

Morphometric Studies on Buffalo Ovary and Corpus Luteum during the Luteal Phase of Estrous Cycle and Pregnancy

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ABSTRACT

Per rectal examination is the most adapted technique to diagnose pregnancy in large ruminants, particularly the palpation of the corpus luteum in buffalo is of great significance. The present study investigated the basic morphological differences between the ovaries along with corpus luteum of the pregnant buffalo and non-pregnant buffalo in the luteal phase of the estrous cycle. For the purpose, ten pregnant (60-70 days of gestation) and ten non-pregnant genitalia (11-17 days of estrous cycle) with the corpus luteum were obtained from freshly slaughtered buffaloes at local abattoir. Morphological parameters such as length, width, weight and thickness of the ovary were measured. Vasculature, colour, length, diameter and weight of the corpus luteum were also recorded. Morphological parameters of right ovaries as well as CL were higher in pregnant genitalia than in non-pregnant ones. Pregnancy CL was more pinkish (more vascularization), well embedded in the ovary (as a diffused swell portion without crown-like elevation) without a demarcated point of ovulation. The study concludes that there were well-demarcated morphological differences between pregnant and non-pregnant buffalo ovary and corpus luteum.

Keywords: Buffalo, Ovary, Corpus luteum, Morphology, Pregnancy diagnosis.

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INTRODUCTION

India has maximum buffalo population in the world and they contribute more than 50% of the country's milk production. Buffalo is dual purpose animal and is reared mainly for milk, meat and drought purposes. It is essential to establish the norms of reproductive parameters. Relatively few references exist concerning the morphological and physiological aspects of the buffalo ovary during early pregnancy. The corpus luteum is a transient endocrine gland formed from the Graafian follicle after ovulation, by a complex mechanism involving morphological and biochemical changes. It performs various vital functions in the reproductive processes as the estrous cycle, embryonic survival, implantation of the embryo, and maintenance of pregnancy (Niswender *et al.*, 2000). It is commonly said as biological clock of the estrous cycle and pregnancy. The corpus luteum is a dynamic endocrine gland showing variation in size, structure, and steroidogenic activities in different stages of the estrous cycle as well as during pregnancy (Fields and Fields, 1996). There is great significance of the ovary and corpus luteum in pregnancy diagnosis in buffalo by per rectal examination. Clean and thorough knowledge about morphometry of the ovary and corpus luteum is necessary to make the pregnancy diagnosis at the field level with clear differences in pregnant and non-pregnant status. The present study was planned to investigate the basic differences regarding some morphological parameters such as gross morphology, size, colour, consistency, weight, and vasculature of the ovary along with corpus luteum of the pregnant buffalo and non-pregnant buffalo in the luteal phase of the estrous cycle.

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MATERIALS AND METHODS

Genitalia of pregnant (60-70 days gestation) and non-pregnant buffaloes were randomly collected from the local slaughterhouse and transported to the laboratory in physiological saline within 45 min. After slaughter. They were washed in running tap water and then placed in a stainless steel tray for examination. Genitalia of ten pregnant (60-70 days pregnancy) and ten non-pregnant (11-17 days of estrous cycle) buffaloes with the corpus luteum of the luteal phase were selected. Paired ovaries from

each genitalia were excised carefully with the help of scissors, thumb forceps and a BP blade. With the help of Vernier caliper, morphological parameters such as length, width and thickness of the ovary were measured as pole to pole, surface to surface and hilus to free border, respectively. After noting the visible vasculature, and colour of the corpus luteum, it was gently squeezed out from the ovarian surface and the weight (g) of right and left ovaries was recorded by using a monopan balance after trimming the extraneous tissue of each ovary. The final length and diameter at the widest portion of the corpus luteum were measured.

RESULTS AND DISCUSSION

Gross morphological studies on buffalo CL are relatively rare as compared to cattle. There are a lot of studies regarding CL at different stages of the estrous cycle (Meur *et al.*, 1999; Chandrahasan and Rajsekaran, 2004) but very fewer studies have been done on comparison between pregnancy and non-pregnancy stages. The main difference observed as CL in cases of non-pregnant ovary was well elevated on the ovarian surface as a crown (Fig. 2), while in pregnancy (60-70 days), the CL was well embedded in the ovary and observed as diffused swell portion without crown like elevation (Fig. 4). The presence of CL was detected because of the vasculature, colour difference and consistency on pressing in case of pregnant genitalia. In CL, there was demarcation of the point of ovulation in non-pregnant genitalia, while it was less or not visualized in case of CL of pregnant genitalia.

Size

The length, width and thickness of right ovaries were found non-significantly higher in the case of 60-70 days pregnant

animals (Fig. 3) than in non-pregnant buffaloes (Fig. 1). The width and thickness of left ovaries were higher in the case of non-pregnant genitalia than pregnant ones, except for length, while there was no significant difference in morphometry between pregnant and non-pregnant left ovaries (Table 1).

The length and diameter of the corpus luteum of pregnant ovaries were found non-significantly higher than non-pregnant ovaries (Table 2). The size of the CL at the mid-luteal phase of the estrous cycle and pregnancy varied from 1 to 2 cm compared with 2 to 2.5 cm and the weight ranged from 1 to 1.9 gm compared with 2 gm to 3.4 gm, respectively, (Singh *et al.*, 1990; Chandrahasan and Rajasekaran, 2004).

