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Comparative Efficacy of Three Different Fixed Timed Artificial Insemination Protocols in Sahiwal Cows

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ABSTRACT

The study evaluated efficacy of three different fixed timed estrus synchronization protocols administered at random stage of estrous cycle in Sahiwal cows. Pluriparous Sahiwal cows (n=30) having normal estrous cycle and genitalia were randomly divided into three groups. Group 1 (Ovsynch group), cows (n=10) were administered standard Ovsynch protocol, Group 2 (Ovsynch + CIDR); cows (n=10) were administered Ovsynch protocol along with intravaginal insertion of CIDR implant on day 0, Group 3 (Estradiol based group); cows (n=10) were injected Estradiol benzoate 2 mg + Cloprostenol 500 µg along with insertion of CIDR on day 0, then Estradiol benzoate 1 mg + Cloprostenol 500 µg + eCG 400 IU (Inj. Folligon 400 IU) on Day 7. CIDR was removed at the time of prostaglandin injection (Day 7) in the group 2 and 3. All the cows were fixed timed inseminated on Day 10 of the protocol. Results revealed that the growth rate of dominant follicle from day 7 to 10 was higher in Estradiol based and Ovsynch + CIDR group than Ovsynch group (1.42±0.14, 1.38±0.13 and 1.13±0.07 mm, respectively). Average size of dominant follicle on day 10 was higher in Ovsynch + CIDR than the Ovsynch and Estradiol based group (11.4±0.48 vs 10.3±0.61 vs 10.2±0.42 mm, respectively). All cows of Ovsynch+ CIDR group underwent ovulation, whereas, 60 percent cows ovulated in the group 1 and 2. The ovulating cows had higher average diameter of preovulatory follicle on day 10 compared to anovulatory cows in the Ovsynch (11.6±0.20 vs 8.4± 0.80 mm, $p<0.05$) and Estradiol based group (10.5±0.56 vs 9.9±0.66 mm, $p>0.05$). Higher pregnancy rate was achieved in the Ovsynch + CIDR group compared to Estradiol based and the Ovsynch group (60 vs 40 and 30 percent, respectively). Results showed that Ovsynch + CIDR protocol was more effective on random day of estrous cycle in inducing ovulatory estrus and subsequent fertility compared to the Ovsynch and Estradiol based protocol. It was also concluded that higher diameter of dominant follicle on day 10 and follicle growth rate from day 7-10 favored ovulation and establishment of pregnancy in Sahiwal cows.

Key words: CIDR, Fixed Timed AI, Ovsynch, Ovulation, Pregnancy, Sahiwal.

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INTRODUCTION

Being a native of Indian sub-continent, Sahiwal is better adapted to survive under harsh tropical climatic conditions than *Bos taurus*. Due to their sturdy nature and premium pricing of milk and milk products, farmers have started rearing Sahiwal cows on commercial scale. However, the success of dairy farming entrepreneur depends upon the efficient management of reproduction component. Precise detection of estrus plays an important role in optimizing reproductive performance of a dairy farm. However, like any other indigenous breed, detection of estrus in Sahiwal is difficult due to poor expression of estrus signs. Estrus detection is also affected by the fact that duration of estrus is shorter (average 10 hours) in Sahiwal as compared to *Bos taurus* cows (Galina and Arthur 1990, Pinheiro *et al*., 1998). The estrus detection aids like pedometer and heat watch etc. have been found useful but require additional expenditure and waiting period till the cow come in estrus and get detected for insemination. This would invariably increase the number of open days and decrease the economic returns of a dairy farm.

