Comparison of sperm morphology and DNA fragmentation in normal and asthenozoospermic dogs

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Sperm motility and morphology have been correlated in various studies with fertility. Recently, the influence of sperm DNA fragmentation on fertility has been investigated. The objective of this study was to compare sperm morphology abnormalities and DNA fragmentation in normal (≥80\% progressively motile sperm; \(n=4\)) and asthenozoospermic (≤60\% progressively motile sperm; \(n=6\)) dogs. Seven breeds of dogs were represented. Mean age per group was not significantly different. Semen (1\textsuperscript{st}/2\textsuperscript{nd} fraction) was collected by manual stimulation in the presence of an anestrous teaser. Sperm were evaluated under light microscopy for total motility, progressive motility and speed. Sperm concentration was determined using a hemocytometer. Sperm morphology was determined following staining with eosin and nigrosin and one individual (HM) counted 200 cells under oil immersion at 1000X magnification. In addition, a semen sample (1 mL) from each dog was frozen and shipped on dry ice for sperm chromatin structure analysis (SCSA).\textsuperscript{1} Briefly, aliquots (2-7 \(\mu\)l) of thawed semen samples were diluted to 200 \(\mu\)l in a Tris buffer solution and mixed with 400 \(\mu\)l of acid-detergent solution. A DNA probe (acridine orange; 1.2 ml) was added to each sample. Using a flow cytometer, 5000 spermatozoa/sample were analyzed at a flow rate of 100-200 cell/sec. The percentage of spermatozoa with altered chromatin structure was identified as those with increased red fluorescence corresponding to increased DNA fragmentation (DFI).\textsuperscript{1} Half (3/6) of the asthenozoospermic dogs had a DNA fragmentation index (DFI) >30\% whereas none (0/4) of the dogs with normal sperm motility had a DFI >30\%. However, with respect to sperm morphology, only dogs with ≥10\% head abnormalities had a DFI >30\%. In addition, there was a significant positive linear correlation between percent sperm head abnormalities and %DFI. In other species, a DFI >30\% is statistically correlated with a decrease in term pregnancies.\textsuperscript{1} However, it is important to note that a DFI value above 30\% does not preclude a normal, full-term pregnancy. Based upon these findings, sperm morphology, specifically head abnormalities, was correlated with poor fertility and increased % DFI, whereas sperm motility was not. Due to the small sample size in this study, further research is needed to corroborate these findings.

Keywords: Asthenozoospermia, DNA, dog, SCSA, sperm

Reference