Faecal Cortisol, Haematological and Serum Biochemical Parameters in Captive Asian Elephants in Three Protected Areas of Madhya Pradesh, India

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Introduction

The Asian elephant (*Elephas maximus*), a charismatic 'flagship species', is threatened by extinction. The development of self-sustainable captive populations is important to prevent captures from the wild.

Most trained captive elephants in India are maintained by the Forest Department and used for purposes such as immobilisation of carnivores, visiting and patrolling protected areas. Captive populations continue to decline due to failure in reproduction, diseases, and poor husbandry practices (Sarma 2011).

Traditionally mahouts manage the captive elephants. However, the skills and quality of mahouts have declined due to reduced monetary benefits, which affect the welfare and management of captive elephants (Vanitha *et al*. 2011). Increased use of non-traditional, unskilled and inexperienced mahouts leads to stress of elephants, which makes them violent and could cause human casualties. Also, stress may negatively impact immunity, agility and strength (Hing *et al*. 2016).

Stress may be evaluated by analysis of excreted hormones, particularly cortisol, which is found in faeces, urine, saliva and tears. Cortisol secretion in elephants is diurnal, with the highest levels in early morning, decline throughout the day and increase after midnight (Paudel *et al*. 2016).

Methodology

The study was conducted on captive elephants in the Kanha, Bandhavgarh and Panna Tiger Reserves of Madhya Pradesh.

Blood and faecal samples were collected from 30 elephants consisting of 14 males and 16 females in different age groups, comprising of 9 calves, 9 sub-adults and 12 adults. Age groups were defined as calves <5 years, sub-adults, 5–15 years and adults >15 years. All the animals sampled were apparently healthy and one female was pregnant. None of the sampled males were in musth but 5 in post-musth.

Blood samples were collected from ear veins (Fig. 1) in sterile vacutainers containing EDTA as an anticoagulant for haematological studies and without anticoagulant for serum biochemical analysis. The serum was separated within 2–3 hours of blood collection and stored at 4ºC prior to analysis. Haematological values were assessed using a semi-auto haematology analyser (PG-6800), liver function and kidney function tests were done using ERBA diagnostic kits and semi-auto analyser mini CHEM 100, ARK diagnostics, Mumbai.

Freshly voided faecal samples were collected in a plastic container and stored at -20ºC prior to analysis of faecal cortisol metabolites (Fig. 2)

Figure 1. Collection of blood sample.

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Table 1. Faecal cortisol metabolite levels of 30 elephants in three Tiger Reserves (mean±SE).

Location	N	Sex		Age group			Total
		Male	Female	Calf	Sub-adult	Adult	
Kanha	10	265.3 ± 22.5	187.5 ± 56.2	220.0 ± 90.2	173.3 ± 96.1	272.7 ± 21.5	234.2 ± 27.4
Panna	10	252.3 ± 22.3	262.0 ± 36.1	176.7 ± 92.6	290.0 ± 12.7	277.0 ± 54.5	250.8 ± 16.8
Bandhavgarh	10	219.4 ± 23.4	168.7 ± 76.8	173.3 ± 96.1	272.7 ± 21.5	246.0 ± 35.5	232.2 ± 19.6
Total	30	245.7 ± 22.7	229.6 ± 44.1	190.0 ± 93.0	280.9 ±19.3	266.8 ±37.1	239.6 ± 12.2

using ELISA diagnostic kits (Cortisol Enzyme Immunoassay Kit, DetectX, ARBOR ASSAYS).

Results and discussion

Cortisol metabolite levels

The mean faecal cortisol metabolite levels observed from Kanha, Panna and Bandhavgarh were similar ($p>0.05$) (Table1). Estimates of faecal cortisol levels by Chichilichi *et al*. (2018) in Odisha, also found no significant variation between three free ranging populations, which however were higher than in captive elephants from a single location. Therefore, faecal cortisol may be similar in populations living in comparable circumstances.

We found no significant difference in cortisol metabolite levels between males and females (Table 1). Cortisol metabolites were significantly higher in sub-adults and adults than in calves (Table 1). Stead *et al*. (2000) reported higher levels of glucocorticoids in juvenile African elephants kept in small enclosures, compared to those housed in a large area. The elephants we sampled were kept under semi-captive conditions, but sub-adults were confined for training and used for patrolling in the dense forest areas inaccessible to vehicles. This may explain their having higher levels of cortisol metabolites among the three age classes in Panna and Bandhavgarh (Table 1). However, this pattern was not observed in Kanha, which also had high levels in calves, due to unknown reasons.

