

## OESTRADIOL - 17 $\beta$ AND PROGESTERONE PROFILES IN PREPUBERTAL BUFFALO-HEIFERS

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### ABSTRACT

Using RIA technique, oestradiol- 17 $\beta$  and progesterone level were estimated in serum of fifteen buffalo-heifers aged 9 to 14 months. Oestradiol showed unsteady level during experiment. It was low ( $5.12 \pm 0.23$  pg/ml) in the 9<sup>th</sup> month of age, but increased non significantly to be  $5.46 \pm 0.18$  and  $5.90 \pm 0.39$  pg/ml in the 10<sup>th</sup> and 11<sup>th</sup> months, respectively. Oestradiol- 17 $\beta$  increased significantly ( $p < 0.01$ ) at 12<sup>th</sup> month of age ( $11.85 \pm 0.78$  pg/ml), then started to decline in the 13<sup>th</sup> month of age ( $9.75 \pm 0.54$  pg/ml) and reached the base line level in the 14<sup>th</sup> month of age ( $5.77 \pm 0.46$  pg/ml).

Progesterone level remains below 1 ng/ml during prepubertal period. It was  $0.118 \pm 0.38$ ,  $0.141 \pm 0.016$ ,  $0.167 \pm 0.021$ ,  $0.152 \pm 0.025$  and  $0.144 \pm 0.012$  ng/ml in the 9<sup>th</sup>, 10<sup>th</sup>, 11<sup>th</sup>, 12<sup>th</sup> and 13<sup>th</sup> months, respectively. A mild increase was recorded at the 14<sup>th</sup> month ( $0.348$  ng/ml).

From the preceding results, it can be concluded that, the onset of puberty in buffalo-heifers accompanied with ovarian activity may be started around a 12-14 month of age.

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#### INTRODUCTION

The onset of puberty in animals results via synchronized and maturation changes in central nervous system, hypothalamus, pituitary and ovaries (Morrow, 1986). Previous studies had mainly focused on measurement of pituitary and plasma gonadotropins during prepuberty stage in cattle (Gonzales-Padilla, 1976 and Royan and Foster, 1980). However, little work has been carried out concerning oestrogens and progesterone levels in prepubertal period in buffalo-heifers.

As puberty approaches the endocrine system developed gradually that, Gn-RH increased significantly in response to the negative feedback threshold of the gradually elevated ovarian estrogens from the ovarian follicular maturation (Schillo et al., 1982).

Jain and Pandey (1981) recorded that, oestradiol 17 B fluctuated between 8.8 Pg/ml and 12 Pg/ml during the first 12 months of age, then slightly declined to 11.76 Pg/ml at puberty. In contrast, Hattab (1988) a lower unsteady level of oestradiol at the prepubertal period that ranged from 2.33 to 7.29 Pg/ml. Moreover, the author added that, the oestradiol peaks were noticed at 12<sup>th</sup> month of age and declined at 16<sup>th</sup> month, where it became steady till puberty. Meanwhile, Morrow (1986) stated that a much higher level of oestradiol- 17 B in prepubertal period at two months prior to ovulation (60 Pg/ml) was observed, then declined to a base line level (15 Pg/ml) by first ovulation.

However, Jain and Pandey (1983 and 1985) and Jain (1988) found that oestradiol- 17 B in buffalo plasma increased from the day of birth up to fifth month of age and then no further increase could be traced up to 30 month of age.

Regarding to prepubertal progesterone level, Jain and Pandey (1985), Jain (1988) and Mecool et al., (1988) reported that, the prepubertal plasma progesterone in growing buffalo-heifers lies below 1 ng/ml, where it increased significantly around puberty. Moreover,

Gonzalez-Padillo et al. (1979) and Kaltenbach (1980) observed also in buffalo heifers that, progesterone level reached 300 pg/ml with two elevations before puberty.

oestrogen and progesterone levels during such period still gained interest for monitoring genetic progress and productivity in buffaloes as they indicate the beginning of ovarian activity, explaining dynamics of maturation process and the attainment of such period (Hattab, 1988).

#### MATERIAL AND METHODS

The data were obtained over 6 months from fifteen buffalo heifers of 9 months age. The animals used in the present study were selected from the farm belonging to Faculty of Vet. Med., Suez-Canal University, Ismailia, Egypt.

