T233 Velocity-related changes in stride variables of the intermediate gait of the Irish Draught horse of North America. Molly Nicodemus*[1], Rachel Fletcher[1], and Jeannette Beranger[2], *[1]Mississippi State University, Mississippi State, MS, [2]Livestock Conservancy, Pittsboro, NC.*

Although originating as a farming breed having a gait derived from the bloodlines of the extinct ambling Irish Hobby horse, the Irish Draught horse today is bred to excel in the sport-horse industry. With around 500 in North America, the breed is on the Livestock Conservancy's (LC) watch list. Study objectives were to determine the relationship between trotting velocities and stride variables. Ten Irish Draught horses selected by the LC and Irish Draught Horse Society of North America were worked at a slow velocity (SV) and fast velocity (FV) at the trot (SV = 4.5 ± 0.2 m/s; FV = 5.2 ± 0.1 m/s). Frame-by-frame analysis using the Ariel Performance Analysis System was performed documenting hoof contact and lift-off for 10 strides for each horse at each velocity with stride variables given as a % of stride duration. Means (SD) were calculated and student's paired *t*-tests were performed ($P = 0.05$). Both velocities demonstrated a leaping diagonal footfall sequence with diagonal limb pairs at hoof contact and periods of bipedal support (FV = 79 ± 4, SV = 87 ± 3%). While neither velocity demonstrated a 4-beat rhythm at hoof contact, the diagonal limbs disassociated at lift-off (4 ± 1%) at the SV with the hind lifting first creating a period of forelimb unipedal support (9 ± 1%). Along with limb support and coupling of diagonal limbs, fore stance duration (FV = 41 ± 3, SV = 48 ± 1%), stride length (FV = 3.8 ± 0.1, SV = 3.2 ± 0.1 m/s), and length of suspension (FV = 21 ± 4, SV = 4 ± 2%) distinguished between velocities ($P < 0.05$). Stride duration (FV = 0.70 ± 0.01, SV = 0.74 ± 0.02 s) and rate (FV = 1.41 ± 0.02, SV = 1.37 ± 0.01 strides/sec) and hind stance duration (FV = 41 ± 4, SV = 43 ± 2%) remained consistent between velocities. The North American Irish Draught horse did not demonstrate an ambling gait at velocities measured. Nevertheless, pattern and timing of the disassociated limbs and resulting limb support are unique compared with breeds that are today used in the breeding of the Irish Draught horse such as the Thoroughbred and Warmblood, and thus, potentially reflecting gait characteristics of the extinct Irish Hobby horse.

Key Words: stride variable, trot, velocity

T234 Influence of fibrolytic enzymes and yeast addition in horse's diet on digestibility, blood chemistry and fecal coliform. M. M. Y. Elghandour[1], A. E. Kholif[2], A. Z. M. Salem*[1], J. C. Vázquez Chagoyán[1], J. S. Martínez Castañeda[1], L. M. Camacho[3], R. Montes de Oca[1], and T. A. Morsy[2], *[1]Facultad de Medicina Veterinaria y Zootecnia, Universidad Autónoma del Estado de México, Toluca, Estado De México, Mexico, [2]Dairy Science Department, National Research Centre, Giza, Egypt, [3]Unidad Académica de Medicina Veterinaria y Zootecnia, Universidad Autónoma de Guerrero, Altamirano, México.*

Improving fibrous feeds utilization ensures low consumption of high-starch grains and may reduce various pathologies. Fibrolytic enzymes (FE) like cellulase and xylanase, and yeast addition can improve fibrous feeds utilization. Therefore, the current study aimed to study the effect of FE (Exp. 1) and yeast (Exp. 2) on the utilization of diet with 11.2% CP and 51.1% NDF. Thirty-two mares of Quarter Horse (450-500 kg BW) were used in a complete randomize design for 15 d, with 10 d for adaptation and 5 d for samples collection. For Exp. 1, mares (n=16) were distributed onto 4 treatments to be fed on the basal diet without FE (control), or plus cellulase at 10 mL/mare/d (CELL), plus xylanase at 10 mL/mare/d (XYL), or plus mixture cellulase and xylanase at 5 ml of each enzyme/mare/d (CX). For Exp. 2, mares (n=16) were distributed onto 4 treatments to be fed on the basal diet without yeast (control), or fed the control diets plus Procreatin 7 (1.5×10^{10} cfu/g *S. cerevisiae*) at 15 g/mare/d (P7), plus Biocell F53 (2×10^{10} cfu/g *S. cerevisiae*) at 11 g/mare/d (F53), or plus Biosaf SC47 (1.5×10^{10} cfu/g *S. cerevisiae*) at 15 g/mare/d (SC47); yeast products were in powder form. Both of enzyme and yeast doses were mix with the 1 kg of concentrate diet at 0400 h. Mares were fed the concentrates twice daily at 0400 and 1600 h, while the forage of oat straw was offered ad libitum at 0500 and 1700 h. Acid insoluble ash concentrations in feed and fecal samples were used for digestibility determination. No effects for FE and yeast were obtained in blood alanine transaminase, aspartate aminotransferase, urea, creatinine, total protein and glucose. Addition of CELL, XYL and CX increased ($P = 0.001$) nutrient intakes from oat straw versus control. Moreover, CELL, XYL and CX increased ($P < 0.05$) digestibilities of DM, OM, NDF and ADF. Enzymes decreased ($P < 0.05$) concentration of fecal coliform. In the contrary, F53 increased nutrients intake of oat and nutrients digestibility ($P < 0.05$) without difference compared with other treatments. Yeast had no effect on fecal coliform concentration. It could be concluded that addition of FE at 10 mL/mare/d or addition of Biocell F53 at 11 g/mare/d improved feed intake and nutrients digestibility without affecting mare's blood parameters.

