Data are limited on how claw development early in life will affect the susceptibility to lameness and claw disorders later in life. Adequate nutrition and bioavailable trace minerals might enhance claw development by ameliorating various stressors early in life. Ninety Holstein bull calves <1 wk old were purchased in 3 groups and transported to the Illinois research facility from Wisconsin. Calves were randomly assigned to treatments in a 2x2 factorial arrangement of plane of nutrition (PN) and trace mineral source (TM). Conventional PN received a fixed amount (568 g/d) of milk replacer (22% CP, 20% fat) plus ad libitum starter (18% CP) and were weaned at 6 wk; they received ad libitum starter to wk 12 and 0.5 kg/d of hay wk 10-12. During wk 13-20 calves were fed 3.2 kg/d of grower (16% CP) plus chopped hay ad libitum. Intensified PN received variable amounts (810, 1136, and 568 g/d for wk 1, 2 to 6, and 7, respectively) of milk replacer (28% CP, 20% fat) plus ad libitum starter (22% CP), were weaned at 7 wk, were fed ad libitum starter to wk 12, and fed ad libitum grower plus 0.5kg/d hay wk 13-20. Feeds contained either inorganic sulfates of Fe, Cu, Zn, and Mn or bioavailable sources (Zinpro Performance Minerals, Eden Prairie, MN). Calves were individually housed in hutches bedded with straw through wk 9 and group-housed by diet wk 10-20. Calves had free access to water. Claws were measured at wk 0, 5, 10, 15, and 20. Calves were born with uneven claw length (P<0.001), with rear and medial claws longer than front and lateral claws. Organic TM tended to increase claw length (P=0.105) after wk 15. Net growth increments were established after wk 5 regardless of treatment, which could be associated with complete establishment of corium tissue and physiological changes such as rumination and thermoregulation. Rear medial claws had more growth and wear (P<0.05) regardless of treatment. Intensified PN increases biological value of organic TM reflected by enhanced hoof dynamics and body growth.

Key Words: trace minerals, claw growth, calves

316 Gene expression profile research of dairy goat mammary gland by Long-SAGE. H. Yan, C. Li, Q. Li*, and X. Gao, Northeast Agricultural University, Harbin, China.

Serial analysis of gene expression (SAGE) is a high-throughput, sensitive and efficient method for global expression profiles analysis that allows the quantitative and simultaneous analysis of large-scale transcripts under different conditions or in certain tissues. The Long-SAGE technique, developed from original SAGE, generates longer tags (21bp) than typical 14bp tags, which are more unambiguous in uniquely identifying with corresponding genes. Therefore, Long-SAGE technique was chosen to study gene expression profile of lactating dairy goat mammary gland, to find candidate genes that could control or regulate function of mammary gland. An improved protocol of Long-SAGE, considering characters of mammary gland, was applied to construct Long-SAGE libraries. Briefly, mRNA were isolated and synthesized into double-strand cDNA (ds cDNA), concatemers were formed by linking tags randomly which were extracted from ds cDNA through a series of restriction enzyme cleaving and ligating processes, sequenced the positive clones of concatemers to get information of tags. An extra heating process was necessary to increase cloning efficiency of concatemers. 7 Long-SAGE libraries of different lactation stages (initiation, peak, stabilization and involution) of healthy dairy goat lactating mammary gland were successfully constructed. Over 21 thousands Long-SAGE tags were obtained by sequencing 7 thousands positive clones of concatemers. Removing the of duplicated and invalid tags, there’re about 10 thousands of 17bp unique Long-SAGE tags, only 12% tags were matched to UniGene data according to limited genome resources. Further gene expression profile research of dairy goat mammary gland is still going on.

Key Words: gene expression profile, mammary gland, Long-SAGE

317 Selection of key gene related to development of mammary gland in dairy goat. C. Li, H. Yan, Q. Li*, and X. Gao, Northeast Agricultural University, Harbin, China.

