

Effect of bull bio-stimulation on the reproductive parameters of post-partum anestrus cows

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ABSTRACT

Background: Post-partum anestrus is a major reproductive challenge that hinders optimal productivity in dairy cattle, especially in tropical systems. Among non-hormonal strategies, bull bio-stimulation has emerged as a natural, low-cost approach to enhance ovarian resumption.

Aim: This study investigated the influence of bull exposure on reproductive hormone profiles, hematological and biochemical parameters, and key fertility outcomes in post-partum Holstein Friesian crossbred cows under controlled farm conditions.

Methods: Twenty-two non-cyclic post-partum cows, 60 days after calving, were divided into two groups: one exposed to a bull (BE; $n = 11$) and the other not exposed (NBE; $n = 11$). Over 21 days, estrus behavior, blood hormonal [estrogen, progesterone, luteinizing hormone (LH), follicle-stimulating hormone (FSH)], hematological, and biochemical parameters were assessed.

Results: Bull exposure significantly accelerated the resumption of ovarian activity (81.82% vs. 27.27%) and improved progesterone levels ($p < 0.05$). In addition, FSH and LH concentrations were lower in bull-exposed cows (0.06 ± 0.02 vs. 0.36 ± 0.1 and 0.13 ± 0.04 vs. 1.22 ± 0.6), indicating a change in gonadotropin regulation. The exposed group showed fewer services per conception (1.33 ± 0.16 vs. 1.85 ± 0.34), and a higher conception rate (90.90% vs. 63.63%). Biochemical analysis revealed lower total protein and globulin levels, while hematological profiles showed elevated erythrocyte sedimentation rate, mean corpuscular volume, and mean corpuscular hemoglobin in the bull-exposed group.

Conclusion: Bull bio-stimulation significantly improves post-partum reproductive efficiency in dairy cows through enhanced ovarian activity and favorable endocrine and hematological changes. The reduction in gonadotropins, alongside increased progesterone, suggests an earlier increase of luteal activity in bull-exposed cows. This technique offers a practical, drug-free solution for improving fertility in resource-limited farming systems.

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Introduction

The dairy sector in Bangladesh plays a crucial role in poverty reduction, improving food and nutrition, and empowering women. The demand for milk and milk products is increasing day by day because of the rapid increase in population, higher income,

and growing nutrition awareness [1]. Although the production of milk has increased over the last 10 years, a gap still exists between the demand and supply. According to the Directorate of Livestock Service [2], Bangladesh produced 15.04 million metric tons of milk in 2023/2025, which is now

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meeting two-thirds of the total demand. Many factors influence the productivity of dairy farms; among these, post-partum anestrus is one of the cardinal factors that negatively affect the milk production of dairy cows. Generally, a calving interval of about 1 year is considered economically optimal for dairy cows [3]. Due to post-partum anestrus, this interval will be longer and will reduce the ultimate milk production in a dairy herd. To achieve such a calving interval, a post-partum cow has to resume ovarian cyclicity as soon as possible, which will be noticed by the timely estrus. Generally, the principal influences on reproductive performance are exerted by environmental and management factors. Therefore, reproductive performance can be improved by applying rational management strategies. One management strategy that has the potential to increase the proportion of post-partum cows that resume cycling is the use of the bio-stimulatory effect of bulls [4]. Bio-stimulation by bull exposure represents a relatively inexpensive management tool that is often underestimated. Bio-stimulation (male or bull effect) can be defined as the stimulus provoked by the presence of males, which induces estrus and ovulation, through genital stimulation, pheromones, or other external clues [5]. It affects reproductive efficiency by hastening sexual maturity, ovulation induction, reduction of post-partum anestrus, and also in mating. Although hormonal treatments appear to be highly effective in bringing a cow into cyclicity [6], non-hormonal methods such as bull bio-stimulation are preferable in small-scale systems to avoid potential future long-term adverse effects. In Bangladesh, many cows are susceptible to post-partum anestrus, and there is a variable degree of success after using various hormonal and management techniques; however, bull bio-stimulation still remains poorly documented in this context. Limited scientific literature exists regarding the application of bull bio-stimulation in tropical climates, despite its potential as a cost-effective strategy to enhance reproductive efficiency in dairy cattle. Therefore, the research work was performed to explore the possible potential of the bull bio-stimulation effect on the reproductive performances and blood parameters of post-partum anestrus dairy cows.