To alleviate requirement of estrus detection, a number of fixed timed artificial insemination (FTAI) protocols are available which can be used irrespective of the stage of estrus cycle. However, FTAI protocols developed in *Bos taurus* when applied in *Bos indicus* did not yield similar success (Pinheiro *et al*., 1998) which could be due to minor differences in endocrinology and physiology of *Bos indicus* and *Bos taurus* cows. *Bos indicus* cows were found to be more sensitive to reproductive hormones like, luteinizing hormone (Griffin and Randel, 1978), progesterone and estradiol (Segerson *et al*., 1984). In addition to this, the differences had also been reported in their follicular dynamics. *Bos indicus* cattle had smaller maximum diameter of the dominant follicle (10–12 mm) and the corpus luteum (17–21 mm; Figueiredo *et al*., 1997) than *Bos taurus* cattle (DF: 14–20 mm and CL: 20–30 mm; Bo *et al*., 1993). These minor differences could impact response of *Bos indicus* cattle to various hormonal interventions. Therefore, the present study was planned to evaluate the available estrus synchronization protocols to select most appropriate one for its use in Sahiwal.

MATERIALS AND METHODS

Experimental animals

The study was conducted on Sahiwal cattle maintained at Regional Research & Training Centre (RRTC), Kaljharani, Bathinda and Dairy farm, DLF, GADVASU, Ludhiana, Punjab. Sahiwal cows (n=30) pluriparous, lactating, 60 to 180 days postpartum, non- pregnant with normal estrous cycle and genitalia, weighing between 320 to 450 kg were used for the study.

Treatment groups

Cows were randomly allocated into three groups and subjected to three FTAI protocols (Fig. 1) beginning at random stage of estrous cycle (referred as Day 0): -

Group 1: (Ovsynch group)

Sahiwal cows (n=10) were administered standard Ovsynch protocol. Briefly, the cows were administered Buserelin acetate 20 µg, a GnRH analogue (Injection Receptal 5ml, MSD Animal Health, Germany) on day 0. On day 7, an injection of Cloprostenol 500 µg; a PGF2α analogue (Injection Estrumate 2 ml; MSD Animal Health, Germany) was administered intramuscularly followed by second dose of Buserelin acetate 20 µg on day 9 i.e. at 48 hours post PG.

Group 2: (Ovsynch + CIDR group)

Sahiwal cows (n=10) were administered standard Ovsynch protocol as described in Group 1 along with placement of intravaginal EAZI-BREED CIDR Cattle Insert (Zoetis India) containing 1.38 grams progesterone on day 0, which was withdrawn at the time of PGF2α administration on Day 7.

Group 3: Estradiol based group

The Sahiwal cows (n=10) were administered Estradiol benzoate 2 mg (Sigma Aldrich , India) + Cloprostenol 500µg; a PGF2α analogue (Inj. Estrumate 2 ml; MSD Animal Health, Germany) along with placement of intravaginal EAZI-BREED CIDR Cattle Insert (Zoetis India) containing 1.38 grams progesterone on random day of estrous cycle. On day 7, cows were administered Estradiol benzoate 1 mg (Sigma Aldrich, India) + Cloprostenol 500 µg; a PGF2α analogue (Inj. Estrumate 2 ml; MSD Animal Health, Germany) + eCG 400 IU (Inj. Folligon 400 IU, MSD Animal Health, Germany). CIDR was withdrawn at the time of PGF2α administration on Day 7.

All cows were subjected to fixed timed artificial insemination (FTAI) performed on day 10 and 11 with good quality frozen semen of Sahiwal bulls.

