

## EFFECT OF EXOGENOUS GnRH AND PGF<sub>2α</sub> ON POSTPARTUM ESTROUS ACTIVITY AND FERTILITY OF BUFFALOES DURING LOW BREEDING SEASON

Rafiq. H. Usmani

Animal Sciences Division, Pakistan Agricultural Research Council, P.O. Box: 1031, Islamabad- 44000, Pakistan

### ABSTRACT

Pluriparous Nili-Ravi buffaloes (n = 18) which calved during the months of November to July were treated with a single injection of 50µg of GnRH on day 30 postpartum followed by an injection of 150 µg of PGF<sub>2α</sub> seven days later. Response to hormonal treatment in terms of induced estrus within 96 hours after the injection of PGF<sub>2α</sub> was 27.8%. Only 3 buffaloes conceived at the induced estrus giving an overall fertility response of 16.7%. During first three months postpartum, 61.1% of buffaloes resumed observable estrous activity with a mean postpartum interval of 53.7±15.6 days. Ten of eighteen buffaloes (55.5%) became pregnant within 90 days postpartum with an average postpartum interval of 63.0±20.0 days. Retrospective grouping of experimental buffaloes that received hormonal treatment during initial stage (n=9) or terminal stage (n=9) of the low breeding season revealed that the estrus response (within 96 hours) was higher during initial than the terminal stage (44.4 vs 11.1%; P<0.05). Cumulative estrous activity during first 90 days postpartum was also higher during the initial stage (77.7 vs 44.4%; P<0.05). The mean postpartum intervals to first observed estrus (47.3 vs 64.5 days) and to conception (56.8 vs 72.5 days) were significantly shorter during the initial than the terminal stage of low breeding season. It is concluded that during the low breeding season, the early postpartum Nili-Ravi buffaloes show poor response to exogenous GnRH plus PGF<sub>2α</sub> treatment, in terms of induced estrus and subsequent fertility. The efficiency of hormonal treatment is, however, comparatively better during the initial than the terminal stage of low breeding season.

**Key words:** Buffalo, GnRH, PGF<sub>2α</sub>, Low breeding season, Postpartum, Reproduction.

### INTRODUCTION

Delayed resumption of postpartum estrous activity is the most vital factor responsible for poor reproductive efficiency of dairy buffaloes. Seasonal trend of breeding plays a significant role in regulating the period of postpartum anoestrus in dairy buffaloes. Depending on the environmental conditions (e.g. ambient temperature, humidity and rainfall), the peak breeding season of buffaloes varies with the geographical location (Shah *et al.*, 1989; Naqvi, 2000). The author examined the breeding records of Nili-Ravi buffalo herd (n=85) maintained at the Livestock Research Station Islamabad for the period of five years (1995-1999) and observed that on yearly basis, more than 70% of non-pregnant breedable buffaloes were bred during the months of September to November. These months, therefore, represented the "peak breeding season" of buffaloes at the research station.

Breeding activity during other nine months (December to August) remained lower than 20% and these months represented "low breeding season" of buffaloes.

It is generally believed that the postpartum anoestrus is caused by the deficiency of gonadotrophic hormones LH and FSH. Owing to its ability to stimulate the release of LH as well as FSH, the gonadotropin releasing hormone (GnRH) has been successfully used in combination with prostaglandin-F<sub>2α</sub> (PGF<sub>2α</sub>) to induce ovulation in anoestrus and early postpartum cows (Britt *et al.*, 1974; Nash *et al.*, 1980; Thatcher *et al.*, 1993). Some work done with non-suckled buffaloes indicated a positive response of exogenous GnRH on the postpartum reproductive performance (Shah *et al.*, 1990). In this study, however, a single injection of GnRH was given on day-14 postpartum and PGF<sub>2α</sub> was not given to synchronize the oestrous activity.

Present study was designed to examine the effect of exogenous GnRH and PGF<sub>2α</sub> on the induction of observable oestrus and subsequent fertility of limited-

suckled postpartum buffaloes during initial and terminal stages of the low breeding season.

## MATERIALS AND METHODS

The study was conducted at the Livestock Research Station of the National Agricultural Research Centre, Islamabad. Pluriparous buffaloes (n=18) of Nili-Ravi breed which calved normally during the months of November to July were included in the study. All experimental buffaloes received exogenous hormonal treatment consisting of intramuscular injection of 50 µg of GnRH on day 30 postpartum followed by 150 µg of PGF<sub>2α</sub> seven days later. The months during which hormonal treatment was applied (December to August) represented the low breeding season of Nili-Ravi buffaloes at the research station.

Age and parity of experimental buffaloes ranged from 6 to 15 years and 2 to 7, respectively. Throughout experimental period, the buffaloes were maintained as a group and were housed in semi-covered sheds. Each buffalo was daily fed with 30-40 kg of available green fodder plus 3-4 kg of a concentrate mixture containing 16% crude protein and 68% total digestible nutrients. The buffaloes were hand-milked twice daily and limited suckling by calf was used for let-down of milk.

