

observed between WTfat and RNfat mice. In WTfat, the gonad fat and skin fat weights were 1.16 ± 0.13 g and 0.74 ± 0.07 g versus 0.77 ± 0.08 g and 0.56 ± 0.04 g respectively ($P < 0.05$) for RNfat mice. Intriguingly, RN mutation enhanced feed consumption ($P < 0.05$) of mice both in normal- and high-energy diets. The high feed intake but increased leanness indicates that RN- allele enhances energy expenditure which warrants further studies.

Key Words: AMPK, skeletal muscle, adipogenesis

T142 Growth hormone does not stimulate IGF-I mRNA expression in bovine skeletal muscle, myoblasts, or myotubes. X. Ge and H. Jiang*, *Virginia Polytechnic Institute and State University, Blacksburg.*

Growth hormone (GH) is a major regulator of muscle growth in animals, including cattle. The growth-stimulating effect of GH had long been thought to be mediated by circulating IGF-I produced from the liver. However, recent knockout mouse studies support the concept that IGF-I produced in the skeletal muscle and IGF-I-independent mechanisms mediate the effect of GH on growth. To clarify the mechanism by which GH stimulates skeletal muscle growth in cattle, we determined whether GH increased IGF-I mRNA expression in bovine skeletal muscle in vivo and bovine myoblasts and myotubes in vitro. Five nonlactating, nonpregnant beef cows were administered subcutaneously with 500 mg of recombinant bovine GH formulated for slow release, and longissimus muscle biopsies and blood samples were taken seven days before and seven days after GH administration. Serum IGF-I concentrations, measured by an ELISA, were four times higher after GH injection than before GH injection ($P < 0.01$). Skeletal muscle IGF-I mRNA abundance, measured by real-time RT-PCR, was not different between before and after GH injection ($P > 0.1$). Western blotting analyses showed that GH administration increased the levels of phosphorylated JAK2 and STAT5, known protein components of GH signaling, in the skeletal muscle ($P < 0.05$). In the in vitro experiments, myoblasts were isolated from bovine skeletal muscle and were allowed to proliferate or induced to form myotubes in culture. Growth hormone at the concentrations of 10 ng/ml and 100 ng/ml did not affect IGF-I mRNA expression in either the myoblasts or myotubes ($P > 0.1$). These in vitro data echoed the earlier in vivo observation that GH did not increase IGF-I mRNA expression in the skeletal muscle. Taken together, the data of this study demonstrate

that GH does not increase skeletal muscle IGF-I mRNA expression in cattle and suggest that GH stimulation of muscle growth in cattle is not mediated by the locally produced IGF-I.

Key Words: growth hormone, skeletal muscle, IGF-I

T143 Early-weaning down-regulates the expression of aminopeptidase N gene in the jejunum of the piglet. D. Lackeyram*, T. Archbold, K. C. Swanson, and M. Z. Fan, *University of Guelph, Guelph, Ontario, Canada.*

Aminopeptidase N (APN) cleaves neutral AA from oligopeptides during digestion in the small intestine. The objectives of this study were to examine the responses of APN activity and protein abundances in the partitioned mucosal homogenate (H), intracellular soluble and the apical membrane (M) fractions as well as APN mRNA abundance during early-weaning in comparison with suckling pigs. A total of 20 Yorkshire piglets, 10 suckling (SU) and 10 early-weaning (WN) with an average BW of 3 kg at the age of 10 d, were used in this study. Weaning piglets were fed a corn and SBM-based diet for 12 d. Proximal jejunal samples from both SU and WN groups were collected. L-alanine-P-nitroanilide hydrochloride (0-16 mM) was used in the enzymatic kinetic experiments. Abundances of APN protein and mRNA were analyzed by Western blot and the real time RT-PCR using β -actin as the housekeeping gene. The jejunal APN maximal specific activity ($\mu\text{mol}/\text{mg protein}\cdot\text{min}$) was decreased ($P < 0.05$) in weaning piglets (H: WN, 0.122 ± 0.01 vs. SU, 0.172 ± 0.01 ; and M: WN, 0.5479 ± 0.02 vs. SU, 0.721 ± 0.01). There were decreases ($P < 0.05$) in the APN protein abundance (arbitrary units) for the WN group in the jejunal fractions (H: WN, 0.655 ± 0.03 vs. SU, 0.992 ± 0.05 ; and M: WN, 0.232 ± 0.02 vs. SU, 0.515 ± 0.01). There were correlations ($P < 0.05$) between the APN maximal specific activity and its protein abundance (H, $r = 0.87$; M, $r = 0.91$, $n = 20$). Furthermore, early-weaning decreased ($P < 0.05$) the relative abundance of APN mRNA by 39% (WN, 0.304 ± 0.02 vs. SU, 0.498 ± 0.02). The APN mRNA abundance was also correlated ($P < 0.05$, $n = 20$) with its protein abundances in both homogenate (H, $r = 0.82$) and membrane (M, $r = 0.86$) fractions. We conclude that the reduced activity of APN during early-weaning can be attributed to a down-regulation of the APN gene at the transcriptional and translational levels.

