

SEASONAL VARIATION IN AI AND PREGNANCY RATE IN BUFFALO AND IMPROVING THEIR FERTILITY STATUS FOLLOWING APPLICATION OF FTAI DURING NON-BREEDING SEASON

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Received: 02.05.2016

Accepted: 01.07.2016

ABSTRACT

Seasonal variation in artificial insemination (AI) and pregnancy rate in buffalo of small farmers was recorded following analysis of AI carried out in 1087 buffalo in a calendar year. On an average, 90.6 ± 11.1 inseminations were carried out per month and the average pregnancy rate in a calendar year in inseminated buffaloes was $40.0 \pm 2.2\%$. The outcome of failure of AI viz. returning to estrus or remaining anestrus was also seasonally variable. In June and January, the pregnancy rate in buffalo was minimum (27%) and maximum (54%), respectively as compared to other months. The overall (FTAI and re-insemination) conception rate in ovsynch (n=24) and estradiol (E) - progesterone (P) based FTAI protocol (n=96) was 25% and 69.8% ($p < 0.05$), respectively. Out of buffalo failing to conceive following FTAI, 62.5% remained anestrus in ovsynch group, whereas, their counterparts in E-P group were 19.7% only ($p < 0.05$). In brief, the non-breeding season influences exhibition of overt estrus and pregnancy establishment in buffalo. For alleviating non-breeding season induced sub-fertility in buffalo, E-P based FTAI protocol is an option.

Keywords: Buffalo, Conception, Estradiol, FTAI, Season

INTRODUCTION

Small dairy farmers raising 1-3 bovines account to 3/4th of livestock wealth and 68% of milk production in India. Moreover, only 19.2% buffalo ownership in India is with large farmers. Thus, to have a major impact on total milk production in India, strategies need to focus on improving the reproductive potential of buffalo reared by small farmers. During summer season in Punjab, true anestrous buffalo exhibit clear follicular wave pattern with dominant follicle attaining ovulatory size (>12 mm) but failing to ovulate (Ghuman *et al.*, 2010). Applying fixed-time AI (FTAI) protocol in buffalo may provide a potential alternative for increasing their life time productive period (de Carvalho *et al.*, 2016). However, the major bottlenecks in wide application of FTAI at small farmer's doorstep are, a) poor conception rate during non-breeding season, and b) failure of non-conceived buffalo to return to estrus following FTAI. In

addition, if a buffalo fails to conceive, a farmer has to bear a loss of \$4.2/d in terms of loss of milk and other managemental expenses. In fact, during summer, conception rates were 26% following spontaneous-estrus and 7% following induced-estrus in buffalo (Baruselli *et al.*, 2007). Thus, the small farmers can be convinced to adopt FTAI if a protocol with good conception rate is available. The present study aimed at, a) recording seasonal variation in AI and pregnancy rate in buffalo of small farmers, and b) comparison of fertility outcome of two FTAI protocols during non-breeding season in buffalo reared by small farmers.

MATERIALS AND METHODS

For recording seasonal variation in AI and pregnancy rate in buffalo of small farmers, the work was carried out in village Badbar, Distt Barnala ($30^{\circ}23'N$, $75^{\circ}33'E$) between November, 2013 to October, 2014 and the monthly climate averages (minimum and maximum temperature, and rainfall) were recorded. During the study period, 1087 buffalo were inseminated,

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Figure 1: Fixed-Time artificial insemination (FTAI) protocol. E - Estradiol, P - Progesterone, EB - Estradiol Benzoate, PD - Pregnancy diagnosis

and subsequently, the buffalo returning to estrus within day 90 post-AI were recorded. The remaining buffalo were subjected to per-rectal pregnancy diagnosis on day 90 post-AI.

The second part of study was conducted on 120 healthy anestrous buffalo in non-breeding season. Group-I buffalo (n=24) were subjected to ovsynch protocol (Protocol cost \$11.63; day 0 and day 9, 20 µg GnRH analogue each; day 7 500 µg PGF_{2α}, both hormones by i.m. route; AI 16h after day 9 GnRH). Group-II buffalo (n=96, Protocol cost \$26.16) were administered (i.m.) 2 mg estradiol benzoate on day 0 and 500 µg PGF_{2α} on day 9. Sustained progesterone

release device (1.38 g progesterone) was placed intravaginally from day 0 to day 9. On day 11, 20 µg GnRH analogue and 500IU eCG were administered (i.m.). AI was done 16h after GnRH analogue administration (Figure 1). In both groups, pregnancy was confirmed by ultrasound aided diagnosis on day 60 post-AI and the buffalo failing to conceive and returning to estrus were re-inseminated at observed spontaneous estrus without any additional hormonal treatment. Statistical analyses were performed using MINITAB release 13.2 statistical software. Numerical data differences were considered significant at p<0.05 using Chi-square (χ²) test.