Materials and Methods

Ethical statement

The study was conducted as per the guidelines of the Animal Experimentation Ethics Committee of CVASU.

Study design

This study was conducted at a dairy farm in Chattogram, Bangladesh, which has well-organized management and regular data-keeping practices. The study was conducted for 21 days. A total of 22 post-partum Holstein Friesian (HF) crossbred cows (61–70 days post-partum) of the same body condition score (BCS), age, and parity were selected from a total of 490 post-partum anestrus cattle. The cows were divided into two groups ($n = 11$). The first group was exposed to bull daily, and the second group was not exposed to bull and used as a control. A pre-check was done 7 days before the study to eliminate the cyclic cows. Animals were checked for cyclicity by rectal palpation, and only non-cyclic animals were selected. All cows were kept under the same environmental and managerial conditions according to the farm routine work. The experimental cows were sorted before randomization based on lactations (2nd–3rd), age (4–6 years), and BCS (ranging from 3.0 to 3.5 on a 5-point scale) to minimize variation between groups. One bull was housed with an exposed group of cows separated by a 5 ft high fence. The non-bull exposed (NBE) group of cows was housed in a similar shed at a distance of about 500 yards from the houses of the bull exposed (BE) group of cows to ward off the effect of airborne bull pheromones.

Detection of cyclicity and pregnancy

Daily visual observation of cows was executed at 6:00 am and at 6:00 pm. Once any sexual behavior signs are demonstrated, cows are considered in estrus. The experimental observation of estrus behavior (like mounting or bullying) for both groups was carried out by visual observation. To confirm it, rectal palpation was also performed, and if the animal was in true estrus, it was inseminated. The cows were kept in the same settings for 42 more days to check their cyclicity. After 60 days of insemination, the cows were rectally palpated again to ensure the pregnancy.

Blood analysis

Blood samples were collected from each cow on four phases: “0” day (before the grouping), 7th day,

14th day, and 21st day and analyzed for hemato-bio-chemical and hormonal parameters. Hematological parameters were performed using an auto hematology analyzer (Celltac Alpha®, Nihon Kohden, Japan). The serum glucose, total protein (TP), Albumin (Alb), globulin, uric acid, blood urea nitrogen (BUN), triglyceride, and cholesterol were assayed using an automated biochemical analyzer (Humalyzer 3000®, Human, Germany). The level of reproductive hormones in the serum sample was evaluated using progesterone, estradiol, follicle-stimulating hormone (FSH), and luteinizing hormone (LH) by the ELISA method. Reproductive hormones (progesterone, estradiol, FSH, and LH) were measured using commercial ELISA kits (BioGenes GmbH, Germany) with the following catalog numbers and detection limits: Progesterone: B233453 (0.001 ng/ml), Estradiol B233453 (0.001 pg/ml), FSH: B233453 (0.001 mIU/ml), and LH: B233453 (0.001 mIU/ml). All assays were conducted according to the manufacturer's instructions.

Statistical analysis

All the data were stored and sorted in Microsoft Excel 2019 and analyzed with R software. The cyclicity rate, pregnancy rate, hematological, biochemical, and hormonal values were measured, and the descriptive analysis was performed. Assumptions of normality and homogeneity of variance were assessed using the Shapiro-Wilk and Levene's tests, respectively. Furthermore, the results were tested to check the difference between groups using a chi-square test, *t*-test, and one-way ANOVA and post hoc test to distinguish among groups, where the significance level was set at $p < 0.05$.

