Np
DOI: 10.30954/2277-3371.01.2018.5

Theriogenology Insight: 8(1): 33-36, April 2018

Effect of Age, Month and Season on Testosterone Levels in Farm Raised Ostrich (*Struthio camelus)*

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ABSTRACT

A study was carried out to assess the effect of age, month and season on blood and seminal plasma teststerone of individual ostrich aged four to nine years for a period of 12 months in tropical climate of India. The birds were grouped into two categories viz., four to six and seven to nine years-old. Highly significant (P<0.01) differences were observed in blood plasma testosterone level in individual birds with a mean value of 9.17 ± 0.28 ng/ml. Similarly, seminal plasma testosterone level also showed significant difference (P<0.05) among individual birds and the mean value was 7.26 ± 0.31 ng/ml. Between age groups (four to six and seven to nine year-old ostrich), there was no significance difference in the levels of blood plasma (9.28 ± 0.71 vs. 9.10 ± 0.29 ng/ml) and in seminal plasma testosterone (7.21±0.29 vs. 7.28 ± 0.54 ng/ml). The blood plasma and seminal plasma testosterone levels showed highly significant differences (P<0.01) among seasons.

Keywords: Teststerone Level, Seasonal variation, Ostrich

Ratite species such as ostrich (*Struthio camelus*), emu (*Dromaius novaehollandiae)* and rhea (*Rhea americana*) are fundamentally attractive for farming to produce leather, meat, oil and feathers. Ostrich are very adaptable and can be farmed in almost all climatic conditions, but are ideally suited to dry and arid climate. Ostrich are considered to be seasonal breeders, although they may also breed all year round (Degen *et al.*, 1994). During the breeding season, male and female ostrich are reproductively active for six to eight months of the year. On farm, ostrich are kept in breeding paddocks, most frequently at a ratio of one male to two or three females, or in large breeding colonies that contain several males and females at a similar ratio. Ostrich farming in India has been established with limited success. Unpredictable egg production, unstable fertility, poor hatchability and poor chick survival are some of the major constraints in viable ostrich farming. Not much work has been done on the testosterone profile in ostrich; hence the present work becomes a pioneering study in India. With these backgrounds, the present investigation has been taken up to assess the both blood and seminal plasma teststerone levels during breeding and non-breeding seasons in tropical climate of India.

MATERIALS AND METHODS

This experiment was carried out at Post Graduate Research Institute in Animal Sciences, Tamil Nadu Veterinary and Animal Sciences University, Kattupakkam, Kanchipuram district in Tamil Nadu during 2014 - 2016. The station is situated approximately at 12.5ºN latitude and 80º to 81ºE longitudes and at the height of 48 meter above mean sea level. Being nearer to East coast of India it enjoys a tropical maritime monsoon climate. During this study period, the average high and low temperature, relative humidity and the annual rainfall were recorded as 34.2ºC, 23.2ºC, 87.3 per cent and

1391 mm, respectively. This station gets most of its seasonal rainfall from the northeast monsoon i.e. during October to December (Indian Metrological Department, Pune).

Estimation of testosterone

Eight adult male ostrich aged four to nine years were taken for this study; and housed in trios i.e. one male with two females providing a floor space of 1500 m^2 per trio. All the birds were housed under standard management conditions. A total of each 96 blood plasma and seminal plasma samples were utilized to assess the testosterone. Semen samples followed by blood collection were carried out for a period of 12 months. Both the seminal and blood plasma sample were centrifuged (2500 rpm) for 15 min and the resulting blood plasma and seminal plasma were collected and stored at -80°C until assayed. The quantitative determination of testosterone in blood and seminal plasma of male ostrich were carried out by competitive immunoassay by using Testosterone ELISA kit (Enzo Life Sciences Inc, Farmingdale, NY 11735). Test procedure was carried out as per the Catalog #: ADI-900-065, Enzo Life Sciences Inc, Farmingdale, NY 11735.