Key Words: enzyme, horse, yeast

T235 Changes in salivary IgA and nasopharyngeal leukocyte populations in response to prolonged head elevation. Jill M. Bobel*, Megan R. Di-Lernia, Jeffrey R. Abbott, Maureen T. Long, and Lori K. Warren, *University of Florida, Gainesville, FL.*

Prolonged head elevation is thought to be a major contributor to the increased risk of respiratory disease associated with transportation in horses. Prior investigations have focused on immunological changes in the lower respiratory tract. The aim of this study was to characterize the response to head elevation in the upper respiratory tract. Twelve horses (mean ± SEM, 552 ± 10 kg; 11.5 ± 1.4 y) were tethered for 12 h with their heads elevated at a height of 1.5 m to induce physiological stress. While tied, horses had unlimited access to bermudagrass hay and were offered water every 2 h. Each horse underwent head elevation on 4 occasions, each separated by 30 d. When not tied, horses were maintained on pasture forage. Nasopharyngeal flush (NPF) and saliva samples were obtained before head elevation, immediately after (0 h), and 12, 24, and 72 h post head elevation. Mucus content and leukocyte populations were quantified in NPF and IgA was measured in saliva. Data were compared using mixed model ANOVA with repeated measures. NPF samples contained more mucus at 0 h post ($P < 0.02$) compared with pre-head tie samples. Percentage and number of neutrophils in NPF increased at 0 h post ($P < 0.0001$) and the number remained elevated through 72 h ($P = 0.04$). Lymphocytes, monocytes, and eosinophils in NPF increased in number ($P < 0.05$) but decreased in percent ($P < 0.05$) in response to head elevation. While the proportion of these cells normalized by 72 h, the numbers declined to levels lower than pre-head tie values. Percentage of CD8+ T cells and B cells in NPF were lower at 0 h post ($P < 0.05$) and returned to baseline by 12 h. Percentage of CD4+ and the ratio of CD4+ to CD8+ cells increased at 0 h post ($P < 0.01$) and remained elevated through 72 h ($P < 0.05$). Salivary IgA

increased at 0 h post ($P < 0.0001$), then decreased below baseline at 12 h post ($P < 0.0001$) and remained lower than pre-head elevation values through 72 h ($P < 0.05$). The immunological changes observed in the upper respiratory tract agree with those reported for lower tract, and highlight increased risk of disease following prolonged head elevation.

Key Words: immunosuppression, upper respiratory disease, horse

T236 Utilization of the equine SNP 70 beadchip in monitoring inbreeding and describing the genetic background in an Arab horse herd. Mohammed Al Abri*[1], Samantha Brooks[2], and König von Borstel[3], *[1]Cornell University, Ithaca, NY, [2]University of Florida, Gainesville, FL, [3]Göttingen University, Göttingen, Germany.*

