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JOURNAL ARTICLE

In vitro and xenogenous capacitation-like changes of fresh, cooled, and cryopreserved stallion sperm as assessed by a chlortetracycline stain

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Like the human female, the mare experiences reproductive tract pathology that may sometimes be circumvented by the use of assisted reproductive technologies (ARTs). One such technology, gamete intrafallopian transfer (GIFT), may be used in mares that exhibit ovulatory, oviductal, or uterine abnormalities that limit the use of common ARTs, such as embryo transfer. Homologous GIFT has been successfully performed in the horse; however, the logistics, costs, and associated risks of surgically transferring gametes to the oviducts of a recipient mare are considerably high. Use of a less costly species in a heterologous or xenogenous procedure would therefore be beneficial. This study represents the preliminary investigation into the use of sheep as recipients for xenogenous GIFT procedures using equine gametes. We investigated the capacitation response of fresh, cooled, or frozen stallion sperm after 1) in vivo incubation in the reproductive tract of estrous and anestrus ewes as well as 2) in vitro incubation in a modified Krebs/ Ringer extender at 37 degreesC with and without the addition of heparin at 10 IU/mL for up to 8 hours. A chlortetracycline (CTC) fluorescent stain was used to assess the capacitation response of sperm. Findings indicated that oviductal fluid samples recovered from estrous ewes had significantly higher numbers of sperm exhibiting capacitation-like staining patterns when compared to samples recovered from anestrus ewes ($P < .05$). Fresh semen yielded higher capacitation-like staining patterns after in vivo incubation than did frozen-thawed or cooled samples. A transition from majority CTC unreacted sperm to majority CTC non-acrosome intact sperm was demonstrated for both in vivo and in vitro studies. In vitro incubation of stallion sperm with heparin did not result in an increased capacitation-like staining response over time when compared with nonheparinized samples. Results from this study suggest that xenogenous capacitation of stallion sperm may occur in the estrous ewe.

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