274 ASAS Centennial Presentation: The history of growth biology research - A reflection on the episodic nature of science. T. Etherton*, *Penn State University*, *University Park*.

Evans and co-workers were the first to demonstrate the presence of a substance in the anterior pituitary that increased the growth rate of rats in the 1920's. Subsequent research established the active compound was growth hormone (somatotropin; ST). In the ensuing 80 years much has been learned about the endocrine system and how it regulates animal growth, and nutrient partitioning. Products of biotechnology, such as growth hormone and beta-agonists, have been developed and approved for commercial application. A "reflection over the past eight decades of research is telling. In the early days of recombinant DNA technology (late 1970's), numerous hypotheses could be tested that were not feasible with purified pituitary preparations of ST. Recombinant bovine ST (rbST) was approved for use by the FDA in 1993, with great excitement in the scientific community. A contemporary reflection on research is revealing. We have developed numerous and wondrous scientific breakthroughs, and the ability to manufacture recombinant proteins in large scale. Yet, the agricultural scientific community has not been actively involved in the "public discussion" about the need for and benefits of "biotechnology in the barnyard". Activist groups have attacked science and the application of science. A recent "episode" that illustrates this is the attack on rbST use in the dairy industry. The point of their misguided attacks being that rbST is a risk to consumers! We are standing at a point in time where the next pulse is not associated with the secretory profile of a circulating hormone, but rather a concerted "push" by the academic community to defend science and the freedom to use products of science.

Key Words: Growth Biology, Endocrine Regulation, Somatotropin

275 ASAS Centennial Presentation: Future needs and directions in animal growth and development research. M. A. Mirando*, *Cooperative State Research, Education, and Extension Service, United States Department of Agriculture*, *Washington, DC*.

As the 21st century progresses, the increased rate of societal, economic and environmental changes occurring at the global, national, regional and local levels will significantly impact animal agriculture and require even greater commitment to research to solve new problems that arise. Research in the area of animal growth and development, in particular, can yield solutions to many of those problems. A variety of needs and approaches will dictate future directions in animal growth and development research. Changes in land use and global climate will provide additional impetus for greater environmental stewardship, reduced nitrogen output and decreased carbon footprint of generating animal products. Research will continue to focus on increasing production efficiency, thereby reducing input of resources, while continuing to improve quality of animal products. Areas of research emphasis will include, but not be limited to, studying mechanisms regulating muscle growth, adiposity and marbling of meat, fetal programming of postnatal growth and development, and removal or replacement of dietary antimicrobial growth promotants. Future studies will take advantage of genome sequences that have been or will be obtained for agricultural animals, and research will include transcriptional profiling and proteomic approaches. However, although the latter approaches are incredibly powerful, they only yield information on which genes are involved but not how those genes products act or are utilized in cellular or tissue responses. Thus, research will continue to include biochemical and physiological studies to elucidate specific actions and functions of individual gene products, as well as their roles in metabolic and signaling pathways. Finally, there will be continued need for a balance between basic and applied research. Because the private sector, including commodity groups, funds primarily applied research, the fundamental discoveries that lay the groundwork for future applied research will need to be supported by Federal funding sources.

Key Words: Development, Growth, Research

276 The role of microRNA on murine mammary epithelial cell and mammary gland. Q. Z. Li* and C. M. Wang, *Northeast Agricultural University*, *Harbin, Heilongjiang, China*.

MicroRNAs (miRNAs) are encoded small RNAs that hybridize with messenger RNAs, resulting in degradation or translational inhibition of targeted transcripts. The potential for miRNAs to regulate cell proliferation or differentiation from murine mammary epithelial cell or mammary gland is unknown, and the mechanism of miRNA on mammary development is also not clear. We used microarrays and qRT-PCR to evaluate miRNA expression in mammary tissue during development and functional states of the murine mammary gland. Physiological stages evaluated were that miRNAs was differentially expressed the different developmental stages of the mouse mammary gland. Bioinformatic programs were used to predict targets for miRNA that were identified as significant feartures of differential expression during developmental and functional cycles of the mammary gland. Antagomirs were used to assess the function and targets of miR-138, miR-292-5p and let-7g in vivo and in vitro. Stage-specific expression patters on miRNA were evident. miRNAs such as miR-138 and miR-431 were downregulated ($P<0.05$) some miRNAs such as miR-133, miR-133a, miR133b were upregulated ($P<0.05$) during pregnancy and lactation compared with expression during mammary involution or expression tissue from virgin mice. It also displays that Prolactin-receptor, STAT5 protein, MAPK protein were up-regulated after treatment with miR-138 antagomir ($P<0.05$), and PKC protein was (spell out up or down)-regulated by antagomirs for miRNA-292-5p and let-7g ($P<0.05$). Treatment with antagomirs demonstrated that miR-138 inhibited viable and proliferation of mouse mammary eptihelial cells.($P<0.01$). Venous injections of antagomir for miR-138 into mice on the second day of lactation increased milk production at 12 h followed by a decrease at 48 h ($P<0.01$). Finally, assembling miRNA networks helped to predict miRNA targets. In conclusion, we demonstrated that different networks of miRNA are expressed during development, lactation and involuation of the mouse mammary gland, and that these miRNAs play important roles during these physiological states.

