whole intestinal tissue is analyzed.

Key Words: Gnotobiotic, Pig, Caspase-3

773 The effect of microbial colonisation on disaccharidase activity in the gnotobiotic pig. L. M. Williams, B. P. Willing*, R. H. Siggers, T. W. Shirkey, B. G. Goldade, J. K. Marshall, and A. G. Van Kessel, *Department of Animal and Poultry Science, University of Saskatchewan, Saskatoon, SK, Canada.*

In previous gnotobiotic pig research we have found that germ free and mono-associated pigs had up to 2-fold longer villi than conventional pigs, chiefly in the distal small intestine (SI). Enterocytes along the distal villi of mono-associated and germ free pigs contained large and apparently empty vacuoles. To characterize the effect of specific bacteria on digestive function in relation to intestinal morphology, brush border disaccharidase activity was determined in SI segments collected from pigs in two separate experiments. Each experiment included 16 pigs derived by caesarian-section and assigned to, germ free (GF), mono-associated with non-pathogenic *Escherichia coli* (EC), mono-associated with *Lactobacillus fermentum* (LF) or conventionalised with sow feces (CV), treatment groups. In experiment 2 the germ free group was contaminated with a *Staphylococcus* sp. (ST). Pigs were fed a 2:1, Similac®.water, (v/v) mixture *ad libitum* and killed at 13 days of age. Lactase, sucrase and maltase activities were determined per unit protein, DNA and wet tissue weight, for segments collected at 25 and 75% of SI length. The effect of treatment and SI location on specific disaccharidase activities were similar whether reported per unit protein, DNA or wet tissue weight, were always higher in the proximal SI, were similar among GF and monoassociated pigs and were always lowest in CV pigs. Lactase activity was highest in all treatment groups at both locations, however, this was most notable (2-fold higher versus maltase and sucrase) in distal SI of mono-associated and GF pigs. In conclusion the vacuolated enterocytes, associated with markedly elongated villi observed in the distal small intestine, of GF and monoassociated pigs demonstrated significant disaccharidase activity, however the pattern of activity was consistent with the very early postnatal period and slow enterocyte turnover.

Key Words: Gnotobiotic, Disaccharidase, Pig

774 Characterization of specific gene expression following differentiation of bovine stromal-vascular cells into adipocytes. M. J. Herson*¹, J. W. Ross², D. Stein², L. McBeth², R. D. Geisert², U. Desilva², C. R. Krebbiel², and J. R. Malayer², ¹University of Florida, Gainesville, ²Oklahoma State University, Stillwater.

The objectives of the present experiment were to 1) determine if gene expression differs during adipocyte differentiation of bovine stromal-vascular (SV) cells from different adipose depots and 2) determine similarity of differentiation of cultured bovine SV to the mouse 3T3-L1 cell line. Bovine SV cells were isolated from s.c., i.m., and kidney-pelvic-heart (KPH) adipose depots, cultured to confluence, then stimulated to differentiate for 0, 12, 24, 48, and 72 h. Expression of bovine specific peroxisome proliferators-activated receptor- γ (PPAR γ), stearoyl CoA desaturase 1 (SCD-1), lipoprotein lipase (LPL), fatty acid synthase (FAS), and acyl-CoA synthetase (ACS) mRNA was determined using quantitative RT-PCR. 18S ribosomal RNA was utilized to standardize RNA loading. Relative quantitative mRNA expression was evaluated using the comparative CT method. PPAR γ gene expression of s.c. adipocytes exhibited a 7.5-fold increase from 12 to 48 h compared to 3-fold and 3.8-fold increase in i.m. and KPH from 6 to 12 or 24 h (D x T, $P < 0.001$). SCD-1 gene expression in s.c. increased 7-fold from 0 to 72 h compared to 1.8 and 2.7-fold increases for i.m. or KPH (D x T, $P = 0.04$). Gene expression of LPL in s.c. increased 8-fold by 48 h compared to 2- and 3-fold increases in KPH and i.m. (D x T, $P < 0.001$). FAS (D x T, $P < 0.001$) and ACS (D x T, $P = 0.002$) gene expression decreased 4.9 and 4.6-fold in i.m. and 3.4 and 3.6-fold in KPH, s.c. gene expressions decreased 1.7 and 1.2-fold, respectively. 3T3-L1 PPAR γ gene expression was minimal at 6 h and was 16 and 223-fold greater ($P < 0.001$) at 24 and 48 h. Differences in gene expression exist among bovine adipose depots. A similarity in response time but not magnitude of PPAR γ in bovine s.c. and 3T3-L1 cells indicates that bovine SV cells are an acceptable model for adipocyte differentiation. Differences between mouse and bovine cells and among depots indicate the physiological importance of adipocyte specificity.