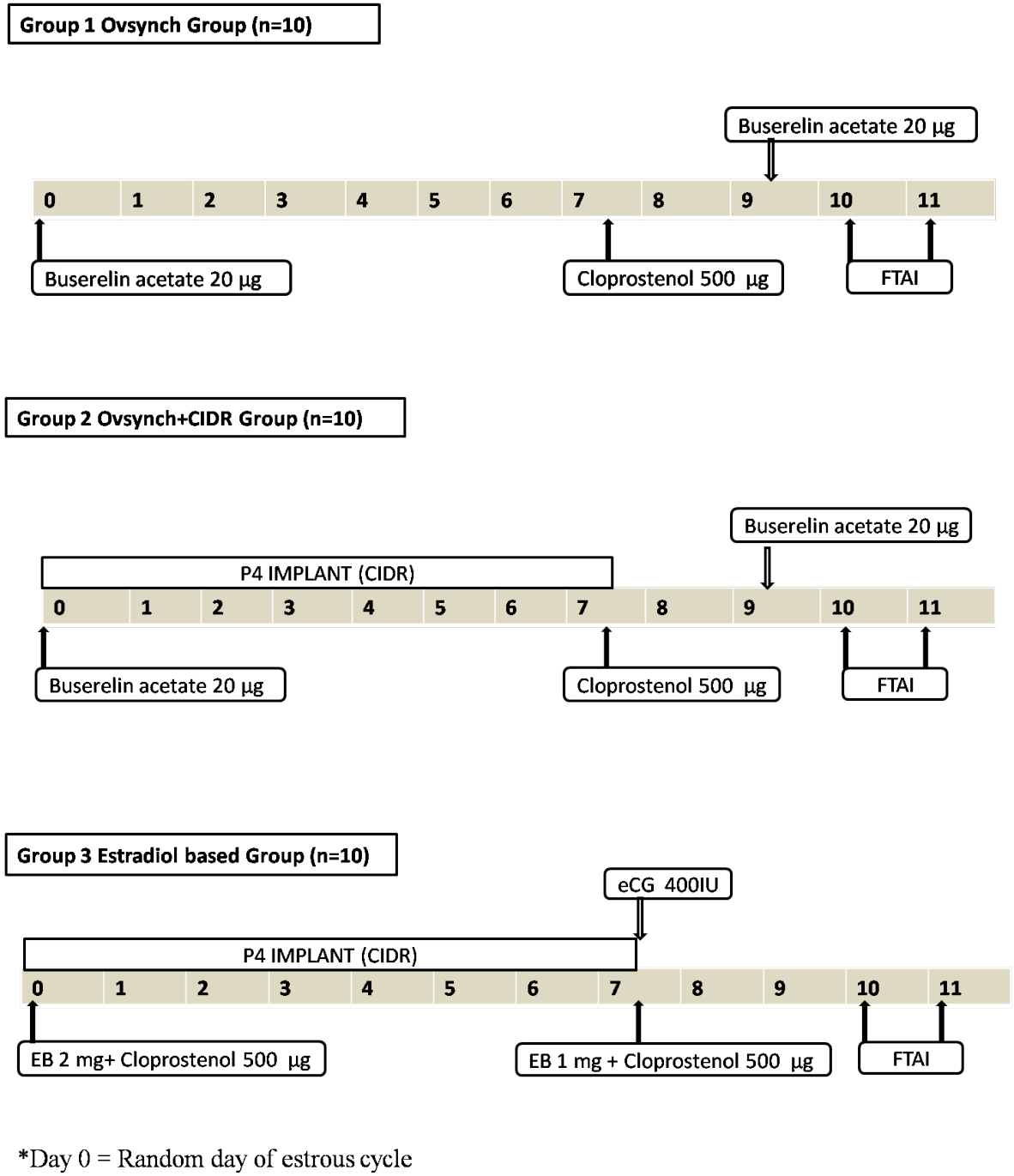

Fig. 1: Schematic representation of Ovsynch, Ovsynch + CIDR and Estradiol based protocols used in the present study.

Estrus detection, ultrasonography and blood sampling

All cows were subjected to visual observations to record externally visible estrus signs like cervico-vaginal mucous (CVM) discharge, mounting activity, bellowing, frequent urination and vulvar swelling (tumefaction) to detect onset and intensity of estrus signs. Trans-rectal ovarian ultrasonography was carried out on days 0, 3, 7, 9, 10, 11, 12 using ultrasound scanner (Exago, ECM, France) equipped with B-mode linear array trans-rectal probe of 7.5 MHz frequency. Location, size and fate of follicles and size of corpus luteum (CL) from the start till the end of experiment were recorded. The ovulation was recorded by the disappearance of dominant follicle that was present on the previous day and subsequent formation of a luteal body at the same spot. Ultrasonographic diagnosis of pregnancy was performed on day 35-40 post insemination.

