

232 Real-time PCR quantification of reproductive hormone receptor gene expression in superovulated MOET donor cows. S. Wise*¹, M. A. Okomo-Adhiambo¹, D. Joos¹, W. Rauw¹, A. Rink², and L. Gomez-Raya¹, ¹University of Nevada, Reno, ²Animal Disease and Food Safety Laboratory, Reno, NV.

Multiple ovulation and embryo transfer (MOET) facilitates production of offspring from cattle of high genetic merit world-wide. Twelve multiparous non-lactating donor cows were synchronized to estrous by intravaginal insertion of 1.38g progesterone (Eazi-Breed™ CIDR®), in addition to intramuscular injection of 1ml combination of progesterone (150mg/ml) and 17-β-estradiol (10mg/ml). Evening of day 5, 5ml FSH (Folltropin® 20mg/ml) was given intramuscular, then every 12 hrs (AM and PM) over the next 4 days. Morning and evening of day 8, corresponding to day 4 of FSH treatment, 10ml prostaglandin (Lutalyse® 5mg/ml) was given intramuscular. Morning of day 9, the last FSH shots were given and CIDR® devices removed. Cows were monitored for heat and bred thrice by AI, and embryos collected 7 days after breeding. Superovulation and embryo flushing protocol was performed twice for all cows, yielding average 0-30 embryos. Blood and serum samples for nucleic acid and hormone biochemical analysis, respectively, were taken throughout the experimental period. Quantitative real-time PCR (QRT-PCR) was used to profile expression of 5 reproductive hormone receptor genes (ESR1, FSHR, LHR, OXTR and PGR), using total RNA isolated from blood sampled over 84 hrs of FSH treatment (0h, 12h, 24h, 36h, 48h, 60h, 72h, 84h), from two cows that yielded the highest number of embryos (30 and 24, respectively) and from the cow that yielded none. All five genes were significantly over-expressed (expression ratio ≥2.0 and t-test p-value ≤0.05) in the best performing cows (#119 and #128) compared to the least performing cow (#134), in at least five of the eight experimental time-points. In cow #119 compared to #134, ESR1 was up-regulated 2- to 4-fold (0-48h) and by 10-fold at 84h, while FSHR, LHR, OXTR and PGR were significantly up-regulated at all time-points. Similar results were observed in cow #128 compared to #134, suggesting that expression of hormone receptor genes is vital to a donor cow's physiological response to reproductive hormone treatments administered for superovulation.

Key Words: MOET, Reproductive Hormone Receptor Genes, Quantitative real-time PCR

Companion Animals: Companion and Comparative Animal Nutrition

234 Effect of gut-loading time on nutrient content of adult feeder crickets. C. L. Dikeman*, S. D. Plesuk, D. L. Klimek, and L. G. Simmons, *Omaha's Henry Doorly Zoo, Omaha, NE.*

Insectivorous amphibians and reptiles in captivity are typically limited to diets consisting primarily of feeder crickets. Without supplementation, farmed feeder crickets lack proper nutrient profiles to sustain the health of preying animals. While gut-loading of crickets is common practice among herpetologists, management of gut-loading crickets is poorly defined. The objective of this experiment was to determine optimal gut-loading time to best improve the nutrient profiles of feeder crickets for amphibians and reptiles in captive environments. Farmed crickets (*Acheta domesticus*) were purchased from a vendor (The Bug Company, Ham Lake, MN) and fed a commercial high-calcium cricket diet (PMI, St. Louis, MO) for 24, 48, 72, or 168 h, in a replicated block design. Dry matter (DM) concentrations of crickets gut-loaded for 24, 48, or 72 h (31.7, 31.5, and 31.8%, respectively) were higher (P<0.05) compared with 168 h (30.1%). No differences were

233 Poisson versus logit models for genetic analysis of mastitis in Norwegian cattle. A. I. Vázquez*¹, K. A. Weigel¹, D. Gianola¹, D. M. Bates¹, and B. Heringstad², ¹University of Wisconsin, Madison, ²Norwegian University of Life Science, Ås, Norway.

