Characterization of follicular wave emergence during estrous cycle in *Bos indicus*

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Abstract

The Bachaur is medium-sized draft-purpose cattle breed, with its breeding tract mainly in the Madhubani, Darbhanga, and Sitamarhi districts of Bihar. At present, the breed is on the verge of extinction. Since no data exist regarding the reproductive performance of Bachaur cows, so keeping in views of the fact the study was under taken with the objective to evaluate follicular growth pattern in Bachaur cows. A total of nine healthy breeding Bachaur cows, aged 2.5 to 5 years, were selected and maintained under identical management conditions. Both ovaries were scanned per rectum using a 7.5 MHz ultrasound probe in multiple planes. The study was aimed to established number of follicular wave either two wave or three wave, Days of emergence of 2nd wave in two wave cycle, Days of emergence of 2nd wave in three wave cycle, Last follicular wave emergence to oestrus interval (Days) in two wave cycle. Results showed that 66.67% of Bachaur cows exhibited two follicular-wave cycles, while 33.33% exhibited three follicular-wave cycles. The average day of emergence of the second wave in a two-wave cycle was 10.50 ± 0.34 days. In three-wave cycles, the average days of emergence of the second and third waves were 11.00 ± 0.58 and 14.00 ± 0.58, respectively. The interval from the emergence of the last follicular wave to estrus was 10.83 ± 0.31 days in two-wave cycles and 6.00 ± 0.58 days in three-wave cycles. Thus, Bachaur cows exhibit both two- and three-follicular-wave patterns during their estrous cycle, similar to other *Bos indicus* cattle breeds.

Key words: Bachaur cow; Estrus cycle; Follicular waves; Follicular pattern

Introduction

The ICAR–National Bureau of Animal Genetic Resources (NBAGR), Karnal, has recently recognized the Bachaur as distinct cattle breed from Bihar (Fig.1), with accession number 3002. Its ancestral tract lies in Border of Nepal region and the middle Gangetic plain areas of Bihar, particularly in North Bihar the districts of Madhubani, Darbhanga, and Sitamarhi (Azad *et al*., 2024). The Koiree and Ahir communities traditionally maintain Bachaur cattle (Specimen of cattle is given below) for draft purposes, with the animals grazing in the adjoining grassland regions. This breed is medium in stature, weighing around 270 kg, compact in body, and typically white or grey in color, with short, stumpy horns. The forehead is flat or slightly convex.

Due to lack of information and scarcity of data related, to Bachaur breed of cattle the study was aimed with the objective to study the follicular wave pattern in the Bachaur cattle. The production traits of this breed in term of milk yield was 250-400 kg per lactation, daily milk yield 1-2 kg/day with lactation length of 180 to 240 days. Previous studies in Bos taurus cattle have indicated the occurrence of two to four follicular waves during the estrous cycle, with two-wave cycles being predominant and four-wave cycles being very rare (Sirois and Fortune, 1988). Follicular wave studies are crucial for success of ARTs in cattle (Jagadheesh Kumar et al., 2025). In *Bos indicus* cattle, Gaur *et al*. (2007) recorded a higher proportion of two-wave cycles (78.57%) compared to three-wave cycles (21.42%). Adams *et al.* (2008) reported that most Nellore heifers (*B. indicus*) exhibited a three-wave pattern (65%). Similarly, Hadiya *et al.* (2016a) observed the occurrence of a two-wave pattern (66.66%, n = 4) and a three-wave pattern (33.33%, n = 2) in postpartum crossbred cows. Imron *et al*. (2016) reported two- (66%) and three-wave (34%) follicular patterns in Peranakan Ongole cattle.

In view of the above findings, the present study was undertaken to evaluate the follicular dynamics and reproductive performance of Bachaur cows.

