## Physiology and Endocrinology: Male reproduction, deers, and poultry

W238 The effect of moringa oleifera leaf meal on follicle stimulating hormone, luteinizing hormone and testosterone of wad goat bucks serum. Abimbola O. Ladokun\*, Khadeejah Kareem-Ibrahim, Bukola D. Adenaike, Oladimeji M. Abioja, and John A. Abiona, *Federal University of Agriculture, Abeokuta, Ogun, Nigeria.* 

Moringa as a plant has been attributed with possessing anti-oxidative properties and lowering of blood cholesterol. It has also been reported to be involved as an endocrine system blocker (ESB). This study investigated the effect of moringa leaf meal supplemented in the feed of West African dwarf (WAD) goat bucks on their serum FSH, LH and testosterone. Sixteen matured WAD bucks were used; they were assigned into 4 groups with 2 replicates and 2 bucks per replicate. They were semi-intensively managed, allowed to graze and their concentrate supplemented with blended moringa leaf at 0g/kg concentrate, 100, 200, and 300 g/kg respectively for 8 weeks. Blood was sampled 3 times (Start, 4th week and end of 8th week) and serum harvested for analyses using EIA for testosterone and ELISA kits and reader for FSH and LH. Data were analyzed using one-way ANOVA of SAS, and significant (P =0.05) means were separated using the Duncan test in SAS. Concentrations of testosterone were similar across the groups (5.65-5.95ng/L). At the 4th and 8th week, the concentrations though greater than start, they were also similar across the groups indicating that moringa did not influence testosterone concentration in WAD bucks. Concentrations of FSH were also similar at the start (4.85–5.0), but there were decreases in concentrations at the 4th and 8th week as compared with start. It is noteworthy that for bucks supplemented with 200g/kg moringa, there was a significant (P = 0.05) decrease in concentration at 4th and 8th week (5.0–4.40–4.30ng/L). The average serum LH at start was similar among groups (3.75-4.10ng/L). At the 4th and 8th week, treatment did not affect concentrations of LH. Results indicate that moringa leaf at the level used in this study did not elicit any response indicative of disrupting the concentration of testosterone, FSH and LH in WAD bucks.

Key Words: moringa, goat buck, hormone

**W239** Proteomic analysis of testicular proteins between yak and its sterile hybrid by iTRAQ labeling mass spectrometry. Wei Fu<sup>1</sup>, Wenlin Bai<sup>2</sup>, Lin Huang<sup>1</sup>, Wenjing Liu<sup>3</sup>, Caixia Li<sup>1</sup>, Suyu Jin<sup>1</sup>, Xiang Qiu<sup>1</sup>, Liang Ren<sup>1</sup>, and Yucai Zheng<sup>\*1</sup>, <sup>1</sup>Southwest University for Nationalities, Chengdu, Sichuan Province, China, <sup>2</sup>Shenyang Agricultural University, Shenyang, Liaoning Province, China, <sup>3</sup>Southwest University of Science and Technology, Mianyang, Sichuan Province, China.

The objective of the present study aimed at identifying differential expression proteins in testes of adult yak (*Bos grunniens*) and its sterile hybrid (cattle-yak), to provide basic data for elucidating the molecular mechanisms of the male sterility of cattle-yak. Cattle-yak is a hybrid of male cattle (*Bos taurus*) with female yak, exhibiting higher body size, milk and meat yields than those of yak, but  $F_1$  to  $F_3$  male sterility, the mechanism is still unknown. In the present study, total proteins were extracted from the testes of adult yaks (n = 6) and cattle-yaks (n = 4), respectively, followed by analysis of the testicular protein profiles (pooled samples for each group) by isobaric tags for relative and absolute quantitation (iTRAQ) technique. Proteins with a fold change of >1.2 and P < 0.05 (*t*-test) were considered to be significantly differentially expressed. Hypergeometric test was used in Gene Ontology analysis. A total of 4464 proteins were identified, of which 714 proteins