To identify key genes involved in the initiation and development of mammary gland in dairy goat, we analyzed the global gene expression profiles of 7 different developmental stages using long serial analysis of gene expression (LongSAGE). We performed LongSAGE in healthy dairy goat mammary at seven different developmental stages (early puberty, late puberty, early pregnancy, late pregnancy, middle lactation, early involution and late involution). Computational analysis was carried out to identify differentially expressed genes in mammary gland between early puberty and late puberty, early pregnancy and late pregnancy which were further validated by real-time quantitative RT-PCR. Approximately 8,000 clones were sequenced for the seven libraries. Totally, 104,995 valid LongSAGE tags were obtained with 12,574 unique tags. Compared with the gene expression profile of early puberty mammary gland, 404 genes were identified to be differentially expressed in late puberty. 83 genes were high-abundance expressed in late pregnancy and early pregnancy contrast with late puberty. These diversely expressed genes were related to cell proliferation, biosynthesis, signal transduction, and cellular transport. In this study, seven LongSAGE libraries of different developmental stages of dairy goat mammary gland were successfully constructed. The genes expression profiles of these seven developmental stages were depicted on a genome-wide scale. For the restriction of annotation databases, we provided novel candidate genes that might be related to mammary gland development.

Key Words: dairy goat, mammary gland, LongSAGE

318 Epigenetic changes during functional differentiation of the mammary gland. M. Rijnkels*, C. Freeman-Zadrowski, and J. Hernandez, USDA/ARS Children’s Nutrition Research Center, Baylor College of Medicine, Houston, TX.

The packaging of the DNA that surrounds a gene, the conformation of chromatin, is an integral part of gene regulation. Chromatin conformation is determined by DNA methylation, the post-translational modifications of the core histones and the proteins that bind to this. Hypermethylated DNA is usually associated with silent genes, whereas actively transcribed genes are hypomethylated. Different histone modifications are associated with open (active) or closed (inactive) chromatin. DNAase hypersensitivity (DHS) indicates an open chromatin conformation and is often associated with regulatory elements. To identify a role for chromatin...
in functional differentiation in the mammary gland we investigated the epigenetic changes (DNA methylation, Histone modifications and DHS) in the casein gene cluster. The casein genes encode the major milk protein genes, which are expressed exclusively in the functionally differentiated mammary epithelial cells. We analyzed the presence of DHS, as well as the presence of an active histone modification (histone H3 Acetylation, H3Ac), using Chromatin immuno-precipitation (ChIP), in lactating mammary gland tissue of mice. We showed that chromatin surrounding the casein genes is in an open chromatin conformation in the lactating mammary gland as compared to liver. In addition we show that the promoters and potential distal regulatory elements are hypomethylated in lactating mammary tissue. Analysis at different time points during post-natal mammary gland development showed that DNA methylation is lost (opening up of chromatin) on promoters in concordance with major transcriptional up-regulation in pregnancy, while some potential distal regulatory elements already acquire an open chromatin structure (loss of DNA methylation) during pubertal development. This implies a model where chromatin conformation is changed at regulatory elements at different stages of mammary gland development to prepare the genomic region for further chromatin changes that will allow full transcriptional activity upon lactation.

Key Words: lactation, epigenetics, casein


The study was conducted to investigate the effects of oligopeptides (di- or tripeptide) and hormones (insulin or prolactin) on expression of peptide transporter PEPT2 gene in cultured bovine mammary tissues. The explants were obtained from the mid-lactating dairy cows and cultured in DMEM/F12 medium containing 10% fetal bovine serum. Contents of other essential amino acids were similar during the whole experiment. Phenylalanine (Phe) was firstly replaced by different ratios (0, 5, 10, 15 and 20%) of dipeptide, phenylalanylphenylalanine to investigate the oligopeptide level for optimal expression of PEPT2 gene. The mRNA expression was determined by SYBR green method of quantitative real-time PCR, and protein by immunocytochemistry, respectively, after the tissues were cultured for 48 h. The PEPT2 protein was successfully detected in the cultured bovine mammary tissue. The abundance of PEPT2 mRNA was highest (P<0.01) when 10% of phenylalanylphenylalanine was included. And then, this level (10%) was also successfully detected in the cultured bovine mammary tissue. The abundance of PEPT2 mRNA was highest (P<0.01) when 10% of phenylalanylphenylalanine was included. And then, this level (10%) was also successfully detected in the cultured bovine mammary tissue. The abundance of PEPT2 mRNA was significantly(P<0.01) increased when the insulin was added into the culture medium for 1 h, but prolactin did not have effect on PEPT2 gene expression. These results indicate that peptide transporter PEPT2 in bovine mammary gland may be influenced through its gene expression by oligopeptides or hormones.