Key Words: Adipose, Bovine, Gene Expression

775 Dexamethasone downregulates glucocorticoid receptor expression in cultured bovine preadipocytes. G. Ortiz-Colón*, A. C. Grant, J. L. Burton, M. E. Doumit, and D. D. Buskirk, *Department of Animal Science, Michigan State University, East Lansing.*

Bovine adipose tissue from different depots have been shown to have distinct physiological responses to glucocorticoids. The objectives of this study were to determine if the glucocorticoid receptor (GR) was expressed in cultured preadipocytes isolated from bovine adipose tissues, and to evaluate the effect of dexamethasone treatment upon the GR abundance. Preadipocytes were isolated from intramuscular (i.m.), perirenal (p.r.) and subcutaneous (s.c.) adipose tissues of an Angus steer (556 Kg, 13.5 mo. old), propagated in culture, and seeded in 35 mm-diameter dishes at a density of 2,600 cells/cm². Preadipocytes were fed every other day with growth media: Dulbeccos modified Eagles medium (DMEM) containing 10% fetal bovine serum. After 8 days, cells were exposed to control growth media, or growth media containing .25 μ M dexamethasone for 48 h. The cells were then solubilized by the addition of hot (95°C) electrophoresis sample buffer. Proteins were electrophoretically separated using 10% polyacrylamide gels and transferred onto polyvinylidene fluoride microporous membranes. The membranes were incubated with an antibody against GR and subsequently exposed to a secondary antibody conjugated to alkaline phosphatase. Immunoreactive bands were visualized upon addition of substrate. The GR was detected as a specific 97 kDa band in the cultured preadipocytes from the three adipose tissues. An antibody against β -actin was used as an internal control. Bands were quantified using Quantity One software (Bio-Rad) and data were analyzed by the GLM procedure of SAS. Glucocorticoid receptor abundance was reduced by dexamethasone exposure in i.m. (7.0%), s.c. (52.7%), and p.r. (35.6%) preadipocytes ($P < .03$). It can be concluded that GR was expressed in these cultured preadipocytes isolated from bovine s.c., p.r., and i.m. adipose tissue and that the synthetic glucocorticoid dexamethasone decreased GR abundance.

Key Words: Bovine, Preadipocytes, Glucocorticoid Receptor

776 Dietary coconut oil and conjugated linoleic acid reduce body fat in mice. K. M. Hargrave* and J. L. Miner, *University of Nebraska, Lincoln*.

