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Research Article

The influence of seasonal variation on the serum progesterone of Black Bengal does (*Capra aegagrus hircus)*

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Abstract: The Black Bengal does of Indian sub-continent generally show a noted seasonal variation during in reproductive activity**.** India has a good quality of Black Bengal goat populations, which has significant role in the lives of the local goat rearers. The objective of the present study is to realize the influence of seasonal variation on the serum progesterone of Black Bengal does (*Capra aegagrus hircus*) in two different agro-climatic regions in India. The highest assessment of temperature (42.6 ± 1.5 ºC) has been reported during the month of April or May in the season of pre monsoon in Purulia. However, the lowest assessment of temperature (8.6 ± 0.9 ºC) has been reported during the month of December or January in the season of post monsoon again in Purulia. Progesterone hormone has been analyzed in the serum samples collected once a week. It has been observed that, from January to April for both of the regions of Purulia and Nadia and the month May has the lowest serum progesterone level in Purulia (0.32 ± 0.06 ng/ml) and the serum progesterone hormone concentration stayed approximately the same from January to March in Purulia as similar to Nadia. However, the serum progesterone reached to its peak level in November in both the region Purulia (4.52 ± 0.28ng/ml) and Nadia (5.55 ± 0.18 ng/ml) respectively**.** It can also be presume that the early months of the post monsoon season may be taken as an alternate breeding seasons for the Black Bengal breeds**.**

Keywords: Progesterone; Doe; Purulia; Nadia; Pre-monsoon; Post-monsoon.

INTRODUCTION:

Progesterone (or P4), additionally called pregn-4-ene-3e,20-dione [1] is an endogenous steroid and progestogen sex hormone concerned in the estrous cycle in goats. It belongs to a group of steroid hormones known as the progestogens [2] and is the predominant progestogen within the body. Progesterone is also a vital metabolic intermediate within the manufacturing of different endogenous steroids, consisting of the sex hormones and the corticosteroids, and plays an essential role in brain as a neurosteroid [3]. Throughout implantation and gestation, progesterone decreases the maternal immune response to allow the pregnancy [4]. In addition progesterone inhibits lactation in the course of pregnancy. The fall in progesterone levels following shipping is one of the triggers for milk production. Progesterone performs a crucial role in mammary gland development in does [5]. In conjunction with prolactin, it mediates lobulo-alveolar maturation of the breasts during in the course of pregnancy to make access for milk production, and therefore lactation and breast feeding after childbirth [6]. Progesterone and its neurosteroid lively metabolite allopregnanolone appear like importantly involved in sex power in females [7].

Bauernfeind and Holtz [8] demonstrate the progesterone levels in serum of goats measured by enzyme immunoassay techniques. Assessment of progesterone degrees throughout distinctive physiological levels in small livestock animals is considered one of the maximum vital parameters to determine their fertility popularity [9]. In goats, patterns of reproduction reveal the appearance of a self persistent endogenous rhythm. Such endogenous rhythm has been synchronized or stimulated by photoperiod [10]. However, other environmental stimuli such as availability of food [11-12], number of does in the folks and the interaction of the doe with them [13-14] may not be ignored as the controllers of seasonality of reproduction [15].

The objective of the present study is to comprehend the influence of seasonal variation on the Progesterone hormone of Black Bengal does **(***Capra aegagrus hircus)* [16] in two different agro-climatic regions in West Bengal, India. Moreover, this will be a probable contribution to this field of research on Black Bengal goats where inadequate researches have been

available and these results can be useful to the determine the best possible season of reproduction alternate to monsoon.