were differentially expressed, including 432 downregulated and 282 upregulated proteins in the testis of cattle-vak compared with that of vak. Interestingly, approximately 30% of the top 50 downregulated proteins in cattle-yak testis are testis-specific or highly expressed in testis, and most likely exert crucial effects on spermatogenesis. Gene Ontology analysis of the differentially expressed proteins demonstrated that 19 major protein categories belong to carbohydrate metabolism and protein folding were highly enriched in the Biological process category, implying the important roles of the 2 processes in spermatogenesis. Further analysis of Biological process category revealed 43 proteins associated with reproductive process, and most of which were downregulated in the testis of cattle-yak. In addition, fatty acid oxidation metabolism was found to be more active in cattle-yak testis than in yak testis. Based on the testicular protein profiles, protein functions and protein-protein interaction analysis, we propose that the abnormal expression of several downregulated proteins of testis-specificity or with high abundance in testis, such as YBX2, PIWIL1, TDRD1, and BAG6, might be closely associated with the sterility of male cattle-yak.

Key Words: yak, hybrid sterility, proteomics

## W240 Effect of androstenone level on the boar testis tran-

scriptome. Dianelys Gonzalez-Pena\*, Robmay Garcia, and Robert V. Knox, and Sandra L. Rodriguez-Zas, *University of Illinois at Urbana-Champaign, Urbana, IL*.

Androstenone is the most abundant steroid produced in boar testis and share a precursor (pregnenolone) with testosterone, the principal male sex hormone needed for sperm production in the testis and its maturation on the epididymis. Androstenone is a pheromone of non-castrate males present in urine, sweat, saliva, and fatty tissues responsible for boar taint, which includes unpleasant odor and meat flavor. The objective of this study was to understand the differences in the transcriptome between high (greater than 1.0  $\mu$ g/g) and low (less than 0.5  $\mu$ g/g) and rostenone boars. Testis mRNA from 10 Duroc crossbred boars was profiled. Singleend reads were mapped to the Sus scrofa reference genome (UCSC susScr3) using Tophat v2.012. In total, 4,969 transcripts from 4,744 genes were tested and 69 transcripts from 72 genes were differentially expressed between the high and low androstenone groups using Cufflink v2.2.1 (False Discovery Rate adjusted P-value <0.05). Among these, steroidogenic acute regulatory protein (STAR), cytochrome P450 family 19 subfamily A polypeptide 1 (CYP19A1), and sulfotransferase family cytosolic 2A dehydroepiandrosterone-preferring member 1 (SULT2A1) were overexpressed, while macrophage receptor with collagenous structure (MARCO) and anti-Mullerian hormone receptor type II (AMHR2) were underexpressed in the high relative to low androstenone groups. CYP19A1 encodes enzymes that participate in the synthesis of cholesterol, steroids, and other lipids and STAR encodes a protein critical in steroid hormone synthesis by enhancing the metabolism of cholesterol into pregnenolone the precursor of testosterone and androstenone. The overexpression of SULT2A1 in high relative to low androstenone groups is consistent with the role of this enzyme in the sulfo-conjugating of androstenone. Functional analysis of the differentially expressed genes using DAVID identified 2 category clusters (enrichment score >2) that include hormone metabolism biological processes and regulation, steroid and lipid biosynthetic and metabolism. These functional categories suggest that practices to reduce androstenone should aim to

minimize transcriptome changes that can negatively impact testosterone and boar fertility.

Key Words: transcriptome, androstenone, testis

## W241 Gonadal sperm characteristics of growing boars fed varying levels of raw or fermented cottonseed cake. Mayowa

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Cottonseed cake (CSC) is a veritable source of protein, energy and fiber for a variety of livestock species. However, it contains a deleterious polyphenolic compound, gossypol that restricts it use to ruminant feeding. Fermentation is one of the biotechnological options for reducing toxicity of many feed ingredients. This study was conducted to determine the effect of different levels of raw or fermented CSC on sperm