Mice raised on a coconut oil (CO)-containing diet for 6 wk prior to the addition of conjugated linoleic acid (CLA) are more sensitive to CLA-induced body fat loss than mice raised on a soy oil (SO)-containing diet. Coconut oil is deficient in essential fatty acids. However the addition of linoleic acid to a CO-containing diet did not alter the enhanced sensitivity to CLA. The objective of this study was to determine if CO enhances sensitivity of mice to CLA-induced body fat loss independent of an essential fatty acid deficiency. Eighty male mice (12 wk old) were fed a purified SO-containing diet for a 1 wk adaptation. Mice were then blocked by body weight and randomly allotted to a treatment diet. Diets were arranged in a 2 x 2 factorial design with SO vs CO and 0 vs 0.5% CLA isomers. Mice were fed for 2 wk and body weight and feed intake were measured weekly. Mice were then killed and body fat and lean mass were determined by dual x-ray densitometry and fat pads and livers were weighed and collected. DNA fragmentation, indicative of apoptosis, was measured in one epididymal fat pad and is expressed as (fragmented DNA/total DNA)*100. CO-fed mice consumed more ($P < 0.05$) feed than SO-fed mice during both weeks of treatment but weighed less ($P < 0.05$) in the second wk. CO+CLA mice consumed less feed than CO mice but SO and SO+CLA mice did not differ in feed intake (CO x CLA interaction, $P < 0.05$). Both CO and CLA reduced ($P < 0.001$) body fat and fat pad weights. There was a trend for a CO x CLA interaction ($P = 0.06$) as CO+CLA mice were leaner than SO+CLA (11.94% vs 15.48% body fat, respectively). There were no differences in lean mass. CLA increased ($P < 0.01$) liver weight. There were no differences in DNA fragmentation but CLA did increase ($P < 0.001$) total DNA content of the fat pad. In summary both CO and CLA reduced body fat with a trend for an interaction. Therefore, feeding CO for a short time appears to increase the sensitivity of mice to CLA-induced body fat loss similar to a longer feeding period. This indicates that CO increases CLA-induced body fat loss in an essential fatty acid deficiency-independent manner.

Key Words: Conjugated Linoleic Acid, Coconut Oil, Body Fat

777 Subcutaneous and abdominal fatty acid composition and CLA profiles in grain finished steers. L. H. Baumgard*¹, S. R. Sanders¹, O. B. Mendivil¹, J. K. Kay¹, J. A. Marchello¹, P. Delmonte², J. M. Griinari³, and M. P. Yurawecz², ¹The University of Arizona, Tucson, ²U.S. Food and Drug Administration, Washington, DC, ³University of Helsinki, Finland.

Subcutaneous (SQ) and abdominal (AB) fat was collected 24 h after harvest from non-implanted Hereford steers (n=3) fed a high concentrate (80%), low forage (20%) finishing diet for 90 d. SQ and AB fatty acid (FA) composition was determined via gas chromatography, and the complete CLA profile (15 isomers) was characterized by triple-column silver-ion high performance liquid chromatography (Ag⁺-HPLC). Levels of total *trans*-18:1 was higher ($P < 0.01$) in AB than SQ (2.37 vs. 1.56%) and the content of *trans*-6,7,8, *trans*-9, *trans*-10, *trans*-11 and *trans*-12 18:1 was 73, 14, 23, 62 and 82% higher ($P < 0.04$), respectively, in AB compared to SQ. Conversely, SQ had a higher ($P < 0.02$) content of unsaturated FA (51 vs. 37%), enhanced CLA levels (0.56 vs. 0.35%) and an increased Δ^9 -desaturase index (47 vs. 30) compared to AB. *cis*-9, *trans*-11 and *trans*-7, *cis*-9 CLA were the first and second most predominant CLA isomers, respectively, in both depots and were higher in SQ compared to AB (69 vs. 59% and 13 vs. 9% of total CLA, respectively). *trans*-10, *cis*-12 CLA was the third and seventh most abundant CLA isomer in SQ and AB depots respectively, but levels did not differ between depots (3.3% of total CLA). *trans*-11, *trans*-13 and *trans*-9, *trans*-11 CLA were the third and fourth most predominant CLA isomers in the AB depot and were higher compared to SQ (5.8 vs 1.5% and 4.9 vs. 2.5% of total CLA, respectively). When adipose depots were combined, the content of total CLA and both the *cis*-9, *trans*-11 and *trans*-7, *cis*-9 CLA isomer (products of Δ^9 -desaturase) were highly correlated with the Δ^9 -desaturase index ($R^2 = 0.97, 0.85$ and 0.81 , respectively). These data indicate marked differences in fatty acid composition including CLA profiles between SQ and AB depots, and suggests the Δ^9 -desaturase system is responsible for *cis*-9, *trans*-11 and *trans*-7, *cis*-9 CLA being the predominant CLA isomers in beef adipose tissue.