All selected heifers were free from any reproductive anomalies. The heifers were fed identically on balanced growing ration. Rectal examination of the genital organs at the start of experiment (9<sup>th</sup> month) revealed the absence of any ovarian activity. the examination was repeated weekly interval. the animals were kept with a healthy bull in an open shelter to detect estrus and rectal examination was applied to assure and confirm the developed ovarian structure in animals showing signs of estrus during the experimental period.

Blood samples were collected at monthly interval from jugular vein. the samples were left one hr. for coagulation, then centrifugated at 3,000 r.p.m. for 1/2 an hour to separate the serum. the serum was carefully decanted and stored at - 20 C° until assay.

Radio-immuno-assay (RIA) technique with diagnostic kits (obtained from Diagnostic products Corporation, Los Angeles, USA) were used for determination of both serum oestradiol- 17  $\beta$  and progesterone. Oestradiol- 17  $\beta$  assay was carried out according to Xing et al. (1983), while progesterone assay was done according to Kubasik et al. (1984).

## RESULTS

At the start of this study, where the ovaries lack any functional structures, the serum oestradiol-17 $\beta$  level was low and averaged 5.12, 5.46 and 5.90 Pg/ml at 9<sup>th</sup> and 11<sup>th</sup> months of age, respectively (table 1 and Fig. 1). meanwhile, a significant increase ( $P < 0.09$ ) was observed at 12<sup>th</sup> and 13<sup>th</sup> months of age (11.85 and 9.75 Pg/ml, respectively), then followed again by a significant decrease ( $P < 0.01$ ) at the 14<sup>th</sup> month (5.47 Pg/ml). However, small follicles (less than 1 cm in diameter) and small corpus luteum were palpated at the 11<sup>th</sup> and the 12<sup>th</sup> months of age. the small follicles has undergone atretic changes, so that Graafian follicle was finally palpated in ovaries at 12 to 14<sup>th</sup> months of age just prepuberty but there was no detectable estrus signs as well as no ovulation.

The present study revealed that, the serum progesterone level of the prepubertal buffaloes (9<sup>th</sup> to 14<sup>th</sup> months of age) showed non-significant changes as their values ranged between 0.141 and 0.348 ng/ml (Table 2 and Fig. 2). Non of the samples reached the level of 1.0 ng/ml.

## DISCUSSION

The low level of estradiol-17 $\beta$  demonstrated during the prepubertal stage sat 9<sup>th</sup>, 10<sup>th</sup> and 11<sup>th</sup> month of age in the present study were in accordance with that reported by Jain and pandey (1981). However, a higher level at 12<sup>th</sup> month was similar to that recorded by Gonzales-Padill et al. (1985), Morrow (1986) and Hattab (1988). before puberty, the growing follicles present in the ovaries start to undergo atresia, but as puberty approaches, these follicles developed to produce mature Graafian follicle. This is dependent upon the stimulus of gonadotrophic hormones (FSH and LH) regulated by Gn-RH secreted from hypothalamus (Hafez, 1987). Although, the affect and stimulus of higher brain center acting on hypothalamus are present, but the hypothalamus does not respond before puberty because of immaturity due to age dependent (Arthur et al., 1989). Thus the high oestrogen level might explain the relationship between the dynamic process of

maturation of hypothalamus, pituitary-ovarian axis and beginning of ovarian follicular maturation (Morrow, 1986). Raheet al. (1980) added that the ovarian steroidogenic activity might regulate hypothalamus, Gn-RH and pituitary gonadotropins release patterns.

The decline level of the oestradiol- 17 $\beta$  observed at 14th month of age might be probably of an indicative value for the age of puberty. This is assured by Schillo et al. (1982) who stated that, as puberty approached, Gn-RH releasing system is regulated by negative feed back mechanism from the ovarian estrogen. Thus secretion of such oestrogen will inhibit pulsatile LH production resulting more frequent LH pulse patterns and accordingly ovarian activity started.

The low mean values of progesterone indicated in the current study in the 9<sup>th</sup>, 10<sup>th</sup>, 11<sup>th</sup> months of age, was in agree with the study of Jain and Pandey (1981), Jain (1988) and Mecool et al. (1988). The lower progesterone level at this period might be attributed to the dysfunction of the luteal tissue in the gonads (Morsy, 1982), Foot et al. (1970) and Bane et al. (1978) who stated that silent or unovulatory heats around puberty period could be observed because the C.N.S. have to be primed by progesterone before it well respond. Moreover, Stiagmiller et al., (1978), Nelson et al., (1985) and Rutter and Randel (1986) observed that, the prepubertal unovulatory heats were not followed by elevation in serum progesterone concentrating.