Clinical mastitis incidence is typically coded as presence/absence during some period of exposure, and records are analyzed with linear or logit models. Since "presence" includes cows with multiple episodes, there is a loss of information when a count is treated as a binary response. Poisson (P) and logit (L) mixed models were fitted to clinical mastitis records on 36,178 first-lactation daughters of 245 Norwegian Red sires distributed over 5,286 herds. An R implementation (lme4 package) was modified to allow for correlations between random effects, such that pedigree information could be included. Since 26% of the cows had lactations shorter than 300 d, the log of days in milk (DIM) was included via the function $\log(\lambda * \text{DIM}) = \text{fixed} + \text{herd} + \text{sire}$ effects, where λ is the per-day Poisson parameter peculiar to a record. Predictive ability of models was assessed via a two-fold cross-validation using mean squared error (MSE) as end-point. On average, there were 0.295 cases of mastitis per cow; 77% of the cows did not have the disease, and only about 1% of the cows had 3 or more episodes during lactation. Between-sire variance estimates were 0.065 in P and 0.093 in L. The ratio between herd and sire variances was 4.6 and 3.7 for P and L models, respectively. The correlation between predicted random effects from the two models was 0.95 and 0.94 for sires and herds, respectively. Predictive MSE for all data was smaller for the Poisson model: 0.346 vs. 0.351. Within healthy animals, MSE was 0.053 (L) and 0.085 (P). For animals with 1 case, MSE values were 0.583 (L) and 0.499 (P). For animals with 2 cases, and 3 or more cases, the P model also had a better predictive performance. The cross-validation suggested a better overall performance of the Poisson model over the logit model, primarily due to improved predictive ability within diseased animals. The P model may be even better in situations in which the disease is more prevalent.

Key Words: Mastitis, Poisson, Threshold

detected for organic matter (OM), crude protein (CP), acid-hydrolyzed fat (AHF), or crude fiber (CF) concentrations among gut-loading treatments. Calcium concentrations were higher (P<0.05) after crickets were gut-loaded for 24 h (1.39%) compared with 48, 72, or 168 h (0.42, 0.37, and 0.66%, respectively). Magnesium concentrations were higher (P<0.05) after gut-loading crickets for 24 or 168 h (0.20 and 0.18%, respectively), compared with 48 or 72 h (0.10 and 0.09%, respectively). Concentration of manganese in crickets was higher (P<0.05) after 24 h (59.5 ppm) compared with crickets gut-loaded for either 48 or 72 h (28.5 and 27.0 ppm, respectively). No differences were detected in concentrations of sulfur, phosphorus, potassium, sodium, iron, copper, or zinc. Overall, gut-loading crickets for 24 h appeared more effective in increasing mineral concentrations compared with other time treatments. Further research is needed to completely elucidate the most effective strategies to increase nutritive value of feeder crickets for captive amphibians and reptiles.

Key Words: Reptile, Amphibian, Minerals

235 Effect of supplement type on mineral content of feeder crickets and growth of leaf-tailed geckos. C. L. Dikeman^{*1}, S. Plesuk¹, A. Koraleski¹, A. DeVries¹, K. Bilof², D. Klimek¹, J. Krebs¹, and L. G. Simmons¹, ¹Omaha's Henry Doorly Zoo, Omaha, NE, ²University of Illinois, Urbana.

