FACTORS AFFECTING SUCCESSFUL FROZEN EMBRYO TRANSFER IN CATTLE UNDER FARM CONDITION

G. K. MISHRA¹, NARINDER SINGH², M. U. SIDDIQUI³, M. K. SHUKLA and S. K. SAXENA

Animal Breeding Centre, Salon - 229 127
A unit of Animal Breeding Research Organization (India)

ABSTRACT

Embryo transfer technology (ETT) helps in the dissemination of best germplasm of both male and female whereas artificial insemination (AI) disseminates only male genotype. The aim of present study is to evaluate the variables affecting pregnancy rate following non-surgical embryo transfer in crossbred and indigenous cattle under farm condition. A total of 48 frozen embryos were transferred into as many recipients which resulted in overall 20.83 per cent pregnancy rate. The study revealed that transfer of embryos into anterior one third of uterine horn resulted in significantly (P < 0.05) higher pregnancy rate (34.62%) compared to the posterior two third of uterine horn (4.55%). The results indicated that the site of embryo deposition in uterine horn has significant influence on the pregnancy rate in cattle.

Key words: Frozen embryo transfer, Pregnancy, Cattle

INTRODUCTION

Embryo Transfer Technology (ETT) is considered to be the most important tool in the hand of reproductive biotechnologist to bring genetic improvement of livestock at a faster rate by utilizing genetic contribution from both male and female simultaneously (Sidiqui et al., 2008).

Pregnancy rate among recipient cows, a key index in measuring successful embryo transfer program depends upon complex series of interrelationships between the embryo, its uterine environment and corpus luteum (Sreenan and Diskin, 1987). Various factors that influences the conception rate in cattle include stage of embryo development (Hasler *et al.*, 1987), embryo quality (Linder and Wright, 1983), synchrony between donor and recipient's estrus (Hasler *et al.*, 1987), skill of operator and ease of transfer (Schneider *et al.*, 1980), transfer location (Hasler *et al.*, 1980), maternal endocrine

profile (Wilmet et al., 1985) and uterine environment (Walton and Stubbings, 1986). Keeping this in view, the present study was conducted to investigate the effect of different factors in establishing successful pregnancies through non-surgical transfer of frozen bovine embryos under farm condition.

MATERIALS AND METHODS

The study was conducted at Animal Breeding Centre, Salon in Rae Bareli district of Uttar Pradesh during August 2006 to April 2008. Embryos were collected by non-surgical method (Newcomb *et al*, 1978) from Holstein Friesian cattle maintained at collaborative project of National Dairy Development Board (NDDB) and Anand Agricultural University (AAU), Anand, Gujarat. The good quality embryos frozen either in ethylene glycol or glycerol freezing media were procured from NDDB-AAU collaborative ET project and used in the study.

Nine crossbred and 18 indigenous healthy and disease free cattle without any genital abnormalities were selected as recipients (Siddiqui *et al.*, 2008). Estrus of selected recipients was either natural or induced (two PGF₂₀ regime - Lutalyse, 5 ml,

^{1* =} Corresponding author, Email: drkodu@rediffmail.com

^{2 =} Assistant Scientist, Deptt. of Animal Breeding & Genetics, GADVASU, Ludhiana

^{3 =} Deputy General Manager (AB), NDDB Anand

intramuscular at an interval of 11 days). Estrus detection of recipients was carried out twice daily (morning – evening) by visual observation and then confirmed by rectal palpation.

On day 7 of estrus, recipients having functional corpus luteum (CL) were considered for non-surgical embryo transfer. The corpus lutea were classified as large (> 10 mm), medium (6-10 mm) and small (< 5 mm or embedded).

Embryos were non-surgically transferred into indigenous (Sahiwal, Red Sindhi, Hariana and Rathi) and crossbred (Holstein Friesian x Sahiwal and Jersey x Red Sindhi) breeds of cattle that were within ± 24 hrs of estrus synchrony with the donors. The recipients were examined for pregnancy 60 days after transfer in heifers and 90 days after transfer in cows, per rectally.

Effect of different factors such as stage and grade of embryos, type of freezing media, estrus synchrony, CL grade, CL side, transfer side and site, transfer grade, recipient's parity and type of recipients (indigenous and crossbred cattle) used, was analyzed by χ^2 test (Snedecor and Cochran, 1994).

RESULTS AND DISCUSSION

In the present study, overall pregnancy rate with frozen embryos was found to be 20.83%.

In our study, higher pregnancy rate was observed with compact morula (CM) stage of embryos (23.08%) in comparison to blastocyst (16.67%) and expanded blastocyst (0.00%) stage of embryos. But it was not found to be statistically significant. The results coincide with the report of Linder and Wright (1983). Whereas, Donaldson (1985) reported higher pregnancy rates with blastocyst stage in comparison to morula.

In the present study, there was no significant difference in pregnancy rates between various grades of embryos. Higher pregnancy rate was observed with Grade-II (25%) than Grade-I (17.86%) embryos. Sometimes, poor quality embryos may produce pregnancy while morphologically good quality embryos failed to do so, suggesting the involvement of various other factors in establishing pregnancy.