Blood sampling were collected on day 0, 7 and 10 of the protocol for progesterone estimation. Plasma progesterone was measured using enzyme linked immunoassay PROG-EASIA (Catalogue number: E0018Bo) manufactured by Bioassay technology laboratory, China.

Statistical analysis

The data was analyzed for means and standard errors (SE) for all variables. Student`s t test (two tailed) was applied to compare average size of follicles in different treatment groups in Excel computer programme. The ovulation and pregnancy rates were compared using chi-square test.

RESULTS AND DISCUSSION

Selecting an appropriate fixed timed artificial insemination (FTAI) protocols for Sahiwal cows will help in optimising farm reproduction. The study evaluated the efficacy of Ovsynch, Ovsynch + CIDR and Estradiol based FTAI protocols in inducing estrus, ovulation and subsequent pregnancy in Sahiwal cows.

Induction and synchronization of estrus

Following administration of FTAI protocols, most of the cows of each group expressed one or more behavioural estrus signs as mentioned in Table 1. The CVM discharge and mounting activity were the two prominent signs expressed by 70, 60 and 30 percent cows administered Estradiol based, Ovsynch + CIDR and Ovsynch protocol, respectively, whereas, percentages of cows expressing other signs viz. vulvar swelling, bellowing and frequent urination was comparatively lower than CVM discharge and mounting activity. The three cows in the Ovsynch group and two cows in the Ovsynch + CIDR group did not show any visual estrus sign. However, per rectal and ultrasonic examinations revealed that two cows not expressing any visual signs in Ovsynch + CIDR group were in estrus, whereas, three cows not expressing sign in Ovsynch group; none was found in estrus. The previous studies had shown that expression of estrus could be influenced by many factors like heritability, postpartum period, lactation number, milking status and health. Environmental factors like nutrition, season, housing, herd size, etc. also play an important role in expressing overt signs of estrus (Roelofs *et al*., 2010).

In addition to behavioural estrus signs, per rectal assessment of uterine tonicity was used to identify cows in estrus. On the day of expected estrus (i.e. day 10); 100, 70 and 40 percent cows had intense uterine tone in the Estradiol based, Ovsynch + CIDR and Ovsynch group, respectively (Table 1). Higher percentage of cows showing estrus signs in Estradiol group could be due to exogenous administration of estradiol (day 7); exerting its effect on hypothalamus and genitalia in low progesterone milieu. Three cows each in the Ovsynch + CIDR and Ovsynch group had moderate uterine tone on day of expected estrus. The three cows having low uterine tone in Ovsynch group were not in estrus as ascertained by per rectal and ultrasonographic evaluation. Similar results in terms of uterine tonicity and sexual behaviour had been reported in crossbred cows by Singh and Kharche (1985) whereas;

Table 1: Expression of estrus signs in the Ovsynch, Ovsynch + CIDR and Estradiol based protocols in Sahiwal cows

SN	Parameters	Estradiol based Group Number (%)	Ovsynch + CIDR Group Number (%)	Ovsynch Group Number (%)
1	Behavioural estrus signs	10 (100)	8 (80)	7 (70)
	i. CVM discharge	8 (80)	8 (80)	4 (40)
	ii. Vulvar swelling	4 (40)	4 (40)	2 (20)
	iii. Mounting	7 (70)	6 (60)	3 (30)
	iv. Bellowing	2 (20)	1 (10)	2 (20)
	v. Frequent urination	3 (30)	2 (20)	2 (20)
2	No visible estrus signs	0	2 (20)	3 (30)
3	Uterine tonicity			
	i. No or Low uterine tone	0	0	3 (30)
	ii. Moderate uterine tone	0	3 (30)	3 (30)
	iii. Intense uterine tone	10 (100)	7 (70)	4 (40)

Values did not differ significantly at 5% confidence interval in different treatment groups (P>0.05).

Gunasekaran *et al.* (2008) reported majority of cows having moderate uterine tone during estrus in crossbred cows.