The increase in the serum progesterone level observed at the 14th month of age coincided with that reported by Kaltenbach (1980), Schams et al. (1981) and Hattab (1988) who stated that, progesterone lead to establishment of the phasic LH release characteristic of the induced ovarian activity and cyclicity.

It was of importance to record that the observed increase in the progesterone level was not accompanied clinically with the presence of palpable corpus luteum, however, Berardinelli et al., (1979) indicated an elevation in the progesterone level at prepubertal period and attributed that to the embedded luteal tissue within the ovary which could not palpated rectally or on the ovarian surface.

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Table (1) : Means and their standard error for the estradiol 17 B in growing buffalo-heifers during prepubertal period (Pg/ml).

Months	N	$\bar{X} \pm$	S.E
9th month	15	5.12 $\pm$	0.23 C
10th month	15	5.46 $\pm$	0.18 C
11th month	15	5.90 $\pm$	0.39 C
12th month	15	11.58 $\pm$	0.78 a
13th month	15	9.75 $\pm$	0.54 b
14th month	15	5.47 $\pm$	0.46 C

Difference between means with different superscripts is significant at (P < 0.01)

Table (2) : Means and their standard error for the progesterone in growing buffalo-heifers during prepubertal period (ng/ml).

Months	N	$\bar{X} \pm$	S.E
9th month	15	0.118 $\pm$	0.039 b
10th month	15	0.141 $\pm$	0.016 b
11th month	15	0.167 $\pm$	0.021 b
12th month	15	0.152 $\pm$	0.025 b
13th month	15	0.144 $\pm$	0.012 b
14th month	15	0.348 $\pm$	0.071 C

Means with different superscripts mean significant different at P < 0.01.

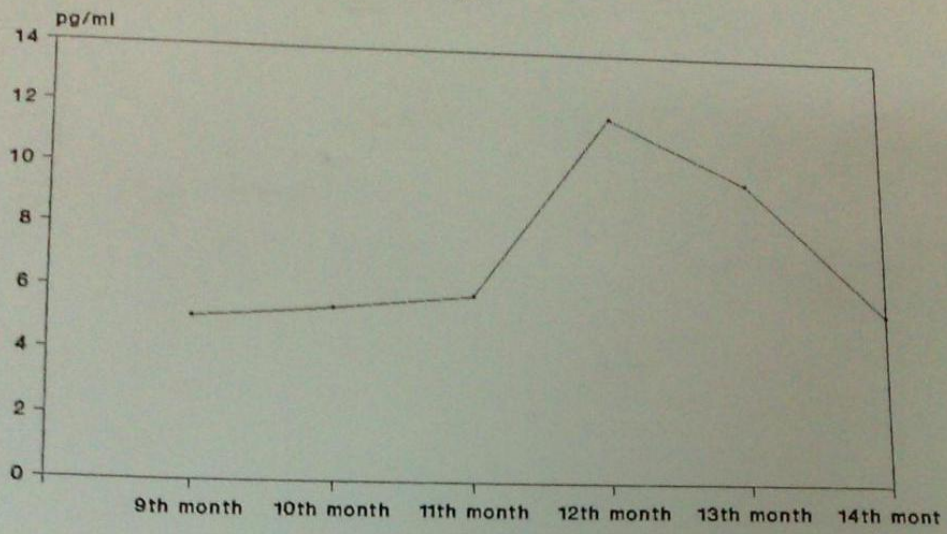


Fig. (1) Estradiol- 17B in growing buffalo-heifers during prepubertal period. (Mean values)