Key Words: bovine mammary gland tissues, oligopeptide, small peptides transporter II

320 Microarray analysis of gene expression profiles in dry period bovine mammary gland. X. Hou and Q. Li*, Northeast Agricultural University, Harbin, Heilongjiang, China.

Mammary gland undergoes dramatically functional and metabolic changes during the transition from lactation to dry period. To better understand the molecular events underlying these changes, the microarray analysis was performed using Affymetrix GeneChip Bovine Genome Array to analyze the gene expression profiles in bovine mammary tissue. Mammary tissues samples were collected in dry period and lactation from Holstein dairy cows. At the cutoff criteria of the signed fold change ≥2 or ≤2, A total of 143 genes were identified as differentially expressed, including 57 up-regulated and 86 down-regulated genes during dry period compared with lactation, and the results was identified by quantitative real-time PCR. Gene ontology analysis showed that the mainly up-regulated genes in dry period were those associated with metabolic process, regulation of apoptosis and immune system process. the mainly down-regulated genes were those associated with cellular components, binding, transport, biosynthetic process and signal transduction. The microarray data provided insight into the molecular events in the dry period bovine mammary gland and established the groundwork for further studies of the basic genetic control in bovine mammary gland apoptosis and reconstruction.

Key Words: mammary gland, microarray, genomics
324 Mammary expression of activating transcription factor 4 (ATF4) and tribbles homolog 3 (TRB3) is up-regulated during CLA-induced inhibition of milk fat synthesis in the dairy cow. K. J. Harvatine*, Y. R. Boisclair1, and D. E. Bauman2, 1Pennsylvania State University, University Park, 2Cornell University, Ithaca, NY.

Milk fat depression (MFD) in dairy cows is caused by unique fatty acids originating from ruminal fatty acid biohydrogenation. The best studied of these is trans-10, cis-12 conjugated linoleic acid (CLA) and its mechanism involves a coordinated decrease in mammary expression of lipogenic enzymes. Endoplasmic reticulum (ER) stress is emerging as a causative mechanism of metabolic diseases, with ATF4 and TRB3 providing a link between ER stress and metabolism. TRB3 inhibits AKT1, MAPK, and acetyl-CoA carboxylase. We conducted in vivo and in vitro studies to examine expression of ATF4 and TRB3 in mammary tissue during MFD and in bovine mammary epithelial cell culture (MECT) during CLA treatment. ATF4 and TRB3 expression was increased in mammary tissue during MFD induced by a low forage, high oil diet fed for 11 d but not by 3 d infusion of CLA. Temporal response was subsequently investigated. An initial priming dose of 7.5 g of CLA was given followed by infusion of CLA for 5 d. TRB3 and ATF4 expression was increased at 12 h but not at 30 h and 5 d. Although TRB3 and ATF4 are associated with ER stress signaling, we did not observe increased expression of the chaperone protein HSPA5 or alternate splicing of XBP1 indicating a non-classical ER stress response. CLA dose dependently increased expression of TRB3 and ATF4 with maximal response at approximately 10 μM, response diminished with increasing doses and no response was observed at 50 μM. CLA doses that increased expression of TRB3 and ATF4 decreased lipogenesis with no decrease in expression of fatty acid synthase. However, ER stress induced by tunicamycin or thapsigargin increased expression of TRB3 and ATF4 but did not decrease lipogenesis. CLA induces TRB3 and ATF4 expression in some conditions, but the inconsistency of the signaling response and the lack of an effect of chemically induced ER stress on lipogenesis calls into question the functional importance of TRB3 and ATF4 in the lipogenic phenotype of classical MFD. USDA-NRI 2006-35206-16643.

Key Words: ER stress, CLA, milk fat depression

325 Lipid transporters and their regulators in the bovine mammary gland in relation to blood serum metabolites during pregnancy, involution, and lactation. O. Mani1, M. T. Sorensen2, K. Sejrsen3, R.M. Bruckmaier*1, and C. Albrecht1, 1Institute of Biochemistry and Molecular Medicine, University of Bern, Bern, Switzerland, 2Department of Animal Health, Welfare and Nutrition, Aarhus University, Tjele, Denmark, 3Veterinary Physiology, Vetsuisse Faculty, University of Bern, Bern, Switzerland.