Transfer of embryos to the recipients after natural and induced heat resulted in almost similar pregnancy rate i.e. 20.49 and 21.53 per cent, respectively.

No significant difference between the types of freezing media, with respect to pregnancy rate was observed. Present study revealed higher pregnancy rate (28.00%) with embryos frozen in ethylene glycol freezing media than those frozen in glycerol freezing media (13.04%).

The pregnancy rates in synchronized estrus and asynchrony within \pm 24 hrs were 21.43 and 16.67 per cent, respectively. The pregnancy rates were not compromised when recipients were in estrus as much as 24 hrs before or after the donor estrus (16.67%). The results of the present study are in accordance with the findings of Sreenan *et al.* (1975).

No significant difference was observed with respect to CL grade and CL side in getting successful pregnancy.

Transfer of embryos into anterior one third of uterine horn resulted in significantly (P < 0.05) higher pregnancy rate (34.62%) compared to the posterior two third of horn (4.55%) which was in agreement with the earlier reports (New Comb and Rowson, 1980). They suggested that the poor survival of embryos might be due inadequate signal to the ovary as a result of suboptimal location (New Comb and Rowson, 1980).

Higher pregnancy rate of 27.27 per cent resulted when embryos were transferred easily and quickly with least manipulation (transfer grade - easy) in comparison to 6.67% where the operator struggled a lot during transfer (transfer grade - difficult). However, they are not significantly different in establishing pregnancy. This finding was in agreement with the finding of Thibier and Nibart (1992). Probably the skill and experience of the operator determine not only the site of embryo deposition within a selected horn but also the degree of trauma inflicted during transfer (Sreenan and Diskin, 1987).

A non significant difference in pregnancy rates between heifers (22.27%) and cows (15.38%) was

observed in this study, similar finding was also reported by Wright (1981).

The pregnancy rates were found to be almost similar between crossbred (20.00%) and indigenous (21.21%) cattle.

, The results indicate that eight calves were born from a total of 10 successful pregnancies and sex ratio was observed to be 50:50.

From these results, it may be concluded that among the various factors, site of embryo deposition was found to be the most important in establishing successful pregnancy and the anterior one third of uterine horn was appeared to be appropriate site for embryo deposition.

ACKNOWLEDGEMENT

Authors are thankful to the General Manager, Animal Breeding Centre Salon, Rae Bareli for providing facilities, necessary support and valuable suggestions for carrying out the study.

REFERENCES

- Christie, W. B., New Comb, R. and Rowson, L. E. A (1980). Non-surgical transfer of bovine eggs: Investigation of some factors affecting embryo survival. Vet. Rec., 106: 190-193.
- Donaldson, L. E. (1985). Matching of embryo stages and grades with recipient estrous synchrony in bovine embryo transfer. Vet Rec., **117**: 489 491.
- Hasler, J. F., Mc Cauley, A. D., Lathrop, W. F. and Foote, R. H. (1987). Effect of donor-embryo-recipient interactions on pregnancy rate in a large scale bovine embryo transfer program. Theriogenology, **27**: 139-168.
- Linder, G. M. and Wright, R. W. Jr. (1983). Bovine embryo morphology and evaluation. Theriogenology, **20**: 407-416.

- New Comb, R., Christie, W. B. and Rowson, L. E. A. (1978). Nonsurgical recovery of bovine embryos. Vet. Rec., **102**: 414 – 417.
- New Comb, R. and Rowson, L. E. A. (1980). Investigation of physiological factors affecting non-surgical transfer. Theriogenology, **13**: 41 49.
- Schneider, H. J. Jr., Castleberry, R. S. and Griffin, I. L. (1980). Commercial aspects of bovine embryo transfer. Theriogenology, **13**: 73-85.
- Siddiqui, M. U., Panchal, M. T. and Kavani, F. S. (2008). Superovulation response, embryo recovery and quality in Sahiwal cows and Heifers. Indian J. Anim. Reprd., **29**(2): 195-198.
- Snedecor, G. W. and Cochran, W. G. (1994). Statistical Methods. 8th Edn. Iowa State University Press, Ames, Iowa, USA.
- Sreenan, J. M., Beeham, D. and Mulvehill, P (1975). Egg transfer in the cow: factors affecting pregnancy and twinning rates following bilateral transfers. J. Reprod. Fertil., **44**: 77 85.
- Sreenan, J. M. and Diskin, M. G. (1987). Factor affecting pregnancy rate following embryo transfer in the cows. Theriogenology, **27**: 99-113.
- Thibier, M. and Nibart, M. (1992). Clinical aspects of embryo transfer in some domestic animals. Anim. Reprod. Sci., **28**: 139- 148.
- Walton, J. S. and Stubbings, R. B. (1986). Factor affecting the yield of viable embryo transfer by superovulated Holstein Friesian cows. Theriogenology, **26**: 167-177.
- Wilmet, I., Sales, D. I. and Ashworth, C. J. (1985). The influence of variation in embryo stage and maternal hormone profiles on embryo survival in farm animals. Theriogenology, **23**: 107-119.
- Wright, J. M.(1981). Non-surgical embryo transfer in cattle. Embryo recipient interactions. Theriogenology, 15: 43-56.