Key Words: MicroRNA, Mammary Pregnancy Cycle, Target

277 Effect of AMP-activated protein kinase (AMPK) and insulin-like growth factor-1 (IGF-1) on expression of muscle-specific ubiquitin ligases in C2C12 myotubes. J. F. Tong*, K. R. Underwood, X. Yan, M. J. Zhu, and M. Du, *University of Wyoming*, *Laramie*.

INTRODUCTION: AMP-activated protein kinase (AMPK) is a key cellular energy sensor. A mutation in AMPK leads to Rendement napole (RN) phenotype in pigs characterized by superior lean growth. AMPK regulates protein synthesis in skeletal muscle, but its role on myofibrillar protein degradation is unclear. Two newly identified muscle-specific ubiquitin ligases (UL), MAFbx and MuRF1, are crucial for myofibrillar protein breakdown. The insulin-like growth factor-1 (IGF-1) pathway induces muscle hypertrophy in part through inhibition of MAFbx and MuRF1 expression, mediated by protein kinase B (PKB). C2C12 myotubes are commonly used for studying skeletal muscle growth. HYPOTHESIS: AMPK interacts with IGF-1 to control the expression of UL in C2C12 myotubes. METHODS: C2C12 cells were incubated in DMEM medium with 10% fetal bovine serum. Fusion was induced by 2% horse serum. Myotubes were treated with 5-Aminoimidazole-4-carboxamide-1-β-D-ribonucleoside (AICAR, 0, 0.1, 0.3 and 1 mM), and/or IGF-1 (100 ng/ml). After 4, 12 and 24 h treatments, myotubes were collected for real-time PCR and immunoblotting analyses. Four independent experiments were conducted (n=4). RESULTS: Activation of AMPK by AICAR, a specific AMPK activator, increased MAFbx and MuRF1 mRNA expression, accompanied by an increase in their protein levels. As expected, IGF-1 activated PKB and reduced the expression of UL ($P < 0.01$). Intriguingly, activation of AMPK synergized IGF-1-induced PKB activation; however, the expression of UL was not attenuated, but strengthened by AMPK activation. These data indicate that a PKB independent mechanism exists through which AMPK induces the expression of UL, surpassing the inhibitory effect of IGF-1/PKB pathway on UL expression. In addition to regulating IGF-1/PKB signaling pathway and protein synthesis in muscle cells, AMPK also mediates protein degradation through enhancing the expression of UL. These data indicate that AMPK may be an important molecular target for enhancing lean growth in livestock.

Key Words: Skeletal Muscle, AMPK, Degradation

278 Metabolic gene networks in longissimus muscle of early-weaned Angus and Angus × Simmental steers fed high-grain or high-byproduct diets during the growing phase. D. E. Graunard*, P. Piantoni, M. Bionaz, L. L. Berger, D. B. Faulkner, and J. J. Loor, *University of Illinois*, *Urbana*.