Key Words: aminopeptidase N, gene expression, weaning pigs

Horse Species

T144 Influence of extension on the stock-type western pleasure jog. M. Nicodemus* and J. Williams, *Mississippi State University, Mississippi State.*

One of the most popular performance classes for the stock-type breed is the western pleasure class. Previous research of the stock-type western pleasure gaits found the jog and lope were performed as 4-beat stepping gaits with periods of quadrupedal support. To encourage breeds to perform with more forward motion, stock-type breed associations added to the class requirements extension of the jog. Study objectives were to determine the influence of extension on the temporal variables of the stock-type jog. 5 registered stock-type horses showing in a western pleasure class were filmed at 60 Hz. Strides of both the jog and extended jog that were judged as desirable by carded judges were evaluated using frame-by-frame analysis. Means (SD) were determined for 5 strides for each horse for both gaits with variables given as % of stride. Effect

of extension for each variable was tested using a one-way analysis of variance ($p < 0.05$). Velocity (Jog: 1.5 ± 0.1 m/s, Extended: 2.2 ± 0.2 m/s) and stride length (Jog: 1.6 ± 0.1 m, Extended: 2.5 ± 0.2 m) and duration (Jog: 917 ± 67 ms, Extended: 873 ± 42 ms) were not significantly influenced by extension ($p > 0.05$). Majority of stride for both the jog (Right: Fore= $67 \pm 2\%$, Hind= $63 \pm 6\%$; Left: Fore= $66 \pm 3\%$, Hind= $64 \pm 4\%$) and extended jog (Right: Fore= $66 \pm 4\%$, Hind= $60 \pm 3\%$; Left: Fore= $64 \pm 2\%$, Hind= $61 \pm 3\%$) was spent in stance with hind stance shortening with extension ($p < 0.05$). Rhythm changed from 4 to 2 beats as extension created diagonal limb pairing during the extended jog (Jog: Right Hind-Left Fore Advanced Placement= $4 \pm 1\%$, Left Hind-Right Fore= $4 \pm 1\%$; Extended: Right Hind-Left Fore Advanced Placement= $0 \pm 0\%$, Left Hind-Right Fore Advanced Placement= $0 \pm 0\%$). While both gaits were performed as stepping gaits with the greatest % of stride spent in diagonal bipedal support (Jog: $63 \pm 7\%$, Extended: $70 \pm 5\%$), extension shortened limb support during tripodal support with 2 hind limbs (Jog: $5 \pm 3\%$,

Extended: 0±0%) and quadrupedal support (Jog: 23±7%, Extended: 20±4%) ($p < 0.05$). Although extension did not create a gait that follows current breed standards, extension did improve upon the timing and limb support of the jogging gait.

Key Words: western pleasure, stock-type horse breeds, jog

T145 Manure management practices on equine farms. M. L. Westendorf*¹, T. Joshua², S. J. Komar¹, C. Williams¹, and R. Govindasamy¹, ¹Rutgers, The State University of New Jersey, New Brunswick, ²USDA National Agricultural Statistics Service, Trenton, NJ.