Key Words: Beef, CLA, Adipose Depot

Lactation Biology: Biology of Lactation

778 Exogenous porcine prolactin stimulates mammary development in prepubertal gilts. C. Farmer* and M.-F. Palin, *Dairy and Swine R & D Centre, Agriculture and Agri-Food Canada, Lennoxville, QC, Canada*.

The impact of injecting gilts with porcine prolactin (pPRL) in the prepubertal period on mammogenesis was investigated. Crossbred gilts received sc injections of saline (controls; n = 12), 2 mg of pPRL (4PRL; n = 12) or 4 mg of pPRL (8PRL; n = 12) twice daily for a period of 28 d, starting at 75.1 ± 0.5 kg BW. Injections of saline or pPRL were given at 0730 and 1530. Jugular blood samples were collected from all gilts the morning before the first injection, as well as 14 and 28 d later. These samples were assayed for PRL and insulin-like growth factor-I (IGF-I). Gilts were slaughtered on the 28th day of treatment and mammary glands were collected for dissection of parenchymal and extraparenchymal tissues, and for determination of DNA, dry matter, protein and fat contents. Treatments did not alter ($P > 0.1$) IGF-I concentrations but concentrations of PRL at slaughter were greater ($P < 0.01$) in both 4PRL and 8PRL compared to controls, while at mid-treatment, they were greater ($P < 0.05$) only in 8PRL gilts. Parenchymal tissue weight increased with exogenous pPRL (199.8, 434.4 and 364.1 ± 28.4 g for controls, 4PRL and 8 PRL, respectively; $P < 0.001$) and DNA concentrations were greater in pPRL-treated gilts (2.3, 6.0 and 6.2 ± 0.3 mg/g for controls, 4PRL and 8PRL, respectively; $P < 0.001$). The percentages of protein and dry matter in parenchyma increased ($P < 0.001$) while that of fat ($P < 0.001$) and the protein to DNA ratio ($P < 0.05$) decreased with exogenous pPRL. Treatment differences were always observed between the 4 mg dose and the controls, and no further differences were seen when increasing the dose to 8 mg daily. Extraparenchymal tissue weight and total parenchymal fat were unaltered by treatments ($P > 0.1$). It is apparent that pPRL can stimulate mammogenesis in prepubertal gilts through hyperplasia. It would be of great interest to determine if this favorable effect translates into greater milk yield dur-

ing subsequent lactations. (Sincere thanks to Monsanto for supplying the pPRL).

Key Words: Mammary Gland Development, Prolactin, Gilts

779 Gene expression profiling of bovine mammary epithelial cells in response to prolactin and extracellular matrix. L. Rowley*¹, D. Baird², T. Wilson³, J. Whitley¹, C. Berends¹, M. Wells¹, and M. Goddard¹, ¹Primary Industries Research Victoria, Attwood, Australia, ²AgResearch, Lincoln, New Zealand, ³AgResearch Molecular Biology Unit, University of Otago, Dunedin, New Zealand.

Dairy farming is a financially important agricultural resource to Australia and further economic gains could be realised through improved milk yield or composition. In order to manipulate milk production however, we need to more fully understand the functional regulation of milk protein synthesis and secretion in the bovine mammary gland. In order to identify regulatory mechanisms involved in the differentiation of the mammary gland, primary mammary epithelial cells were cultured in the presence and absence of both prolactin (1 μ g/mL) and extracellular matrix. Culture medium was analysed by Western blotting to evaluate milk protein synthesis and secretion. RNA was isolated from cells in culture to assess gene expression using cDNA microarrays containing 23k bovine expressed sequence tags. Microarray data was statistically analysed by mixed model analysis using REML (restricted maximum likelihood). When cultured in the presence of extracellular matrix, mammary epithelial cells were able to assume three-dimensional spherical structures similar to mammary secretory alveoli in vivo. Western blot analysis showed that the secretion of the milk proteins, β -lactoglobulin, α_{S1} - and β -casein was greatly upregulated in the presence of both prolactin and extracellular matrix. Microarray analysis has identified genes