Beginning on the day of PGF<sub>2α</sub> injection, all buffaloes were observed for oestrous activity twice daily at 6 AM and 6 PM with the help of a penile deviated teaser bull. The experimental buffaloes were continuously observed for oestrous activity until day 90 postpartum. A buffalo was considered to be in "standing oestrus" when she accepted the teaser bull and stood willingly for being mounted. Any buffalo detected in standing oestrus was inseminated twice at 12th and 20th hour after the onset of oestrus using good quality fresh semen collected from buffalo bulls maintained at the research station. Inseminated buffaloes were checked for pregnancy diagnosis through rectal palpation of the uterus on days 50-60 postbreeding.

Buffaloes observed in oestrus within 96 hours after the treatment with PGF<sub>2α</sub> were considered as respondents to the hormonal treatment. Number of buffaloes responding to hormonal treatment, showing standing estrus and becoming pregnant during first 90 days postpartum were recorded. Experimental buffaloes were retrospectively grouped by the stage of low breeding season during which hormonal treatment was applied. The periods from December to February and June to August were considered as initial and terminal stages of low breeding season, respectively. Percentages of buffaloes responding to hormonal

treatment, showing observable oestrus and becoming pregnant were compared between initial and terminal stages of low breeding season by the use of Chi-square test. Student t-test was employed for the comparison of mean values of variables like postpartum interval to first observed oestrus, services per conception and postpartum interval to conception (service period) between the two stages of low breeding season (SYSTAT, 1996).

## RESULTS

Results regarding overall response of experimental buffaloes to the hormonal treatment, postpartum oestrous activity and fertility during first 90 days after calving are summarized in Table 1. Five of eighteen buffaloes (27.8%) responded to hormonal treatment and exhibited estrous behaviour within 96 hours after the injection of PGF<sub>2α</sub>. Three buffaloes conceived after insemination at the induced oestrus and the overall fertility response to hormonal treatment was 16.7%.

The cumulative percentage of buffaloes showing observable oestrous activity during first 90 days postpartum was 61.1 (11/18) and the average interval to first estrus in these buffaloes was 53.7±15.6 days. The remaining 7 buffaloes remained anoestrus during first 3 months after calving. The cumulative fertility rate in experimental buffaloes until day 90 postpartum was 55.5%. The inseminated buffaloes required 1.4 ± 0.5 services per conception and the mean interval from calving to conception was 63.0±20.0 days.

The comparative results for initial and terminal stage of low breeding season in terms of response to hormonal treatment and oestrous activity and fertility of experimental buffaloes during first 90 days postpartum, are presented in Table 2. The buffaloes receiving hormonal treatment during initial stage of low breeding season (December to February) showed a better oestrus response than those treated during the terminal stage (June to August; P<0.05). Fertility response to hormonal treatment was, however, not different between the two stages of low breeding season.

For the period of first 90 days after calving, the cumulative percentage of buffaloes showing estrous activity was higher during the initial than the terminal stage of low breeding season (77.7 Vs 44.4%; P<0.05). The postpartum interval to first observed estrus was shorter (P<0.05) during the initial than the terminal stage. Cumulative percentage of buffaloes conceiving during first 90 days postpartum was slightly higher during the initial stage of low breeding season but the difference from terminal stage was not significant (P>0.05). Similarly, the mean number of services per conception did not differ between the initial and

Table 1: Effect of hormonal treatment on estrous activity and fertility of postpartum buffaloes during low breeding season.

Reproductive Variable	Value
<b>Response to Hormonal Treatment<sup>a</sup></b>	
Number of buffaloes receiving hormonal treatment	18
Oestrous response within 96 hours after PGF <sub>2α</sub> injection	
Number	5
Percent	27.8%
Fertility Response:	
Number	3
Percent	16.7%
<b>Cumulative Oestrous Activity During First 90 Days Postpartum</b>	
Buffaloes showing observable oestrus	
Number	11
Percent	61.1%
Mean postpartum interval to observed oestrus (days) <sup>b</sup>	53.7±15.6
<b>Cumulative Fertility During First 90 Days Postpartum</b>	
Buffaloes becoming pregnant:	
Number	10
Percent	55.5%
Number of services per conception <sup>b</sup>	1.4±0.5
Mean postpartum interval to conception (days) <sup>b</sup>	63.0±20.0

<sup>a</sup>GnRH on day 30 postpartum followed by PGF<sub>2α</sub> on day 37 postpartum ; <sup>b</sup>Values are Mean ± SD

Table 2: Comparison of initials and terminal stage of low breeding season for response to hormonal treatment, oestrous activity and fertility of postpartum buffaloes.