mRNA abundance and expression profiles of lipogenic and intracellular energy metabolism gene networks were evaluated via quantitative PCR in longissimus muscle (LM) of early-weaned (~142 d age) Angus (n = 3/diet) and Angus × Simmental steers (AxS; n = 3/diet) fed a high-grain (HiE) or high-byproduct (HiF) diet for 112 d. LM biopsies were collected at 0, 56, and 112 d of feeding. We evaluated 32 genes associated with lipogenesis (e.g., *G6PD*, *ACACA*, *FASN*), intracellular FA activation (*ACSL1*), esterification (*GPAM*, *DGAT1*, *DGAT2*), desaturation (*SCD*, *FADS2*), intracellular energy metabolism (AMPK subunits, *PRKAA1* and *PRKAA2*), TCA-cycle (*MDH2*), mitochondrial biogenesis (*PPARGC1A*), nuclear receptor signaling (*PPARG*, *PPARD*), transcriptional regulation of lipogenesis (*SREBF1*, *THRSP*, *INSIG1*), as well as insulin-regulated glucose transport (*GLUT4*) and signaling (*INSR*, *IRS1*). *MDH2*, *SCD*, *GLUT4*, *IRS1*, *ACSL1*, *FASN*, and *PRKAA2* had the greatest relative mRNA abundance averaging 26, 16.5, 7.9, 6.9, 5.4, 5.3, and 4.8%, respectively, of total genes measured. Among those, there was a diet × steer type × time ($P < 0.05$) effect for *PRKAA2* whose expression was 2-fold greater in AxS steers namely on d 56. A similar response was observed for *THRSP* in AxS steers fed HiE during the first 56 d (~60-fold greater mRNA relative to d 0). A diet × steer type effect was found for *SREBF1*, *PPARG*, and *PPARGC1A*, with HiE inducing greater expression in Angus and HiF in AxS steers. Angus steers had greater ($P < 0.05$) expression of lipogenic genes (*ACACA*, *FASN*, *G6PD*, *GLUT4*, *SREBF1*), which might have been associated with numerically greater marbling score (4.49 vs. 4.16, $P = 0.20$) during the 112 d feeding period. Feeding HiE resulted in greater ($P = 0.05$) *IRS1*, *PPARGC1A*, and *ACSL1*, as well as a tendency ($P < 0.15$) for greater *SREBF1*, *THRSP*, and *GLUT4*. Results indicate that lipogenic and intracellular energy metabolism gene networks have different temporal expression patterns in crossbred vs. purebred steers. Further, these patterns could be altered by level of dietary energy during the rapid-growth phase.

Key Words: Genomics, Nutrition, Skeletal Muscle

279 Enhanced skeletal muscle protein synthesis rates in pigs treated with somatotropin requires fed amino acids levels. F. A. Wilson*, A. Suryawan, R. A. Orellana, H. V. Nguyen, A. S. Jeyapalan, M. C. Gazzaneo, and T. A. Davis, *Baylor College of Medicine*, *Houston, TX*.

Chronic somatotropin (pST) treatment in pigs increases skeletal muscle protein synthesis and circulating insulin, a known promoter of protein synthesis. Previously, we showed that the pST-mediated rise in insulin alone could not account for the pST-induced increase in protein synthesis. This study aimed to determine whether the pST-induced increase in insulin promotes skeletal muscle protein synthesis when amino acids are not limiting and are provided at fed levels, and whether the response is associated with enhanced translation initiation factor activation. Growing pigs were treated with pST (180 μg/kg/day, n=18) or saline (n=18) for 7 days, then pancreatic-glucose-amino acid clamps were performed to obtain plasma insulin levels equivalent to fasted or fed-pST treated pigs. Amino acids were raised to fed levels at both fasted and fed insulin concentrations; glucose was maintained at fasting levels throughout. Treatment with pST decreased plasma urea nitrogen and increased insulin-like growth factor^{-1} levels ($P<0.001$), confirming effectiveness of treatment. Skeletal muscle protein synthesis was increased by pST treatment and by insulin and/or amino acid infusion ($P < 0.001$). When amino acids were raised to fed levels, in the presence or absence of fed insulin concentrations, muscle protein synthesis rates were higher in pigs treated with pST than saline ($P < 0.02$). Fed amino acids, with or without raised insulin concentrations, increased the phosphorylation of S6K1 and 4EBP1, decreased the inactive 4EBP1•eIF4E complex association and increased active eIF4E•eIF4G complex formation ($P < 0.02$). However, treatment with pST did not alter translation initiation factor activation. We conclude that the pST-induced stimulation of muscle protein synthesis requires fed levels of amino acid, but this response is not mediated by mRNA binding to the ribosomal complex. (Supported by USDA NRI 2005-35206-15273).

Key Words: Growth Hormone, Mammalian Target of Rapamycin

280 Changes in the transcriptome of adipose tissue of the dairy heifer during late pregnancy and lactation as measured by gene array analysis: Global changes and cell control. J. Sumner*, C. Schachtschneider, and J. McNamara, *Washington State University*, *Pullman*.

