

Morphometric characteristics, testicular histology, and semen parameters in mature hybrid bucks born from white-tailed deer dams sired by a mule deer buck

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Despite white-tailed deer (WTD) and mule deer (MD) sharing similar number of chromosomes ($n = 70$), the 2 species have distinct features making the crossing of species unsuccessful in wild or captive conditions. Across mammals, hybrids are regarded as infertile, yet anecdotal reports suggest that WTD-MD hybrids are fertile. However, there have been limited reports of hybrid animals producing fully formed sperm, though none on hybrids of WTD-MD. We aimed to describe the somatic morphometric features, testicular histology and semen features of captive hybrids of WTD-MD. Four 1.5-year-old bucks were enrolled in this study, 2 hybrids WTD-MD, 1 WTD, and 1 MD from a captive farm. The hybrid animals were born from different WTD, but were sired by the same MD. Morphometric profile included thorax circumference, crown-rump length, metacarpus and metatarsus diameter, tail length, tail color, location of the metatarsal gland, antler configuration, antler inside spread, ear length, metatarsal tuft color, scrotal circumference and length of the penis. Semen collected via electroejaculation was evaluated for the presence of sperm and the concentration of alkaline phosphatase (ALP). Testicular biopsies were collected from both testes using a split needle biopsy tool. The scrotal circumference was 22 cm for the WTD, 19 cm for the MD, and both hybrids measured 12 and 14 cm, respectively. The penis length was 28.6 for the WTD, 27.4 for the MD and 12 and 14 cm for the hybrids. It has been suggested that testosterone regulates penis growth after puberty; perhaps these animals had lower testosterone production hence the shorter penis. Semen collection yielded ~ 1 ml of yellow and viscous fluid. No sperm were visualized under the phase-contrast microscope. Histologic evaluation revealed the presence of hypoplastic seminiferous tubules in both animals populated with spermatogonia in the basal compartment and normal Sertoli cells. One animal had primary spermatocytes in the adluminal compartment and scattered spermatids could be seen in a few seminiferous tubules. The basal membrane of the seminiferous tubules was surrounded by dense, irregular connective tissue. The Leydig cells were present in the interstitium and appeared morphologically normal; this explains why the hybrids were able to produce intermediate sized antlers that hardened at the peak of rut. Concentrations of ALP in hybrid 2 was 1620 U/l. The color of the tail of the hybrids was brown on the dorsal surface, but white on the ventral part and resembled 1 of the WTD. The metatarsal gland was in the proximal segment of the metatarsus in the hybrids and WTD, whereas it was below in the MD. Only the MD presented dichotomous antlers. The metatarsal tuft color was brown in the hybrids and MD, but white in the WTD. In conclusion, hybrid 1 male was unable to complete spermatocytogenesis, and the second could not

complete spermiogenesis, making then unable to fully form sperm. The high concentrations of ALP confirmed ejaculation in these 2 animals; however, they were deemed infertile.

Keywords: Azoospermia, infertility, mule deer, white-tailed deer, hybrids

Association of metabolic status with uterine diseases and reproductive outcomes in lactating Holstein dairy cows

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Selection of high yielding dairy cows has predisposed them to develop metabolic and uterine diseases. While the association of hyperketonemia (HRK) with metritis and decreased reproductive performance is known, little data exists examining the association of HRK and concurrent hypoglycemia with metritis and reproductive outcomes. Our hypothesis was that cows with HRK have a higher incidence of puerperal metritis and poorer reproductive outcomes than cows without HRK and those effects are more profound in cows with hypoglycemia. The objectives of this study were to analyze the association of metabolic status with metritis incidence and reproductive outcomes in lactating dairy cows. Cows ($n = 2651$) had blood samples collected between 3 - 9 days postpartum (DPP) and whole blood beta-hydroxybutyrate (BHBA) and whole blood glucose was measured using a hand-held cow-side device validated in dairy cows (Precision Xtra, Abbott, Mississauga, ON, Canada). Hyperketonemia was defined as BHBA ≥ 1.2 mmol/liter and hypoglycemia was defined as glucose ≤ 2.2 mmol/liter. Cows were then categorized into the following 4 groups: first those having no metabolic abnormality (Norm, $n = 1996$), those having HRK only (BHBA, $n = 260$), those having hypoglycemia only (HG, $n = 181$), and those having both HRK and hypoglycemia (BHBA + HG, $n = 214$). Incidence of puerperal metritis (defined as watery, fetid discharge present at time of blood collection), and the reproductive outcomes first insemination pregnancy per AI (P/AI), pregnancy loss, average days open (DOPN), and proportion of cows pregnant at 150 DIM (P150) were compared for the 4 metabolic statuses enrolled in the study. The cow-level prevalence of hyperketonemia was 17.9% (474/2651), and the cow-level prevalence of hypoglycemia was 14.9% (395/2651). The cow-level prevalence for each metabolic category was as follows: Norm, 75.3% (1996/2651); BHBA, 9.8% (260/2651); HG, 6.8% (181/2651); and BHBA + HG, 8.1% (214/2651). Statistical analysis was performed using ANOVA and logistic regression with JMP Pro 13 (SAS Institute Inc. Cary, NC, US). Parity (Parity 1 (P1) versus Parity ≥ 2 (P2)), season, and farm were retained in the model. P2 prevalence of puerperal metritis was significantly less for cows in the HG group compared to cows in the BHBA and BHBA+HG groups

