Using color flow Doppler ultrasonography to estimate progesterone concentrations at embryo transfer and during early pregnancy in recipient mares

Department of Equine Sciences, Faculty of Veterinary Medicine, Yalelaan 114, 3584 CM Utrecht, The Netherlands

Color-flow Doppler sonography (CF) has been described as a means of rapidly assessing corpus luteum (CL) function in cycling mares because luteal blood flow correlates with circulating progesterone (P₄) concentrations. The hypothesis was that CL size and blood flow would provide an indication of luteal function as represented by circulating levels of P₄ in mares at and following embryo transfer (ET). Day 8 equine embryos (n=48) were transferred into available recipient mares (ovulated 1 day before to 3 days after donor) as part of a commercial program. B-mode and CF sonography were performed using a MyLab™Five (Esaote, Maastricht, The Netherlands) immediately prior to ET (ET+0), and blood was collected from the jugular vein to measure plasma [P₄] (Coat-A-Count TKPG; Siemens Healthcare Diagnostic BV, Los Angeles, CA). For every detectable CL, fixed settings of 10 cm depth in B-mode (frequency 7.5MHz, maximum gain) and color mode (frequency 5.0MHz, 70% gain, pulse repetition frequency 1.0kHz) were used to examine size and vascularity. Three cross-sectional images at the position of maximal size and area of blood vessels of the respective CL were captured and stored. Measurements were repeated at day four after ET for all mares, and days 11, 18 and 25 in pregnant mares. The cross sectional area (corrected for presence of lacunae) and area of color pixels within the cross-section were analyzed using ImageJ software (National Health Institutes, Bethesda, MD), and statistical analysis was performed using SAS® (Version 9.4, SAS Inst., Cary, NC). None of CL area, area of color pixels or [P₄] at the time of ET were predictive of pregnancy outcome when analyzed with binary logistic regression. The total area of color pixels in the CL correlated significantly (r = 0.35 to 0.45), if only moderately, with [P₄] at all-time points except day 18 after ET (Spearman’s rank-order correlation). A significant correlation between CL area and [P₄] was evident until day 11 (r = 0.37 to 0.60). Corpus luteum vascularity (area of blood vessels) decreased significantly after day 18, whereas CL area had already decreased from day four (Wilcoxon signed rank test). These findings confirm that area of color pixels in the CL cross-section is a reasonable index of circulating [P₄] at the time of ET and during early pregnancy and can be used to indicate luteal insufficiency.

Keywords: Equine, mare, color flow Doppler, embryo transfer, progesterone