MATERIALS AND METHODS

Animals:

The animals were taken from the local rearers of Brindabanpur (23˚20´5´´N, 86˚3´29´´E); Chatambari (23˚20´36´´N, 86˚4´4´´E); Jaharhatu (23˚21´6´´N, 86˚2´36´´E); Lakshmipur (23˚15´N, 86˚5´5´´E) and Murguma (23˚21´11´´N, 86˚31´34´´E) Purulia district and from the Mohanpur farm (22˚56´N, 88˚31´E) and Gayeshpur (KVK) farm (22˚57´19´´N, 88˚28´45´´E) of Nadia district both from the state of West Bengal but in two different agro-climatic regions, there are no feed restriction to the goats. Animals had been free to choose their feed requirements by foraging and grazing on natural vegetation and pasture lands. According to Nandi *et al*. [17], cent percent of goat in West Bengal have been reared through grazing. Addition to that each goat is feed with 1-2 kg of green fodder (like maize), 0.5-1 kg of dry fodder, 100 -200 grams of readymade seeds as concentrates and 20-50 grams of mineral mixtures (Argemin Forte) for a day. Animals were maintained in its ambient condition for four weeks prior to blood sampling [18]. For each month and for each region ten (n=10) animals has been taken.

Study areas:

Planning Commission of India, during 2006 has demarcated the geographical area of India into 15 agro-climatic regions**.** The present studies have been carried out into two agro-climatic zones of India. These are as follows:

Purulia, fall under Eastern Plateau and Hills region of India [19]. This agro-climatic zone is Located at the southern tip of Bihar. It receives about 1,200 mm of rainfall annually. The climate is moist sub-humid to sub-humid and the soil is red loamy, red and yellow. Average annual rainfall is varies from 1100 to 1500 mm. The humidity is high during in monsoon season, from seventy five percent to eighty five percent. But in scorching summer it goes down from seventy thirty percent to twenty five percent. Temperature varies over a wide range from 7˚C in winter to 46.8˚C in the hot summer (Table 1). Due to undulated topography just about fifty percent of the total rainfall flows away as run off [19]. The total goat population of Purulia has been recorded as more than 813 thousand **[**20**]**.

Nadia, fall under Lower Gangetic Plains region of India **[**21**]**. About 68% of the land is cultivated. The soil of this sub-zone is deltaic alluvial and the climate is per humid to humid. Annual rainfall is ranging between 1200 mm to 1700 mm. The zone has a tropical climate with a short spell of winter season. The hot season lasts from mid-March to mid-June, with the day temperature ranging from 38°C to 45°C in different parts of this region. The monsoon arrives by the month of middle June. Winter extends about three months; the average minimum temperature not goes down below 10°C (Table 1). Average rainfall of this area is 1,435.8 mm **[**21**]**. The total goat population of Nadia has been recorded as more than 952 thousand **[**20**]**.

Climatological measurement:

The three year data on temperature of the study area has been collected from the state meteorological department and the mean of the three years with standard deviation was calculated (Table 1) using MS-Excel 2007 and shown here in a tabular form (Table 1) **[**22**]**.

Table-1: Mean maximum and minimum temperature (2011-2016)

			March	April	May	June	
Temperature	Purulia	Max (ºC)	40.4 ± 2.3	42.6 ± 1.5	42.6 ± 1.5	38.6 ± 5.0	Pre-Monsoon
		Min (ºC)	21.6 ± 10.9	21 ± 0.7	22.4 ± 1.1	23.2 ± 0.8	
	Nadia	Max (ºC)	37 ± 2.45	38 ± 1	39.2 ± 1.5	36 ± 4.7	
		Min (ºC)	16 ± 3.9	19.4 ± 3.3	23.4 ± 1.5	23.6 ± 1.3	
			November	December	January	February	Post-Monsoon
	Purulia	Max (ºC)	32.0 ± 1.0	30.8 ± 2.2	30.0 ± 2.5	34.6 ± 2.5	
		Min (ºC)	13.6 ± 1.1	9.4 ± 1.7	8.6 ± 0.9	11 ± 2.5	
	Nadia	Max (ºC)	31.6 ± 1.2	28.75 ± 0.5	28.6 ± 1.5	32.2 ± 3.6	
		Min (ºC)	14.4 ± 2.8	11.5 ± 1.3	10.2 ± 1.6	12.6 ± 3	

Blood collection and clinical analysis:

The blood have been collected from apparently healthy goats using purposive sampling technique **[**23**]** for the year and categorized into two seasons. The seasons include pre-monsoon and post-monsoon. About 4 ml of blood was collected by jugular venipuncture from each goat between 12 o'clock to 2 pm under the intense sun using disposable needles (SRS^{TM} Sterivan) and vacutainer tubes (Vacutech) **[**24**]**. Blood samples have been collected once in a week. The blood has been collected always at the same time of the day to avoid circadian variations [25]. The collected blood has been dispensed into vials and labelled accordingly.

