Bio-stimulatory Effect of Bull on Postpartum Estrus Interval in Nili-Ravi Buffaloes

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Abstract
The objective of this study was to determine the effect of bull exposure after calving on postpartum interval to estrus, in Nili-Ravi buffalo. Buffaloes were divided into 4 groups; BEC & BEI; exposed continuously or intermittently to the physical presence of bull, EPB & NE; exposed or not exposed to discharge waste (urine & feces) of bull. Buffaloes were exposed on d 5 after parturition. Postpartum interval to first behavioural estrus was shorter in BEC than BEI, EPB and NE treatments. The mean serum progesterone concentration did not differ significantly between BEC and other (BEI, EPB and NE) treatments. It was concluded that presence of bull has positive effect in reducing calving interval in Nili Ravi buffalo.

Keywords: Calving interval, Biostimulation, Buffalo, Bull exposure

INTRODUCTION
Reproductive efficiency is hampered in female buffaloes due to their inherent vulnerability to environmental stress, distinct seasonal reproductive patterns, delayed puberty, poor estrus expression, delayed age at first calving and prolonged inter calving intervals. Fertility is affected by poor body condition which is resulted from poor nutrition at calving, characterized by reduced conception rates, more services per conception and prolonged postpartum interval. As in postpartum buffaloes, the resumption of ovarian cyclicity after calving plays a major role in obtaining a satisfactory reproductive performance, the extremely long postpartum interval (400 to over 600 d) in this species results in extensive economic loss [1].

Bio-stimulation or bull impact can be characterized as the stimulus incited by the nearness of male, which actuate estrus and ovulation through genital incitement, pheromones, or other outer signals [2]. Male existence...
provoked stimulus which can effect on estrus cycle. These signals include olfactory, optical, and acoustic signs. In cattle, presence of male enhance ovarian activity after parturition through discharging wastes and cervical secretion of females having estrus also take part in post-partum ovarian function \[5\]. Bio-stimulation can apply intense effects on reproductive function involving hypothalamic process. The bio-stimulatory effect is considered to be facilitated through pheromones, which are airborne chemical materials secreted externally in the excretory product or feces or integumentary secretions and detected by the olfactory sense provide basis for behavioural and hormonal responses in the animal of same species \[6\].

Bio-stimulatory effect of bull is a low-cost and appropriate technique and can be described as “control system aimed to improve or manage propagation of animal” \[5\]. In small ruminants, the part of bio-stimulation is well explained. It has extreme importance that in a herd of pre mature heifers the existence of a teaser bull did not change the ovaries or accelerate the adolescence \[5\].

The phenomenon of the male influence on postpartum recommencement of ovarian action is well documented in cattle \[7\], sheep and goats. Continuous contact of postpartum cows to a vasectomized bull restrains the occurrence of silent ovulation and improves the conception rate \[8\]. In Pakistan, since no such study has been undertaken, so presently, there is scanty information in literature regarding effects of bull exposure or its excretory products in buffalo and on physiological mechanisms by which bull may influence postpartum reproductive functions. The objective of this study was to determine the effect of bull exposure continuously or intermittently or its excretory products after calving on postpartum interval to estrus in Nili-Ravi buffalo.

**MATERIAL and METHODS**

The current study was performed at a livestock farm located between 31.0167° N latitude and 73.8500° E longitudes. The experimental period was extended from June 2014 to November 2015. Forty-eight Nili-Ravi breed buffaloes were used in this experiment. All buffalo were allocated one of the four treatments in a totally randomized plan using a 4 x 1 factorial design. Influences were exposure form 1) exposed continuously to the physical presence of a bull (BEC; n=12); 2) exposed intermittently to the physical presence of bull (BEI; n=12); 3) exposed to discharge waste (urine & feces) of bull (EPB; n=12) and 4) not exposed to a bull or discharge waste of bulls (NE; n=12). Buffaloes were exposed on d 5 after parturition. Day 5 postpartum represented d 0 for each treatment.

Buffalo exposed continuously or intermittently to bulls and its waste material do not have interaction with bulls during pregnancy and later giving birth till they were sited into treatments approximately 5 d (d 0) after calving. Postpartum detection of estrus was observed five d later in each treatments group. Estrus was detected two times every day (07.00 and 18.00) in the course of research. A buffalo stood for mounting considered to be in estrus.

BEC and BEI treatments, the buffalo which were exposed to continuous physical presence of bull (BEC) treatment, the bull was present in the herd all time. The ratio of buffalo to bull (12:1) was maintained throughout the study period. The buffalo that was exposed to bull intermittently (BEI) treatment, the bull was introduced for 2 h daily.

EPB and NE treatments, the buffalo exposed to the excretory (EPB) products of bulls, Buffalo in this treatment were sited into the shed at nearly 07.00 and were expelled at 20.30 (approximately 13.5 h). Bulls (n=10) were then moved into the shed overnight between 20.30 and 07.00 (approximately 10.5 h) each d throughout the experiment. The sheds were cleaned daily. Buffalo in the EPB treatment have no visual contact with these bulls. Buffalo assigned to the NE treatment considered as control group and have no interaction with bull and its excretory products.

Buffaloes were given free access to good-quality ration (green fodder and Vanda) according to their requirement that was available in their respective treatment sheds. The criteria used to determine interval from calving to estrus includes, behavioural estrus and existence of corpus luteum. Blood samples were collected from buffalo of each treatment by puncturing jugular vein starting on d 0 and calculations were done after every four d till the end of experiment. For the assessment of progesterone, serum was collected and saved at -20°C. In order to decide the interval from calving to estrus, serum progesterone value was calculated by using ELISA. The sandwich type of ELISA was used in this experiment. Once an ascent of progesterone over 1 ng/mL was measured and a corpus luteum was detected by using rectal palpation technique, interval from calving to the comparing d of the progesterone rise was computed. The bioethical committee of Bahauddin Zakariya University approved the methodology and procedures used in present study. The data regarding interval from calving to estrus was determined by Statistix 8.1 (Tukey HSD All-pair wise comparison test of days by group) and SPSS was used to find out (Mean ± SE) of serum progesterone concentration.