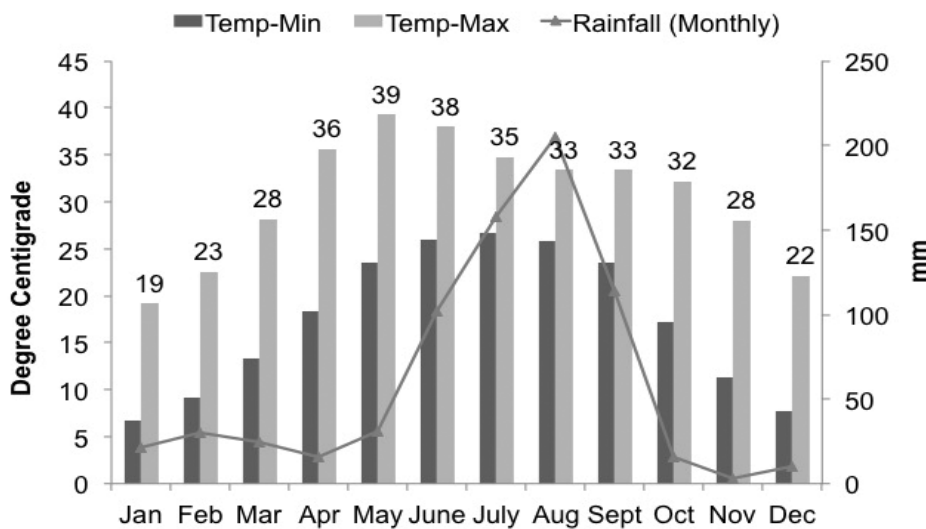


Figure 2: The climate averages in a calendar year in Punjab state

RESULTS AND DISCUSSION

The climate averages revealed the period between May to September with high temperature and high rainfall leading to hot-humid conditions (Figure 2). The data on seasonal variation in AI in buffalo suggested that on an average 90.6 ± 11.1 (Monthly range: 41-151) buffalo were inseminated per month. The maximum inseminations per month were in October and November, whereas, the minimum inseminations per month were in June and July (Figure 3). This suggested the adverse impact of hot season on efficiency of buffalo to exhibit estrus. In Indian buffalo, maximum percentage of heats was observed during short day-length (74%) and the minimum during long day-length (26%; Tailor *et al.*, 1990).

Furthermore, the average pregnancy rate subsequent to AI of 1087 buffalo in a calendar year was $40.0 \pm 2.2\%$ (Monthly range: 27-54%). The minimum and maximum pregnancy rate was observed in June and January and was respectively recorded as 27% and 54% (Figure 3). This further strengthened the adverse impact of heat stress on reproductive efficiency of buffalo (de Carvalho *et al.*, 2016). In an earlier study in buffalo, conception rates were lower

and number of services per conception was higher during summer than in other seasons (Qureshi *et al.*, 1999). In summer season, high plasma prolactin secretion contributed to poor fertility in buffalo by lowering production of gonadal hormones (estradiol- 17β , total estrogens and progesterone (Das and Khan, 2010). Moreover, embryonic mortality is increased following dam's exposure to elevated ambient temperatures especially in tropical areas. During winter, early embryonic mortality in buffalo was 20% which increased to 45% in summer (Abdoon *et al.*, 2001).

Out of inseminated buffalo, $36.5 \pm 2.2\%$ (Monthly range, 26-51%; $n=396/1087$) returned to estrus within day 90 post-AI. The buffalo not returning to estrus within day 90 post-AI ($n=691/1087$) were subjected to per-rectal examination for pregnancy diagnosis and it was revealed that $36.6 \pm 3.4\%$ (Monthly range, 17-56%; $n=240/691$) buffalo were non-pregnant (anestrus) and remaining $63.4 \pm 3.4\%$ ($n=451/691$) buffalo were pregnant. This concluded that out of the inseminated buffalo reared by small farmers, a good proportion of buffalo failing to get pregnant goes to anestrus condition. This warrants an early pregnancy diagnosis