Statistical Analyses

The effect of various sources of variation on testosterone were analysed by SPSS 20.0 (SPSS Inc., Illinois, USA) programme using One-way ANOVA as per the procedure of Duncan'smultiple comparison test (Duncan, 1955). A value of $P < 0.05$ was considered statistically significant.

RESULTS AND DISCUSSION

The mean blood plasma and seminal plasma testosterone levels of ostrich observed in this study were 9.17 ng/ml and 7.26 ng/ml, (Table 1) which are higher than the value observed by Walsangkar (2010) with corresponding value of 9.09 ng/ml and 1.07 ng/ml, respectively. Between age groups (four to six and seven to nine year-old ostrich), there was no significance difference in the levels of blood plasma (9.28 ± 0.71 vs. 9.10 ± 0.29 ng/ml) and in seminal plasma testosterone (7.21±0.29 vs. 7.28 ± 0.54 ng/ml) (Table 2). Further, noticeable lower level of blood plasma testosterone was also reported by Degen *et al.* (1994) in ostrich (2 to 4 ng/ml), Malecki *et al.* (1997) and Blache *et al.* (2001) in emu. The difference observed in this study may be due to genetic makeup of the birds, seasonality of breeding, geographical location, nutrition and availability of feeding resources and environmental factors such as variation in climate and rainfall.

Table 1: Blood plasma and seminal plasma testosterone levels (mean ± SE) of individual ostrich

Bird No.	Blood plasma testosterone (ng/ml) (n=12)	Seminal plasma testosterone (ng/ml) (n=12)
01	$9.59^{bc} \pm 0.80$	$7.40^{ab} \pm 0.72$
02	$7.90^{bc} \pm 0.39$	$5.60^{c} \pm 0.29$
03	$9.54^{bc} \pm 0.70$	$8.05^{a} \pm 0.68$
04	$9.67^{bc} \pm 0.58$	$8.07^{a} \pm 0.40$
05	$7.12^{c} \pm 0.55$	$5.45^{c} \pm 0.33$
06	$12.16^{a} \pm 0.90$	$8.24^{a} \pm 0.57$
07	$9.89^{b} \pm 0.50$	$8.60^{a} \pm 0.53$
08	$7.98^{bc} \pm 0.57$	$6.56^{c} \pm 0.43$
Over all mean	9.17 ± 0.28	7.26 ± 0.31
F value	4.516	2.761
Significance	**	*

Figure in parenthesis indicate number of observation/bird; Means bearing different superscripts within the same column differ significantly; **Highly significant (P<0.01); * Significant (P<0.05).

Table 2: Effect of age on blood plasma and seminal plasma testosterone levels (mean ± SE) in ostrich

Age group	Blood plasma Testosterone (ng/ml) (n=48)	Seminal plasma testosterone(ng/ml) (n=48)
4-6 year	9.28 ± 0.71	7.21 ± 0.29
7-9 year	9.10 ± 0.29	7.28 ± 0.54
Overall mean	9.17 ± 0.28	7.26 ± 0.31
F value	1.225	1.834
Significance	Not significant	

Figure in parenthesis indicate number of observation/age group.

Print ISSN: 2249-6610

Effect of month on blood plasma and seminal plasma testosterone

Blood plasma testosterone level showed highly significant difference (P<0.01) between months (Table 3). Higher level of blood plasma testosterone was observed between September and April (9.9 ng/ml) than from April to October (7.95 ng/ml), which was comparable with earlier findings of Degen *et al.* (1994) in ostrich where maximum level was found between April to July (4.0 ng/ml) and minimum level between August to December (2.0 ng/ml). Further, complementary results were also observed by Walsangkar (2010) in ostrich with the highest level (14.89 ng/ml) during peak season i.e. January to March. Similarly, Valdez *et al.* (2014) had observed a significant differences on blood plasma testosterone level among months with maximum concentration during August to January (5.0 ng/ml) than April to May (2.0 ng/ml) in rhea.

