

Comparison of Pregnancy Outcome in Mares among Methods of Sperm Selection for Deep-Horn Insemination

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Project Aims. The aim of this study was to compare pregnancy outcome among three methods of selecting sperm for insemination deep in the uterine horn near the utero-tubal junction (UTJ).

Methods. Thirty mares and two stallions of various light-horse breeds were used in this study. The stallions had achieved acceptable conceptions per cycle in past seasons. Mares were equally randomized between stallions and bred in successive cycles with sperm selected for absolute number (AN) only, by filtration through a glass wool/sephadex (GWS) column, or by separation over a percoll gradient (PS). A total of 90 cycles are represented in this study, 45 for each stallion. All mares were bred to the assigned stallion with sperm from each of the selection methods. A total of 30 cycles are represented for each of the three sperm selection methods, 15 for each stallion. Sperm selection method was randomized for each mare in the first cycle. Mares were inseminated with 25 million sperm selected by the method assigned for the cycle and standardized for concentration (50 million/ml) and volume (0.5 ml). When a ≥ 35 mm follicle was detected hCG was administered and the mare was inseminated approximately 24 hours later. Sperm cells were deposited deep in the uterine horn near the UTJ ipsilateral to the dominant follicle using a 65 cm pipette fitted with a smaller diameter inner catheter for delivery of a low volume (Deep Uterine Insemination Kit, Mini Tube of America). Pregnancy exams were conducted 14 days following ovulation. Following the exam, each mare was short-cycled with cloprostenol and monitored for breeding with the next assigned sperm selection method. Proportions of pregnancies among numbers of breedings were contrasted between treatments and stallions and pairwise comparisons of treatments were performed using vector-specified linear contrasts in PROC CATMOD (Statistical Analysis System, SAS Institute, Cary, NC).

Findings. Overall mares were pregnant in 10/30, 15/30 and 13/30 cycles for AN, GWS and PS selection methods, respectively. Individually, mares were pregnant for Stallion A in 6/15, 9/15 and 8/15 cycles and Stallion B in 4/15, 6/15 and 5/15 cycles for AN, GWS and PS selection methods, respectively. Sperm selection method did not effect pregnancy outcome in this study ($P=0.422$). However, the GWS sperm selection method showed a tendency toward better results than the AN method ($P=0.105$). It was also interesting that the proportion of pregnancies was greatest when sperm were selected by GWS and lowest by AN, for both stallions. Another interesting observation was that none of the mares required lavage post-breeding. Several of the mares used in this study had required post-breeding uterine lavage following the use of standard insemination doses in past seasons. Fertility results were within acceptable ranges for the group of stallions and mares used in this study. Specifically selecting sperm for deep-horn insemination may allow more efficient use of stallion sperm as well as reducing the post-breeding inflammatory reaction in mares. **Keywords.** Deep-horn insemination, glass wool/sephadex filtration, percoll separation, utero-tubal junction, sperm