characteristics of growing boars. Thirty (30) cross-bred (Landrace  $\times$ Large White) with average weight of 7 kg, weanling boars (n = 6 per treatment) were randomly assigned to 5 dietary treatments arranged in a  $2 \times 2$  factorial with a control within a completely randomized design (CRD). The CSC was included in diets at 0 (control), 10 and 20% (raw or fermented). Animals were fed for 12 weeks after which they were killed at average weight of 17.1 kg and the reproductive organs removed and processed for sperm characteristics. Sperm concentration was significantly (P < 0.05) higher for the control ( $48.50 \times 10^6$ /mL) than other treatments. Sperm motility for the raw CSC (68.55%) was similar to that of the control (68.01%). Boars that were fed 10% fermented CSC recorded lowest sperm motility (56.05%). Normal sperm was highest (71.34%) (P < 0.05) for the control while the lowest value was obtained at 10% fermented CSC (52.76%). Gonadal sperm reserve (GSR) was the highest (P < 0.05) for the control group ( $1.928 \times 10^9$ ), while the lowest value was observed at 20% fermented CSC (0.610  $\times$ 10<sup>9</sup>). Boars fed raw CSC, irrespective of level, had higher GSR than those fed fermented CSC. Daily sperm production (DSP) was significantly (P < 0.05) higher for the control than 10% and 20% fermented CSC groups. Daily sperm production/gram (DSP/g) was significantly (P < 0.05) higher for 10% raw CSC (40.44 × 10<sup>6</sup>) compared with other treatments. This study shows that growing boars could tolerate raw CSC up to 10% in diets on a short-term basis without adverse effects on the gonadal sperm characteristics.

**Key Words:** boar, cottonseed cake, fermentation and sperm characteristics

## **W242** GameteGuard treatment improves post-thaw sperm quality and pregnancy per insemination in dairy cows. Lisa Herickhoff\*<sup>1</sup>, Allison Lindsey<sup>1</sup>, Amanda Fritts<sup>1</sup>, and Patrick Burns<sup>2</sup>, <sup>1</sup>Membrane Protective Technologies Inc., Fort Collins, CO, <sup>2</sup>School of Biological Sciences, University of Northern Colorado, Greeley, CO.

GameteGuard is a blend of plant extracts containing antioxidants and membrane stabilizers. Previous studies have shown improvements in post-thaw sperm health with GameteGuard addition to the extender, and therefore it was hypothesized that the addition of GameteGuard to sperm extender would result in increased pregnancy rates per AI in lactating cows. Sperm from 2 ABS Global Inc. production Holstein bulls was extended in egg yolk citrate or egg yolk citrate plus GameteGuard to 30 ×106 sperm/mK (15 × 10<sup>6</sup> sperm/dose) in a split ejaculate study. Sperm was packaged and frozen according to industry standards. The effectiveness of GameteGuard on pregnancy was assessed by first postpartum insemination of 208 Holstein cows. Over 6 mo, cows were cleared for insemination by the farm veterinarian then 2 farm personnel determined estrus status using visual cues such as disturbed tail chalk. The cows were inseminated within 12 h by one trained farm employee. Cows were inseminated with either Bull 1 or 2, treated or control sperm, where treatment was blind to farm personnel. Bull and treatment combinations were dictated by a calendar day. For example, all cows to be bred on May 15 would use bull 1, red straw (control) while May 16 would use bull 1, blue straw (treated). Cows having postpartum problems and those with reproductive issues, such as ovarian cysts, were excluded from the study. Thirty-five to 45 d post-insemination pregnancy was assessed using transrectal ultrasonography and data were analyzed using Chi-squared. GameteGuard improved pregnancy per AI by 31% (from 38 to 50%; P < 0.05; ~100 cows/treatment) across both bulls. Importantly, of the 124 animals bred within 83 d of calving, GameteGuard improved pregnancy per AI 36.1%.

**Key Words:** artificial insemination, sperm quality, pregnancy per insemination

**W243** Evaluation of factors affecting pregnancy rate to AI in pen-raised white-tailed deer. Kyle J. Stutts\*, Jessica L. Leatherwood, Christopher R. Stewart, Mark J. Anderson, Marcy M. Beverly, and Stanley F. Kelley, *Sam Houston State University, Huntsville, TX.* 