Several ATP-binding cassette (ABC) transporters are known to play a pivotal role in cellular lipid efflux. To determine if they are involved in maintaining lipid homeostasis of mammary tissue, we investigated the expression and localization of candidate ABC transporters and their regulators during different physiological stages of the bovine mammary gland (MG). MG biopsies were repeatedly taken from ten dairy cows during the pregnancy-lactation cycle. mRNA levels were determined by current research. The marked up-regulation of HK1 and HK2, were very dramatic, indicated some unclear roles of HK2 in lactation.

Key Words: glucose transport, regulatory proteins, lactation, cell culture systems.
quantitative RT-PCR. In parallel, blood serum metabolite levels were measured; proteins were localized by immunohistochemistry. Relative mRNA levels of the lipid efflux transporters ABCA1 and ABCA7 were elevated during involution as compared to lactation (P=0.0197, <0.0001, resp.). The intracellular cholesterol transporter NPC1 and the regulatory genes LXRα, PPARγ, SREBP1, SREBP2 were increased post partum as compared to the dry period (P=0.0003, 0.0271, <0.0001, 0.038 resp.). Correlation analysis of ABCA1, ABCA7 and ABCG1 mRNA profiles with blood cholesterol levels revealed significant inverse relationships (r=-0.39, -0.51, -0.29, resp.; P<0.05). ABCA1 and ABCG1 showed different localization and activity in mammary epithelial cells (MEC) during involution and lactation. This study demonstrates that lipid transporters and their regulatory genes are differentially expressed in the MG during the pregnancy-lactation cycle and correlate with blood serum cholesterol profiles. Our results suggest that ABCA1 and ABCG1 are involved in the removal of cholesterol from MEC. ABCA7 may play a role in phagocytosis of apoptotic MEC during involution. Regulation of lipid transporters in the MG is only partially associated with transcription factors that control lipid homeostasis because the induction of lactation is triggered by lactogenic hormones that may interfere with regulators of lipid homeostasis.

**Key Words:** ABC transporters, cholesterol, mammary gland

### Meat Science and Muscle Biology: Symposium: Effects of By-product Feeding on Meat Quality Traits


The objective of this study was to evaluate the effects of modified wet distillers grains with solubles (MDGS) on partitioning of fat between various depots. Crossbred cattle (n = 168) were used in a randomized complete block design with 7 pens per treatment. Treatments consisted of 0, 20, or 40% (DM basis) MDGS, as a replacement for high-moisture corn in the basal diet. The basal diet consisted of 38% high moisture corn, 12% corn silage, and 5% protein-mineral supplement. Ultrasound scans were taken every 56 d throughout the study to estimate intramuscular fat (IMF) and subcutaneous fat deposition. Cattle were fed for 139 d and harvested when 12th rib fat thickness (SQF) was estimated to be 1.01 cm. Performance and carcass data was evaluated. Final weight was calculated by adjusting carcass weights to a constant dressing percent. The subsequent weight was used to calculate ADG. Shrink dressing percent was calculated by adjusting live weights to 4% shrink. Inclusion of MDGS had no effect on DMI, however carcass adjusted G:F tended to increase (P = 0.08) with the inclusion of MDGS in the diet. The inclusion rate of 20 and 40% MDGS tended to increase shrink dressing percent (P = 0.09) and yield grade (P = 0.06) when compared to the control diet. The ratio of IMF:SQF (IMR) was lower (P < 0.01) in cattle fed MDGS when compared to the control diet. The inclusion of MDGS within the diet may affect energy partitioning between IMF and subcutaneous fat deposition. This data suggests that feeding MDGS at inclusion levels of 20 or 40% may result in greater subcutaneous fat deposition with minimal effects on marbling.