Ovulation and Pregnancy rate

All the ten cows administered Ovsynch + CIDR protocol ovulated, whereas, in the Estradiol based group despite all cows showing estrus signs (CVM discharge and intense uterine tone) only 60 percent ovulated (Table 1). Out of 10 cows inseminated in each group, six, four and three got pregnant in Ovsynch + CIDR, Estradiol based and Ovsynch group, respectively. The ovulation and pregnancy rate achieved in the Ovsynch group was lower than the Ovsynch + CIDR and Estradiol based group. In the Ovsynch group, lower ovulation rate could be due to failure of three cows to respond to the protocol. Hassan *et al* (2017) reported 50 percent ovulation rate following Ovsynch protocol which was comparable to the ovulation rate achieved in the present study.

Khade (2010) observed 100% ovulation in Gir heifers treated with Ovsynch + CIDR protocol, which was in agreement with the findings of the present study. However, Naiko *et al* (2016) reported 50 percent estrus induction and 17 percent conception in Ovsynch + CIDR protocol in Kankrej cattle which was lower than the present study. Higher pregnancy rate in Ovsynch + CIDR group (71.42 and 85.71) compared to PGF2α group (23.8 and 61.8%) had been reported by Rad and Ajam (2008). Higher pregnancy rate with eCG at progestin withdrawal compared to present study was also reported by Huguenine *et al.* (2013) and Bó *et al.* (2001). SaFilho *et al* (2010) observed significant increase in pregnancy rates in eCG treated Nellore lactating anestrus cows (46.2% and 25%; P = 0.07) and Nellore cyclic and acyclic heifers (50.0% vs. 36.8% P = 0.04) than non treated group.

Ovarian status, dominant follicle growth *vis-a-vis* efficacy of FTAI protocols

The influence of ovarian status at the time of beginning of protocol, dominant follicle size and growth rate were evaluated and presented in Table 2.

a) Ovarian status on Day 0

Out of the 5 cows having ≥8mm Follicle + CL at the start of protocol, ovulatory estrus was recorded in 4, whereas; out the remaining 5 cows only 2 ovulated in the Ovsynch group. It was observed that successful ovulatory response to first GnRH was the key for inducing ovulatory estrus following Ovsynch protocol (Vasconcelos *et al*., 1999). Accordingly, it appeared that the presence of DF measuring ≥8mm Follicle + CL favoured subsequent ovulation in the Ovsynch protocol. Previous studies had shown that efficiency

Table 2: Ovarian status at the beginning of protocols (Day 0) and efficacy of Ovsynch, Ovsynch + CIDR and Estradiol based protocols in Sahiwal cows.

SN	Ovarian status Day 0	No. of cows	Size of Largest Follicle (mm)	Ovulated cows (n)	Pregnant cows (n)
1.	**Ovsynch Protocol (n=10)**				
	≥8mm Follicle + CL	5	9.2±0.80	4	1
	≥8mm Follicle + No CL	1	12.0	0	0
	<8mm Follicle + CL	0	-	0	0
	<8mm Follicle + No CL	4	4.75±1.03	2	2
2.	**Ovsynch + CIDR Protocol (n=10)**				
	≥8mm Follicle + CL	7	9.7±0.68	7	4
	≥8mm Follicle + No CL	1	8.0	1	1
	<8mm Follicle + CL	2	5.0±2.00	2	1
	<8mm Follicle + No CL	0	0	0	0
3.	**Estradiol based Protocol (n=10)**				
	≥8mm Follicle + CL	4	8.75±0.47	2	1
	≥8mm Follicle + No CL	2	10.0±2.00	0	0
	<8mm Follicle + CL	2	6.5±0.50	2	1
	<8mm Follicle + No CL	2	3.0±0.00	2	2

Values did not differ significantly at 5% confidence interval in different treatment groups (P>0.05).

of first GnRH in inducing ovulation was depended on the maturation stage of the follicles at the time of treatment (Perry *et al*., 2005, Bello *et al*., 2006). The proposed potential indicators of follicular maturational or ovulatory capacity include follicle size, life span, duration of dominance (Vasconcelos *et al*., 1999, Burns *et al*., 2005), circulating concentrations of progesterone and estradiol (Sartori *et al*., 2004, Souza *et al*., 2005). It was reported that the growing follicles when reached a size of around 9 mm become responsive to luteinising hormone (Ginther *et al*., 1996; Yaniz *et al*., 2004).