(HG, 1.9%; BHBA, 16.8%; BHBA+HG, 9.4%). P2 prevalence of puerperal metritis was significantly less in the Norm group compared to cows in the BHBA group but similar to cows in HG group (Norm, 6.7%; BHBA, 16.8%; HG, 1.9%). For primiparous cows there was no difference in incidence of puerperal metritis amongst metabolic categories. There were no differences between metabolic groups for P/AI, pregnancy loss, average DOPN, or P150. In conclusion, hyperketonemia of multiparous cows was associated with increased puerperal metritis; however, hypoglycemia alone was associated with decreased puerperal metritis compared to cows with elevated BHBA with or without concurrent hypoglycemia.

Keywords: Hyperketonemia, hypoglycemia, dairy cows, uterine disease

Effect of GnRH at artificial insemination for dairy cows detected in estrus by an activity monitoring system or by conventional estrus detection

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Artificial insemination (AI) after detected estrus constitutes a substantial proportion of AI's that occur in the US. Moreover, AI after detected estrus may be increasing with the use of activity monitors. We hypothesized that GnRH treatment at AI increases both ovulation and circulating progesterone concentrations, thereby improving pregnancies per AI (P/AI); additionally, we hypothesized that this effect is higher in farms using activity monitors. The objectives of the study were to determine if GnRH treatment at AI increases P/AI for lactating dairy cows detected in estrus on farms using activity monitors (AM) or not (NAM). Holstein cows were blocked by parity and randomly assigned to receive an injection of GnRH at AI (G-AI) or to receive no injection of GnRH (NG-AI) at AI on a farm using AM for estrus

detection and on a group of 4 farms using NAM. On the farm with AM, 409 cows were enrolled (G-AI, n = 207; NG-AI, n = 202) and for the farms using NAM, 398 cows were enrolled (G-AI, n = 197; NG-AI, n = 201). Ovarian structures and plasma progesterone concentrations were assessed in a subset of cows (G-AI, n = 52; NG-AI, n = 55) detected in estrus by conventional methods at the time of AI and 7 days later. Data were categorized by milk production quartiles, genomic daughter pregnancy rate (High: > the median, Low: < the median), activity level (AL) for the farm using AM (High: AL > the median AL versus Low: AL < the median AL) and DIM (> 150 DIM versus < 150 DIM). Statistical analyses were performed using logistic regression and a Student's *t*-test. There were no differences in ovulation rate (G-AI = 83.2 ± 6.1%; NG-AI = 77.9 ± 5.5%) between G-AI and NG-AI. There were no differences in plasma progesterone concentrations at day of estrus detection (day 0) (G-AI = 0.16 ± 0.11 ng/ml; NG-AI = 0.09 ± 0.10 ng/ml) nor at day 7 after enrollment between G-AI and NG-AI (G-AI = 2.17 ± 0.15 ng/ml; NG-AI = 2.04 ± 0.15 ng/ml). Data for all farms were analyzed together for P/AI; no difference for P/AI at first pregnancy diagnosis (G-AI = 38.7 ± 3.9%; NG-AI = 40.9 ± 3.9%) or second pregnancy diagnosis (G-AI = 35.1 ± 4.1%; NG-AI = 35.7 ± 4.2%) was identified. No difference in P/AI between G-AI and NG-AI when farms were analyzed separately based on estrus detection method (AM separate from NAM) at first pregnancy diagnosis (AM: G-AI = 39.1 ± 5.0%; NG-AI = 38.6 ± 5.1%; NAM: G-AI = 38.3 ± 5.2%; NG-AI = 43.3 ± 5.2%) or second pregnancy diagnosis (AM: G-AI = 36.3 ± 5.1%; NG-AI = 33.8 ± 5.2%; NAM: G-AI = 33.8 ± 5.3%; NG-AI = 37.8 ± 5.2%) was identified. There was no interaction between treatment and method of estrus detection. For the farm using AM, there was a significant interaction between treatment and AL with the injection of GnRH having a greater impact on cows with high AL. In conclusion, GnRH treatment did not enhance P/AI. Additional studies are warranted to understand the interaction between treating cows with GnRH at AI and AL in herds using activity monitoring systems.

Keywords: Dairy cows, estrus detection, activity monitors, GnRH