Results

Effect of exposure to bulls on reproductive performance

At the end of the 21-day experiment, 81.82% and 27.27% of cows exposed to bull (BE) and not exposed to bull (NBE) came into estrus. Among the cows, the BE cows required significantly less services per conception than the NBE cows. The conception rate was also higher in BE cows (90.9%), where 10 out of 11 cows conceived, whereas in NBE cows, only 7 out of 11 cows became pregnant (63.6%). The number of services per conception in BE and NBE bulls has been shown in Table 1.

Effects on the hormonal level of BE and NBE

The result reported that the reproductive steroid hormones (FSH and P4) were significantly ($p < 0.05$) varied among the BE and NBE groups (Table 2). On the other hand, LH showed a non-significant trend ($p = 0.09$), suggesting a possible biological effect despite statistical insignificance. A significant ($p < 0.05$) negative correlation was observed with blood FSH between NBE and BE cows.

Day-wise variations of reproductive hormones (N = 88)

According to Table 3, the mean progesterone concentrations (ng/ml) of the BE cow group at days 0, 7, 14, and 21 were 5.59 ± 1.12 , 5.06 ± 2.63 , 11.83 ± 3.6 , and 18.56 ± 4.48 , respectively. Again, the mean progesterone concentrations (ng/ml) of the NBE cow group at the same intervals were 4.61 ± 1.05 , 6.16 ± 1.6 , 6.3 ± 2.14 , and 2.97 ± 1.2 , respectively. The progesterone concentration rose sharply in BE cows on days 14 and 21, indicating the establishment of luteal function, whereas P4 levels in the NBE group rose only modestly on days 7 and 14 and declined on day 21.

Table 1. Effect of exposure to bulls on the reproductive aspect of cows [*N* (total cows) = 22].

Items	BE (Mean ± SE) (n = 11)	NBE (Mean ± SE) (n = 11)	<i>p</i> value (<i>t</i> -test)
Number of services per conception	1.33 ± 0.16	1.85 ± 0.34	0.009

Services per conception = artificial insemination.

Table 2. Effects of bull bio-stimulation on the hormonal level of BE and NBE cows [*N* (blood samples) = 88].

Hormone	BE (n = 44) Mean ± SE	NBE (n = 44) Mean ± SE	<i>p</i> value
FSH (mIU/ml)	0.06 ± 0.02	0.36 ± 0.1	<0.01
LH (mIU/ml)	0.13 ± 0.04	1.22 ± 0.6	0.09
Progesterone (ng/ml)	10.3 ± 1.76	5.01 ± 0.78	<0.01
Estrogen (pg/ml)	22.7 ± 1.36	22.6 ± 1.62	0.9

Table 3. Day-wise hormonal changes of the experimental cows [*N* (blood samples) = 88].

Hormone	Group	Mean ± SE			
		Day 0	Day 7	Day 14	Day 21
FSH (mIU/ml)	NBE	0.25 ± 0.11	0.27 ± 0.11	0.30 ± 0.14	0.62 ± 0.36
	BE	0.05 ± 0.04	0.08 ± 0.07	0.04 ± 0.04	0.09 ± 0.04
LH (mIU/ml)	NBE	0.26 ± 0.16	0.27 ± 0.14	2.2 ± 1.93	2.15 ± 1.61
	BE	0.21 ± 0.13	0.09 ± 0.08	0.09 ± 0.06	0.12 ± 0.07
Progesterone (ng/ml)	NBE	4.61 ± 1.05	6.16 ± 1.6	6.3 ± 2.14	2.97 ± 1.2a
	BE	5.59 ± 1.12x	5.06 ± 2.63x	11.83 ± 3.6xy	18.56 ± 4.48by
Estrogen (pg/ml)	NBE	18.8 ± 2.54	23.9 ± 2.4	26.5 ± 3.59	21.27 ± 4.05
	BE	19.6 ± 2.39	21.4 ± 2.73	25.6 ± 1.88	24.02 ± 3.57

^{x,y}Means in a row lacking common superscript differ in between ($p < 0.05$).

^{a,b}Means in a column lacking common superscript differ in between ($p < 0.05$).

Table 4. Variations of hematological parameters in BE and NBE cows [*N* (blood samples) = 88].