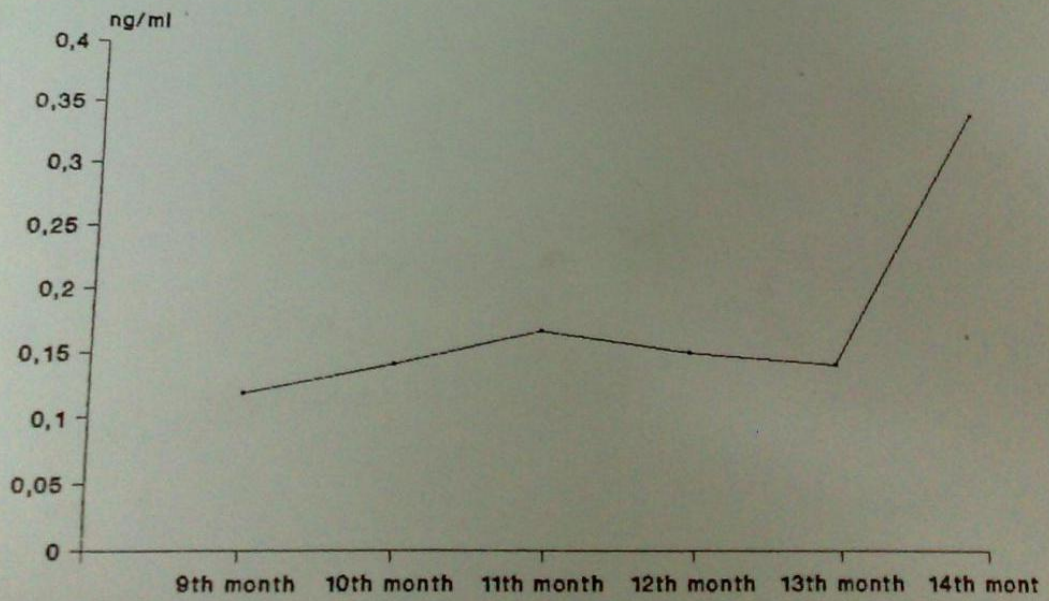


Fig. (2) Progesterone in growing buffalo-heifers during prepubertal period. (Mean values)

## الملخص العربي

« أنماط هرموني الإستراديول ١٧ بيتا والبروجستيرون في مرحلة

ما قبل البلوغ لعجلات الجاموس »

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كان الهدف من إجراء هذا البحث هو تحديد عمر البلوغ فى عزبات الجاموس المصرى وكذلك النضج الجنسى وذلك عن طريق التغيرات فى مستوى هرمون الأستراديول ١٧ بيتا والبروجستيرون فى الدم ولقد تم إجراء هذا البحث على عدد ١٥ عذبة جاموس عمر ٩ أشهر تم اختيارها من مزرعة كلية الطب البيطرى جامعة قناة السويس بالإسماعيلية وكانت هذه الحيوانات خالية من الإضطرابات الصحية والتناسلية أثناء فترة إجراء التجربة وقد تم فحص الأعضاء التناسلية عند بداية التجربة ( الشهر التاسع ) حيث أظهرت الفحوص عدم وجود أى نشاط للمبايض وقد تكرر هذا الفحص مرة كل أسبوع .

لقد تم تجميع عينات الدم على فترات كل شهر من بداية التجربة ولمدة ستة شهور ( أى حتى الشهر الرابع عشر من العمر ) وتم فصل المصل لقياس مستوى كلاً من هرمون الأستراديول ١٧ بيتا والبروجستيرون وذلك باستخدام طريقة المناعة الأشعاعية .

وقد أسفرت النتائج على الآتى :

كان هناك إنخفاض فى مستوى هرمون الأستراديول ١٧ بيتا فى الشهر التاسع من العمر مع وجود زيادة غير معنوية فى الشهر العاشر والحادى عشر . أما عند الشهر الثانى عشر من العمر كان هناك زياده معنويه فى مستوى الأستراديول ١٧ بيتا ثم بدأ فى الأنخفاض فى الشهر الثالث عشر ووصل إلى النقص المعنوى فى الشهر الرابع عشر من العمر .

أما بالنسبة لهرمون البروجستيرون فقد أثبتت النتائج عن عدم وجود أى تغيرات معنوية أثناء فترة التجربة ( من الشهر التاسع إلى الشهر الرابع عشر ) . بإستثناء وجود زياده طفيفه فى مستوى هرمون البروجستيرون فى الشهر الرابع عشر .

ومن هذه النتائج نستنتج أن بداية فترة البلوغ فى عجلات الجاموس مصحوبة بنشاط المبايض ربما تبدأ حول الفترة بين الشهر الثانى عشر إلى الشهر الرابع عشر من العمر .