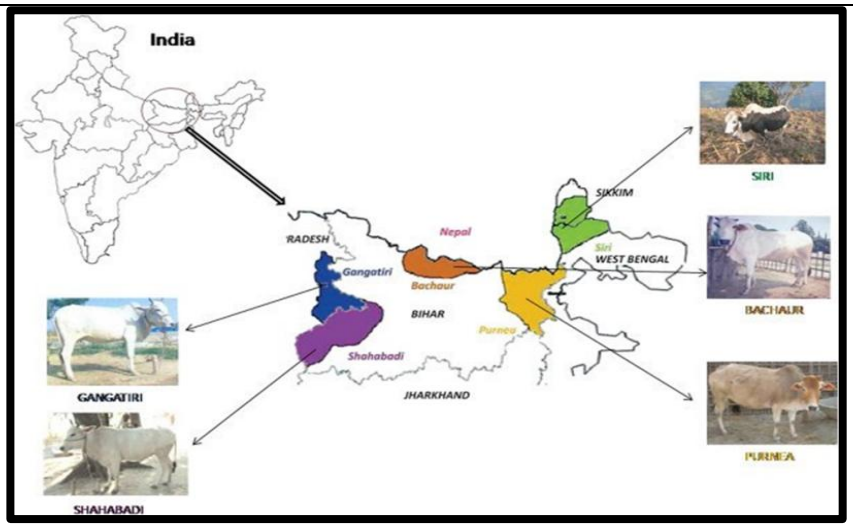

Fig.1 Geographical Map denoting habitat of Bachaur Cattle

Fig.1 Geographical Map denoting habitat of Bachaur Cattle

Fig 2. Bachaur cow

Fig 3. Bachaur Bull

Materials and methods

Bachaur cows (n=9) aged 3 to 5 years with body condition score ranging from 3 to 5 (0-5), free from external and internal parasitic infestation and normal haematological parameters were selected from its native breeding tract at Sitamarhi district of Bihar. The cows were in 2nd to 3rd lactation and calved at least 2 to 3 month before. The cows were monitored for six consecutive milking and selected with average milk yield exceeding 4 litres/day. These cows were subjected to gynaecological examination twice with an interval of 10 days to ensure normal genitalia with active ovaries and regular oestrous cycle.

The present study was carried out under Rashtriya Gokul Mission (RGM, Government of India) for ETT project after the approval of the institutional animal ethics committee.

The study was conducted at Livestock Farm Complex (L.F.C.) Bihar Veterinary College, Bihar Animal Science University, Patna -800014, Bihar, India. The Livestock Farm Complex is located at an altitude of 170 feet above the sea level and latitude of 25^0 North and a longitude of 85^0 East. The average temperature is 26^0C and the relative humidity range between 60 to 65 %.

Synchronization of estrus was done by two Intramuscular injection of 500 µg Cloprostenol Sodium (Pragma®, Intas, India) 11 days apart following which heat was detected at every morning and evening by external signs such as bellowing, mucus discharge from vulva, mounting, frequent micturition, swollen and edematous vulva. Corpus luteum was detected by per rectal examination of ovary on day 7 of cycle.

Both the ovaries were scanned per-rectally with 7.5 MHz ultrasound probe with multiple aspect of experimental design. The follicles having more than three millimeters diameter were recorded in such a way that the upper and lower lines of the greater lengths of the follicle is visible as thin white lines within the image of the ovary. Immediately after the ultrasonography of ovaries image was freeze and the diameter were then determined by using in built electronic calipers of the scanner.

1. Emergence of follicular wave

The day of wave onset was defined as the day at which the dominant follicle was retrospectively traced to be 4 to 5 mm in diameter (Burke *et al*., 2000)

2. Duration of follicular wave

Duration of follicular wave was calculated as the period between the day of initiation of follicle recruitment having more than three millimeter diameter to the day of disappearance of follicle either due to atresia or ovulation.

3. Number of follicle waves

The synchronized growth of a group of small ovarian follicles, from which a single dominant follicle emerges while the others regress, is referred to as a "follicular wave." The emergence of these waves may occur once, twice, three times, or four times during the estrous cycle, corresponding to the first, second, third, and fourth follicular waves, respectively. Usually, during each follicular wave, one or two dominant follicles (Fig. 4) along with several subordinate follicles develop.