**RESULTS**

The postpartum interval from calving to first behavioural estrus was 55.2±0.78 d in BEC, 66.71±0.93 d in BEI, 68.25±0.87 d in EPB and 68.57±0.93 d in NE treatments. Significant difference was seen between BEC and other treatments (BEI, EPB and NE) for interval from parturition to first behavioural estrus. Postpartum interval to first behavioural estrus was shorter in BEC than BEI, EPB and
NE treatments. The buffalo assigned BEC treatment showed estrus 13 d earlier than EPB and NE treatments and 11 d earlier than EPI treatment. There was no significant difference in postpartum interval to estrus between BEI, EPB and NE treatments (Table 1). In present study, the percentage of buffaloes showing estrus during experimental period was 83.33%, 58.33%, 66.66% and 58.33% in BEC, BEI, EPB and NE treatments, respectively. The mean serum progesterone concentration did not differ significantly between BEC and other (BEI, EPB and NE) treatments (Table 1). Progesterone concentration was measured from d-1 (d 0 = 5 after calving) to first rise in progesterone above 1 ng/mL.

**DISCUSSION**

In the present study we determined whether the proportion of buffalo and postpartum interval to estrus could be changed in continuously bull exposed buffalo (BEC), the buffalo that was exposed to bull intermittently (BEI), the buffalo exposed to excretory product of the bull (EPB) and the buffalo treated as control have no interaction with bull and its excretory materials.

Exposing Nili-Ravi buffalo on d 5 after parturition to mature bull continuously (BEC) increased the percentage of buffalo showing estrus and reduced the postpartum interval to estrus than buffalo that have no visual contact with bull (NE). This result agrees with the result of Custer et al. and Fernandez et al. In present study buffalos exposed continuously to bull returned to estrus 13 d prior than buffalo without exposure to bull which is agreed with Naasz and Miller. Postpartum interval to estrus did not alter in buffalo exposed intermittently to bull (BEI) for 2 h daily. Postpartum estrus interval differed non-significantly (P>0.05) among BEI, EPB and NE buffaloes. Petropavlovskii and Rykova reported that postpartum interval to conception decreased with Berardinelli and Joshi. Berardinelli and Joshi reported that postpartum interval to estrus interval differed non-significantly (P>0.05) among BEI, EPB and NE treatments (Table 1). Progesterone concentration enhanced conception rate and in turn curtails the postpartum interval to estrus.

The proportion of buffalo displayed estrus in BEC was more than BEI, EPB and NE. Exposing postpartum Nili-Ravi buffalo continuously to bull increased the proportion of buffalo that showed estrus. The results of present study show compatibility agreement with other studies. The present work also agrees with Rekwot. The proportion of buffalo exposed intermittently to bull in BEI was equivalent to the proportion of buffalo not exposed to bull in NE (Table 1). This result agrees with the result of Fernandez et al. and in contrast with Petropavlovskii and Rykova due to the difference of duration and period of exposure. The buffalo exposed to excretory product of bull (EPB) show greater proportion (66.66%) of displaying estrus than NE. This result also supported the result of Berardinelli and Joshi.

The mechanism exactly how bulls hasten resumption of ovarian activity and estrus interval is unknown. The bio-stimulatory effect of bull alters the LH secretion that reached at highest point in ovulation. Bio-stimulatory effect of bull on estrus interval is associated with pheromonally activated signals that stimulate release of GnRH through hypothalamus and increased the release of LH. Continuous presence of bull involves in signaling trigger as well as other cues that enhance estrus. Bio-stimulatory pheromones are present in excretory product of bull reported by. Pheromonal activity in the bio-stimulatory effect is most likely to have in urine of the bull.

The progesterone concentrations (ng/mL) were apparently higher in the buffalo of BEC treatment than buffalo not exposed to bull (NE) treatment. Several researches observed transient increase in progesterone concentration from postpartum to estrus. Increase in progesterone concentration enhance conception rate and in turn curtails the postpartum interval to estrus.

**Table 1. Interval from Calving to Estrus and Serum Progesterone Concentration in BEC, BEI, EPB and NE Buffaloes (Mean ± SE)**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>BEC (n=12)</th>
<th>BEI (n=12)</th>
<th>EPB (n=12)</th>
<th>NE (n=12)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Percentage of buffaloes showing estrus per treatment</td>
<td>83.33% (10/12)</td>
<td>58.33% (07/12)</td>
<td>66.66% (08/12)</td>
<td>58.33% (07/12)</td>
</tr>
<tr>
<td>Interval from calving to first behavioural estrus (days)</td>
<td>55.2±0.78a</td>
<td>66.71±0.93b</td>
<td>68.25±0.87b</td>
<td>68.57±0.93b</td>
</tr>
<tr>
<td>Progesterone concentration from calving to first behavioural estrus</td>
<td>0.88±0.1a</td>
<td>0.86±0.06a</td>
<td>0.84±0.08a</td>
<td>0.82±0.06a</td>
</tr>
</tbody>
</table>

Values lacking common superscripts letters differ significantly (P<0.05)
It was concluded that buffalo exposed to the bull continuously showed reduced postpartum interval to estrus. Presence of bull has positive effect in reducing calving interval in Nili Ravi buffalo. Although bulls are considered burdens by some farmers due to the cost of nutrition and management, it is clear that presence of bull is beneficial for improving the reproductive efficiency of buffalo.

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