In Ovsynch + CIDR group, irrespective of the ovarian status at the beginning of the protocol all the cows ovulated (Table 2). The previous studies had shown that inclusion of an exogenous progesterone implant/device like CIDR/PRID between first GnRH and PGF2α injection not only prevented premature estrus but also improved estrus synchronization and pregnancy rate in cows (Kojima *et al*., 2000; Pursley *et al*., 2001; Martinez *et al*., 2002). Higher pregnancy rates (61.8%) irrespective of season were recorded in Ovsynch protocol when it was used in combination with CIDR to that of Ovsynch alone in Holstein heifers, (Ambrose *et al*., 2008). Stevenson *et al.* (2006) stated that use of CIDR insert during the Ovsynch protocol improved fertility in lactating dairy cows having low serum progesterone. Sahiwal cows showed 2-wave and 3-wave estrous cycle (Dodiyar *et al*., 2022). As stated earlier, presence of DF measuring ≥8mm Follicle + CL at the time of beginning of protocol probably favoured subsequent ovulation in Ovsynch and Ovsynch + CIDR group. However, this trend was not observed in Estradiol based protocol, where, out of six cows having ≥8mm Follicle + with or without CL, only two ovulated following administration of the protocol. This difference could be attributed to the different action of exogenously administered estradiol compared to GnRH on ovarian follicles. The administration of exogenous estradiol resulted in suppression of FSH and LH leading to regression of FSH- and LH-dependent follicles. Once exogenously administered estradiol got metabolized, the FSH surge came and a new follicle wave emerged four day later in cattle (Siqueira *et al*., 2009). The studies had shown that treatments with estradiol and progesterone-releasing devices resulted in synchronous emergence of a new follicular wave (Bo *et al*., 2002) and when a second estradiol treatment was given 24 h after device removal, synchronous ovulation and high pregnancy rates to fixed-time AI were obtained.

b) Follicle Size and Growth Rate in different protocols

The average size of the largest ovarian follicle was smaller on Day 7 in the Estradiol based group than observed in the Ovsynch and the Ovsynch + CIDR group (6.0±0.39 vs 6.9±0.53 & 7.2±0.59mm, respectively, Table 3). Following administration of protocols, higher ovulatory response was recorded in Ovsynch + CIDR group which had higher size of dominant follicle at the time of PG injection than other two groups. The difference in dominant follicle size among different groups could be due to administration of estradiol at the beginning of the protocol in Estradiol based group and GnRH in the Ovsynch and Ovsynch + CIDR group. In the group 1 and 2, the cows having >8 mm follicle probably responded to the first GnRH favourably resulting in emergence of new follicular wave 48 hrs later. Therefore, the day 7 dominant follicle would be from 5 day old follicular wave in the cows responding to first GnRH in the Ovsynch and Ovsynch + CIDR group. Whereas, administration of estradiol resulted in regression of follicles present on ovary resulting in emergence of new follicular wave after 4 days in the Estradiol group. Therefore, the dom-

Table 3: Effect of size of dominant follicle (Day 7) and its growth rate on ovulation and pregnancy rate in Ovsynch, Ovsynch + CIDR and Estradiol based protocols in Sahiwal cows

SN	Parameter	Ovsynch Group (n=10)	Ovsynch + CIDR Group (n=10)	Estradiol Group (n=10)
1	Av. Dominant Follicle size Day 7 (mm)	6.9±0.53	7.2±0.59	6.0±0.39
2	Av. Dominant Follicle size Day 10 (mm)	10.3±0.61	11.4±0.48	10.2±0.42
3	Growth Rate (Day 7 to 10) (mm per day)	1.13±0.07	1.38±0.13	1.42±0.14
4	Preovulatory follicle size (mm)	11.9±0.27	11.6±0.52	12.2±0.20
5	Interval from PG to Ovulation (hours)	86±2	85.2±1.2	106.5±4.36
6	Ovulation rate (%)	60	100	60
7	Pregnancy rate (%)	30	60	40