Blood Parameters	BE (<i>n</i> = 44) (Mean ± SE)	NBE [<i>n</i> = 44] (Mean ± SE)	<i>p</i> value
TEC (million/cu.mm)	4.77 ± 0.2	5.04 ± 0.2	0.35
TLC (thousand/cu.mm)	7.9 ± 0.7	7.1 ± 0.23	0.28
Hb (gm/dl)	8.13 ± 0.2	7.7 ± 0.16	0.09
PCV (%)	32.66 ± 0.7	31.91 ± 0.59	0.42
ESR (mm/hour)	1.07 ± 0.12	0.76 ± 0.07	0.02
Neutrophil (%)	28.7 ± 1.06	27.6 ± 1.38	0.52
Eosinophil (%)	6.25 ± 0.62	6.18 ± 0.48	0.93
Basophil (%)	0.29 ± 0.07	0.32 ± 0.08	0.83
Lymphocyte (%)	60.54 ± 1.32	61.23 ± 1.52	0.73
Monocyte (%)	4.25 ± 0.37	4.93 ± 0.44	0.24
MCV (fl)	73.44 ± 3.54	64.64 ± 2.41	0.04
MCH (pg)	18.29 ± 0.92	15.69 ± 0.65	0.02
MCHC (g/dl)	25.10 ± 0.56	24.37 ± 0.57	0.37

PCV = packed cell volume; TEC = total erythrocyte count, TLC = total leukocyte count.

Hematological parameters in BE and NBE cows

Table 4 shows that the mean erythrocyte sedimentation rate (ESR), mean corpuscular volume (MCV), and mean corpuscular hemoglobin (MCH) were significantly ($p < 0.05$) higher in BE cows (with bull contact) compared to NBE (cows without bull contact group). No significant difference ($p > 0.05$) was found between BE and NBE cows for packed cell volume, hemoglobin (Hb), total erythrocyte count, total leukocyte count, lymphocyte, monocyte, neutrophil, and mean corpuscular hemoglobin concentration (MCHC) (Table 4).

Day-wise variations of hematological parameters in experimental cows

According to Table 5, the ESR values were significantly higher in the BE cows than the NBE cows in

all 4 days (0,7,14,21). Hb was higher in BE than in NBE, except on day 21. The MCV, MCH, and MCHC values progressively increased throughout the study period, especially in BE cows.

Variations of blood biochemical parameters of BE and NBE cows

Table 6 reveals that TP and globulin were significantly higher in the NBE group, while triglyceride (TG) levels were significantly higher in the BE cows. No significant differences were observed for glucose, Alb, BUN, uric acid, and cholesterol between the groups.

Day-wise variations of biochemical parameters in experimental cows

According to Table 7, almost all parameters were lower in the BE cows than the NBE cows on day 0,

Table 5. Day-wise changes of blood parameters [*N* (blood samples) = 88].

