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RESEARCH ARTICLE

Determination of Ovsynch and CIDR Efficiency for Estrus Synchronization by Conception Rate in Nili Ravi Buffaloes during Low and Peak Breeding Seasons

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A B S T R A C T

Efficiency of Ovsynch and CIDR protocols for estrus synchronization was determined by conception rate in Nili Ravi buffaloes in low and peak breeding seasons under farm condition. The protocol with better conception rate (%) was then tested under field condition in the respective season. Ovsynch protocol comprised of administration of GnRH analogue (Lecirelin, d 0) PGF2α analogue (Cloprostenol, d 7) and Lecirelin (d 9). Animals were inseminated at 12 and 24 hours after last Lecirelin injection using chilled semen. In CIDR protocol, the device (containing 1.38 g progesterone) was used for 8 days and Cloprostenol was administered on day 7. Animals were inseminated twice at 48 hours and 60 hours after CIDR removal using chilled semen. Conception rate was higher (P=0.07) in CIDR group (30.0%) than that in Ovsynch group (0.0%) under farm condition in low breeding season, however this difference was statistically non significant. Conception rate did not differ (P=0.47) between Ovsynch (70.0%) and CIDR (54.5%) groups under farm condition in peak breeding season. In field trial, a conception rate of 36.8% was achieved with CIDR in low breeding season and a conception rate of 54.2% was achieved with Ovsynch in peak breeding season (P=0.26). In conclusion, CIDR as compared with Ovsynch protocol seemed to be more effective for estrus synchronization during low breeding season in Nili Ravi buffalo. Ovsynch and CIDR were equally effective for estrus synchronization during peak breeding season.

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INTRODUCTION

Productivity of buffalo is affected by prolonged calving interval, silent estrus and seasonality of breeding (El-Wishy, 2007). To maximize the productive life of a buffalo, it should be bred during 80 to 90 days after parturition (Abdalla, 2003). Applying estrus synchronization protocols along with timed artificial insemination (TAI) is a potential line of action for decreasing the service period and overcome seasonality of breeding in this important dairy animal (Ghuman *et al.,* 2012). Prostaglandin F2α alone and in combination with GnRH or progestagens has been used for this purpose (Luthra *et al.,* 1994; Brito *et al.,* 2002) with varying results.

In Murrah buffaloes pregnancy rates of 33% after TAI and 31% for those inseminated after spontaneous estrus detection have been reported (Paul and Prakash, 2005). In Egyptian buffaloes a conception rates of 18% in cyclic and 0% in non-cyclic animals was reported during summer (Karen and Darwish, 2010). In Nili Ravi buffaloes, Ovsynch protocol resulted in a significantly better response in terms of luteinizing hormone (LH) peak and progesterone (P4) rise in blood plasma during peak breeding season compared to that in low breeding season (Jabeen *et al.,* 2013). Implantation of progesterone in the form of a controlled internal drug release (CIDR) intra-vaginally for 10 to 14 days induced resumption of estrus among 83% of anestrus buffaloes within 12 to 120 hours after implant removal and of those detected estrus, 80% of buffaloes conceived following natural service (Singh, 2003). Naseer *et al.* (2011) noted that CIDR treated Nili Ravi buffaloes gave a conception rate of 37 and 30% after TAI during low and peak breeding seasons respectively. Based on our previous findings that Ovsynch was not as effective in low breeding season as that in peak breeding season (Jabeen *et al.,* 2013) and that of Naseer *et al.* (2011) that CIDR was almost equally effective over these two periods, conception rate in Nili Ravi buffaloes was compared using CIDR and the Ovsynch protocols.

MATERIALS AND METHODS

Efficiency of Ovsynch and CIDR protocols for estrus synchronization was determined by conception rate in Nili Ravi buffaloes under farm condition in low and peak breeding seasons. The protocol with better conception rate (%) was then further tested under field condition in the respective season.

