in fact, it has been reported that fT3 directly regulates testosterone production and sperm motility. Decrease of fT4 correlated with the augmentation of fT3 may be due to the fT3 demand of tissues and to the increase of fT4 deiodinase.

Reference

Anderson, R.R., Nixon, D.A., Akasha, M.A., 1988. Total and free thyroxine and triiodothyronine in blood serum of mammals. Comp. Biochem. Physiol. A 89, 401–404.

Maran, R.R., 2003. Thyroid hormones: their role in testicular steroidogenesis. Arch. Androl. 49, 375–388.

doi:10.1016/j.anireprosci.2008.05.112

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Factors associated with fertility in horses in a Danish equine practice after artificial insemination with frozen–thawed semen

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Timing of insemination, mare selection and stallion fertility have been described as important in the successful outcome of frozen semen inseminations (Vidament, 2005). To optimize the use of frozen–thawed semen, factors influencing pregnancy rates following insemination (AI) with frozen-thawed semen were studied in a Danish equine practice. Records of 490 mares (971 oestrus cycles), inseminated at one commercial insemination centre, were used in this study. The mares were examined by transrectal ultrasound during oestrus with 8-h intervals until ovulation, and inseminated post ovulation with frozen-thawed semen from 133 different stallions. Ovulation was not induced. In a total of 389 mares (789 oestrus cycles) pregnancy result was known. The design of the study was a retrospective observational accumulated cross-sectional study. Multivariable logistic regression was used to identify significant factors associated with the pregnancy result. Data were analysed using Statistical Analysis System (SAS vers. 9.1.3). The significance level was set at $P = 0.05$. The pregnancy rate per season was 61.4% and per cycle 39.1% with in average 1.98 cycles per mare. Following factors had a significant negative influence on pregnancy rates: (1) one or more positive endometrial culture ($P = 0.007$); (2) oestrus induction with prostaglandin ($P = 0.007$); (3) no or poor signs of oestrus ($P = 0.032$); (4) AI with one single mini straw (0.5 ml) compared to AI with macro straws (2.5–5.0 ml) or multiple mini straws (0.5 ml) ($P < 0.001$); (5) breed of the mare ($P = 0.005$); (6) number of cycles inseminated ($P < 0.001$); (7) accumulation of fluid before insemination ($P = 0.043$). Following factors had a significant positive influence on pregnancy results: (1) Caslick's operation ($P = 0.009$), (2) year of insemination ($P = 0.049$). The factors (1) accumulation of fluid after insemination; (2) time of the year for first insemination; (3) uterine treatment or lavage; (4) positive endometrial cytology; (5) twin pregnancy; (6) uterine cysts, had no influence on pregnancy rates. The negative influence of oestrus induction with prostaglandin is not believed to be due to prostaglandin by itself, since mares only were treated with prostaglandin in the case of poor oestrous signs. Endometrial culture was only performed on clinical indication or after more than two barren oestrous cycles. This could explain the negative

influence of culture on the pregnancy result. It should be noted, that if an endometrial pathogen was identified and treatment conducted, the chances of the mare to become pregnant was the same, as if no endometrial culture was performed.

Reference

Vidament, M., 2005. French field results (1985–2005) on factors affecting fertility results of frozen stallion semen. Anim. Reprod. Sci. 89, 115–136.

doi:10.1016/j.anireprosci.2008.05.113

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Transferrin concentrations in seminal fluid in horses: a marker for fertility

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Transferrin (Tf) is an iron-binding protein found in cells and physiologic fluids of vertebrates. In the human and bull, abnormal levels of Tf in seminal plasma have been associated with testicular dysfunction and reduced fertility (Gilmont et al., 1990; Zalata et al., 1996). In the horse, significantly higher Tf concentrations were found in the cauda epididymis (Parlevliet et al., 2004). The aim was to investigate seminal fluid Tf concentrations in post-pubertal colts and older stallions and to determine if these concentrations were correlated with fertility. Seminal fluid Tf concentrations (μg/mg protein) in the first and second ejaculates of post-pubertal colts ($n = 21$; 2–4 yrs) and in the ejaculates of breeding stallions ($n = 18$; 4–15 yrs) were measured during the breeding season and the latter were related to pregnancy rates of mares. The ejaculates were centrifuged for 15 min at 600 × g at 4 °C and the seminal fluid was stored at −20 °C for analysis of Tf by a validated enzyme-linked immunosorbant assay (ELISA) (Bidstrup et al., 2002). The Bio-Rad protein kit was used to measure protein content in all tissue sample extracts. Tf concentrations were normalized to protein concentrations in the sample. Results of Tf concentrations are expressed as the mean ± S.E.M. For statistical analysis the paired T-test and Pearson correlation test were used. There were no significant differences in seminal fluid Tf concentrations between the first (0.9 ± 0.2 μg/mg protein) and second ejaculate (0.6 ± 0.1 μg/mg protein). The seminal fluid Tf concentration in the ejaculates of the breeding stallions was 0.4 ± 0.1 μg/mg protein and the pregnancy rate was 78.3 ± 8.0%. No significant correlation between Tf and pregnancy rates was observed. However, in stallions ($n = 5$) older than 5 years with known semen parameters, a correlation of 0.7 with Tf concentrations and normal morphology was found. The presence of high concentrations of Tf in seminal fluid is in support of previous work showing Tf production by Sertoli cells from pubertal and postpubertal horses and high Tf concentrations in the epididymis (Parlevliet et al., 2004; Bidstrup et al., 2002). No correlation between seminal fluid Tf concentrations and pregnancy rates was observed but breeding stallions with poor fertility were not included in this study. In conclusion, Tf is present in stallion seminal fluid but the effect on fertility needs further research.