Current methods of supplementation for feeder crickets to insectivorous reptiles in captivity include gut-loading and/or dusting with powders. Two experiments were conducted to address effects of supplement type on mineral content of feeder crickets and growth of captive geckos. In a replicated complete block design, farmed crickets were purchased (The Bug Company, Ham Lake, MN) and assigned to 1 of 4 dietary treatments including: fasted for 24 h (F), gut-loaded with Mazuri[®] high-calcium cricket diet (PMI, St. Louis, MO) for 24h (GL), gut-loaded plus dusted with Herptivite[™] (RepCal Research Labs, Los Gatos, CA) (GLH) or gut-loaded plus dusted with MinerAll[™] (Sticky Tongue Farms, Romoland, CA) (GLM). In Experiment 2, 6 newly hatched geckos (*Uroplatus sikorae*) were randomly assigned to GL, GLH, or GLM treatments. Geckos were fed ad lib, weights and lengths were measured weekly for 4 months. Concentrations of sulfur and iron were higher ($P < 0.05$) in GLH crickets (0.72 %, 1,168 ppm, respectively) compared with other treatments. Calcium concentration was higher ($P < 0.05$) in GLM crickets (6.37%) compared with other treatments (2.3, 2.4, and 2.9% for GLH, GL, and F, respectively). GL crickets had higher ($P < 0.05$) concentrations of phosphorus (1.28%) compared with F (0.98%). Potassium was higher ($P < 0.05$) in GLH (1.78%), GL (1.75%) and GLM (1.44%) compared with F (1.18%). Sodium concentrations were higher ($P < 0.05$) in GLH and GL treatments. Manganese concentrations were higher ($P < 0.05$) in both GLH (148.5 ppm) and GLM (128.5 ppm) compared with GL (49 ppm). No other differences were detected. Average weight of geckos fed GLH (2.5 g) was lower ($P < 0.05$) than average weights of geckos fed GL or GLM (2.8 g). Mean percent gain ranged from 33 to 38.5% for GLH, and GL, respectively. All treatments supported weight gain; however, enough data are not available to conclusively demonstrate benefits of these treatments in this species.

Key Words: Gecko, Cricket, Mineral

236 Serum nutrient concentration comparisons between free-ranging and captive giraffe (*Giraffa camelopardalis*). D. A. Schmidt^{*1,2}, M. R. Ellersieck³, and M. E. Griffin⁴, ¹Lincoln Park Zoo, Chicago, IL, ²Zoological Society of San Diego, San Diego, CA, ³University of Missouri, Columbia, ⁴Purina Mills, LLC, Saint Louis, MO.

Serum concentrations of amino acids, fatty acids, lipoproteins, vitamins A and E, and minerals in captive giraffe were compared to values obtained from free-ranging giraffe in an effort to identify potential nutrient problems in the captive population. Captive giraffe have a specific set of maladies, including peracute mortality, energy malnutrition, pancreatic disease, urolithiasis, hoof disease, and severe intestinal parasitism, which may be related to basic nutritional inadequacies. Dietary requirements for giraffe are not known; invasive studies used with domestic animals can not be performed on zoo animals. Though domestic animal standards are often used to evaluate nutritional health of exotic animals, they may not be the most appropriate standard to use. Twenty serum samples from captive giraffe

at 10 zoological institutions in the United States were compared to previously collected samples from 24 free-ranging giraffe in South Africa. Thirteen of the captive animal samples were collected from animals trained for blood collection. Seven were banked samples obtained from previous serum collections while animals were under anesthesia. Dietary information was also collected on each captive giraffe. Most captive giraffe diets consisted of alfalfa-based pellets and alfalfa hay. Differences between captive and free-ranging giraffe, males and females, and adults and sub-adults were analyzed using a $2 \times 2 \times 2$ factorial and Fisher's LSD for mean separation. Of the 84 parameters measured, 54 (60%) were different ($P \leq 0.05$) between captive and free-ranging giraffe. Nine (11%) items were different ($P \leq 0.05$) between adult and sub-adult animals. Only one parameter, sodium concentration, was found to be different ($P \leq 0.05$) between genders. Diets for captive giraffe need further investigation to address the differences seen in this study and the potentially related health problems.

Key Words: Giraffe, Nutrition

237 Nutrient digestibility and fecal characteristics of exotic felids fed a beef-based raw diet. B. M. Vester^{*1}, S. L. Burke², C. L. Dikeman², L. G. Simmons², and K. S. Swanson¹, ¹University of Illinois, Urbana, ²Henry Doorly Zoo, Omaha, NE.