Fig. 4. Sonogram of Bachaur cow showing wave emergence in the form of small follicle

Results and discussion

Wave Prevalence and Emergence

Out of the nine Bachaur cows evaluated, six animals exhibited a two-wave follicular cycle (66.67%), while the remaining three showed a three-wave follicular cycle (33. 33%). In the two-wave cycle, the average day of emergence

Table 1. Characteristics of follicular dynamics of oestrous cycle of Bachaur cow

Wave Character	Values in Mean ± S.E.
Number of animals in two wave	6 (66.67 %)
Number of animals in three wave	3 (33.33 %)
Days of emergence of 2nd wave in two wave cycle	10.50 ± 0.34
Days of emergence of 2nd wave in three wave cycle	11.00 ± 0.58
Days of emergence of 3rd wave in three wave cycle	14.00 ± 0.58
Last follicular wave emergence to estrus interval (Days) in two wave cycle	10.83 ± 0.31
Last follicular wave emergence to estrus interval (Days) in three wave cycle	6.00 ± 0.58

for the second wave was 10.50 ± 0.34 days. For the animals displaying a three-wave cycle, the average days of emergence for the second and third waves were 11.00 ± 0.58 days and 14.00 ± 0.58 days, respectively. The interval from the last follicular wave emergence to the onset of estrus was markedly longer in the two-wave cycle (10.83 ± 0.31 days) compared to the three-wave cycle (6.00 ± 0.58 days) (Table 1).

Duration of Follicular Waves

The lifespan and structural duration of individual waves varied by cycle type. In the two-wave cycle, the average duration of the first and second waves was 9.50 ± 0.34 days and 10.83 ± 0.31 days, respectively. Conversely, in the three-wave cycle, the average durations of the first, second, and third waves were recorded as 10.00 ± 0.58 days, 3.00 ± 0.58 days, and 6.00 ± 0.58 days, respectively.

The distribution of follicular waves observed in this study (66.67% two-wave vs. 33.33% three-wave) aligns closely with previous findings in other tropical and crossbred cattle. Hadiya et al. (2016b) reported an identical proportion (66.67% two-wave and 33.33% three-wave) in HF × K crossbred heifers. Similarly, Imron *et al*. (2016) reported a comparable wave frequency in Peranakan Ongole cattle, with 66% showing two waves and 34% showing three waves.

Regarding wave emergence timeline, the second wave in the two-wave cycle of Bachaur cows emerged on day 10.50 ± 0.34. This is earlier than the day 12.0 ± 0.91 reported in Nelore cattle by Figueiredo *et al*. (1997) and the day 12.3 ± 1.0 observed in Bangladeshi Zebu cows by Akter et al. (2010). However, our timing is consistent with Ginther *et al.* (1989), who reported second-wave emergence on day 10 of the estrous cycle in Holstein heifers.

For the three-wave cycles, Figueiredo *et al.* (1997) observed second-wave emergence on day 9.14 and third-wave emergence on day 15.14 in Nelore cattle. In comparison, Bachaur cows exhibited a delayed second-wave emergence (day 11.00) but an earlier third-wave emergence (day 14.00). When evaluating wave durations, the second-wave duration in the two-wave cycle (10.83 ± 0.31 days) was closely comparable to the 10.45 ± 0.98 days reported in Rathi cattle by Gaur and Purohit (2007). In three-wave cycles, Gaur and Purohit (2007) noted a third-wave duration of 7.67 ± 1.80 days, which is higher than the 6.00 ± 0.58 days found in Bachaur cattle.

Furthermore, comparison with HF × K crossbred heifers studied by Hadiya *et al*. (2016a) reveals distinct variations. They reported higher durations for two-wave cycles (10.00 ± 0.40 days for the 1st wave; 12.00 ± 0.70 days for the 2nd wave). In their three-wave cycles, durations were 10.00, 6.00, and 7.00 days for the first, second, and third waves, respectively. Compared to Bachaur cows, these crossbred timelines are identical for the first wave, but feature a substantially longer second wave (6.00 vs 3.00 days) and a slightly longer third wave (7.00 vs 6.00 days).

Conclusion

In conclusion, the Bachaur breed exhibits a clear predominance of two-wave follicular cycles (66.67%) over three-wave cycles (33.33%). Although external clinical signs of estrus like vulvar edema and mucous discharge are less pronounced in this draught breed compared to other indigenous milch types, their underlying ovarian follicular wave patterns remain highly consistent with other zebu, taurine, and crossbred cattle populations

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