ELISA:

To analyse the progesterone hormone, the serum has been taken for the ELISA using the standard bovine progesterone ELISA kits (containing 96 well ELISA plate) obtained from Biosource (MBS026041). Serum progesterone (ng/ml) has been performed using standard sandwich ELISA technique as directed by Sogorescu *et al.* [15] using automated (Thermo-scientific) ELISA reader.

Statistical analysis:

The statistical analysis of the data was performed using SPSS 21.01 **[**26**]**. A one-way analysis of variance (ANOVA) test has been used with *post doc* modification as Duncan's Multiple Range Test (DMRT) to determine the effects of seasons on the Black Bengal bucks serum progesterone parameters studied here [**27**]. Mean separation and standard error has been calculated using MS-Excel 2013.

RESULTS:

During in the month of March, in the season of pre-monsoon (Table 2), the progesterone hormone level in the serum of Black Bengal does has been found significantly ($p<<0.01$) lower in Purulia (2.5 ± 0.8 ng/ml) than the data obtained in Nadia (3.31 ± 0.19 ng/ml). During in the month of April, the P4 level in the serum is found significantly ($p<<0.01$) lower in Purulia (0.42 ± 0.9 ng/ml) than that of Nadia (0.82 ± 0.05 ng/ml) (Figure: 1). During in the month of May the progesterone hormone level in the serum of Black Bengal does in Purulia have been reduced to 0.32 ± 0.06 ng/ml (Table 2), which is the lowest value of the progesterone hormone level obtained in the serum of Black Bengal does in the present study (Figure 1). The progesterone hormone level in the serum of Black Bengal does obtained during this time in Purulia has been significantly lower ($P<<0.01$) than the value obtained (0.73 ± 0.05 ng/ml) in Nadia. Similar kind of results have been found during in the month of June, here also the progesterone hormone level in the serum of Black Bengal does in Purulia (0.76 ± 0.06 ng/ml) have been found significantly ($P<<0.01$) lower than the progesterone hormone level found in the serum of Black Bengal does in Nadia (1.33 ± 0.19 ng/ml) (Table 2).

On the other hand, during in the season of post-monsoon (Table 3) the progesterone hormone level in the serum of Black Bengal does in Nadia (5.55 ± 0.18 ng/ml) in the month of November has been found significantly ($P<<0.01$) higher than that of Purulia (4.52 ± 0.28 ng/ml). The value obtained in Nadia i.e. 5.55 ± 0.18 ng/ml, has been the highest value of the progesterone hormone in the serum obtained during the present study (Figure 1). The value of the progesterone hormone found in Purulia (4.52 ± 0.28 ng/ml), during in the month of November also the highest value among the months studied here in Purulia (Figure 1). More or less similar kind of result has been obtained during the month of December, where the progesterone hormone level in the serum of Black Bengal does in Nadia (4.52 ± 0.19 ng/ml) has not been found significantly differs from the progesterone level found in Purulia (4.43 ± 0.29 ng/ml) (Table 3). However the data collected on the progesterone hormone level in the serum of Black Bengal does obtained during the months of January and February are more or less similar in Purulia and Nadia and there are no significant difference have been observed between them (Table 3).