Horse breeders rely heavily on the accurate identification of individual ancestry through pedigrees. Errors in such pedigrees may inaccurately assign horses to false lineages or breed memberships, and can result in inaccurate estimates of inbreeding. Discrepancies in pedigree records can lead horse owners into making misguided purchasing and breeding decisions. Genome-wide SNP data provide a robust and precisely quantitative tool to resolve many lineage assignments and provide genomic measures of inbreeding values. The aim of this project was to pilot a comparison between pedigree and genomic relatedness measures. Here, we describe a herd of 36 pedigreed Egyptian Arabian horses genotyped using the Equine SNP70 (Geneseek, Inc.) Genomic inbreeding values and pair-wise relatedness between horses within the herd were estimated from the genotypic data. Pedigree derived inbreeding values were significantly correlated with the genomic inbreeding values across the population ($r = 0.44$, $P = 0.007$), although within an individual the 2 values could differ substantially. Genomic inbreeding values were also positively correlated with the year of birth ($r = 0.29$, $P = 0.086$), demonstrating a trend in inbreeding over the years. A multi-dimensional scaling analysis (MDS), a phylogenic analysis and a clustering analysis were preformed to compute the relationships between the horses and the results were compared with the pedigree information. These same analyses were also conducted for the herd among US, Polish, and Egyptian Arabian horses to examine their Arabian ancestry. The within herd analysis was successful in recapturing much of first-degree relationships, although more distant relationships were not entirly reconstructed. The herds' Egyptian lineage was successfully assigned among Arabian horse sub-groups. It can be concluded that genome-wide genotypes are good alternatives to compute inbreeding and verify the integrity of pedigrees in horses in cases where records are unavailable or in doubt. Although costly, application of this tool in a breeding program can enable informed mating decisions and assess inbreeding in lines of horse already at risk of loosing genomic diversity.

Key Words: Arabian horse, pedigree inbreeding, genomic inbreeding

T237 Evaluation of single nucleotide polymorphisms effects on injury predisposition in a population of multi-discipline athletically trained horses. Sarah Mercer[1], Neely Walker[2], and Matthew Garcia*[1,2], *[1]Louisiana State University, Baton Rouge, LA, [2]LSU AgCenter, Baton Rouge, LA.*

Tendon and ligament injuries (TLIs) in the performance horse represent a significant burden to the equine industry. Furthermore, treatment is often unsuccessful and re-injury common, prevention of TLIs is a major goal. The objective of the current study was to evaluate a population of athletically trained horses from multiple disciplines for SNP located 5 candidate genes in association with increased injury predisposition or injury resistance. A total population of 63 performance horses with documented injury history or injury resistance was utilized for the current study. Specifically, the study was comprised of 25 horses of various ages with at least one injury and 33 horses of various ages with no injury history. A 5mL blood sample collected from all horses via jugular venipuncture and DNA was subsequently extracted from white blood cell buffy coats for SNP genotyping. The 5 candidate genes selected for evaluation included the Angiotensin 1 converter enzyme gene (ACE), the ATPase α 2 peptide gene (ATP1A2) the Bradykinin receptor B2 gene (BDKRB2), the Collagen type 1 α 1 gene (COL1A1) and the Collagen type 5 α 1 gene (COL5A1). Candidate genes in the current study have been previously reported to be associated with jumping ability (ACE), racing ability (ATP1A2), and ligament and tendon injuries (BDKRB2, COL1A1 and COL5A1). A total of 64 single nucleotide polymorphisms (SNP) were selected across all candidate genes (ACE = 11, ATP1A2 = 14, BDKRB2 = 9, COL1A1 = 14, COL5A1 = 16). A mixed model analysis was utilized for the current data set with independent variables of sex, breed, age, discipline, training start age, first competition age, first age of injury, and number of years in competition. Independent effects included injury status (injured or not injured) and inherited SNP genotype for each unique SNP located on the previously described candidate genes. Although multiple SNP were identified in the current study as being associated with injury susceptibility/resistance, these SNP and a greater number of candidate genes must be evaluated in larger more diverse populations before implementation into selection strategies.

Key Words: equine, injury, SNP

T238 Effect of oil supplementation on milk IgG, serum insulin, glucose, placental efficiency, and immune status of foals. Lauren B. Hodge*, Brian J. Rude, Caleb O. Lemley, and Toree L. Bova, *Mississippi State University, Starkville, MS.*