Experiment Design: Two estrus synchronization protocols **(***vis-à-vis* Ovsynch and CIDR) were compared during two seasons (low and peak breeding season) to induce fertile estrus in buffalo under farm condition.

Estrus Synchronization Protocols

Ovsynch: Synthetic analogue of GnRH (Lecirelin 50 µg i.m., Dalmarelin; Fatro, Italy) was administered on day 0. Seven days later, synthetic analogue of PGF2α (Cloprostenol 150 µg i.m., Dalmazin, Fatro) was administered and on day 9 buffaloes received second injection of the Lecirelin. Animals were inseminated twice (at 12 and 24 hours) after second GnRH injection with freshly collected buffalo bull semen diluted in skim milk and stored at 4°C. Each insemination dose of 0.5 ml diluted semen contained 10 million progressively motile spermatozoa.

Controlled Internal Drug Release (CIDR): On day 0, EAZI BREED CIDR (DEC Int. NZ Ltd, Hamilton, New Zealand) containing 1.38 g progesterone in molded silicone was inserted in vagina of each buffalo. The cloprostenol was administered on day 7 and CIDR was removed on day 8. Animals were inseminated twice (at 48 hours and 60 hours after CIDR removal) using chilled semen as described for the Ovsynch protocol. Animals in the control group were not given any heat inducing treatment.

Animals: The farm trial during low and peak breeding season was conducted at National Agricultural Research Center, Islamabad, Pakistan (33° 42' N, 73º 10' E). Non pregnant and healthy buffaloes (with body condition score 3 plus) in their first to fourth lactation were used for the trial. Last calving had taken place at least more than 60 days ago. These buffaloes were separated from rest of the herd (of 100 buffaloes) and kept in a well facilitated shed where they were either treated with Ovsynch or CIDR or given no heat inducing treatment (Control). Animals were offered seasonal green fodder (40 kg/buffalo/day). A concentrate comprising of cotton seed cake (2 kg) and wheat bran (1 kg) mixed with 2 kg wheat straw was offered per animal/day. Animals were sent for grazing natural grasses for two h (8 am to 10 am) every day. Clean water was available *ad lib* for drinking. Animals were also given a 30 min shower twice daily at 10:30 am and 02:30 pm.

Pregnancy Test: Pregnancy test in buffaloes was performed via palpation per rectum 45 days after insemination.

Conception rate under field conditions with CIDR and Ovsynch in the respective season: Better evolved estrus synchronization protocols under farm condition in low and peak breeding seasons were further tested in the field trial conducted in Layyah (30° 58' 18" N and 70° 58' 09" E), Punjab, Pakistan. The number of buffaloes with each farmer in the field was 5-10. Animals were in good body condition and in their 2nd to 5th lactation having calved 3 to 14 months earlier. During day time, animals were kept tied on the manger under tree shade (summer) or in the sun (winter). Animals were kept in door during winter only at night hours. There was a ‘cut and carry’ system of feeding green fodder. Egyptian clover and lucerne were the major green fodders offered during winter, and millet and sorghum were fed during summer. Each buffalo was fed 2-3 kg cotton seed cake per day. Buffaloes included in field trial were spread over a radius of 10 km during low (n=19) and peak breeding season (n=24). Animals were treated with CIDR or Ovsynch protocol and then inseminated as described above. However, in Ovsynch protocol, animals were inseminated only once (at 16 hours) after 2nd GnRH injection (Baruselli *et al.,* 2003) under field condition. Pregnancy test in buffaloes was performed via palpation per rectum 45 days after insemination.

Statistical analysis: Conception rate among the treatment groups was compared by Chi square method. Data are presented as mean±SEM. Probability of $P<0.05$ was considered to be statistically significant (McKillup, 2012).