Color

The colour of corpus luteum (CL) in the non-pregnant ovary was observed as reddish tan to orange yellowish (Fig. 2). There was not much difference in the color of 60-70 days pregnant animals CL where it was observed reddish brown to pinkish (Fig. 4). In pregnancy CL, it was observed that the colour is somewhat more pinkish to reddish tan, which may be because of the good to very good vascularization present all over the CL. Vascularization in CL of the non-pregnant ovary is good at the periphery with a pinkish apex. In some of the non-pregnant CL, the colour was somewhat more reddish at the tip. It may be due to the less advanced stage of the CL, as there was well demarcation for the point of ovulation. Mature CL (day 6-15) has been reported to be orange yellowish, while late CL (day 11-17) is characterized by the presence of surface vasculature and colour is brown/orange to tan (Chandrahasan and Rajsekaran, 2004; Thangeval and Nayeem, 2004).



Fig. 1: Ovaries in luteal phase of estrous cycle in non pregnant buffalo



Fig. 2: Corpus luteum of luteal phase of estrous cycle in non pregnant buffalo

Table 1: Morphometry of ovaries of buffaloes

Ovary	Stage	Length (cm)	Width (cm)	Thickness (cm)	Weight gm)
Right	Non-pregnant (n=10)	2.36±0.11	1.55±0.09	1.94±0.09	3.71±0.36
	Pregnant (n=10)	2.37±0.04	1.58±0.05	1.98±0.06	3.87±0.20
Left	Non-pregnant (n=10)	2.23±0.13	1.41±0.09	1.76±0.12	2.96±0.43
	Pregnant (n=10)	2.27±0.10	1.37±0.05	1.56±0.07	2.38±0.21

Figures in parentheses are the number of ovaries



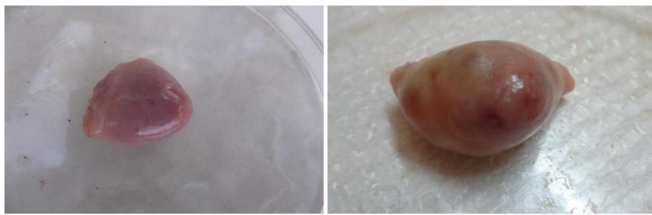


Fig. 3: Buffalo ovaries during pregnancy (day 60-70)

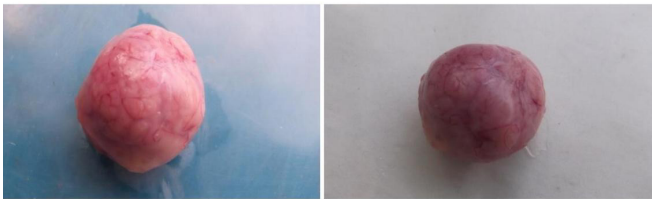


Fig. 4: Corpus luteum from the ovaries of pregnant buffalo (day 60-70)

Table 2: Biometry of CL during pregnancy and non-pregnancy (luteal phase) of buffaloes

Stage	Length (cm)	Diameter (cm)	Weight (gm)
Non pregnant (n=10)	1.68±0.08	1.49±0.06	1.35 ^b ±0.16
Pregnant (n=10)	1.84±0.05	1.63±0.05	1.88 ^a ±0.12

n= number of ovaries Means with different superscripts differ significantly within a column ($p < 0.05$).

Weight

The data revealed that the weight of right ovaries was non-significantly higher in the case of pregnant genitalia (60-70 days pregnancy) than non-pregnant, however in the case of left ovaries, weight was non-significantly higher in non-pregnant than pregnant genitalia (Table 1).

The weight of CL observed was significantly higher for the pregnant than the non-pregnant corpus luteum (Table 2). Shah and Mehta (1992) also observed the maximum weight of the CL, between 1.60 g to 1.79 g.

The weight of the CL changes according to the different stages of the estrous cycle. Corpus luteum of the buffalo increases in weight progressively with the advancement of luteal development and decreases with the regression process. The CL became larger and firmer with advancing pregnancies along the trimesters of gestation. The higher weight of the CL at pregnancy is because of the larger cell population along with hypertrophy of granulosa and theca cells as well as some mitotic division. The luteal cell population was observed greater in the case of pregnancy CL than the CL of the 3rd Stage of estrous cycle (Baithalu *et al.*, 2013).

Consistency

In the present study, the consistency of corpus luteum was observed almost similar in pregnancy CL as well as luteal

phase non-pregnancy CL. The consistency was almost semi-soft in both types of CL. Recent reports suggested that the bovine CL might be loose, and soft during the early luteal phase (day 1-6); compact, soft or compact during mid luteal phase (day 13-18) and firm during the follicular phase (day 19-21) of estrous cycle (Arosh *et al.*, 2002). According to Roy *et al.* (2006), the buffalo CL tissue is soft in texture during the mid luteal phase (day 5-10); moderately hard texture at the late luteal stage and has hard texture in the cut surface at follicular phase with regressed CL.

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