NON-INVASIVE MONITORING OF OESTROUS CYCLE IN SRI LANKAN ELEPHANT *(Elephas maximus maximus)* BY FAECAL PROGESTERONE MEASUREMENTS

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Introduction

In Sri Lanka, no other animal has been associated for so long with the people in their traditional and religious activates as elephants do. This association dates back to pre-Christian era, approximately over 5,000 years. Elephants are used for all-important ceremonial occasions, especially where pomp and pageantry are required, and thus, assume a significant position in Sri Lankan culture. It was estimated that about 3000 elephants are still found in the jungle, and about 500 are in captivity (Santiapillai & Jackson, 1990). However, the Captive Elephant Owners Association of Sri Lanka (CEASL) claims that only less than 200 elephants are in captivity at present. Since the capture of wild elephants is no longer practiced to replenish captive stocks it is essential and crucial to develop a successful breeding program for the captive population.

Routine monitoring of reproductive hormones is viewed as a valuable tool for making informed decisions about reproductive management of elephants in captivity (Brown, 1999). Thus, in the present study the relationship between blood and faecal progesterone (P4) profiles during the oestrous cycle were evaluated to monitor the estrus cycle.

Materials and Methods

Blood samples (50-60 samples/ animal) from ear vein were collected from privately owned elephants (n=5) without chemical immobilization, at 3-day intervals for 5-6 months. Serum was separated by centrifugation at 900 g for 20 min at 4°C, and stored at -30°C. Concentration of P4 in serum was determined in duplicate by second antibody enzyme immunoassay (EIA) after Diethyl ether extraction using 96-well ELISA plates.

Fecal samples (50-60 samples/ animal, about 50 g each) were collected from 10 she elephants at Pinnawala Elephant Orphanage, at 3-day intervals for 5-6 months, and were stored at -30°C. Samples were thawed and dried in an oven at 60°C for 18 hrs. A mixture of 0.4 ml methanol and 0.2 ml distilled water (4:1; v:v) was added to 1 g of each sample, and thoroughly mixed for 10 min. The mixture was centrifuged at 1200 g for 25 min. at 4°C. The supernatant was taken, evaporated and the residue was re-suspended in assay buffer (40mM PBS, 0.1% BSA, pH

7.2). Samples were then stored at -30°C until EIA was performed for P4.

The data on blood and fecal serum P4 profiles in different time points were compared using ANOVA followed by the Tukey-Kramer test for mean separation. Pearson Correlation coefficient (r) between blood and fecal P4 level was calculated using SAS computer package (2002).

Results

The data on serum P4 levels indicate that Sri Lankan elephants have an estrous cycle of 108 days with 66 days long luteal phase and a 42 days long follicular phase (Figure 1-A). Mean P4 level in fecal samples was 28.6 pg/g during the luteal phase, and it was undetectable during the follicular phase (Figure 1-B). A Strong positive correlation of 0.86 ($P<0.0001$) was observed between blood and fecal P4 profiles.

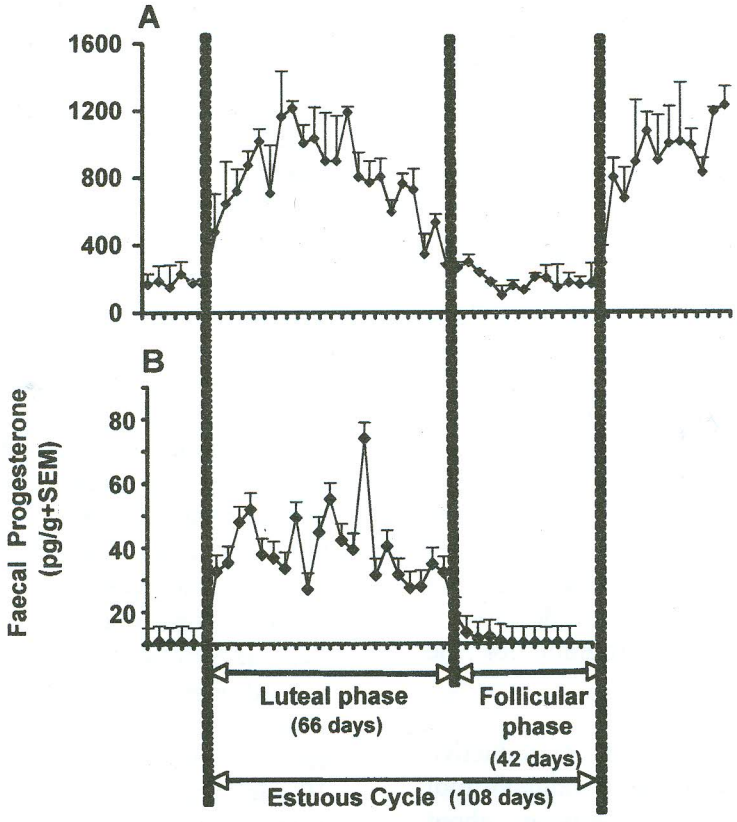

Fig.1. Progesterone profiles in blood (A), feces (B) during the estrous cycle of Sri Lankan elephants.

Discussion

The estrous cycle of an elephant consists of a follicular phase of approximately 6 weeks, followed by a non-pregnant luteal phase of approximately 8 weeks (Hodges *et al.*, 1998). The follicular period is characterized by two distinct waves or "phases" of follicular growth. Both phases of follicular growth are terminated by a surge of luteinizing hormone (LH) but, the first is an-ovulatory, while the second ends with ovulation (Hodges, 1998; Brown, 1999). The two surges of LH are consistent and occur approximately at 18 and 38 days, respectively after the end of the luteal phase (Brown, 1999). Thus, determination of drop in blood or fecal P4 levels at the end of luteal phase established in the present study can be efficiently utilized to an accurate prediction of ovulatory LH surge (or ovulation).

Conclusion

A non-invasive method of monitoring the reproductive cycle was established in this study. This method can be efficiently utilized to determine the best time of breeding, and to assist increasing the number of captive births to promote the *ex-situ* conservation of Sri Lankan elephants. Knowledge gained in the present study will help the wildlife conservationist, reproductive biologists and policy makers to plan future conservation strategies for Asian elephant.

Acknowledgement

This study was supported by the National Science Foundation of Sri Lanka (Grant No RG/2006/EB/03). Authors wish to thank Professor Akio Miyamoto, Obihiro University of Agriculture and Veterinary Medicine, Japan for the P4 EIA Kits.

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