According to the 2007 USDA Census of Agriculture there are over 65,000 equine farms with 465,000 horses, ponies, mules, etc. in Northeastern and Mid-Atlantic states. These farms may influence environmental and water quality due to equine manure collection, storage, spreading, and disposal practices. A manure management survey was mailed to 2000 New Jersey equine farms during the winter of 2006-2007. The study tested the hypotheses that equine producers who store or spread manure will also implement certain best management practices. Four hundred and seventy-two surveys were returned; 18.5% were training or performance farms, the remainder (81.5%) were breeding, boarding or pleasure farms. Fifty-four percent of all farms indicated that they spread manure on farm. Of those who spread, only 27% indicated having more than 8.09 hectares (20 acres) available for spreading. Seventy-four percent had a designated area for manure storage. Eighty-three percent said their manure storage was greater than 200 feet from water or wetlands; 86% percent said storage was greater than 200 feet from neighbors. Data were modeled to determine the relationship of manure storage or manure spreading with other management practices. The storage model showed that farms with 6-10 horses were more likely to have manure storages than farms not included in the model. This model had a predictive accuracy of 83.3%, and R^2 of 0.35 ($P < 0.01$). The manure spreading model showed that those who spread manure were also likely to credit manure for its fertilizer value. The spreading model had an overall predictive accuracy of 95.5%, and R^2 of 0.795 ($P < 0.01$). These results indicate that while most equine farms did not pose a direct risk to water quality or to a neighbor, some do not currently utilize best management practices in managing, spreading, or storing manure.

Key Words: manure storage, manure spreading, equine manure management

T146 Temporal variables of the Marsh Tacky intermediate gait. M. Nicodemus*¹ and J. Beranger², ¹Mississippi State University, Mississippi State, ²American Livestock Breeds Conservancy, Pittsboro, NC.

Marsh Tacky is listed as a critical horse breed according to the American Livestock Breeds Conservancy (ALBC) due to its scarce population. Originating from Colonial Spanish horses the breed can be found in the coastal marshlands of South Carolina and Georgia. Due to the breed's heritage it is thought to be gaited, but research concerning the gaits is lacking. Study objectives were to define the Marsh Tacky intermediate gait using temporal variable measurements. 10 Marsh Tacky horses selected by the ALBC as representatives of the breed were filmed performing their intermediate gait. 10 strides that were consistent with visible hoof contact and lift-off were evaluated using frame-by-frame analysis. Descriptive statistics comprising mean and standard deviation were computed for each stride variable with variables given as % of stride. Coefficient of variation (CV) expressed as a % of mean was

calculated as an indicator of variability. Velocity of the Marsh Tacky intermediate gait was 3.9±0.2 m/s (CV 5.1%) with a stride length of 2.9±0.3 m (CV 10.3%) and duration of 727±46 ms (CV 6.3%). The majority of stride was spent in stance (Right Fore: 52±3%, CV 5.8%; Left Fore: 53±5%, CV 9.4%; Right Hind: 54±2%, CV 3.7%; Left Hind: 52±4%, CV 7.7%). The gait demonstrated disassociation of the diagonal limb pairs so that the forelimb made impact before the diagonal hind (Diagonal Advanced Placements: Right Hind-Left Fore-3±1%, CV 17.6%; Left Hind-Right Fore-3±1%, CV 18.9%) and the hind limb lifted off before the diagonal fore (Diagonal Advanced Lift-Offs: Right Hind-Left Fore-5±2%, CV 26.7%; Left Hind-Right Fore-4±1%, CV 15.0%). The gait was a stepping gait alternating between periods of diagonal bipedal (85±11%, CV 12.9%), quadrupedal (7±2%, CV 28.5%), and tripedal (8±1%, CV 12.5%) support. Due to the 4-beat rhythm and the lack of suspension of the intermediate gait, the Marsh Tacky can be considered gaited. The durations, timing, and limb support are similar to the marcha batida of the Mangalarga Marchador in which both breeds share similar Spanish bloodlines. These gait variables will assist in identifying characteristics that are unique to the breed in comparison to other gaited horse breeds.

Key Words: Marsh Tacky, locomotion

T147 The use of Doppler ultrasonography to measure vasoconstriction in horses consuming endophyte-infected tall fescue. K. C. Gradert*¹, J. M. Bormann¹, S. F. DeWitt², L. W. Lomas³, J. M. Kouba¹, and T. L. Slough¹, ¹Kansas State University, Manhattan, ²Woodside Equine Clinic, Ashland, VA, ³Southeast Agricultural Research Center, Parsons, KS.