RESULTS

A higher conception rate (%) was achieved in CIDR group as compared to that in Ovsynch group (30.0 *vs.* 0.0; P=0.07) under farm condition in low breeding season, however this difference was statistically non-significant (Table 1). No control animal showed heat during low breeding season. During peak breeding season, Ovsynch resulted in a 70.0% conception rate as compared to a 54.5% conception rate with CIDR under farm condition (P=0.47, Table 1). Three animals out of four were served and become pregnant over a period of 21 days in control group during peak breeding season but their service-days were scattered (not synchronous). A conception rate of 36.8% was achieved with CIDR and 54.2% with Ovsynch under field condition in low and peak breeding seasons respectively (P=0.26) (Table 2).

DISCUSSION

CIDR treatment gave better (30.0%) conception rate than Ovsynch treatment (0.0%) in buffaloes under farm condition during low breeding season although the difference was non significant. Under field condition,

Table 1: Conception rate (%) in Nili Ravi buffaloes after estrus synchronization with Ovsynch or CIDR under farm condition in low and peak breeding seasons.

Breeding season	Estrus synchronization Protocol	Number of buffaloes	Conception rate at farm (%)
Low	Ovsynch	9	0.0
	CIDR	10	30.0
	Control*	4	0.0
Peak	Ovsynch	10	70.0
	CIDR	11	54.5
	Control*	4	75.0

*Animals in the control group were not given any heat inducing treatment. Conception rate did not differ between Ovsynch and CIDR groups in low breeding season (P=0.07). Conception rate did not differ between Ovsynch and CIDR groups in peak breeding season (P=0.47).

Table 2: Conception rate (%) under field condition in Nili Ravi buffaloes synchronized with CIDR during low breeding season or Ovsynch during peak breeding season.

Breeding season	Synchronization protocol	Number of buffaloes	Conception rate at field (%)
Low	CIDR	19	36.8
Peak	Ovsynch	24	54.2

Conception rate did not differ between two seasons (P=0.26).

CIDR gave a conception rate of 36.8% in the low breeding season. Use of progestogens for estrus synchronization during low breeding season has been shown to improve pregnancy rate in Nili Ravi (Naseer *et al.,* 2011; 2013), Mediterranean (Barile, 2005) and Brazilian buffaloes (Carvalho *et al.,* 2013). Similarly, very low conception rate (6.9 and 4.7%) has been reported in buffaloes synchronized by the Ovsynch in low breeding season (Baruselli *et al.,* 2003; De Rensis *et al.,* 2005). This indicates that progesterone based estrus synchronization protocols can stimulate hypothalamo-pituitary-ovarian axis of buffalo in low breeding season both under farm and field conditions. So they may be used to overcome seasonality of breeding in buffalo. Moreover, the presence of a dominant follicle (>9 mm) at the beginning of the Ovsynch protocol is a determining factor for a successful synchronization of ovulation and optimum conception rates (De Rensis *et al.,* 2005; Warriach and Ahmad, 2007), and this factor is missing in low breeding season when buffaloes are usually acyclic.

In the present study, conception rate in buffalo after Ovsynch and CIDR protocols (70.0 and 54.5%, respectively) under farm condition in peak breeding season were similar. However due to a higher trend in conception rate with Ovsynch, the same was further verified in field condition by a 54.8% conception rate in peak breeding season. Similar optimum conception rate has been reported with Ovsynch protocol in Mediterranean (48.8%: Berber *et al.,* 2002 and 43.7%: De Rensis *et al.,* 2005), Brazilian (56.5%, Baruselli *et al.,* 2003) and Egyptian buffalo (60%: Ali and Fahmy, 2007). It has been observed that the improved response to Ovsynch in peak breeding season is due to the presence of large ovarian follicles in buffalo (De Rensis *et al.,* 2005). The present study confirms our previous findings that Ovsynch protocol is more effective during peak than in low breeding season (Jabeen *et al.,* 2013).

In conclusion, CIDR as compared with Ovsynch protocol seemed to be more effective for estrus synchronization during low breeding season in Nili Ravi buffalo. Ovsynch and CIDR were equally effective for estrus synchronization during peak breeding season.

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