Factors known to influence pregnancy rate in livestock species were evaluated in white-tailed deer. Pen-raised white-tailed does (n = 53)ranging in age from 1.5 yr to 6.5 yr were utilized to evaluate the effects of BCS, BW, disposition, and serum cortisol concentration on fawning rate to AI. All does received a CIDR for 14 d and an injection (i.m.) of 300 IU PMSG at CIDR removal. Laparoscopic insemination was conducted at approximately 60 h following CIDR removal. Prior to insemination, does were restrained in a drop-floor chute and were administered sedative. At this time, BW was collected and disposition scores ranging from 1 to 5 (1 = docile and 5 = aggressive) were assigned by independent observers to evaluate deer behavior as an indicator of stress while restrained in the chute. Immediately following insemination, BCS was evaluated and blood samples were obtained via jugular venipuncture to determine cortisol concentration by RIA analysis. Previous research indicated a moderately positive relationship (r = 0.30) between disposition score and serum cortisol concentration. Does were placed with cover bucks one week post AI. During fawning season, DNA samples were collected from all fawns to identify the sire of each fawn and determine fawning rate to AI. Data were analyzed using the LOGISTIC procedure of SAS. Pregnancy rate to AI was 67.9% and final pregnancy rate was 94.0% following natural service. Mean BCS was  $3.02 \pm 0.52$  (range of 2 to 4), and mean disposition score was  $2.16 \pm 1.21$  (range of 1 to 4). There was no significant effect of BCS (P = 0.59), BW (P = 0.19), cortisol concentration (P = 0.30), or disposition score (P = 0.64) on pregnancy rate to AI. Results of this study indicate that body condition and BW may not have as great of an influence on pregnancy rate in white-tailed deer as observed in livestock species. These data also indicate that the stress response to being restrained may not influence pregnancy rate to AI in white-tailed deer. Given the small sample size of this study, further research is needed to fully elucidate the influence of these factors on pregnancy rate to AI in white-tailed does.

Key Words: white-tailed deer, artificial insemination, disposition

W244 Blood profiles of Ross 308 broiler and indigenous Venda chickens aged 42 and 90 days fed a similar diet. Monnye Mabelebele\*, David Norris, Jones Ng'ambi, and John Alabi, *University of Limpopo, Polokwane, Limpopo, South Africa.*  A study was conducted to assess blood profiles of Ross 308 broiler and indigenous Venda chickens. A total of 120 Ross 308 broiler and Venda chickens were raised for a period of 90 d on a commercial grower diet containing 20% crude protein and 13 MJ ME/kg DM. Ten chickens per breed were randomly selected at 42 and 90 d of age and 3 mL of blood samples from both chicken breeds were collected from the jugular vein into 2 test tubes per bird to determine the blood profiles. A *t*-test was used to test the significance of differences between breeds (P <0.05). Differences among hematological parameters of Ross-308 and Venda chickens at 42 d of age were not significant (P > 0.05) except for platelets ( $\times 10^3 \mu/L$ ) for Ross 308 broiler (59.2 ± 10.13) and Venda chickens (24.0 ± 10.00); serum creatinine ( $\mu$ mol/L) for Ross 308 broiler (17.5 ± 50.05) and Venda chickens (9.0 ± 5.02) and total protein (g/L) of Venda ( $25.5 \pm 3.20$ ) and Ross 308 broiler chickens ( $20.0 \pm 3.12$ ). At 90 d of age, white blood cells ( $734.4 \pm 18.97 \times 10^3$  cell/µL), red blood cells ( $2.3 \pm 0.15$ ;  $\times 10^6$  cell/µL), mean corpuscular volume ( $121.1 \pm 4.10$  f/L), serum calcium ( $2.6 \pm 0.01$  mmol/L), creatinine ( $10.7 \pm 0.76$  µmol/L), cholesterol ( $3.9 \pm 0.12$  mmol/L) and albumin ( $9.5 \pm 0.23$  g/L) were higher for Ross 308 broiler chickens. However, platelets ( $\times 10^3$ µ/L) were higher in Venda ( $14.4 \pm 3.29$ ) than in Ross 308 broiler ( $12.2 \pm 3.00$ ) chickens. In the adult stage, broiler chickens have better hematological properties than indigenous chickens. Further investigation on immunological parameters of these breeds needs to be conducted.

Key Words: hematology, chicken, disease