

Development and Use of 5 α -Dihydroprogesterone Enzyme Immunoassay for the Pregnancy Diagnosis in Asian Elephants

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Abstract

Three progestogen metabolites (5 α -Pregnane-3,20-dione, 3 α -hydroxy-5 α -pregnan-20-one, 17 α -hydroxyprogesterone) are identified during pregnancy in Asian elephants. However, lack of reliable hormonal assay prevents studying 5 α -DHP dynamics in Asian elephants. The present study attempted to develop an enzyme immunoassay (EIA) to measure the 5 α -DHP concentrations in serum of female Asian elephants, to understand the progestogen dynamics as an aid in diagnosing the pregnancy by measuring both 5 α -DHP and progesterone in the same sample, to find the relationship between 5 α -DHP and progesterone concentrations. Serial blood samples were collected from 4 pregnancy suspected Asian elephant cows in Pinnawala elephant orphanage, starting from 1-3 weeks after the date of mating. Serum samples (n=19) were tested for 5 α -DHP using a novel enzyme immunoassay, and for progesterone using an existing EIA. Progestogen concentrations of each elephant cow were plotted against the date of sampling. Log-transformed hormonal data were analyzed by GzLM of SPSS 25.0. The relationship between progesterone and 5 α -DHP was assessed by Spearman's correlation coefficient. Detection range of 5 α -DHP assay was 0.39-400 ng/mL. The intra-assay coefficient of variation and recovery rate were 15.02% (n=5) and 103.57 (n=5), respectively. Serum 5 α -DHP and progesterone concentrations ranged from 1.16-10.75 ng/mL and 0.07-2.80 ng/mL, respectively. Estimated overall means of 5 α -DHP concentrations were 5-times higher (p<0.001) than that of progesterone. There were differences (p<0.05) in individual animals for tested hormones. For each individual animal, 5 α -DHP concentrations were higher than progesterone concentration. There was a positive relationship between 5 α -DHP and progesterone (r =0.544; p<0.05; n = 19). In conclusion, an EIA to measure 5 α -DHP in Asian elephants was developed. In Asian elephant cows, 5 α -DHP concentrations were markedly higher than progesterone concentrations. Circulating 5 α -DHP and progesterone concentrations are positively correlated in female Asian elephants. 5 α -DHP appears to be a better alternative during the pregnancy diagnosis of Asian elephant cows, however, extended sampling period is required for the confirmation of findings.

Keywords: 5 α -DHP, Asian elephant, Enzyme immunoassay, Pregnancy, Progesterone

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