The objective of this research was to evaluate the effects of supplementing pregnant mares with omega-3 fatty acids and how this may affect the suckling foal. Pregnant mares (n = 18) were randomly assigned to 1 of 3 diets beginning 28 d before their expected foaling date until 84 d after foaling. Diet 1 was a commercial feed fed to meet NRC requirements, based on forage analysis; diet 2 was diet 1 plus a blended fish oil; and diet 3 was diet 1 plus a blend of fish and soybean oil. Blood samples were collected 28 d before their expected foaling date, and at 14 d increments. Placental efficiency was calculated as a ratio of placenta weight:foal weight. Milk samples were obtained at foaling and on remaining blood collection days. Body weights were recorded the same day blood samples were collected. Placentas were weighed and ~5 g sample taken. No differences were found for mare plasma IgG ($P = 0.1318$), serum insulin ($P = 0.3886$), plasma glucose ($P = 0.2407$), or milk IgG ($P = 0.1262$) concentrations for treatment or time period of sampling. Mare packed cell were not different ($P = 0.0885$) among treatments; however, decreased ($P < 0.0001$) as the trial progressed. Mare body weight and body weight change were not different ($P = 0.5704$; $P = 0.08265$, respectively) among treatments, or relative to time of sampling. Mare body weight change did differ ($P < 0.0001$) relative to time. Foal plasma IgG ($P = 0.2767$), serum insulin ($P = 0.4843$), or plasma glucose ($P = 0.1204$) were not affected by treatment or time of collection. Foal packed cell was not different ($P = 0.6275$) among treatments, however, there was a difference ($P = 0.0005$) relative to time of sampling. Foal body weight change and total gain were not different among treatments ($P = 0.6825$; $P = 0.8220$, respectively); however foal body weight were least ($P = 0.0041$) for foals consuming diet 1 and greatest for foals consuming diet 3, with foals consuming diet 2 being intermediate. Foal body weight change decreased as the trial progressed ($P < 0.0001$). Placental efficiency and nitrites were not different ($P =$

0.1631; $P = 0.5604$, respectively) among treatments. Research should be conducted to evaluate supplementation earlier in gestation.

Key Words: horse nutrition, fat, supplementation

T239 Testis tissue explant culture supports the viability of equine spermatogonial stem cells. Kyle C. Caires*[1], Louie Y. Chen[2], Rachel A. Lemcke[1], and Laurie A. Seigler[1], *[1]Berry College, Department of Animal Sciences, Mount Berry, GA, [2]RDBL, National Institute of Environmental Health Sciences, National Institutes of Health, Research Triangle Park, NC.*

Spermatogenesis is a stem-cell dependent process that supplies an indefinite supply of spermatozoa during adult male life. Sperm production is well characterized in rodents and depends on a milieu of endocrine and growth factors, but little is known regarding the regulation of male fertility and spermatogonial stem cells (SSC) in other domestic animals. The objective of the present study was to develop an organ culture system to investigate spermatogenesis in horses. To accomplish this aim we obtained testicular parenchyma from pre- and post-pubertal stallions (n = 3 for each age group) and cultured those tissues upon 0.4-μm membranes in XC medium (DMEM supplemented with 1-, 5- and 10% fetal bovine serum) over a weeklong period with alternate-day media changes. All data sets were evaluated using ANOVA and differences between means were considered significant at $P < 0.05$. Immunohistochemical analysis of *PLZF*, *GFRA1*, and *DDX4* expression were used to confirm the identity of undifferentiated spermatogonia, differentiated spermatogonia and meiotic germ cells, whereas *GATA4* was used as a marker for Sertoli cell and Leydig cell populations. Histological analysis of fresh and cultured testis tissue indicates the survival of germ-line stem cells and somatic cells populations within morphologically normal seminiferous tubules at 24, 48, and 72 h during the culture period with different levels of serum. Sertoli cell and Leydig cell populations remained stable ($P > 0.05$) throughout the culture period, but the appearance of pyknotic nuclei in germ and somatic cells observed following 120 h of culture indicates limitations of our approach for long-term tissue maintenance, and this notion is confirmed by TUNEL analysis of apoptosis. Investigating the effect of specific growth factor and hormone treatment combinations on the biological activity and stem cell behavior of undifferentiated spermatogonia is ongoing. Collectively, these results demonstrate successful culture of equine spermatogonial stem cells, and provide a useful ex-vivo model for investigating spermatogenesis in stallions without the costs typically associated with whole-animal experimentation.

Key Words: equine testis, stem cells, spermatogonial stem cells

T240 Valerenic acid detection in equine urine after administration of calming supplement. Celina M. Checura*[1], Nikki McGreevey[1], Travis J. De Wolfe[1], Simon F. Peek[1], Greg A. Barrett-Wilt[1], Richard G. Godbee[2], and Benjamin J. Darien[1], *[1]University WI-Madison, Madison, WI, [2]Central Garden & Pet, Phoenix, AZ.*