Blood parameters	Group	Mean ± SE			
		Day 0	Day 7	Day 14	Day 21
PCV (%)	NBE	30.09 ± 1.24	33.82 ± 1.24	31.36 ± 1.12	32.36 ± 1.01
	BE	31.64 ± 0.85	31.82 ± 1.92	35.18 ± 1.05	32 ± 1.48
ESR (mm/hour)	NBE	0.6 ± 0.15 ^{xyz}	0.95 ± 0.12 ^{xy}	0.45 ± 0.12 ^{xz}	1.05 ± 0.1 ^{axy}
	BE	1.09 ± 0.2 ^{xy}	0.82 ± 0.15 ^y	0.68 ± 0.15 ^y	1.73 ± 0.3 ^{bx}
Hb (g%)	NBE	6.62 ± 0.17 ^x	7.53 ± 0.28 ^{xy}	8.36 ± 0.19 ^{xyz}	8.29 ± 0.31 ^{yz}
	BE	7.14 ± 0.26 ^x	8.05 ± 0.39 ^{xy}	9.16 ± 0.28 ^{by}	8.14 ± 0.37 ^{xy}
TEC (million/ cu.mm)	NBE	4.99 ± 0.59	4.89 ± 0.38	5.05 ± 0.28	5.21 ± 0.34
	BE	4.67 ± 0.46	4.87 ± 0.48	5.06 ± 0.32	4.49 ± 0.35
TLC (thousand/ cu.mm)	NBE	6.6 ± 0.47	6.99 ± 0.44	7.34 ± 0.48	7.48 ± 0.47
	BE	8.98 ± 2.74	7.52 ± 0.42	7.27 ± 0.42	7.83 ± 0.47
Neutrophil (%)	NBE	29.54 ± 3.92	24.91 ± 1.79	26.73 ± 2.16	29.18 ± 2.8
	BE	30 ± 1.23	27.64 ± 2.82	28.91 ± 2.87	28.27 ± 1.1
Eosinophil (%)	NBE	6.45 ± 0.92	5.64 ± 0.85	5.73 ± 0.7	6.91 ± 1.34
	BE	7.73 ± 1.03 ^{xy}	5 ± 0.96 ^{xy}	8.27 ± 1.73 ^x	4 ± 0.59 ^y
Basophil (%)	NBE	0.54 ± 0.16	0.18 ± 0.12	0.36 ± 0.20	0.18 ± 0.12
	BE	0.36 ± 0.15	0.27 ± 0.14	0.27 ± 0.14	0.27 ± 0.14
Lymphocyte (%)	NBE	58.27 ± 3.94	65.64 ± 2.43	61.18 ± 2.51	59.82 ± 2.96
	BE	58 ± 1.7	63.18 ± 2.86	59 ± 3.93	62 ± 1.36
Monocyte (%)	NBE	5.18 ± 0.83	4.54 ± 0.98	6.09 ± 1.06	3.91 ± 0.61
	BE	4 ± 0.54	3.09 ± 0.43	4.36 ± 0.66	5.54 ± 1.08
MCV (fl)	NBE	56.62 ± 3.39	72.14 ± 5.14	64.49 ± 5.24	64.58 ± 4.51
	BE	78.25 ± 11.8	69.88 ± 5.89	71.27 ± 3.35	74.37 ± 5.19
MCH (pg)	NBE	12.78 ± 0.93	16.27 ± 1.44	17.21 ± 1.32	16.5 ± 1.09
	BE	17.83 ± 3.02	17.79 ± 1.56	18.75 ± 1.32	18.78 ± 1.11
MCHC (g/dl)	NBE	22.39 ± 1.17 ^{xyz}	22.78 ± 1.08 ^{xyz}	26.85 ± 0.72 ^z	25.76 ± 1.02 ^{xyz}
	BE	22.66 ± 0.79	25.81 ± 1.35	26.15 ± 0.81	25.78 ± 1.25

PCV = packed cell volume; TEC = total erythrocyte count, TLC = total leukocyte count.

^{x,y}Means in a row lacking common superscript differ ($p < 0.05$).

^{a,b}Means in a column lacking common superscript differ ($p < 0.05$).

Table 6. Biochemical parameters of the experimental cows [*N* (blood samples) = 88].

Parameters (mg/dl)	BE (Mean ± SE)	NBE (Mean ± SE)	<i>p</i> -value
Glucose	50.2 ± 1.46	55.06 ± 1.13	0.067
Total protein	6.82 ± 0.07	7.24 ± 0.06	0.002
Albumin	3.96 ± 0.04	3.86 ± 0.04	0.29
Globulin	2.86 ± 0.08	3.37 ± 0.08	0.002
Uric Acid	48.26 ± 1.18	47.09 ± 1.06	0.61
BUN	22.58 ± 0.55	22.04 ± 0.49	0.61
Triglyceride	81.49 ± 11	40.88 ± 0.99	0.012
Cholesterol	169.61 ± 4.77	178.44 ± 3.29	0.28

and this trend remained consistent through day 21, except for cholesterol, which showed no significant

variation. Notably, TP and globulin remained lower in BE cows across all time points.