Values did not differ significantly at 5% confidence interval in different treatment groups (P>0.05).

inant follicle observed on day 7 in the Estradiol group would be from a 3 day old follicular wave and would be of smaller size compared to Ovsynch and Ovsynch + CIDR group. In literature, intravaginal progesterone implant had been used for 8 days along with estradiol administration at the time of implant removal or 24 hours later (Ayres *et al*., 2008; SaFilho *et al*., 2011), whereas, in the present study the progesterone implant was removed on day 7 and estradiol was also administered at the time of implant removal this could partially explain the reasons for smaller size of dominant follicles on day 7 and 10 and lower pregnancy rate in estradiol based group compared to other groups. In a similar estradiol based protocol, Ayres et al. (2008) observed administration of estradiol benzoate at the time of progesterone implant removal resulted in lower pregnancy rate compared to its administration at 24 hour after implant removal when FTAI was performed 54 hour after implant removal.

In the present study, the growth rate of day 7 dominant follicle was higher in Estradiol based and Ovsynch + CIDR group compared to Ovsynch group (1.42±0.14, 1.38±0.13 and 1.13±0.07 mm, respectively, Table 3). Corresponding to growth rate and size of day 7 dominant follicle, the size of dominant follicle on day 10 was higher in the Ovsynch + CIDR group followed by Ovsynch and Estradiol based group (11.4 ± 0.48, 10.3 ± 0.61 and 10.2 ± 0.42 mm, respectively). Day 7 dominant follicle in Estradiol based group had higher growth rate than other two groups. This could be due to administration of eCG@400 IU in Estradiol based group. Higher ovulatory response was observed in the Ovsynch + CIDR group having larger dominant follicles on day 10 than the other two groups. It was observed that full ovulatory capacity was acquired when the DF reached 10 mm size in *Bos indicus* and 12 mm size in *Bos taurus* cattle (Sartori *et al.,* 2001; Gimenes *et al.,* 2008). However, anestrous beef cows treated with eCG had low ovulatory response despite large follicle diameters (Tortorella *et al.,* 2013).

c) Follicle Size and Growth Rate in Ovulated *versus* Anovulated cows

Sahiwal cows having ovulatory estrus had higher average size of the DF on day 7 compared to anovulatory cows in Ovsynch group (8.0±0.37 vs 5.3±0.48 mm; p<0.05, Table 4). All cows of the Ovsynch + CIDR group ovulated and the average size of day 7 dominant follicle was 7.2±0.59 mm. In the Estradiol based group, the average size of day 7 dominant follicle was comparatively smaller in cows undergoing ovulation than not ovulating but the difference was non-significant (5.6±0.42 vs 6.6±0.69 mm), however, the average growth rate of day 7 dominant follicle was significantly higher in ovulatory cows than non-ovulatory cows (1.64±0.14 vs 1.08±0.20 mm, p<0.05). Due to higher growth rate of day 7 dominant follicle of ovulatory cows, the average size of day 10 dominant follicle reached comparatively higher size in ovulatory cows than anovulatory cows (10.5±0.56 vs 9.9±0.66 mm, respectively). Pitaluga *et al.* (2013) reported that the diameter of largest follicle at progesterone device removal was 7.8±0.31 and at the time of insemination it was 11.2±0.52 mm, respectively in eCG + ECP treated cows and this resulted in 90 percent ovulation rate.