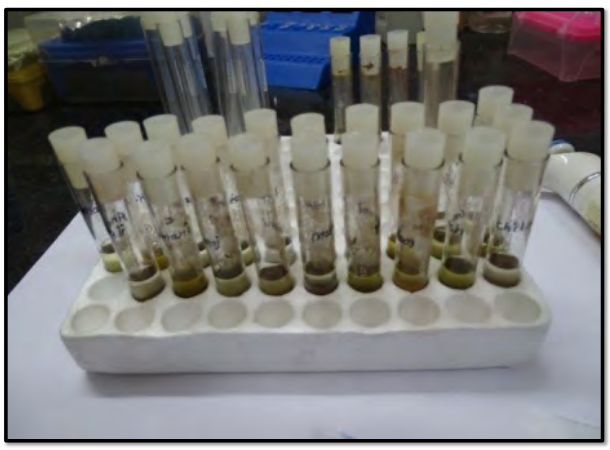

Figure 2. Preparation of faecal samples for cortisol analysis.

Haematological and biochemical parameters

Overall, elevated values of total erythrocyte, leukocytes and neutrophils as well as SGPT, SGOT, alkaline phosphatase and blood urea nitrogen were recorded (Table 2). High RBC and neutrophil counts may be correlated to secretion of glucocorticoids in stressful conditions (Benjamin 2013). Elevation of SGPT and SGOT may indicate parasitic infestation.

There were no significant differences between males and females in haematological and biochemical parameters ($p>0.05$, Table 2). Similar findings were observed by Silva & Kuruwita (1993), Wijesekera *et al*. (2008) and Jayamethakul *et al*. (2017).

The observed mean values of TLC, TEC, HB, HCT, monocytes and neutrophils were significantly above ($p<0.05$) normal values. Similarly,

Table 2. Haematological and biochemical values of male and female elephants.

Parameters	Male	Female
TLC ($x10^3\mu l$)	44.85 ± 9.39	46.79 ± 11.05
TEC ($x10^6\mu l$)	8.58 ± 0.61	7.40 ± 0.37
Monocytes (%)	5.50 ± 2.08	6.55 ± 2.02
Neutrophils (%)	59.55 ± 11.13	55.25 ± 8.05
SGPT ($\mu g/l$)	8.43 ± 1.24	7.43 ± 1.20
SGOT ($\mu g/l$)	42.15 ± 5.02	28.96 ± 4.46

Table 3. Age wise haematological and biochemical values (mean ± SE)

Parameters	Calves	Sub-adults	Adults
TEC (x$10^6\mu$l)	$4.40^{b} \pm 0.47$	$9.01^{ab} \pm 0.88$	$8.66^{ab} \pm 0.71$
HB (g/dl)	$13.94^{ab} \pm 2.77$	$17.35^{ab} \pm 4.45$	$16.98^{a} \pm 3.69$
HCT (%)	$43.66^{b} \pm 4.95$	$58.73^{ab} \pm 8.74$	$58.73^{ab} \pm 8.74$
Monocytes (%)	$03.99^{a} \pm 2.33$	$6.21^{a} \pm 2.25$	$5.08^{b} \pm 2.83$
Lymphocytes (%)	$41.22^{a} \pm 10.07$	$74.88^{b} \pm 11.43$	$63.91^{a} \pm 8.59$
Neutrophils (%)	$44.88^{a} \pm 7.03$	$68.22^{b} \pm 9.63$	$65.00^{a} \pm 13.73$
SGPT (µg/l)	$7.43^{a} \pm 0.79$	$7.06^{ab} \pm 0.87$	$7.1^{a} \pm 1.52$
SGOT (µg/l)	$18.56^{a} \pm 0.56$	$49.98^{a} \pm 4.75$	$42.51^{ab} \pm 5.29$
Uric acid (mg/dl)	$3.84^{a} \pm 0.21$	$4.88^{ab} \pm 0.17$	$3.99^{a} \pm 0.25$

[a,b,c] Means with different superscripts are significantly different ($p<0.05$).

biochemical parameters, SGPT, SGOT, blood urea nitrogen, uric acid, alkaline phosphatase were significantly ($p<0.05$) higher than normal (Table 3). Possible reasons for increased haematological and biochemical values may be gastrointestinal parasitism and stress.

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