Valerian root (*Valeriana officinalis*) is an herbal tranquilizer used in horses. However, many plants and herbs are classified as forbidden by the USEF Equine Drugs and Medications Rule due to their potential to affect performance. The aim of this study was to establish a reliable withdrawal period before competition for trainers considering valerenic acid supplements as a training aid. Ten mares between 10 and 20 years of age were administered an oral paste of valerian root extract (36 mg) daily for 5 consecutive days. Urine samples were collected by sterile catheterization before the first and all subsequent treatments. Urine was also collected at 4, 8, 24, 48, and 72 h after the last treatment. Urine samples were coded for blinding and stored at 4°C until testing. Valerenic acid in urine was analyzed using a triple-quadrupole mass spectrometer coupled to an Agilent 1100 capillary HPLC system. Quantitation of urine valerenic acid was done by generating a calibration curve using the peak areas of 3 replicate injections for each calibration point. Two different calibration curves for quantitation of valerenic acid in urine were created. One curve was generated by spiking valerenic acid at 0 (blank), 25, 50, 100, 200, and 400 ng/mL into known blank urine before protein precipitation and solid-phase extraction. A second curve was generated for later samples by pooling 2 mL from each of 4 blank urine samples and then adding valerenic acid as described. Regression analysis of a representative calibration curve generated from these standards yielded a linearity of $R^2 = 0.9969$. Based on the signal-to-noise ratio of the 25 ng/mL calibration standard, the limit of quantitation was estimated at 20 ng/mL. From a total of 90 post-valerenic acid treatment urine samples, 3 samples were positive. Urine samples obtained from one horse was positive for valerenic acid on d 2 (21 ng/mL) and 3 (29.5 ng/mL) of treatment. A different mare tested positive (67.8 ng/mL) 4 h after the last dosage (d 5). Based on our results, horses should be withdrawn from competition for at least 5 d after the oral administration of 36 mg valerenic root extract.

Key Words: valerian, herbal, tranquilizer

T241 Influence of α-linolenic acid supplementation in mature horses at maintenance: Body composition. Jessica L. Leatherwood*[1], Emily D. Lamprecht[2], Mark J. Anderson[1], Kyle J. Stutts[1], Marcy M. Beverly[1], and Stanley F. Kelley[1], *[1]Sam Houston State University, Huntsville, TX, [2]Cargill Incorporated, Elk River, MN.*

Twenty mature horses (455 to 457 kg and 5 to 10 yr) were utilized in a randomized complete block design to evaluate incorporation of α-linolenic acid into plasma and the effects of supplementation on performance variables in mature horses. Horses were blocked by BW, age, and sex and randomly assigned to treatment within block for a 112 d trial. Dietary treatments included control (no α-linolenic acid; CON) or 150 mg/kg BW/d α-linolenic acid (ALA) derived from a flaxseed oil (TRT; Clear Valley Omega 3 Oil; Cargill, Inc., Eddyville, IA). Diets consisted of CON horses (n = 10) fed 0.25% BW (as-fed) concentrate only or TRT horses fed (n = 10) the same concentrate with additional ALA offered at 12 h intervals. Horses were housed by block and maintained in adjacent dry lots with ad libitium access to coastal Bermudagrass (Cynodon dactylon) hay. Body weight and BCS were obtained every 14 d and concentrate was adjusted accordingly. Rump fat (RF), longissimus dorsi area (LD), and longissimus dorsi fat thickness (LDF) were obtained every 28 d via ultrasonography. Blood samples were collected on d 0, 56, and 112 to determine plasma fatty acid concentrations by gas chromatography. Data were analyzed using the PROC MIXED procedure of SAS. Fatty acid levels in the oil supplement remained stable over the course of the trial. Plasma concentrations of ALA were greater ($P \leq 0.01$), and plasma levels of arachidonic acid were lower ($P \leq 0.01$) in TRT compared with CON beginning at d 56 to d 112 of the study. Body weight and BCS were not influenced ($P = 0.96$ and $P = 0.14$, respectively) by dietary treatment; however, all horses gained BW and BCS throughout the trial ($P \leq 0.01$). Rump fat, LD and LDF were not influenced ($P \geq 0.54$) by dietary supplementation although, performance variables increased ($P \leq 0.01$) over time across treatments. These results indicate that this source of ALA is stable and will incorporate into circulation through targeted supplementation; however, further studies are needed to fully elucidate effects of dietary ALA supplementation to mature horses.

Key Words: equine, α-linolenic acid, longissimus dorsi

T242 In vitro evaluation of protein content on forage digestion using equine fecal inocula. Tayler L. Hansen*, Brooke M. Eubanks, Emily K. Rizzo, and Lori K. Warren, *University of Florida, Gainesville, FL.*

