The Indian Journal of Veterinary Sciences & Biotechnology ***(2017) Volume 12, Issue 4, 30-33***
ISSN (Print) : 2394-0247 : ISSN (Print and online) : 2395-1176, abbreviated as IJVSBT
http://dx.doi.org/10.21887/ijvsbt.v12i4.7675

Submitted : 09-02-2017 ***Accepted : 21-03-2017*** ***Published : 05-05-2017***

Comparative Efficacy of Different Medicaments for Induction of Estrus in True Anestrus Jafarabadi Buffalo Heifers

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Abstract

The relative efficacy of different treatment modalities was evaluated for induction of estrus in 32 post-pubertal true anestrus Jafarabadi buffalo heifers randomly divided into four equal groups. Animals of Group I received intravaginal CIDR and i/m injection of 1.0 mg estradiol valerate on day 0, i/m injection of 500 µg PGF2α on day 7 while removing CIDR, and 0.75 mg estradiol valerate on day 8. Fix timed insemination (FTAI) was performed at 48 and 72 h following PGF2α injection. Animals of Group II received standard Ovsynch protocol with FTAI. In Group III, Prajana HS 3 caps/day for 3 days along with i/m injection of Vitamin AD3E (5 ml) and Tonophosphan (15 ml) on first day were administered. Group IV animals received no treatment and served as control. Among all inseminated heifers, in non-return cases pregnancy was confirmed per rectum 60 days post-breeding. The estrus induction response in Group I and II animals was 100 %, whereas in Group III and IV it was 37.5 % and 12.5%, respectively. In Group I, one animal conceived at induced estrus and another four at second service while in Group II, five animals conceived at second service giving overall conception rate (CR) of 62.5% in each group within 77 days of treatment. In Group III, three animals conceived by 70 days and in Group IV, only one animal (12.5%) showed estrus and conceived by 50 days of treatment with overall CR of 37.5% and 12.5%, respectively. Mean plasma progesterone concentration was significantly ($p<0.05$) higher on day 7 as compared to day 0, at estrus and on day 20 post-AI in all the animals. It was concluded that both CIDR and Ovsynch protocols are better than herbal heat inducer + supplements in inducing fertile estrus in anestrus buffalo heifers.

Key Words: Estrus induction, Conception, CIDR, GnRH, PGF2α, Prajana, Jafarabadi heifers

Introduction

The productivity of buffaloes is essentially affected by the animal's reproductive efficiency, which is, in turn, reduced by the late maturity, poor estrus expression, and longer inter calving intervals and reduced ovarian activity**.** In fact, the low reproduction potential of the buffalo has been a major concern for decades, but now protocols have been developed that are able to precisely control the time of ovulation and thus avoid the need for estrus detection. A clinical survey by Tanwar *et al*. (2003) revealed higher incidences of anestrus and inactive ovaries in buffaloes (55.5 and 19.4%)

than in cows (43 and 17.2%, respectively). To synchronize the estrus cycle, ovarian activity is manipulated so that the time of ovulation can be predicted. Exogenous administration of progesterone exerts a negative feedback effect over the hypothalamus and pituitary and blocks the release of pituitary gonadotropin. Upon withdrawal of progesterone, the block is removed and larger quantities of gonadotrophins are released which in turn ensures growth and maturation of ovarian follicle and thus onset of estrus. In view of the above, this study was aimed to evaluate and compare efficacy of various protocols like use of CIDR, Ovsynch and herbal heat inducer along with Phosphorus and Vit. AD3E injection on fertility response in post-pubertal anestrus buffalo heifers and to study the plasma progesterone profile at day 0, day 7, day of estrus and day 20 post-AI in them.

Materials and Methods

Post-pubertal anestrus Jafarabadi buffalo heifers (n=32) from the Cattle Breeding Farm, JAU, Junagadh, aged 3 to 6 years weighing 350-500 kg, were used. All the animals were healthy and kept under strict periodic deworming and prophylactic vaccination against the endemic diseases. All the heifers were subjected to rectal examination and were found to have normal genital tract with smooth small inactive ovaries, and atonic uterine horns (true anestrus). They were divided in to four equal group each of 8 animals, and were managed as under.

In group-I (CIDR protocol), CIDR (1.38 g progesterone spine, Pfizer) was inserted intravaginally in combination with IM injection of 1.0 mg estradiol valerate (Progynon depot, German remedies) on day 0, i/m inj. PGF2α 500 µg (Cyclix, 2 ml) on day 7 while removing CIDR, and estradiol valerate 0.75 mg was given 24 h later, with fixed time insemination (FTAI) performed at 48 and 72 h after PGF2α injection. In group-II (Ovsynch protocol), IM injection of Buserelin acetate 20 µg - GnRH analogue (Receptal, 5 ml, Intervet) on day 0, followed by an injection of PGF2α 500 µg (Cyclix, 2 ml) on day 7 and a second injection of GnRH 20 µg on the day 9 were administered, and FTAI was performed twice at 12 and 24 h later. The animals once inseminated were followed for recurrence of estrus, if any, and bred again for three cycles, and in non-return cases pregnancy was confirmed per rectum 60 days of last AI.

