

INDUCTION OF CYCLICITY IN MURRAH BUFFALO-HEIFERS DURING SUMMER USING DIFFERENT HORMONAL PROTOCOLS

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ABSTRACT

Twenty four sexually mature, acyclic buffalo heifers were randomly and equally divided into four groups. Group A was kept as control in which no treatment was given. Group B received CIDR on day 0 (start of experiment), 500 I.U. PMSG (day 9), removal of CIDR (day 10) and 1500 I.U. eCG at the time of insemination. Group C animals were subjected to modified Ovsynch protocol where 500 I.U. PMSG was administered (day 0) followed by 16 µg GnRH analogue (day 3), 500 µg cloprostenol (day 10) and second injection 16 µg GnRH (day 12). Group D animals received 500 mg hydroxyprogesterone and 10 mg estradiol valerate (day 0) followed by 500 µg cloprostenol and 500 I.U. PMSG on day 7 and 9, respectively. Estrus was exhibited in 100% (Group B), 66.7% (Group C and D) and 33.3% (Group A). The duration of estrus was also higher in group B (28.2±6.12 hrs) as compared to group A (16.0±2.00 hrs) and group D (17.0±2.52 hrs). The pregnancy rate from insemination at induced estrus was 50.0, 50.0, 16.7 and 50.0 per cent in group B, D, C and A, respectively. The results of the present investigation revealed that different hormonal combinations were effective in inducing cyclicity and conception at first service as compared to untreated animals. Therefore, it is concluded that use of progesterone based protocols during summer season is good for induction of estrus and improved conception rates in acyclic sexually mature buffalo heifers.

Key words: Buffalo-heifers, Cyclicity, Fertility, Hormonal protocols, Summer

INTRODUCTION

Productive and reproductive efficiency of animals are complementary to each other. A number of reproductive problems, especially delayed puberty, silent estrus, summer anestrus affect buffaloes and cause huge economic losses to the farmers. The effect of summer on animal is two fold. Firstly, it reduces feed intake and alters the profiles of reproductive hormones. Lower circulating concentration of FSH and LH (Razdan *et al.*, 1982) has been reported during summer than during winter months. During summer, when ambient temperature and photoperiod are at their

maximum, prolactin levels are highest and plasma progesterone levels are lowest (Kaker *et al.*, 1982). Various managerial strategies and hormonal regimens have been used to overcome the problem of seasonal anestrus in buffaloes. Favorable effects of GnRH and its analogues in inducing early postpartum ovarian activity and estrus have been reported in buffaloes (Saini and Lohan, 2003). Hormonal preparations including Crestar, CIDR followed by PMSG and progesterone (Duraprogen: Tetragon Chemi, India) plus estradiol valerate (Progynon depot: German Remedies) might provide encouraging results in postpartum anestrus cattle. However, the information on the effectiveness of these hormonal treatments in Murrah buffalo-heifers during summer season is limited. Hence, the present study was taken up to investigate the effect of different hormonal regimens in sexually mature acyclic buffalo-heifers (i) to induce cyclicity and (ii) to study fertility response of treated animals at induced estrus.

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MATERIALS AND METHODS

Twenty four apparently healthy acyclic buffalo-heifers having more than 300 kg body weight and more than 30 months of age maintained at animal farm of Buffalo Research Centre of College of Animal Sciences, Haryana Agricultural University, Hisar were selected. They were confirmed to be in anestrus by rectal examination twice at an interval of eleven days and were randomly and equally divided into four groups (A, B, C and D) having six animals in each group. The study was conducted during low breeding season (May-June). The animals included in the study were stall-fed collectively with sufficient quantity of green fodder and concentrates according to their individual requirements. Water showering of animals was also done 1-2 times a day during the period of experiment.

Group A was kept as control in which no treatment was given. Group B animals received Controlled Internal Drug Release device (Eazi-Breed CIDR™, Inter Ag, Hamilton, New Zealand) intra-vaginally on day 0 (start of experiment). On day 9 (one day before CIDR removal), 500 I.U. PMSG (Folligon®, Intervet International, The Netherlands) was administered intramuscularly. Each heifer of this group was also administered 1500 I.U. eCG (Chorulon®, Intervet International, The Netherlands) at the time of insemination. Animals of group C received 500 I.U. PMSG intramuscularly on day 0 and an injection of 16 µg GnRH analogue (Receptal® VET, Intervet International, Germany) and 500 µg cloprostenol (Vetmate™, Vetcare, Provimi, Holland) on day 3 and 10, respectively. A second injection of 16 µg GnRH analogue was administered on day 12 of the start of experiment. Fixed time insemination was done at 12 and 24 hours after second GnRH injection if the animals were not detected in heat by the teaser bull. Each buffalo-heifer of Group D was administered a single injection of 500 mg Hydroxy-progesterone caproate (Duraprogen®, Tetragon Chemi Pvt. Ltd., Bangalore, India) subcutaneously and an injection of 10 mg Estradiol valerate (Progynon depot®, German Remedies Ltd., Mumbai, India) intramuscularly on day 0. Subsequently, each animal of Group D also received an injection of 500 µg cloprostenol and 500 I.U. PMSG intramuscularly on day 7 and 9, respectively. The

experimental buffalo-heifers were observed visually frequently after the end of treatment for external estrus signs aided by a vasectomized bull parading thrice in morning, noon and evening. The animals were confirmed in estrus by per rectal examination and two inseminations at 12 hours apart were performed using good quality Murrah bull frozen semen. Thereafter, the animals were observed for estrus twice daily (morning and evening) by parading vasectomised bull as per farm routine till 60 days of insemination.