Nutrient digestibility has been well studied and characterized in many of our domestic species; however, little has been done to evaluate digestibility in exotic felids. Large exotic felid species in captivity are often fed diets extrapolated from the requirements of the domestic cat. Furthermore, little research has been published determining the nutrient content of diets fed to these animals. The objective of this experiment was to evaluate differences in nutrient digestibility and fecal characteristics in five species of large exotic captive felid species at the Henry Doorly Zoo. The five species evaluated included Bobcats (n=2), Jaguars (n=4), Cheetahs (n=5), Indochinese Tigers (n=4), and Siberian Tigers (n=5). All animals were individually housed and adapted to beef-based raw diet (Nebraska Brand[®] Special Beef Feline) for 16 d. Total fecal collections were conducted on d 17 through 20. Fecal samples were weighed and scored upon collection. Scores were determined on a 5 point scale (1= hard, dry pellets; 2, dry, well formed stools; 3, soft, moist, formed stool; 4, soft, unformed stool; 5, watery, liquid that can be poured). Diet and fecal samples were evaluated for dry matter, organic matter, protein, fat, and energy to determine digestibility. A fresh fecal sample was collected to determine fecal pH. Fecal scores were greater ($P < 0.01$) in Indochinese Tigers when compared to all other species, and Cheetahs had greater ($P < 0.01$) fecal scores than Jaguars and Bobcats. Dry matter, organic matter, and protein digestibility were not different among species. Fat digestibility was greater ($P < 0.01$) in Siberian Tigers, Indochinese Tigers, and Bobcats (0.96) compared to Cheetahs and Jaguars (0.94). Digestible energy tended to be lower ($P < 0.10$) in Jaguars and Cheetahs (0.92) compared to Bobcats and Indochinese Tigers (0.93). Fecal pH was greater ($P < 0.01$) in Bobcats (pH=8) compared to all other species evaluated (pH=6). Overall, the beef-based raw diet was highly digestible in all species. However, differences in fat and digestible energy, suggests that further work should be completed to elucidate the differences between these species.

Key Words: Digestibility, Exotic Felid

238 Influence of dietary protein content and source on digestibility patterns and fecal osmolality in dogs differing in body size. J. Nery*¹, C. Tournier², V. Biourge², H. Dumon¹, and P. Nguyen¹, ¹*École Nationale Vétérinaire de Nantes, Nantes, France*, ²*Royal Canin, Aimargues, France*.

Large breed dogs have frequently poorer fecal quality than smaller ones when given the same diet. Previous work indicated that this difference would be due, at least in part, to differences in fermentation and a higher osmolality in the hindgut chyme of large dogs. As we hypothesized that diet formulation could alter these differences, the aim of this study was to assess the effect of protein source and level on digestibility, fecal quality and osmolality in dogs differing in body size. 24 female dogs (2.75 to 32.10 kg BW) were used. Two diets were tested in a cross-over design. The main protein source of diet A was poultry and poultry by-products (ME=15.7 MJ/kg, CP=35.2%, fat=16.0%, TDF=7.7%, Na=3.45 and K=5.17mg/g DM) and the one of diet B was wheat gluten (ME=16.2 MJ/kg, CP=19.9%, fat=18.0%, TDF=9.0%, Na=3.85 and K=8.67mg/g DM). Fecal scores and DM, energy, fat, CP, ash, Na and K apparent digestibility coefficient (ADC)

were determined. Fresh stools were analyzed for fecal osmolality. Data was statistically analyzed using ANOVA. Fecal score and moisture were higher in dogs fed on diet A and larger dogs had softer stools than smaller ones. ADC of DM, energy, fat, CP and ash was consistently higher for diet B. Differences among dogs' size were found to be higher for DM ($p \leq 0.0001$), energy ($p \leq 0.001$), CP ($p \leq 0.001$) and ash ($p \leq 0.05$) considering diet B. ADC of Na did not vary with dogs' size nor with diet while ADC of K varied both with dogs' size ($p \leq 0.05$) and diet ($p \leq 0.0001$), being higher for diet B. Osmolality was consistently higher for diet A ($p \leq 0.0001$) with differences also found between dogs' size ($p \leq 0.05$). This study showed that lower ADC of K and higher fecal osmolality would stimulate a lower water absorption in the hindgut promoting softer stools. A lower content and higher ADC of protein in the diet ameliorated fecal quality. Decreasing protein content in the diet and increasing its ADC would thus improve large dogs' feces quality.

Key Words: Dog, Protein, Digestibility

Dairy Foods: Cheese I

239 Chemical changes that predispose smoked cheddar cheese to calcium lactate crystallization. P. Rajbhandari*, J. Patel, E. Valentine, and P. S. Kindstedt, *University of Vermont, Burlington*.