In the Ovsynch group, average size of DF on day 10 was also higher in cows having ovulatory estrus as compared to anovulatory estrus (11.6±0.20 vs 8.4±0.80 mm, p<0.05). In the Ovsynch + CIDR group, average diameter of Day 10 dominant follicle was 11.4±0.48 mm and all cows expressed ovulatory estrus. Although, average size of day 7 dominant follicle was smaller in ovulatory cows of the estradiol based group, however, the average preovulatory follicle size was higher than the Ovsynch and Ovsynch + CIDR groups (12.2±0.20 vs 11.9±0.27 and 11.6±0.52 mm, respectively, Table 3). This difference could be due to higher growth rate and higher interval from PG to ovulation in the Estradiol group than other two groups.

d) Plasma Progesterone levels between ovulated and anovulated

Plasma progesterone concentration was higher on Day 0 and 7 in the ovulated cows compared to anovulated in the Ovsynch and Estradiol based group (Table 5). On Day 10 of the protocol (day of expected estrus), the progesterone concentration was higher in anovulatory cows as compared to cows undergoing ovulation in the Ovsynch and Estradiol based group (1.37±0.27 vs 1.04±0.15 and 1.52±0.17 vs 1.15±0.16 ng/ml, respectively) which indicated poor response of non-ovulatory cows to estrus synchronization protocols. Jyothi (2011) observed that the mean serum progesterone concentration was lowest on the day of AI and ranged from 0.77 to 1.17 ng/ml in Ovsynch protocol, 0.04 to 1.46 ng/ml in Ovsynch + CIDR and 0.62 to 1.69 ng/ml CIDR+PG protocol.

CONCLUSIONS

The results of present study suggested that Ovsynch + CIDR protocol was more effective on random day of estrous cycle in inducing ovulatory estrus and subsequent fertility compared to the Ovsynch and Estradiol based protocol. In addition, efficacy of the fixed timed insemination

Table 4: Dominant follicle size and its growth rate in cows having Ovulatory vs. Anovulatory estrus following Ovsynch, Ovsynch + CIDR and Estradiol based protocols

		Ovsynch Protocol		Ovsynch + CIDR Protocol		Estradiol based Protocol	
SN	Parameter	Ovulated cows	Anovulated cows	Ovulated cows	Anovulated cows	Ovulated cows	Anovulated cows
1	No. of cows	6	4	10	Nil	6	4
2	Av. Dominant Follicle size Day 7 (mm)	8.0± 0.37	5.3± 0.48	7.2± 0.59	Nil	5.6± 0.42	6.6± 0.69
3	Av. Dominant Follicle size Day 10 (mm)	11.6± 0.20a	8.4± 0.80b	11.4± 0.48	Nil	10.5±0.56	9.9±0.66
4	Growth Rate from Day 7 to 10 (mm per day)	1.19± 0.07	1.04± 0.14	1.38± 0.13	Nil	1.64±0.14[a]	1.08±0.20[b]
5	Pregnancy rate (%)	50		60		66.7	

Values with different superscript within the same row differed significantly at 5% confidence interval ($p<0.05$).

Table 5: Progesterone levels in Ovulated vs. Anovulated cows following Estradiol based, Ovsynch + CIDR and Ovsynch protocols.

			Progesterone (ng/ml)		
S. No.	Parameter		Day 0	Day 7	Day 10
1	Estradiol Based Protocol	Ovulated cows (n=6)	4.33±0.72	6.6±0.46	1.15±0.16
		Anovulated cows (n=4)	3.52±0.55	5.2±0.48	1.52±0.17
2	Ovsynch Protocol	Ovulated cows (n=6)	4.43±0.63	4.38±0.32	1.04±0.15
		Anovulated cows (n=4)	3.07±0.90	2.92±0.75	1.37±0.27
3	Ovsynch + CIDR Protocol	Ovulated cows (n=10)	4.89±0.35	6.08±0.39	1.11±0.16
		Anovulated cows (n=nil)	-	-	-

Values did not differ significantly at 5% confidence interval in different treatment groups ($P>0.05$).

protocols to induce ovulatory estrus was affected by the ovarian status at the time of beginning of protocol, follicle growth rate and size of dominant follicle on day 7 and day 10.

CONFLICT OF INTEREST

None.

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