Metabolic adaptations in adipose tissue are a critical part of establishment and maintenance of lactation in dairy cattle. Adipose tissue stores and releases energy and also secretes a number of metabolic regulators and cytokines. The objective was to obtain a more in-depth understanding of the transcriptomic adaptations in adipose tissue of Holstein heifers from 30 d prepartum to 14 DIM. Adipose tissue was obtained at 30 d prepartum and 14 DIM, extracted for RNA, and hybridized to the Affymetrix Genechip® Bovine Genome Array. There were 20 animals and of those 12 animals provided a quantity and quality of RNA for gene array analysis. Animals averaged 29.8 (SEM = 1.3 kg/d of milk for the first 60 DIM (range 18.6 to 44.8 kg/d). They lost 42.6 kg of BW (SEM 8.4, range +9.1 to -113.6) and 0.38 BCS units (SEM 0.10, range 0 to -1.0) from 0 to 14 DIM. Chip quality backgrounds averaged below 50 units, and 3/5 bias on control genes < 2.0. Correlations among replicates were > 0.85. Approximately 433 genes increased 100% or more, 3406 increased 25 to 100%; 1951 decreased 25 to 50 %, 337 decreased 75% or more. Genes expressed in greatest amounts (signal > 6000, average signal 125) included collagen, and ribosomal proteins, and FABP4. Lipoprotein lipase was expressed at 4261 (SEM 509), the most highly expressed gene regulating nutrient flux. Leptin receptor expressed at 734 (50) prepartum and fell 12 % at 14 DIM. Genes involved in cell synthesis, transcriptional control and inflammation increased 5-fold or more, including beta-defensin, 10-fold, cytokine inducible nuclear protein, 8-fold; chromosomal reading frame 4; 6-fold; sarcoplasmic Ca ATP-ase, 4-fold; leucine-rich repeat-containing 2, 3.5-fold; voltage-dependent calcium channel subunit, 3.5-fold. Bos taurus uncoupling protein 3 increased 3-fold, indicating possible proton uncoupling in white adipose tissue. These data provide some initial insight into the global transcriptomic response of adipose tissue to lactation.

Key Words: Lactation, Adipose, Transcriptome

281 Changes in the transcriptome of adipose tissue of the dairy heifer during late pregnancy and lactation as measured by gene array analysis: changes in specific metabolic control genes. J. Sumner*, C. Shachtschneider, J. Vierck, and J. McNamara, *Washington State University*, *Pullman*.

Metabolic adaptations in adipose tissue are a critical part of establishment and maintenance of lactation. Adipose tissue stores and releases energy and secretes a metabolic regulators and cytokines. Previous work determined that several enzymes and pathways are controlled gene transcription for enzyme synthesis, and hormonal and neurocrine regulation of enzyme activity. Our objective was to obtain a more in-depth understanding of the gene transcriptome changes underlying the adipose response to lactation. We tested the hypothesis that genes encoding for proteins regulating metabolism changed expression in adipose tissue of Holstein heifers from 30 d prepartum to 14 DIM. Adipose tissue biopsies were obtained, tissue extracted RNA, and hybridized to the Affymetrix Genechip® Bovine Genome Array. Animals averaged 29.8 (SEM = 1.3 kg/d of milk for the first 60 DIM (range 18.6 to 44.8 kg/d)). They averaged 33.0 (1.6) kg/d for d 53-60 (range 25 to 46 kg/d). They lost 42.6 kg of BW (SEM 8.4, range +9.1 to -113.6) and 0.38 BCS units (SEM 0.10, range 0 to -1.0) from 0 to 14 DIM. Anabolic pathway genes decreased ($P < 0.05$), including (mean (% change), (SEM)): SREBP, -25.1, (6.2); GLUT1, –57.3 (14.1); THRSP14, -30.8 (7.4); LPL, -48.4 (7.7) and AcCoA Carboxylase, -60.6 (13.0). The regression of transcript change on milk production was 0.18 for AcCoA carb and 0.26 for ATP-CL ($P <0.05$). Lipolytic control elements increased, with much variation among animals, including Ca channel subunit 338 % (203); B2AR 52.0 (8.8); PKC receptor 10.1 (2.6) and HSL mRNA 23.0 (17.9). The regression of transcript change on milk was 0.30 and 0.25 for B2AR and HSL mRNA. We now have a more complete picture of the adaptive mechanisms to lactation. Reductions in lipogenesis are primarily due to a systemic reduction in enzyme synthesis, while increases in lipolysis are a combination of increases in transcription and metabolic flux control through previously reported changes in hormonal and nervous system activity.

Key Words: Lactation, Adipose, Transcriptome