Reproductive Variable	Initial Stage (Dec. to Feb.)	Terminal Stage (Jun. to Aug.)
<b>Response to Hormonal Treatment<sup>a</sup></b>		
Number of buffaloes receiving hormonal treatment:	9	9
Oestrous response within 96 hours after PGF <sub>2α</sub> injection :		
Number	4	1
Percent	44.4 <sup>c</sup>	11.1 <sup>d</sup>
Fertility Response:		
Number	2	1
Percent	22.2	11.1
<b>Cumulative Estrous Activity During First 90 Days Postpartum</b>		
Number	7	4
Percent	77.7 <sup>c</sup>	44.4 <sup>d</sup>
Mean postpartum interval to observed estrus (days) <sup>b</sup>	47.3±6.4 <sup>c</sup>	64.5±20.8 <sup>d</sup>
<b>Cumulative Fertility During First 90 Days Postpartum</b>		
Number	6	4
Percent	66.6	44.4
Number of services per conception	1.5±0.5	1.2±0.5
Mean postpartum interval to conception (days) <sup>b</sup>	56.8±12.4 <sup>c</sup>	72.5±26.7 <sup>d</sup>

<sup>a</sup>GnRH on day 30 postpartum followed by PGF<sub>2α</sub> on day 37 postpartum

<sup>b</sup>Values are Mean ± SD

<sup>cd</sup>Values within the same row with different superscript differ at P>0.05.

terminal stage of low breeding season. However, the postpartum interval to conception, was significantly shorter in buffaloes during initial stage as compared to the terminal stage of low breeding season ( $P < 0.05$ ).

## DISCUSSION

The overall efficiency of hormonal treatment in terms of induction of detectable estrous activity in buffaloes during low breeding season was only 27.8% (5/18). Similar low response (30%) have been reported by Shah *et al.* (1990) in Nili-Ravi buffaloes treated with a single injection of 100µg of GnRH on day 14 postpartum. Early postpartum stage at the time of treatment with GnRH appears to be responsible for the low response because late postpartum (> 120 days) but acyclic buffaloes showed higher response to gonadotrophic treatment during summer season (Chohan *et al.*, 1995). In dairy cows, use of exogenous GnRH followed by an injection of PGF<sub>2α</sub> seven days later is a well established system of estrous induction in which follicular development is stimulated by GnRH and regression of corpus luteum is synchronized by PGF<sub>2α</sub> (Thatcher *et al.*, 1993). Delayed resumption of ovarian activity coupled with higher incidence of follicular atresia in early postpartum suckled buffaloes (Usmani *et al.*, 1985) might, therefore, lower the efficiency of exogenous GnRH to synchronize follicular development and subsequent induction of oestrus.

In addition to its limited ability to induce estrus in early postpartum buffaloes during low breeding season, the exogenous hormonal treatment system (GnRH followed by PGF<sub>2α</sub>) had carry over effect in terms of subsequent estrous cyclicity and fertility. This was evident from the shorter postpartum intervals to first observable estrus and conception recorded in the present study when compared with results of previous studies in which exogenous hormonal treatment was not applied (Chaudhary & Pasha, 1988; Shah *et al.*, 1989). The positive carry-over effect of GnRH-PGF<sub>2α</sub> system, however, appears to be confounded with the effect of use of fresh semen and double insemination of oestral buffaloes. The conception rate in buffaloes after natural service or use of fresh semen has been reported to be higher than that observed after the use of frozen-thawed semen (Qureshi *et al.*, 1988). Moreover, double insemination of oestral buffaloes (8-hours apart) is expected to cover any deficiency in the accuracy of detection of onset of estrus, thus resulting into better chances of fertilization.

Stage of low breeding season during which experimental buffaloes received hormonal treatment,

had significant effect on certain variables of postpartum reproduction. Buffaloes receiving hormonal treatment during initial stage (December to February) showed better response in terms of percentage of induced estrus within 96 hours after PGF<sub>2α</sub> injection, percentage of overall observed estrous activity during 90 days postpartum and mean postpartum intervals to first observed estrus and conception, than those receiving hormonal treatment during terminal stage (June to August) of low breeding season.

Effect of stage of low breeding season on the efficiency of exogenous GnRH - PGF<sub>2α</sub> treatment, as observed in the present study, could be of considerable practical significance for the managers of dairy buffalo farms. Due to seasonal trend of breeding, majority of buffaloes (>60%) at the farm calve during the months of July, August and September (Usmani & Mirza, 2000). After completing their lactation of 8-9 months, these buffaloes become dry and the farm management faces the problem of severe shortage of milk production during the months of May, June and July.

One of the possible options to solve this problem is to shift the calving season of some selected buffaloes either forward (during October to December) or backward (during April to June) by the use of exogenous GnRH - PGF<sub>2α</sub> treatment. Data of the present study indicates that efficiency of hormonal induction of estrus and breeding in buffaloes during June to August (to induce calvings during April to June) will be low. Instead, the forward shifting of calving season is recommended because the chances of success are better if the attempt of hormonal induction of estrus is made during December to February in order to synchronize the calving of buffaloes during the months of October, November and December.

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