On the other hand, through the DMR test it has been observed that the highest serum progesterone hormone concentration in Black Bengal goat has been observed during the month of November in Purulia. This level of progesterone hormone in November has been significantly ($P<0.01$) higher than the other months of the year. However, the serum progesterone hormone concentration has been found in the month November in Purulia is the highest serum progesterone hormone level has been recorded for this study. The month of December and January also shows higher serum progesterone hormone concentration, which has been lower than the concentration level of November and higher than the rest of months of the year. The February shows significantly ($P<0.05$) higher rate of expression than the months of pre monsoon season. The month of March shows a higher (but comparatively more lowly than the months of post monsoon) level of serum progesterone hormone concentration than the other months of pre monsoon season. During in the season of pre monsoon in Purulia, the month of April and May show similar level of serum progesterone hormone concentration, which are shows significantly ($P<0.05$) lowest level of serum progesterone hormone concentration, than the month of June in Purulia (Figure: 2**)**.

Through the DMR test it has also been observed that the highest serum progesterone hormone concentration in Black Bengal goat has been observed during the month of November in Nadia. This level of progesterone hormone in November has been significantly ($P<0.01$) higher than the other months of the year. The month of December and January also shows higher serum progesterone hormone concentration, which has been lower than the concentration level of November and higher than the rest of months of the year but the difference between them ($p>0.05$) is not significant. The February shows higher rate of expression than the months of pre monsoon season. The month of March shows a higher (but comparatively lesser than the months of post monsoon) level of serum progesterone hormone concentration than the other months of pre monsoon season. During in the season of pre monsoon in Nadia, the month of April and May show similar level of serum progesterone hormone concentration, which are shows significantly ($P<0.05$) lower level of serum progesterone hormone concentration, than the month of

June in Nadia (Figure: 3**)** but the difference between them is not significant (p>0.05).

Table 2: The one-way ANOVA showing the effects of regional variations on the serum concentration of progesterone (ng/ml) in the Black Bengal does during in the different months of pre-monsoon season

Months	Purulia	Nadia	Over all	*P-value*
March	2.5 ± 0.8	3.31 ± 0.19	2.9 ± 0.44	0.0000004 **
April	0.42 ± 0.9	0.82 ± 0.05	0.62 ± 0.23	0.0000038 **
May	0.32 ± 0.06	0.73 ± 0.05	0.53 ± 0.22	0.000000004 **
June	0.76 ± 0.06	1.33 ± 0.19	1.05 ± 0.32	0.0000054 **

*: P < 0.05; **: P < 0.01; NS: not significant

Table 3: The one-way ANOVA showing the effects of regional variations on the serum concentration of progesterone (ng/ml) in the Black Bengal does during in the different months of post-monsoon season

Months	Purulia	Nadia	Over all	*P-value*
November	4.52 ± 0.28	5.55 ± 0.18	5.03 ± 0.58	0.0000038**
December	4.43 ± 0.29	4.52 ± 0.19	4.47 ± 0.24	0.504 ^{NS}
January	4.34 ± 0.8	4.36 ± 0.29	4.35 ± 0.21	0.849 ^{NS}
February	4.31 ± 0.21	4.29 ± 0.2	4.3 ± 0.2	0.939 ^{NS}