In group-III, Indigenous preparations Prajana HS (Powders of Patola/*Trichosanthes dioica* seed 561.00 mg, Marichu/*Piper nigrum* fruit 61.875 mg, Pipali/*Piper longum* fruit, Sunthi/*Zingiber officinale* Rhizome 61.875 mg, Excipients q.s. preservatives: Methyl Paraben Sodium, Propyl Paraben Sodium, Bronopol), 3 caps/day for 3 days (Indian Herbs) were give PO (Per os/orally), along with i/m injection Vitamin AD3E, 5 ml (Zydus), and Tonophosphan 15 ml (Pfizer) on first day. Prajana HS was repeated again for days 11-13, if no estrus was exhibited by the animal. In Group IV, eight anestrus heifers of similar nature kept without any treatment served as control. The heifers exhibiting estrus were inseminated and followed for repeat AI or pregnancy as above.

Blood samples were collected in heparinized vials from heifers of all three treatment groups on day 0, i.e., the day of beginning of the experiment, day 7, day of estrus (first AI), and on day 20 post-AI. In control group, blood samples were collected on day of estrus and again on day 20 post-AI. Plasma progesterone concentration was estimated by employing standard RIA technique of Kubasic *et al.* (1984). The results, thus obtained, were compiled and analyzed statistically using ANOVA and students 't' test.

Results and Discussion

The induction of pubertal estrus at the earliest possible through exogenous hormonal supplementation in healthy Jafarabadi buffalo heifer was attempted. There was 100 % estrus induction response in Group I and Group II, but 37.5 % and 12.5% in Group III and Group IV, respectively. Saini *et al.* (1988) using PRID plus PMSG in non-cycling buffalo heifers reported similar estrus response. 100% estrus induction response using CIDR and Ovsynch protocol was also reported by Dhami *et al.* (2015) in crossbred cows. GnRH induces preovulatory LH surge in pre-pubertal heifers and

consistently induces ovulation of large ovarian follicles (>10 mm) if present at the time of treatment. Heat induction protocols and timed AI approaches are being considered as the recent reproductive technologies and practiced by several research workers (Pursley *et al*., 1995). Derar *et al.* (2012) found that 87.5% and 100.0% of buffalo heifers and buffalo, respectively, ovulated after the first GnRH injection of an ovsynch treatment.

In Group I, 5/8 (62.50%) heifers conceived - one at induced estrus and remaining four with 2nd cycle at the interval of average 70.33 days. This is in accordance with the findings of Nikam *et al*. (2002), Caesar *et al* (2011) and Sarabi and Mosaferi (2015). Progesterone induces follicular development with cyclicity and fertile estrus through its withdrawal effect. The present shortest duration for the onset of estrus with 100% induction in CIDR + estradiol group might be due to the administration of estradiol valerate on the day of implantation and withdrawal of CIDR. The estradiol 17 ß is necessary for the pulsatile LH secretion that is prerequisite for maturation and ovulation of follicle and expression of estrus. It also induces premature regression of CL and enhances response to progestagens (Jainudeen *et al.,* 2000).

In Group II also, 5/8 (62.5%) animals conceived with 2nd service at interval of average 77.20 days after induced estrus. Treatments with GnRH at day 0 and PGF2α at day 7 induced ovulation of the dominant follicles and regression of the CL, respectively, and, thereafter, the second administration of GnRH on day 9 controlled the ovulation of the new dominant follicle. The surge of LH at a random stage of the estrus cycle was induced by the administration of GnRH, which subsequently caused luteinization of the predominant follicles (Hoque *et al*., 2011). Researchers have found that ovsynch protocols can synchronize ovulation in 70-90 % of buffalo (Baruselli, 2001; Paul and Prakash, 2005; Hoque *et al*., 2011).

In Group III, 3/8 (37.50%) animals conceived at 1st observable heat at an average 70.67 days after initiation of treatment. Indigenous herbal preparations such as Prajana HS is commercially available and effective in restoring cyclicity with good success rates (Kodagali *et al.,* 1991). Nutritional deficiency or imbalance play a major role in causing anestrus and hence, specific vitamin-mineral supplementation should be considered for treatment of true anestrus in buffalo heifers. In Group IV, only one animal (12.5%) showed heat symptoms after 50 days of initiation of treatment and conceived at 1st service.

Table 1: Mean (± SE) plasma progesterone profile (ng/ml) at day 0, 7, day of estrus and day 20 post-AI of treatment and control groups

Group / Days	Treatment groups			
	CIDR	Ovsynch	Prajana HS + Vit. ADE+Tonophosphan	Control
0 day	0.40 ± 0.08	0.34 ± 0.07	0.45 ± 0.10	0.39 ± 0.06
7 day	4.03 ± 0.45**	3.88 ± 0.33**	1.17 ± 0.24**	---
Day of estrous	0.31 ± 0.08	0.43 ± 0.09	0.45 ± 0.20	0.23
20 day post A.I.	1.02 ± 0.6	0.51 ± 0.12	3.30 ± 0.43	3.2

** Highly significant ($p<0.01$)

The mean plasma progesterone profile at the initiation of treatment (0 day) was low in most of the animals. The plasma progesterone concentrations were significantly higher on day 7 as compared to corresponding values on day 0, at estrus/AI and on day 20 post AI in all treatment groups (Table 1). Antal *et al*. (1987) reported that conception could only be expected if the progesterone level at the time of insemination (day 0) is low and gradually increased up to at least 24 days after AI. This phenomenon was true for the present study since the progesterone concentration of the pregnant heifer was low (<1.0 ng/ml) at day of AI and then gradually increased (>1.0 ng/ml) until day 7 of treatment and day 20 post-AI.

Acknowledgment

We thank the Principal & Dean, College of Veterinary Science & Animal Husbandry and Research Scientist (AGB), Cattle Breeding Farm, JAU, Junagadh for providing necessary facility for carrying out this work.

Conflict of Interest: All authors declare no conflict of interest.

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