Previous research has indicated crude protein (CP) and neutral detergent fiber (NDF) concentrations are related to forage digestibility by horses. However, it is unclear if greater CP concentrations are simply correlated with higher quality forage or if CP amounts influence digestion and microbial fermentation in the hindgut by increasing nitrogen substrates to cellulolytic bacteria. The objective of this study was to determine the influence of varying amounts of CP on forage digestibility using an in vitro hindgut fermentation model. We hypothesized increasing CP presence would increase in vitro dry matter digestibility (IVDMD), NDF digestibility (NDFD), and acid detergent fiber digestibility (ADFD). Quadruplicate 0.5 g forage samples (alfalfa, bermudagrass, and orchardgrass) and cellulose (Sigma-Aldrich) were incubated at 37.5°C for 48 h in an ANKOM DaisyII Incubator. Casein (Sigma-Aldrich) was added to the digestion jars at 1 of 4 levels (no added casein, 66.1, 125.9, and 188.8 g) to represent control, NRC CP recommendations, industry-typical diets, and 3× NRC recommendations, after adjusting for 51% prececal CP digestibility. Freshly voided fecal samples were collected on 4 separate days from mature horses with ad libitum access to bermudagrass hay to serve as microbial inoculum for DaisyII runs (n = 4). Data were analyzed using ANOVA (SAS, v 9.3) as a split-plot design with the main plot as casein level and subplot as forage type with an error term of casein level × block. Means were separated using LSD comparisons. In vitro DMD decreased ($P < 0.0001$) as added casein increased. Control IVDMD across all forage types (47.5 ± 9.6%, mean ± SE) was greater than ($P < 0.05$) IVDMD for all added casein treatments (30.7 ± 13.0, 32.3 ± 13.6, 28.0 ± 11.2%, as casein level increased). Both NDFD and ADFD were greater ($P < 0.0001$) in control compared with casein-added treatments. In vitro DMD, NDFD, and ADFD differed ($P < 0.0001$) by forage type. Casein as a protein source may have affected results with this closed-system in vitro model. Further evaluations are needed to determine the relationship between protein in the equine hindgut and fiber digestion in vivo.

Key Words: DaisyII, fiber, horse

T243 Changes in plasma calcium and phosphorus concentrations in mares fed decreasing amounts of dietary Ca and P just prior to weaning. Ashley L. Fowler*, Brittany E. Harlow, Morgan B. Pyles, Susan H. Hayes, Andrea D. Crum, and Laurie M. Lawrence, *University of Kentucky, Lexington, KY.*

During lactation, the concentrations of Ca and P in milk are highly conserved, frequently at the expense of bone mineral. Replacement of bone mobilized during lactation and dietary changes in the post- weaning period may further alter Ca and P metabolism, but it is unknown if these changes influence the ability to achieve Ca and P homeostasis. The objective of this study was to examine changes in plasma Ca and P in lactating mares fed decreasing amounts of Ca and P just before weaning. Four mares in late lactation (12 ± 5.7 yr; 527 ± 26 kg) were transitioned to a diet containing Ca and P in amounts appropriate for nonlactating mares for 14 d, weaned and then maintained on the same diet for 7 d. Four nonlactating mares (11 ± 4 yr; 552 ± 33 kg) received the same diet and served as the controls. We hypothesized that low mineral intake during late lactation would stimulate bone resorption (increasing blood Ca and P) and that in the post-weaning period there would be bone deposition (decreasing blood Ca and P). Blood samples for P and Ca analysis were obtained 14 d before weaning (dietary Ca and P equal to or exceeding requirements), at 2 d before, and 7 d after weaning (dietary Ca and P adequate for nonlactating mares). Saliva samples were taken 7 d post-weaning for P analysis. Data were analyzed using ANOVA with repeated measures with group (lactating or nonlactating) and sample date as main effects. Plasma P increased from baseline to pre-weaning ($P < 0.05$) across groups and did not change further post-weaning. The increase in plasma P at 2 d pre-weaning might reflect an increase in bone resorption as a result of decreasing amounts of dietary P, but a decrease in plasma P 7 d post-weaning was not observed. Salivary P was not different between groups post-weaning. Plasma Ca decreased from baseline to pre-weaning ($P = 0.053$) across groups, but did not change further post-weaning. The Ca:P ratio in the blood decreased from 3.7:1 at baseline to 2.8:1 at pre-weaning ($P < 0.05$). From these data, it appears that changes in Ca and P intakes influence blood Ca and P more than the physiological change of lactation cessation.

