



Effect of Different Synchronisation Protocols on Embryo Transfer in Sahiwal Cows

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

The present study was conducted to compare CIDR + Ovsynch, Double PG protocol's synchronisation effect on recipient cows after IVF-ET (Invitro Fertilization - Embryo Transfer). Since there is a positive effect of synchronisation on conception rate, protocol with higher synchronisation efficiency and conception rate can be determined under this study. Invitro Fertilization technology increases the genetic gain by reducing the generation interval. It helps to utilize a female cattle genetic potential fully and produce greater number of embryos in short duration with utilization of less semen. In vitro Production of embryos can be used to produce genetically proven animals with a good performance for improving dairy production. 85 animals were selected randomly and assigned into 3 groups. Group 1 (CIDR+ Ovsynch), Group 2 (Double PG) and Group 3 (control). Embryo transfers were done into these recipient cows by using nonsurgical method after day 7 of estrus by depositing the embryos in cranial or middle third of uterine horn on ipsilateral side of ovary having CL. The estrus response rates, overall conception rate and cervical transfer scores were found to be 60.60%, 18.75% and 66.67 % in CIDR+ Ovsynch protocol, 60.00%, 15.78% and 16.67 % in Double PG protocol, 58.33%, 0% and 16.67 % in natural estrus in recipient cows. The mean BCS and mean CL diameter was 3.07 ± 0.13 and 21.56 ± 1.13 mm, 2.53 ± 0.07 and 14.90 ± 0.59 in pregnant and non pregnant recipients respectively. Conception rate was found higher in animals having vacuolated CL (18.18%) when compared to animals having Compact CL (13.79%) and those receiving Expanded blastocyst (23.80%) than blastocyst (5.26%) when the transport time was within 3hrs (19.04%) when compared to 5-7hrs (10.52%). Overall, higher synchronisation, eligible animals for transfer and conception rates were observed with timed ET in CIDR + Ovsynch protocol than in Double PG and natural estrus animals.

Keywords: *Estrus synchronisation; CIDR + Ovsynch; double PG; embryo transfer; In vitro fertilization - embryo transfer.*

1. INTRODUCTION

Embryo transfer technology (ETT) is important tool for genetic improvement. It has been widely used for calf production in both dairy and beef industry around the world. It is the most widely applied reproductive technique to rapidly multiply animals with desirable genetics (Bo and Mapletoft, 2013). It can be used to improve the accuracy of selection, increase the intensity of superior sire and dam combinations, allow genomic selection as early as the embryo stage. Despite the increased prevalence and usage of Embryo Transfer (ET) programs over the last five decades, Invitro fertilization (IVF) and ET shown a constant conception rates at a range of 30-60% (Erdem et al., 2020). Therefore, more research is required to evaluate factors influencing the success of embryo transfer in terms of recipient utilization, quality of embryo produced and pregnancy outcome.

2. MATERIALS AND METHODS

A total of 85 animals were selected and randomly assigned into three groups irrespective of their stage of estrus cycle. Group 1 (CIDR+ Ovsynch) (n = 33) recipients of this group were estrus synchronized by CIDR insertion intravaginal and Inj. GnRH (Buserelin acetate) @ 10µg given IM (Intramuscular) on day 0, removal of CIDR and

Inj. Cloprostenol (Estrumate) @ 500 µg given IM on day 7, second dose of GnRH on day 9 given IM. Group 2 (Double PG) (n = 40) Recipients of this group are synchronised by using two injections of Inj. Cloprostenol (Estrumate) @ 500 µg, 11 days apart (on day 0 and day 11) given IM. It is followed by 10 µg of Inj. GnRH (Buserelin acetate) given IM on day 13. Group 3 (control) (n = 12) cows in natural estrus were included in control group.

Following estrus synchronisation protocols, estrus response rates were recorded. Embryo transfers were done into these recipient cows by using nonsurgical method after day 7 of estrus by depositing the embryos in cranial or middle third of uterine horn on ipsilateral side of ovary having CL. After 45 days of embryo transfer, pregnancy was confirmed by rectal palpation and transrectal ultrasonography.