*: P < 0.05; **: P < 0.01; NS: not significant

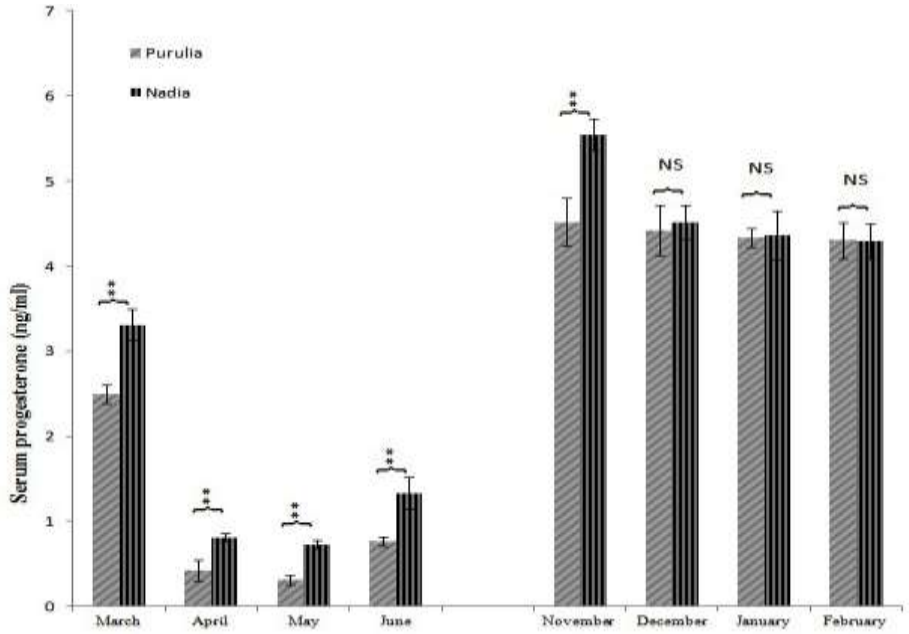

Fig-1: A graphical representation of the one-way ANOVA showing the influence of seasonal variation on the progesterone hormone of Black Bengal does in the different months (mean ± SD) of pre-monsoon and post-monsoon seasons. Here ** denotes P<0.01, * denotes P<0.05 and NS denotes no significant difference has been found between means.

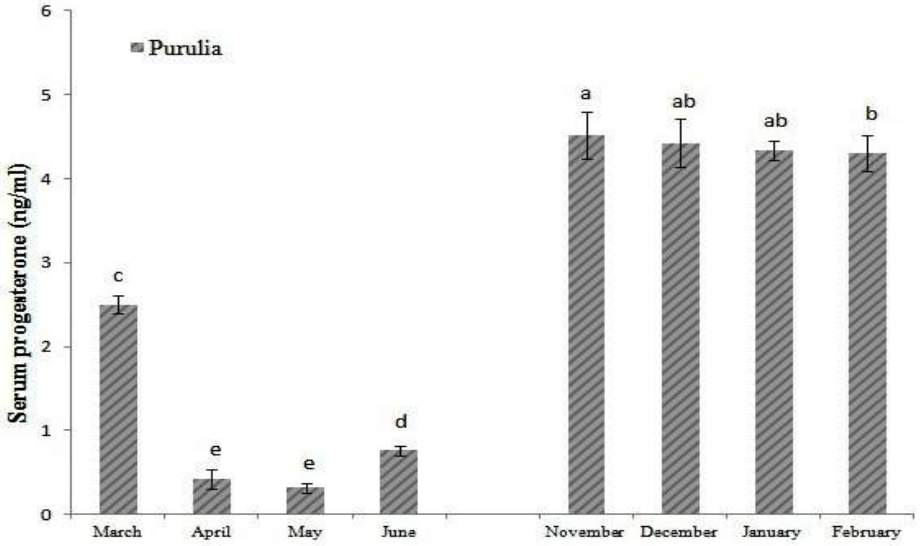

Fig-2: A graphical representation of the one-way ANOVA with *post doc* modification (DMRT) showing the effects of seasonal variations on the serum concentration of progesterone (ng/ml) in the Black Bengal does during in Purulia. Means bearing different superscripts (a–e) differ significantly (P < 0.0001) at different months where the study animals are exposed to different ambient temperatures.