**Table 1.**

<table>
<thead>
<tr>
<th>Item</th>
<th>0%</th>
<th>20%</th>
<th>40%</th>
<th>SEM</th>
<th>Probability</th>
</tr>
</thead>
<tbody>
<tr>
<td>MDGS</td>
<td>7</td>
<td>7</td>
<td>7</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Carcass adjusted ADG, kg</td>
<td>1.33</td>
<td>1.46</td>
<td>1.37</td>
<td>0.05</td>
<td>0.22</td>
</tr>
<tr>
<td>DMI, kg/d</td>
<td>8.63</td>
<td>8.85</td>
<td>8.75</td>
<td>0.27</td>
<td>0.85</td>
</tr>
<tr>
<td>Carcass adjusted G:F</td>
<td>0.154a</td>
<td>0.164b</td>
<td>0.156ab</td>
<td>0.003</td>
<td>0.08</td>
</tr>
<tr>
<td>Shrink dressing %</td>
<td>61.61a</td>
<td>62.63b</td>
<td>61.83ab</td>
<td>0.32</td>
<td>0.09</td>
</tr>
<tr>
<td>Calculated yield grade</td>
<td>2.6a</td>
<td>2.9b</td>
<td>2.9</td>
<td>0.08</td>
<td>0.06</td>
</tr>
<tr>
<td>SQF, cm</td>
<td>1.08</td>
<td>1.22</td>
<td>1.22</td>
<td>0.05</td>
<td>0.12</td>
</tr>
<tr>
<td>Marbling (400 = slight, 500 = small)</td>
<td>505.4</td>
<td>515.7</td>
<td>492.6</td>
<td>9.7</td>
<td>0.27</td>
</tr>
<tr>
<td>IMF ultrasound (IMF:SQF)</td>
<td>11.69a</td>
<td>9.97b</td>
<td>9.76b</td>
<td>0.40</td>
<td>0.01</td>
</tr>
</tbody>
</table>

*Means with unlike superscripts differ.*

**Key Words:** beef, distillers grains, fat deposition


The objective of this study was to evaluate dietary inclusion of DDGS at 30% and CLA at 0.6%. Forty barrows were assigned to a 2×2 factorial arrangement within a completely randomized design with a total of 10 replications. Pigs received ad libitum access to water and feed. Pigs were slaughtered at an average live weight of 129.88 ± 1.21 kg. Data was collected for growth performance, carcass and meat quality and fatty acid profile analysis. The inclusion of CLA in the diet did not significantly affect any of the parameters analyzed for carcass and meat quality. However, the inclusion of DDGS decreased the redness (P<0.05) of the longissimus muscle and increased the flexibility (P<0.05), both vertical and horizontal, of the belly. A significant interaction was observed for jowl and belly samples for total saturated fatty acids (SFA) and total polyunsaturated fatty acids (PUFA) (P<0.05). For both jowl and belly samples, the maximum amount of SFA (41.06 and 39.32%, respectively) were obtained by the DDGS only diet. The maximum amount of PUFA was obtained by the treatment DDGS + CLA for both jowl and belly samples (22.53 and 23.76%, respectively). The interaction of CLA and DDGS did not affect (P>0.13) the amount of monounsaturated fatty acids (MUFA) for both belly and jowl samples. However, the inclusion of CLA and also, the inclusion of DDGS in the diet, affected (P<0.05) the MUFA content in both jowl and belly samples. The maximum amount of MUFA (49.21% for the jowl samples and 50.50% for belly samples) was obtained by the treatments of pigs fed the control diet, with no CLA and DDGS. The iodine value of the samples were affected (P<0.05). For both jowl and belly samples, the maximum amount of SFA (41.06 and 39.32%, respectively) were obtained by the inclusion of CLA, DDGS and the interaction between them for both jowl and belly samples. The highest value was obtained from the samples of pigs fed DDGS with no CLA (71.18 and 72.19 for jowl and belly, respectively). Overall, this experiment indicates that DDGS and CLA and their interaction do not affect in a large proportion the carcass and meat quality when fed to finishing barrows. However, significant changes can be achieved in the fatty acid profile as measured in the belly and jowl.

**Key Words:** CLA, DDGS, iodine value

**328 Effects of distillers grains on beef carcass quality and palatability.** C. R. Calkins*, A. S. de Mello Jr., and L. S. Senaratne, *University of Nebraska, Lincoln.*

Distillers grains, mostly obtained from ethanol production, can be used in cattle diets. Results of 3 experiments show that feeding wet distillers