We have observed a high incidence of calcium lactate surface crystals on naturally smoked Cheddar cheese in the retail marketplace. The objective of this study was to identify chemical changes that may occur during natural smoking which render Cheddar cheese more susceptible to calcium lactate crystal formation. Nine random weight (ca. 300 g) retail-packaged samples of smoked Cheddar cheese were obtained from a commercial manufacturer immediately after the samples were smoked for ca. 6 h at ca. 20°C in a commercial smokehouse. Three similarly sized samples that originated from the same 22-kg block of cheese and that were not smoked were also obtained. Within 2 d after smoking (0 wk), 3 smoked and 3 non-smoked samples were sectioned into 5 sub-samples at different depths representing 0-2, 2-4, 4-6, 6-8, and 8-10 mm from the cheese surface. Six additional smoked cheese samples were similarly sectioned at 4 wk and again at 10 wk of storage at 5°C. Sample sections were analyzed for moisture, pH, L(+) and D(-) lactate, and water soluble calcium. The effects of treatment (smoked, non-smoked), depth from cheese surface and their interactions were analyzed by ANOVA according to a repeated measures design with 2 within subjects variables. Smoked samples contained significantly lower moisture and lower pH, and higher lactate-in-moisture (LIM) and water-soluble calcium-in-moisture (WSCIM) than non-smoked samples at 0 wk. Smoked samples also contained significant gradients of moisture, pH, LIM and WSCIM, with lower moisture and pH, and higher LIM and WSCIM, occurring at the cheese surface. Gradients of moisture were still present in smoked samples at 4-and 10 wk of storage. In contrast, the pH, LIM and WSCIM equilibrated and showed no gradients at 4 wk and 10 wk. The results indicate that calcium and lactate in the serum phase of the cheese were elevated as a result of smoking, especially at the cheese surface immediately after smoking treatment, which presumably predisposed the smoked cheeses to increased susceptibility to calcium lactate surface crystallization.

Key Words: Cheddar Cheese, Calcium Lactate, Crystals

240 Nucleation and growth rates of calcium lactate crystals on smoked cheddar cheese. 1. Effect of storage temperature. J. Patel*, P. Rajbhandari, E. Valentine, and P. S. Kindstedt, *University of Vermont, Burlington*.

Previous studies have shown that storage temperature influences the formation of calcium lactate crystals on Cheddar cheese surfaces. However, the mechanisms by which crystallization is modulated by storage temperature are not completely understood. The objectives of this study were to evaluate the effect of storage temperature on: 1) the number of discrete visible crystals formed per unit of cheese surface area; 2) growth rate and shape of discrete crystals (as measured by radius, area and circularity); 3) percentage of total cheese surface area occupied by crystals. Three vacuum packaged random weight (ca. 300 g) retail samples of naturally smoked Cheddar cheese, produced from the same vat of cheese, were obtained from a retail source. The samples were cut parallel to the longitudinal axis at a depth of 10 mm from the 2 surfaces to give six 10-mm thick slabs, 4 of which were randomly assigned to 4 different storage temperatures: 1, 5, 10°C, and weekly cycling between 1 and 10°C. Samples were stored for 30 wk. Following the onset of visible surface crystals, digital photographs of surfaces were taken bi-weekly and evaluated by image analysis for number of discrete crystal regions and total surface area occupied by crystals. Also, specific discrete crystals were chosen and evaluated bi-weekly for radius, area and circularity. The entire experiment was conducted in duplicate. The effects of storage time and temperature on crystal number and total crystal area were evaluated by ANOVA according to a repeated measures design. Crystal number and total crystal area increased significantly during storage in a temperature-dependent manner as follows: 5°C < 1°/10°C < 10°C < 1°C. However, storage temperature did not appear to have a major effect on the growth rates and shapes of the individual crystals that were chosen for analysis. The data indicated that storage temperature primarily affected the number of nucleation sites on the cheese surface that subsequently developed into discrete visible crystals.

Key Words: Cheddar Cheese, Calcium Lactate, Crystal