2.1 Statistical Analysis

The Chisquare test was used to compare the estrus response rate, impact of parity, cervix transfer score, embryo transfer location, embryo transport time, stage and grade of embryo on conception rate in various groups. One way ANOVA was used to ascertain the impact of BCS and CL diameter on conception.

3. RESULTS AND DISCUSSION

The similar estrus response rates were reported with CIDR synchronization protocol by Jyothi et al. (2012) and Raut et al. (2020). Whereas Buhecha et al. (2015), Dhama et al. (2015) and Devi et al. (2021) reported higher estrus response rate than the present CIDR+Ovysynch group.

The similar estrus response rates were reported with Double PG synchronization protocol by Kawate et al. (2007) and Mehar (2022). Whereas Hirole et al. (2018), Kebede et al. (2024), Siregar et al. (2024) reported higher estrus response rate.

Similar Conception rates were observed by Steichen and Larson (2019) and Salman et al. (2023) with CIDR based protocol. Contrary to this, higher pregnancy rate was recorded by Bonacker et al. (2020) and Crowe et al. (2024) after recipients are synchronized with CIDR based protocol.

Similar results were obtained by, Benyei et al. (2006), Jyothi et al. (2012) and Shakkarpude et al. (2013) after Double PG protocol. Whereas higher conception rate reported by Kawate et al. (2007), Kose et al. (2012) and Manokaran et al. (2023).

Table 1. Effect of synchronisation protocols on conception rate

Synchronisation protocol	Synchronisation efficacy	Conception rate
CIDR+ Ovynch	60.60%	18.75 %
Double PG	60.00%	15.78%
Control	58.33%	0 %

Table 2. Showing mean CL diameter and BCS among pregnant and non pregnant animals

Measurements	Pregnant animals	Non-Pregnant animals
Mean CL diameter (mm)	21.56±1.13	14.90±0.59
Mean BCS	3.07±0.13	2.53±0.07



Fig. 1. Images showing compact CL and Vacuolated CL



Fig. 2. Transrectal ultrasonographic image of foetus with gestational sac on day 52 of gestation

Table 3. Effect of parity, CL quality, cervical transfer score, deposition site in uterus, embryo quality and grade and Transportation time on conception rate

S.no	Parameter	Comparative studies	Conception rate	P value
1.	Parity	Heifer	7.69 %	0.43
		Cows	18.51 %	
2.	CL Quality	Compact CL	13.79 %	0.76
		Vacuolated CL	18.18 %	
3.	Cervical transfer score	Easy	20.00 %	0.44
		Moderate	11.11 %	
		Hard	09.09%	
4.	Deposition site in Uterus	Cranial third	21.77%	0.22
		Middle third	5.88%	
5.	Embryo quality and grade	Expanded Blastocyst (7-1)	23.80%	0.15
		Blastocyst (6-1)	5.26 %	
6.	Embryo transport time	3hrs	19.04 %	0.42
		5-7hrs	10.52 %	

Reason for lower conception rate Double PG protocol might be due to transfer of embryos to repeat-breeder female, inadequate maternal environment, Due to lack of experience in embryo transfer while deposition of the embryo in the reproductive tract The CIDR+ Ovsynch group's conception rate is more with the addition of exogenous progesterone.

4. CONCLUSION

The smaller size of the treatment group, nutritional status, heat stress that causes damage to early embryo, recipient animals managemental conditions and early embryonic death due to transfer-related stress could all be contributing factors to the low conception rates. From the present study it was concluded that CIDR+Ovsynch protocol was efficient synchronisation protocol to improve the estrus synchronisation rate and conception rates in embryo transfer programme. Further, there is a need to standardise the estrus synchronisation protocols in recipient animals and consider the factors such as Estrus response rate, BCS, CL diameter, CL quality, parity of recipient animals. Cervical transfer score, embryo stage and quality, embryo deposition site, embryo transfer time and embryo transportation time to improve the overall portion of suitable recipients for embryo transfer and conception rate.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image

generators have been used during writing or editing of this manuscript.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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