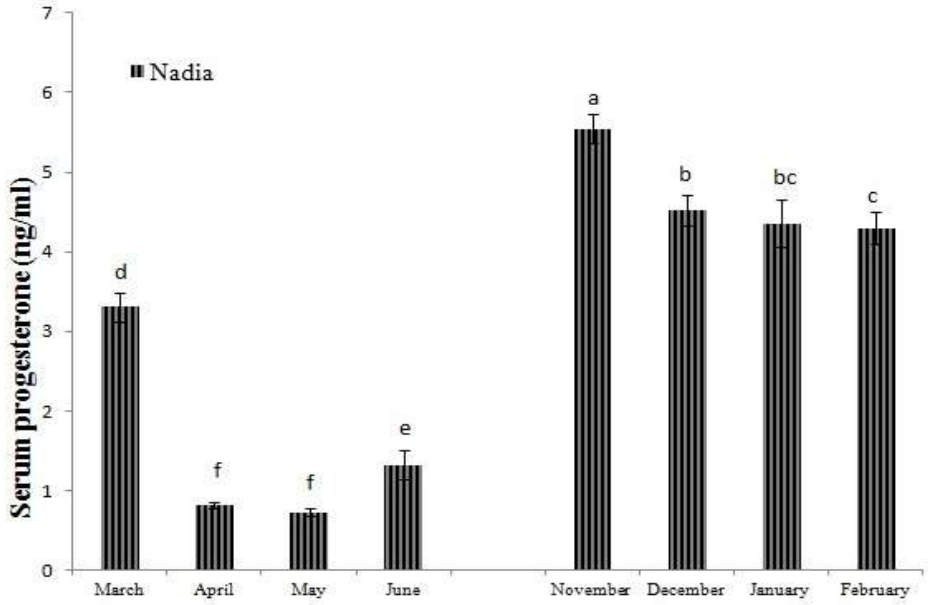

Fig-3: A graphical representation of the one-way ANOVA with *post doc* modification (DMRT) showing the effects of seasonal variations on the serum concentration of progesterone (ng/ml) in the Black Bengal does during in Purulia. Means bearing different superscripts (a–e) differ significantly (P < 0.0001) at different months where the study animals are exposed to different ambient temperatures.

DISCUSSION:

Reproduction in goats has differences in seasonality between breeds and locations. Increasing duration of days starting from early to spring reduces the reproductive activity to the goats. The progesterone level is found lower during in May in both of the regions and found lowest in Purulia (0.32 ± 0.06 ng/ml). Decreasing duration of day lengths starting from pre monsoon to early post monsoon had the inverse effects, stimulated the progesterone level beginning with November. The progesterone 1evel found higher between Novembers to February when the goats present the cyclic sexual activity and the highest progesterone concentration is found during in the month of November in Nadia (5.55 ± 0.18 ng/ml). Similar findings have been reported by Bhatta et al. [28] on the testosterone hormone in the same breed of Black Bengal goats. Calculation of progesterone levels during different physiological phases on goats or any types of livestock animal is thought to be the best significant parameter to determine the ovarian activity [9]. Concentration of progesterone hormone plays a crucial

role to determine reproduction efficiency [29]. The hormonal profile of progesterone has been studied for reproductive physiology for various breeds of goats like Nubian [30], Angora [31], Boar [32] and Dutch White breeds of goat [33].

In temperate regions the breeding period was observed in the autumn and winter. In France, the breeding season probably starts in September when day length is declining and persists until March [34]. In Australia, goats have a period with spontaneous ovulatory activity, from April to August with a peak in June [11]. In the Alpine and local goats bred in subtropical Mexico. The breeding season begins in the near the beginning of autumn and ends in the delayed winter [35].

CONCLUSIONS:

During the present study it has been observed the lowest progesterone levels were observed from April to June for both of the regions of Purulia and Nadia and the month May has the lowest level of the progesterone hormone level in the serum of Black Bengal does in Purulia (0.32 ± 0.06 ng/ml). However, the serum progesterone level reached to its peak level in November in region Nadia (5.55 ± 0.18 ng/ml). In this breed i.e. Black Bengal goat, the higher serum progesterone level has been reached during the decreasing photoperiod and it can be concluded that month of November and December i.e. early the periods of the post-monsoon season can be taken as an alternate breeding seasons for Black Bengal doe. Similar results have been found out by Bhatta et al. [28] on the testosterone hormone in the same breed of Black Bengal goats.

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Conflict of the interest statement:

None of the authors has any financial or personal relationships that could inappropriately influence or bias the content of the article.

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