While fescue foot has not been described in horses, endophyte-produced toxins have been reported to induce vasoconstriction in equine tissue *in vitro*. This study was designed to evaluate whether the consumption of endophyte-infected tall fescue in horses would alter digital circulation *in vivo*. Twelve clinically sound, three-year-old Quarter horses with a mean BW of 459 + 31 kg were blocked by weight, sex, and HYPP status and divided into two groups: control horses ($n = 6$) received an endophyte-free diet (E-) and treatment horses ($n = 6$) received an endophyte-infected diet (E+). Fescue seed was integrated into the concentrate at a rate sufficient to bring daily ergovaline consumption in the E+ diet to a minimum of 0.20 ppm. The E- concentrate contained an equal amount of endophyte-free fescue seed. Horses were also allowed *ad libitum* access to native prairie hay, which was replaced by fescue hay from d 30 to d 60. Control horses received a low-endophyte fescue variety and treatment horses received a high-endophyte fescue variety. Based on assumed daily DM consumption of 2.0% BW, total daily ergovaline consumption was calculated to be 0.19 ppm (E-) and 1.04 ppm (E+) with the inclusion of fescue hay. Doppler ultrasonography was utilized to record velocity of flow and diameter of the medial palmar artery on d 0, d 30, d 60, and d 90. Five serial readings were taken from the left forelimb on each horse following the morning feeding. Regardless of treatment on d 30 and d 90, the regression coefficient for weight was positive ($P < 0.05$), indicating heavier horses had an increased rate of flow compared to lighter weight horses. When changes from d 0 to d 90 were compared, E+ horses had increased ($P = 0.04$) velocity and reduced ($P = 0.04$) cross-sectional diameter of the medial palmar artery. Horses consuming endophyte-infected tall fescue demonstrated vasoconstriction of the medial palmar artery as measured by Doppler ultrasonography.

Key Words: fescue, Doppler ultrasonography, horse

T148 Genistein does not work through estrogen receptors to reduce lipopolysaccharide stimulated tumor necrosis factor α release from equine peripheral blood mononuclear cells (PBMC). A. Taylor*, C. Paulson, and J. Clapper, *South Dakota State University, Brookings.*

Previously we have demonstrated that genistein, a non-steroidal tyrosine kinase inhibitor, decreased the release of tumor necrosis factor (TNF) from circulating PBMC in response to lipopolysaccharide (LPS) in the horse. Genistein also has estrogenic properties and estrogens have been shown to reduce LPS stimulated TNF production in other species. To further examine if the estrogenic properties of genistein are involved in reducing LPS stimulated TNF release in the horse the following experiment was performed. Blood (180 mLs) was collected by jugular venipuncture from a mature healthy gelding and PBMC were isolated. PBMC were then added to polystyrene tubes (4×10^6 cells/tube) and incubated at 37°C in 5% CO₂ for 2 h to allow PBMC to adhere. Non-adherent cells were decanted and PBMC were covered with cell culture media containing 1% fetal bovine serum and various concentrations of estradiol (E2), genistein, and an estrogen receptor antagonist (ICI) with and without 100 pg/mL LPS (*E. coli* O55:B5) and incubated for 6 h at 37°C in 5% CO₂ after which the supernatants were collected by centrifugation and frozen at -80°C until assayed for TNF by ELISA. Triplicate tubes were prepared for each treatment combination. Differences in supernatant concentrations of TNF were determined using the Proc Mixed procedure of SAS. Supernatant concentrations of TNF were not different ($P > .05$) in tubes containing 0.1 pM to 10 nM E2+LPS vs LPS. Supernatant concentrations of TNF were increased ($P < .05$) in tubes containing 1 nM to 10 μ M ICI+LPS vs LPS. Supernatant concentrations of TNF were increased ($P < .05$) in tubes containing ICI+E2+LPS vs LPS. Supernatant concentrations of TNF were decreased ($P < .05$) in tubes containing ICI+genistein+LPS vs LPS. These preliminary data suggest that estradiol does not inhibit LPS stimulated TNF in the horse. However, genistein does not appear to work through estrogen receptors to decrease LPS stimulated TNF production in horse.

Key Words: equine, genistein, LPS

T149 The evaluation of the miniature horse as a nutritional model for full size horses fed various levels of dietary fat. J. S. Pendergraft*¹, B. Gutierrez¹, and M. J. Arns², ¹*Sul Ross State University, Alpine, TX*, ²*University of Arizona, Tucson.*