Table 7. Day-wise variation of the biochemical parameters [*N* (blood samples) = 88].

Biochemical Parameters(mg/dl)	Group	Mean ± SE			
		Day 0	Day 7	Day 14	Day 21
Glucose	NBE	47.49 ± 3.04 ^{x*}	53.24 ± 1.57 ^{xy*}	59.94 ± 2.75 ^y	59.56 ± 3.74 ^y
	BE	35.70 ± 3.75 ^{x*}	58.64 ± 1.39 ^{y*}	55.56 ± 2.66 ^y	50.92 ± 4.42 ^y
Total protein	NBE	6.99 ± 0.15	7.34 ± 0.21	7.18 ± 0.08	7.43 ± 0.2 [*]
	BE	6.59 ± 0.15	7.06 ± 0.18	7.05 ± 0.15	6.57 ± 0.24 [*]
Albumin	NBE	3.97 ± 0.20	3.90 ± 0.08	3.77 ± 0.07 [*]	3.81 ± 0.13
	BE	3.87 ± 0.10	4.13 ± 0.09	4.08 ± 0.11 [*]	3.74 ± 0.12
Globulin	NBE	3.01 ± 0.24	3.44 ± 0.23	3.41 ± 0.11	3.62 ± 0.25
	BE	2.72 ± 0.17	2.93 ± 0.21	2.97 ± 0.22	2.83 ± 0.32
Uric acid	NBE	48.61 ± 3.18 ^x	48.42 ± 2.2 ^{xy}	52.26 ± 2.53 ^y	39.1 ± 2.75 ^{xy}
	BE	50.31 ± 3.01	50.84 ± 1.95	45.23 ± 2.47	46.65 ± 5.17
BUN	NBE	22.75 ± 1.49 ^{xy}	22.66 ± 1.03 ^{xy}	24.46 ± 1.18 ^x	18.30 ± 1.29 ^y
	BE	23.54 ± 1.41	23.79 ± 0.91	21.17 ± 1.16	21.83 ± 2.42
Triglyceride	NBE	41.62 ± 1.33	41 ± 1.57 [*]	44.32 ± 1.86 [*]	36.57 ± 4.83 [*]
	BE	130.71 ± 60.73	67.18 ± 7.23 [*]	66.23 ± 7.23 [*]	61.85 ± 5.35 [*]
Cholesterol	NBE	164.51 ± 6.28	175.94 ± 6.09	188.44 ± 11.22	184.88 ± 11.68
	BE	168.09 ± 13.86	179.74 ± 14.19	177.58 ± 13.84	153.04 ± 12.41

Columns having different letter superscripts are significantly different from each other. Rows having similar symbol superscripts are significantly different from each other.

Discussion

Effect of exposure to bulls on resumption of ovarian activity

There was a marked difference between BE and NBE cows for the interval from calving to resumption of ovarian activity. These findings are in agreement with those of Berardinelli and Tauck [7] and Tauck et al. [8] while differing from Shipka and Ellis [9], who reported that in high-producing multiparous Holstein dairy cows, post-partum ovarian reactivation occurred later in bull-exposed treatments as compared to cows not exposed to bull treatment. The percentage of cows that resumed ovarian activity post-partum was 81.82% in BE cows and 27.27% in NBE cows. This suggests that bull exposure significantly enhances the return to cyclicity, likely through olfactory and pheromonal stimulation of the hypothalamic–pituitary–gonadal axis. The obtained result is in agreement with that reported by Landaeta-Hernández et al. [10]. The percentage of cows with repeated ovarian activity was greater in cows exposed to a bull (57%) than in unexposed cows (7%) [11].

