Expression of anti-Müllerian hormone in equine endometrium

M. S. Blanes, B. A. Ball
Department of Population Health and Reproduction, School of Veterinary Medicine, University of California, Davis, CA, USA

Classically, anti-Müllerian hormone (AMH) has been characterized for its role in the regression of the Müllerian ducts during male sexual differentiation. However, it is clear that AMH plays an important role after birth in regulation of normal function of the testis and ovary, and a recent publication described the expression of AMH and its receptor in human endometrium. The objective of the present study was to examine the expression of AMH in equine endometrium based upon immunohistochemistry and reverse transcription-PCR (RT-PCR). Mares (n = 6) were examined daily by palpation per rectum and transrectal ultrasound to determine stage of the estrous cycle. Endometrial samples obtained by biopsy at estrus and at Day 10 postovulation were fixed in formalin and snap frozen for subsequent mRNA isolation. Fixed tissues were embedded in paraffin and sectioned prior to analysis by immunohistochemistry using a goat polyclonal primary antibody directed against a C-terminal peptide antigen from human AMH (Santa Cruz Biotechnology; Santa Cruz, CA; USA) followed by a biotinylated second antibody (donkey anti-goat IgG) and detection using the Vectastain ABC detection kit (Vectorlabs; Burlingame, CA, USA). Specificity of the immunolabel for AMH was demonstrated by use of the corresponding blocking peptide. To confirm expression of AMH in the equine endometrium, RT-PCR was performed using equine specific oligonucleotide primers for AMH (forward: 5’-GAGCTGCAGGCGGCGGCG-3’; reverse: 5’-GGCCCCCGCGTGCGCTG-3’). Based upon immunohistochemistry, AMH was localized primarily in the glandular epithelium and beneath the luminal epithelium. There did not appear to be differences in AMH expression between estrous and diestrous endometrial samples. RT-PCR revealed the expected 230 bp amplicon confirming that the equine endometrium expresses the gene for AMH. Although the role of AMH in the equine endometrium has not been defined, we speculate that AMH may regulate cellular proliferation or apoptosis in the endometrium possibly by acting in a paracrine/autocrine manner.

Acknowledgements
The authors thank Lauren Mathewson and Jo Corbin for technical support. This research was supported by the John P. Hughes Endowment.

Keywords: Equine, endometrium, anti-Müllerian hormone