

Investigations into the Endocrine Regulation of Horse Testicular Polyunsaturated Fatty Acids Synthesis via Experimental Inhibition and Stimulation of Testicular Function.

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Polyunsaturated fatty acids (PUFA) metabolizing enzymes are expressed in the horse testis and epididymis suggesting an active PUFA metabolism during spermatogenesis and epididymal sperm maturation. The endocrine regulation of sperm PUFA synthesis is, however, unknown. The aim of the study was to fill this gap via an experimental inhibition (after GnRH vaccination) and re-stimulation (subsequent treatment with a GnRH agonist) of testicular function in stallions.

Shetland stallions were assigned to a GnRH vaccination group (Improvac; 400 µg/animal at 4 weeks interval; n=6) or a saline control group (n=6). Each vaccinated stallion was hemicastrated together with an age-matched control when testosterone concentration was <0.3 ng/ml (C1). Treatment of vaccinated stallions with the GnRH agonist buserelin (4 µg/day for 4 weeks and 8 µg/day for 6 weeks) started three weeks thereafter. The remaining testicle was removed when testosterone concentration increased to >0.5 ng/ml in vaccinated stallions (C2). Testicular tissues were stored at - 80°C for RT-qPCR and Gas chromatography–mass spectrometry (GC-MS) and fixed in formalin for histology. Statistical analyses were performed by non-parametrically by Mann-Whitney U test.

Relative mRNA abundance for the $\Delta 5$ and $\Delta 4$ -desaturases (*FADS1* and *DEGS1*) was lower in testicular tissue of vaccinated than control stallions at C1 ($p < 0.05$ and $p < 0.01$ respectively). At C2, *FADS1* and *DEGS1* mRNA abundance no longer differed between groups. No difference in mRNA abundance for the elongases 5 and 2 (*ELOVL5* and *ELOVL2*) was determined. Immunolabelling for *ELOVL5*, *FADS1* and *FADS2* in testicular tissue did not differ between the group. There were no significant differences between groups with regard to testicular tissue omega-3 and omega-6 PUFA content and the omega-3/omega-6 ratio at any time.

In conclusion, GnRH-vaccination changed expression of PUFA metabolizing enzymes at the mRNA level in testicular tissue, but this did not influence omega-3 and omega-6 PUFA content of testicular tissue. These results suggest that the regulation of testicular PUFA synthesis in stallions may not depend solely on the functional hypothalamic–pituitary–gonadal axis.