Key Words: horse, lactation, mineral

T244 Relationship between training difficulty and aggression in horses. M. J. Anderson*, J. L. Leatherwood, K. Jones, K. J. Stutts, M. M. Beverly, and S. F. Kelley, *Sam Houston State University, Huntsville, TX.*

Training of horses involves a balance of trust and dominance between animal and trainer. Yet in a herd, horses will establish a social hierarchy, which lends to antagonistic behaviors. The objective of this research was to characterize behavior and interactions between horses and determine the implications on the trainability of the horse by trainers of varying skill levels. To accomplish this test, horses (n = 10) from the Sam Houston State University behavior and training course were randomly selected and individually introduced in 9.14-m round pens to control horses (n = 5) over a 2-wk period. The interaction between the horses was recorded by 3 independent observers and scored on a 15 cm line scale ranging from submissive (1) to aggressive (15). Horses were individually worked by one of 5 trainers of varying skill levels. Activities included ground work only, and maneuvers began with haltering and leading to advanced maneuvering through obstacle courses. Horses were blindly scored by the same 3 observers using a 15 cm line scale ranging from obedient (1) to resistant (15). Horses were then placed into 2 groups (Difficult, Easy) based on average difficulty of training, and the aggression scores were compared across the 2 groups using the PROC GLM procedure in SAS 9.2. Correlations were calculated between the average aggression and training scores for all test horses. Results from the training scores showed a difference ($P < 0.01$) between the 2 selected groups, indicating that the separation of the groups was valid. However, no difference was detected ($P = 0.22$) in aggression scores between the Difficult and Easy training groups. This illustrates that the aggression of a horse in a herd or in establishing a social order may not be a good indicator of the difficultly to subsequently train the horse. This is further demonstrated in the weak correlation (r = 0.408) between the average aggression and training scores for the test horses. While in establishing social order aggression may be observed, but it is not the sole factor in determining dominance in a herd. Most likely the trait of dominance may be more important in terms of trainability than aggression and should be the focus of future behavior research.

Key Words: horse, behavior, training

T245 The occurrence of different mycotoxins (aflatoxins, fumonisins, zearalnenone, ochratoxin, deoxynivalenol, ergot alkaloids) in horse feed. Nicole Reisinger*[1], Paula Kovalsky[2], Verena Starkl[2], Simone Schaumberger[2], Michael Sulyok[3], and Ursula

Hofstetter[2], [1]*Biomin Research Center, Tulln, Austria,* [2]*Biomin Holding GmbH, Herzogenburg, Austria,* [3]*Center for Analytical Chemistry, Department for Agrobiotechnology (IFA-Tulln), University of Natural Resources and Life Sciences (BOKU), Tulln, Austria.*

Mycotoxins are toxic secondary metabolites of fungi, which can be found in cereal grains and animal feeds. The occurrence of mycotoxins in feed of livestock animals is well investigated. In contrast, only limited information is available on the occurrence of mycotoxins in horse feed as well as on their effects on horse health. The aim of this study investigate the occurrence of the mycotoxins aflatoxins (Afla), fumonisins (FUM), zearalenone (ZEN), ochratoxin A (OTA), deoxynivalenol (DON) and ergot alkaloids in horse feed in Europe. Hay, straw and concentrate feed samples (n = 135) were collected from different countries of Europe in 2014. Samples were analyzed by liquid chromatography – tandem mass spectrometry (Streit et al., 2013). In total, mycotoxins could be found in 83% of all tested samples. Ergot alkaloids (e.g., ergotamine, festuclavine) were detected in 63% of the samples and therefore represented the most frequently occurring mycotoxin in our survey. In the individual samples, concentrations up to 1,230 µg/kg were determined. In addition, a high prevalence of DON (60%) and ZEN (54%) was observed. Maximum concentrations of 3,800 µg/kg and 200 µg/kg were measured for DON and ZEN, respectively. FUM was detected in 16% of the feed samples, maximum concentration measured was 330 µg/kg. In contrast, Afla (1.5%) and OTA (3%) were detectable in only a few of the feed samples. Results of this study clearly show that mycotoxins are frequent contaminants of horse feed, although usually only high quality feed is given to horses. There are only scattered reports on the effects of mycotoxins on horses. However, it is known that horses are extremely sensitive to FUM (Caloni et al., 2011) and also negative effects of ZEN have been reported (Minervini et al., 2006). Furthermore, ergot alkaloids are described to induce dystocia, agalactia and lameness in horses (Cross et al., 1995; Douthit et al., 2012). Therefore, an effective mycotoxin risk management program can help to protect horses against negative effects of mycotoxins.