Eight mature male horses, 4 Quarter horses (F) and 4 Miniature horses (M) were used in a split-plot design with repeated measures to evaluate the miniature horse as a nutrition model for full size horses fed various concentrations of supplemented fat. Horse size was the main-plot treatment, and 3 fat treatments were the sub-plot. Horses were considered replicates. Horse types were paired and then randomly assigned to fat treatments over time. The dietary treatments were: 0% fat supplementation (NF); 3% fat supplementation (LF); and 7% fat supplementation (HF). Soybean oil was used to supplement the treatment diets. All horses were housed individually and received a basal diet of coastal bermudagrass hay (1.37 kg/100kg BW) and a textured concentrate (.4kg/100kg BW) over three equal feedings a day and had ad libitum access to water. The supplemental fat was evenly top-dressed on the concentrate during the 0700 and 1800 feeding. The experiment periods lasted 14 d in length consisting of a 9 d adaptation period, followed by a 5 d total fecal and urine collection period. Concentrate, forage, ort, and

feces samples were analyzed for DM, CP, GE, NDF, and ADF. Urinary subsamples were analyzed for GE. Data were expressed on a body weight basis. Blood samples were obtained prior to feeding at 0600 h on d 1, 7, and 14 during each experimental period. Blood samples were analyzed for serum glucose (GLU), cholesterol (CHOL), lipase (LIPA), and triglycerides (TRIG). All data were analyzed by the PROC GLM procedure using SAS statistical software. Apparent digestibilities were not different between horse sizes when diets were supplemented with 3% and 7% fat. However, M horses had higher ($P < .02$) apparent digestibilities for DM, DE, CPADF, NDF for diets not supplemented with fat, and higher ($P < .04$) NDF digestibilities for high fat diets compared to F. There were no differences between size for ME, serum GLU, TRIG, CHOL, or LIPA. Results suggest that miniature horses consuming diets supplemented with 3-7% fat could be used to measure nutrient utilization for full size horses.

Key Words: miniature, fat, model

T150 The effect of forage nonstructural carbohydrate on glucose, insulin and lipid response in Morgan horses. L. A. Perry*^{1,2}, B. A. Younge², K. W. Cotanch¹, K. N. Lassell¹, and C. S. Ballard¹, ¹*William H. Miner Agricultural Research Institute, Chazy, NY*, ²*University of Limerick, Castletroy, Limerick, Ireland.*

Research evaluating dietary nonstructural carbohydrates (NSC) has focused on effects of varying NSC levels of concentrate meals fed horses. The objective of this study was to evaluate the metabolic response of horses fed a forage-only diet differing in NSC content (water soluble carbohydrate + starch). Six Morgan horses were assigned randomly to one of two treatments: (1) Low NSC hay (<100 g/kg) or (2) High NSC hay (~200 g/kg). Multiple hay sources were core sampled and analyzed prior to study to identify low NSC and high NSC hay. Study was conducted using a crossover design consisting of two 21-d treatment periods with 7-d washout between periods when all horses were fed Low NSC hay. Horses consumed hay (1.75% BW) twice daily (0800 and 1700). Blood and forage samples were taken prefeeding (0730) on days 1, 7, 14, and 21. On days 1 and 21 blood was sampled at 0, 1, 2, 4 and 8 h post feeding. Plasma was analyzed for glucose, insulin, and triglycerides. Neck and girth circumference, BW, and BCS, were measured on days 1 and 21. Forage composites were analyzed for starch, water soluble carbohydrate (WSC) and ether soluble carbohydrate (ESC). Data were analyzed using MIXED procedure of SAS. Forage NSC varied greatly (low NSC: 94-152 g/kg; high NSC: 137-196 g/kg). On day 1, horses fed high NSC had higher plasma glucose and insulin 2 h post-feeding (glucose 104.8 vs. 94.1 \pm 2.1 mg/dl, $P < .01$; insulin 25.0 vs. 17.5 \pm 2.2 mIU, $P < .01$). On day 21, plasma glucose was similar for both diets, however insulin levels were lower 2 h postfeeding for horses fed high NSC hay (glucose 96.8 vs. 96.7 \pm 2.6 mg/dl, $P = 0.96$; insulin 21.6 vs. 31.4 \pm 2.7 mIU, $P < .01$). High forage NSC significantly increased plasma triglycerides from 1 to 21 days while low NSC diet had no effect ($P = 0.01$). There was no effect of diet on physical measurements. This study demonstrated the variability that exists in forage NSC levels. Initially horses responded to high NSC forages with elevated glucose and insulin levels. However, by 21 d there was no difference in glucose response to diet. Forage with high NSC, particularly fructans, may increase plasma triglyceride concentrations.

Key Words: carbohydrate, forage, horse