Effects of bull bio-stimulation on the hormonal level

The endocrine profile in the present study revealed that progesterone concentrations

were significantly higher in BE cows, indicating a stronger luteal phase and establishment of cyclicity. However, FSH and LH concentrations were significantly lower in BE cows than in NBE cows, which appears counterintuitive (Table 2). The study revealed that the mean FSH (mIU/ml) concentration in BE and NBE cows was 0.06 ± 0.02 and 0.36 ± 0.10 mIU/ml, respectively. A possible explanation is that the early re-initiation of cyclicity in BE cows led to feedback inhibition of FSH and LH via elevated progesterone and estradiol levels, as supported by the literature [12]. The LH levels, while not statistically significant ($p = 0.09$), were numerically lower in BE cows (0.13 ± 0.04 mIU/ml) compared to NBE (1.22 ± 0.06 mIU/ml). This suggests that bull exposure may have triggered earlier preovulatory LH surges, leading to a subsequent drop in basal LH levels by the time of sampling. It can be hypothesized that the bio-stimulation effect of bull exposure to beef or dairy cows during the early post-partum period has increased the LH secretion and hastened the resumption of ovulatory activity [13,14]. The mean P4 concentration in BE cows (10.3 ± 1.76 ng/ml) was significantly higher than in NBE cows (5.01 ± 0.78 ng/ml). It was also reported that FSH was significantly ($p \leq$

0.05) decreased in the BE group than in the NBE group.

Effect of exposure to the bull on P4 (ng/ml) at 0, 7, 14, 21 days of experiment

This study revealed that progesterone concentration throughout the experiment in BE cows was higher compared to the NBE group (Table 3). This observation indicates a more consistent luteal activity and corpus luteum development in bull-exposed cows, confirming ovarian cyclicity. The present results are in agreement with Berardinelli and Tauck [7], Rekwot et al. [15], Stevenson and Lamb [16], and Roelofs et al. [17], who found elevated progesterone in cows exposed to bulls during the post-partum period.

Hematological and biochemical parameters of experimental dairy cows

The serum Hb concentration showed no significant variation between BE and NBE cows (Table 4), which aligns with the findings of Pariza et al. [18]. The plasma glucose level (mg/dl) for anestrus dairy cows with bull contact (BE) and cows without bull contact (NBE) shows no significant variation (Table 6). Plasma glucose also did not differ significantly, consistent with previous findings that glucose is not a major metabolic regulator of ovarian cyclicity [19,20]. On the other hand, the TG level was significantly higher in BE cows ($p < 0.05$), suggesting increased lipid mobilization possibly due to metabolic adjustments during early cyclic resumption. Elevated TG level in the blood is an indicator of fat mobilization due to negative energy balance [21,22]. Although TGs do not directly regulate steroidogenesis, they serve as energy substrates during oocyte maturation [23]. Cholesterol levels remained similar between groups, consistent with its non-limiting role in this context [24,25]. TP and globulin were significantly lower in BE cows than NBE cows, which may reflect hepatic protein metabolism influenced by endocrine shifts. Previous reports have noted similar trends during active reproductive phases [26–29].

In alignment with the findings of Talukdar et al. [30], who reported improved conception rates (87.81%) and elevated post-treatment progesterone and estrogen levels in postpartum anestrus cows following a modified Select-Synch protocol, our study demonstrates that bull exposure can similarly accelerate ovarian resumption (81.82%) and enhance reproductive performance, albeit through a natural neuroendocrine pathway characterized

by altered gonadotropin dynamics and hematobiochemical shifts.

Conclusion

The present study elucidated the beneficial effect of the continuous presence of bulls near postpartum dairy cows in reducing the interval from calving to the resumption of ovarian activity and in improving key reproductive outcomes in HF crossbred cows. Bull bio-stimulation appears to influence the hypothalamic–pituitary–gonadal axis through sensory and pheromonal cues, leading to earlier luteal function establishment and hormonal modulation. These physiological changes were accompanied by favorable shifts in hematological and biochemical parameters, further supporting the role of bull exposure in promoting reproductive recovery. Therefore, it is recommended that bulls be strategically used as a bio-stimulatory tool in low-input dairy systems as a natural and drug-free alternative to hormonal interventions. This practice offers a promising avenue for enhancing reproductive performance and improving fertility management in resource-limited farming conditions. This study, though, comprised a smaller sample size—a notable limitation—can pave the way for future studies in this domain, especially in Bangladesh.

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