In group B animals, where CIDR was inserted intravaginally, the implant was found intact in all but one animal. One animal in Group C, died after the insemination and before the pregnancy could be confirmed.

Estrus response was observed as percentage of females showing estrus of those treated. First service conception rate was calculated as percentage of animals that conceived to fixed time insemination at induced estrus in each group. Pregnancy was confirmed by palpation per rectum at 60 days post insemination. Comparative study of all the treatment regimens was carried out by comparing their effectiveness and efficacy for estrus induction and successful conception. Chi-square was used to compare per cent conception rate and one way analysis of variance (ANOVA) followed by Duncan's Multiple Range Test was used to compare effect of various treatments on other observations.

RESULTS AND DISCUSSION

All animals of group B exhibited prominent symptoms of estrus. In groups C and D, 66.7% animals were detected in estrus by the teaser bull. It has been observed that edema of vulva, congestion of vulvar mucus membranes, tone of uterine horn and open os-cervix were some of the important heat symptoms in buffaloes.

The estrus response in group B is significantly ($P < 0.05$) higher than group A and is higher than results reported by Andurkar and Kadu (1995). Good estrus response with Crestar+PMSG in postpartum anestrus buffaloes in low breeding season has been reported by Lohan *et al.* (2001). The PMSG along with Crestar might be beneficial as PMSG leads to induction of behavioral

estrus signs (Singh *et al.*, 2004) due to enhanced ovarian stimulation. The higher estrus response in group B might be due to PMSG supplementation in the treatment protocol. Two-third (66.7%) animals of group C were induced to estrus in the present study which is in close agreement with the findings of Baruselli *et al.* (1999) and Singh *et al.* (2004). In the current study, behavioral estrus (swollen vulva, uterine tone and mucus discharge) was present in 66.7% buffalo-heifers. Similar results have also been reported by Neglia *et al.* (2003) with Ovsynch protocols.

The results on induction of estrus by using hydroxyprogesterone and estradiol valerate combinations followed by PMSG are in consonance with those reported by Shrivastava *et al.* (1997) whereas, Andurkar and Kadu (1995) reported an estrus response varying between 23 and 83 per cent. The present study supports the hypothesis that some amount of progesterone is necessary to sensitize the hypothalamic-hypophyseal system as it plays negative role on its withdrawal by day 7 or 9. The duration of estrus was higher in group B and group C as compared to group A and group D. Markandeya and Bharkad (2002) reported an average duration of estrus (18.4 hrs) by using norgestomet ear implant in anestrus buffaloes whereas, Rajveer (2004) reported duration of estrus as 30 hrs which is in close agreement with the findings of this study.

Reproduction in buffaloes is influenced by season and fertility is reduced during summer due to an environmental effect on the activity of the hypothalamo-hypophyseal axis (Santos *et al.*, 2004). The unpredictability in the timing of ovulation requires that at least two inseminations have to be performed in order to obtain acceptable conception rates. In the present investigation 50.0, 33.3, 16.7 and 50% animals of group B, D, C and A, respectively conceived at first service. The pregnancy rate at first service in group B animals in the present study is higher than reported by Markandeya and Bharkad (2002) and Rajveer (2004) and this might be because in addition to standard CIDR and PMSG protocol, hCG was also supplemented in this study. The administration of hCG at the time of breeding could result in the synchronization of ovulation,

fertilization and subsequent events required for successful maintenance of pregnancy (Breuel *et al.*, 1989). An overall pregnancy rate of 20.0% at induced estrus observed in our study in group C animals is comparable to the pregnancy rates (18.2%) reported by Irikura *et al.* (2003) after using Ovsynch protocol in buffalo-heifers. Earlier studies by Baruselli *et al.* (1999) with Ovsynch protocol in cyclic beef buffaloes reported higher pregnancy rate (50.0%) to first insemination which might be related to the use of cyclic animals and seasonal factors. A very low conception rate of 4.7% has been reported by De Rensis *et al.* (2005) after synchronized ovulation with fixed time Artificial Insemination in non-cyclic buffaloes. Overall pregnancy rate of 33.3% at induced estrus observed in the present study is in conformity with the fertility rates around 30.0 to 35.0% reported by Andurkar and Kadu (1995), however, higher fertility rates of 42 to 70% have also been reported by Lohan *et al.* (2001) and Rajveer (2004).

The results of the present study have shown that different hormonal combinations used in the study are quiet effective in inducing cyclicity and yielded higher estrus response and pregnancy rate at the induced estrus as compared to untreated animals. Thus, it can be safely concluded that use of progesterone based protocols during low breeding season allows the induction of estrus and improved the conception rates in acyclic sexually mature buffalo-heifers that would otherwise be non-productive.

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