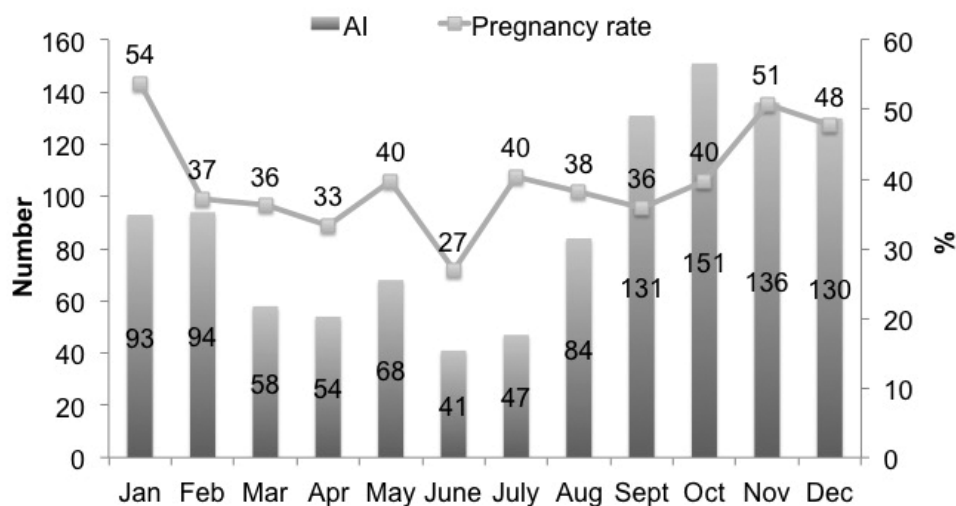


Figure 3: Seasonal variation in AI and pregnancy rate in buffalo reared by small farmers in Punjab state (India)

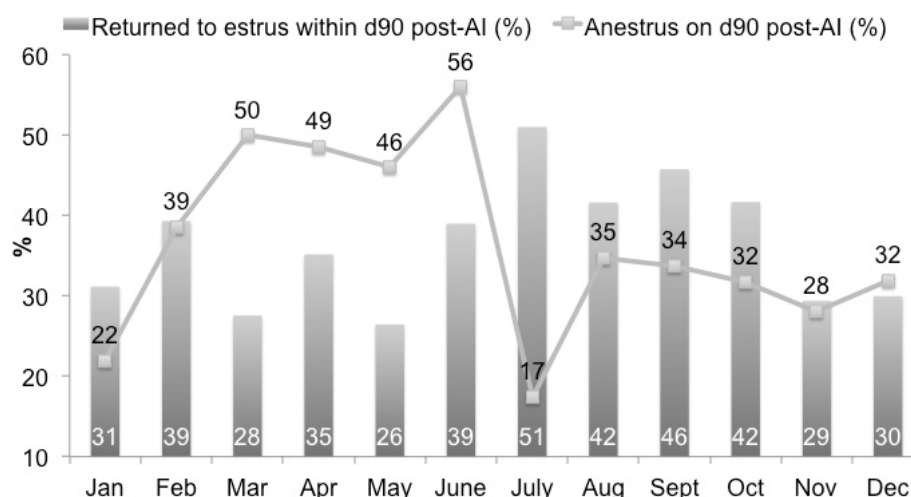


Figure 4: Seasonal variation in the outcome following failure of conception in inseminated buffalo reared by small farmers in Punjab state (India)

of inseminated buffalo to reduce the non-productive period in the life-span of a buffalo (Baruselli *et al.*, 2007).

In the present study in buffalo, it was also observed that the outcome of failure of conception following AI viz. returning to estrus or remaining anestrus is seasonally variable (Figure 4). In most of the buffalo inseminated between March to June, the outcome following failure of conception was diagnosed as anestrus, whereas majority of the buffalo inseminated between July to October and failing to conceive returned to estrus within day 90 post-AI ($p < 0.05$, Figure 4). Between November and February, there was not much difference in the outcome of failure of conception ($p > 0.05$, Figure 4).

The conception rate following FTAI in group-I and group-II was 20.83% ($n=5/24$) and 53.50% ($n=51/96$; $p < 0.05$), respectively. Out of the buffalo failing to conceive to FTAI, about 62.5% ($n=15/24$) remained anestrus in group-I, whereas, their counterparts in group-II were only 19.7% ($n=19/96$). The remaining non-conceiving buffalo that exhibited spontaneous estrus followed re-insemination in group-I and group-II and the respective conception rate was 25% ($n=1/4$) and 61.65% ($n=16/26$). Overall (FTAI and

re-insemination) conception rate in buffalo of group-I and group-II was 25% ($n=6/24$) and 69.8% ($n=67/96$; $p < 0.05$), respectively. Similar results in non-breeding season using estradiol-progesterone based FTAI protocol were also reported in Brazilian Murrah buffalo (de Carvalho *et al.*, 2016).

In summary, the non-breeding season has impact on buffalo exhibiting overt estrus and subsequently getting conceived. Estradiol/progesterone-based FTAI protocol is an option for alleviating non-breeding season induced sub-fertility in buffalo. Although estradiol/progesterone-based FTAI protocol is costlier compared to ovsynch, the former is economical due to much better conception rate in buffalo during non-breeding season.

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