Key Words: horse, equine, mycotoxins

T246 Effectiveness of a brewer's yeast supplement with or without fat for performance horses. Jeneva R. Seidl*[1], Toree L. Bova[1], J. Latham Brister[1], Lauren B. Hodge[1], Angela R. Mays[2], and Brian J. Rude[1], [1]*Mississippi State University, Starkville, MS,* [2]*F.L. Emmert Company, Cincinnati, OH.*

The objective of the current trial was to evaluate the effect of additional fat to a brewer's yeast supplement on hoof, coat, and body condition of performance horses. Twelve performance geldings randomly allotted to one of 3 dietary treatments: 1) a commercially available horse feed (10% CP, 4.5% fat) at 0.9% BW/d; 2) diet 1 plus a brewer's yeast product at 113g/d; 3) diet 2 plus vegetable oil at 5% of the diet. Diet 1 was given to meet basic nutrient requirements of 10% CP and 0.6% LYS. All geldings were fed half of their diet treatment twice a day for 84 d. Geldings had ad libitum access to bermudagrass pasture and hay throughout the trial. Body weight (BW) measurements and body evaluations were collected at initiation of the trial and every subsequent 28 d until 84 d. Body evaluations included coat condition, body condition score (BCS), and hoof condition. Coat and hoof condition were evaluated on scale ranging from 1 to 5 (1 reflecting poor or damaged and 5 reflecting glossy) accounting for condition, texture and appearance. Body condition was based on the standard BCS scale of 1 to 9. Four total evaluations were taken during the experiment, an initial evaluation and then every 28 d. Data were subjected to ANOVA using the GLM procedures of SAS. No effect of diet was found for hoof (3.5, 3.4, and 3.1; $P = 0.6207$), coat (3.3, 3.3, and 3.3; $P = 0.0826$), or BCS (5.3, 5.6, and 5.6; $P = 0.9967$) for diets 1, 2, and 3, respectively. Body weights were not different (509, 524, and 560 kg; $P = 0.4602$) among diets 1, 2, and 3, respectively, nor was the change in body weight during the trial different (1.9, −6.8, and 4.5 kg, respectively; $P = 0.6815$). Addition of fat to brewer's yeast supplement did not enhance body scores or weight change. Diet 1 (basal diet fed to all treatments) was a concentrate based supplement containing a large amount of available energy. Feeding diet 1 at 0.9% BW/d may have masked the effects of increased energy from fat. Further research should be conducted to evaluate brewer's yeast product and fat with horses while being fed a less nutritious basal diet.

Key Words: equine, brewer's yeast, fat supplementation

T247 Effectiveness of a brewer's yeast supplement with or without fat for weanling horses. J. Latham Brister*[1], Toree L. Bova[1], Jeneva R. Seidl[1], Lauren B. Hodge[1], Angela R. Mays[2], and Brian J. Rude[1], [1]*Mississippi State University, Starkville, MS,* [2]*F.L. Emmert Company, Cincinnati, OH.*

The objective of this study was to evaluate the effect of additional fat to a brewer's yeast supplement on hoof, coat, and body condition of weanling horses. Twelve Quarter Horse weanlings (7 colts and 5 fillies) were utilized in an 84 d feeding trial using 3 dietary treatments: 1) a commercially available horse feed (10% CP, 4.5% fat) at 0.9% BW; 2) diet 1 plus a brewer's yeast product at 113g/d; 3) diet 2 plus vegetable oil at 5% of the diet. Weanlings were fed half of their ration twice daily (0600 and 1800 h) with all 3 treatments receiving the diet 1 portion of their treatment at 0.9% BW/d, to meet NRC requirements based on forage analysis. Body weight (BW) measurements and body evaluations were collected at initiation of the trial and every subsequent 28 d until 84 d. Body evaluations included coat condition, BCS, and hoof condition. Coat and hoof condition were evaluated on a scale ranging from 1 to 5 (1 reflecting poor or damaged and 5 reflecting glossy), with coat evaluation scale accounting for condition, texture, and appearance. Body condition score was based on the standard BCS scale of 1 to 9. During the first 7 d post weaning, weanlings were divided into groups of 3 (1 weanling randomly assigned to each treatment) and housed together in open lot to reduce stress. Weaned groups were offered ad libitum access to hay and water while introduced to the diet 1 portion of their dietary treatment. Diets were determined as a percent of the weanling's BW, which was adjusted every 28 d. Weanlings were also provided ad libitum access to grass, bermudagrass hay, and water. Data were analyzed through ANOVA using the GLM procedures of SAS. As expected, BW increased throughout the trial for all 3 treatments. After 28 d foals on diet 2 and 3 had increased BW ($P = 0.0102$; 211 and 207 kg, respectively) compared with foals on diet 1 (194 kg). There were no differences for BCS ($P = 0.2768$), coat score ($P = 0.3243$), or hoof score ($P = 0.5014$). Supplementation with brewer's yeast increased BW gains, but additional fat did not affect horses on the concentrate diet. In conclusion, added fat did not increase brewer's yeast affects in weanling horses.

Key Words: equine, brewer's yeast, fat supplementation