Table 3: Effect of month on blood plasma and seminal plasma testosterone levels (mean ± SE) in ostrich

Months (2015)	Blood plasma testosterone (ng/ml) (n=8)	Seminal plasma testosterone (ng/ml) (n=8)
January	$9.54^{ab} \pm 0.98$	8.28 ± 0.71
February	$12.54^{a} \pm 1.45$	9.88 ± 2.26
March	$11.07^{ab} \pm 1.32$	9.93 ± 2.54
April	$8.84^{ab} \pm 0.10$	7.30 ± 0.95
May	$7.38^{b} \pm 0.71$	6.81 ± 0.79
June	$8.08^{b} \pm 0.73$	5.92 ± 0.41
July	$8.16^{b} \pm 0.50$	5.94 ± 0.36
August	$8.21^{b} \pm 0.81$	6.52 ± 0.35
September	$8.57^{ab} \pm 0.76$	5.72 ± 0.44
October	$8.90^{ab} \pm 0.85$	7.95 ± 0.65
November	$9.65^{ab} \pm 0.53$	7.58 ± 0.40
December	$10.09^{ab} \pm 0.65$	8.81 ± 0.49
Overall mean	9.17 ± 0.28	7.26 ± 0.31
F value	2.515	1.930
Significance	**	NS

Figure in parenthesis indicate number of observation/month; Means bearing different superscripts within the same column differ significantly; **Highly significant (P<0.01); NS- Not significant.

Effect of season on blood plasma and seminal plasma testosterone

Seasonal influence on blood plasma and seminal plasma testosterone of ostrich are highly significant (P<0.01). Both blood plasma and seminal testosterone levels escalated to increase from northeast monsoon (9.55 ng/ml and 8.12 ng/ml) and maintained peak during winter (11.04 ng/ml and 8.85 ng/ml) and summer (9.10 and 7.95 ng/ml), thereafter the level gradually reduced during southwest monsoon (8.31 ng/ml and 6.00 ng/ml) (Table 4).

Table 4: Effect of season on blood plasma and seminal plasma testosterone levels (mean ± SE) in ostrich

Season	Blood plasma testosterone (ng/ml)	Seminal plasma testosterone (ng/ml)
Winter (Jan-Feb) (n=16)	$11.04^{a} \pm 0.93$	$8.85^{a} \pm 0.90$
Summer (March-May) (n=24)	$9.10^{b} \pm 0.65$	$7.95^{a} \pm 0.91$
Southwest monsoon (June-Sep) (n=32)	$8.31^{b} \pm 0.34$	$6.00^{b} \pm 0.19$
Northeast monsoon (Oct-Dec) (n=24)	$9.55^{ab} \pm 0.39$	$8.12^{a} \pm 0.31$
Overall mean (n=96)	9.17 ± 0.28	7.26 ± 0.31
F value	3.919	4.173
Significance	**Highly significant (P≤0.01)	

Figure in parenthesis indicate number of observation/season; Means bearing different superscripts within the same column differ significantly.

This pattern is coinciding with observations of Walsangkar (2010) in ostrich who found, increasing level of blood and seminal plasma testosterone during early (3.5 ng/ml and 0.45 ng/ml) and peak seasons (14.89 ng/ml and 1.7 ng/ml). Further, the present finding was also in agreement with earlier reports of Sundaresan (2014) in emu, who observed increasing level

of serum testosterone level from early to peak seasons (2.47 to 11.69 ng/ml) and thereafter the level reduced (7.43 ng/ml) at the end of the breeding season. However, comparatively higher levels of blood testosterone during off-season and breeding season were also observed by Bertschinger *et al.* (1992) in ostrich (19.81 and 144.90 ng/ml, respectively) and Goes *et al.* (2010) in captive rhea (5.57 and 53.28 ng/ml, respectively).

CONCLUSION

The study concluded that, the elevated level of both blood plasma and seminal plasma testosterone observed during winter and summer season than southwest and northeast monsoon seasons suggested that, testosterone were highly influenced by season and it gives an insight to the physiological signaling in ostrich reproduction.

ACKNOWLEDGEMENTS

We greatly acknowledge Tamil Nadu Veterinary and Animal Sciences University, Chennai, Tamil Nadu, India for providing financial assistance for the accomplishment of the doctoral study.

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