

Effect of repeated Superovulation on superovulatory response and embryo recovery in Sahiwal cows

U.K. MISHRA, O.P. MISHRA and J.R. KHAN

Department of Veterinary Physiology
College of Veterinary Sci. & A.H., Anjora,
Durg (M.P.) 491001

ABSTRACT

Twelve elite Sahiwal donor cows were repeatedly superovulated four to five times with an interval of 75 to 100 days between two superovulations to determine the effect of repeated gonadotrophic superstimulation of ovaries on superovulatory response and embryo recovery during a period of one year. The mean numbers of corpus luteum/ovulations observed were 10.16 ± 0.62 , 9.75 ± 0.64 , 7.75 ± 0.56 , 5.91 ± 0.37 and 4.66 ± 0.87 in five attempts respectively. The mean number of embryo recovery was 6.83 ± 0.70 , 4.83 ± 0.52 , 3.41 ± 0.35 , 3.08 ± 0.39 and 2.33 ± 0.87 . The mean number of transferable embryo was 5.16 ± 0.65 , 3.5 ± 0.37 , 2.5 ± 0.31 , 2.0 ± 0.27 and 2.0 ± 0.57 . Repeated superstimulation of ovaries by exogenous gonadotropin treatments had a tendency towards decreasing number of ovulations, embryo recovery and transferable embryos per cow during a period of one year. It is also observed that in one year five repeated superovulations and flushing were possible only in 25% Sahiwal donors whereas in 75% cows only four repeated superovulation and embryo recovery were done successfully.

—x—x—x—

In recent years embryo transfer technology has emerged as one of the important tools for the fast multiplication of superior germ plasm and its conservation. However, its success depends upon the response of superovulation on repeated treatments of gonadotropins. The indigenous cows have better superovulatory response at lower doses of gonadotropin than those of exotic cows (Munro, 1986), however,

response of varies to repeated superstimulation treatment in Sahiwal cows which is one of the best Indian milch breed has not been yet reported. The reports are also not available on possible attempts at which these cows could be flushed in one year duration. Although the reports on *Bos taurus* cows have shown that number of ovulation, fertilization rate embryo production decreases after repeated superovulations. (Saumande and Chupin 1977, Donaldson and Perry, 1983). The reports in crossbred indicated that repeated superovulation has a tendency towards decreasing number of corpus luteum, embryo recovery and transferable embryos per cows (Totey *et al.*, (1992). Therefore the present study aims at determining the repeated superovulation response and embryo recovery in Sahiwal cows in one year.

MATERIALS AND METHODS

Twelve mature pure elite Sahiwal donor cows reared in the research dairy unit of the Department were used for four to five repeated superovulations. The cows were provided adequate nutrition, housing and management.

The synchronization of estrus was done by injecting 500ug of estrumate 11 days apart. Superovulation was carried out by injecting 28mg of FSH-p twice daily for four days in descending dose levels starting from 9th days of estrus. All the donors

received 500ug estrumate 48 hrs after the start of superovulation treatment and were inseminated 48 hrs after estrumate injection at 0, 12, 24 hrs on estrus detection with frozen semen of progeny tested Sahiwal bulls purchased from NDRI, Karnal. Before starting next superovulation, donor cows were observed for atleast two normal estrus cycles. The donors were also examined for evidence of possible reproductive disorders. The interval between two superovulatory treatment ranged from 75 to 100 days.

Embryos were recovered on 7th day of estrus. A total of 1.0 litre Dulbecco's Phosphate buffered saline (DPBS) with 0.1% Bovine serum albumin was used as flushing media employing embryo flushing catheter (18 FR Minitub) for interrupted syringe flushing method for each donor. The embryos were collected by filtering the flushed media through Emcon embryo filter. The embryos were examined, evaluated and graded morphologically under the zoom stereo microscope. The data were analyzed by one way analysis of variance.

RESULTS AND DISCUSSION

The repeated superstimulation of ovaries by exogenous gonadotropin given in one year duration has indicated that the total ovulations, total embryo recovered and number of transferable embryos have been affected by each attempt of embryo flushings (Table 1). The mean numbers of CL/ovulations were 10.16 ± 0.62 , 9.75 ± 0.64 , 7.75 ± 0.56 , 5.91 ± 0.37 in I, II, III and in IV flush respectively. The number of ovulations decreased significantly ($P < 0.05$) from 2nd flush onwards. It was also observed that V flushing was possible only in 25% cows of this breed. No other data are currently available related to similar trend in Sahiwal cows. Bastidas and Randel (1987) reported decrease in total ovulation

form 9.4 at the first to 2.9 at fifth flushing in *Bos indicus* cows. Similar observations have been given in *Bos taurus* cattle (Saumande and Chupin, 1977) and in crossbred cows (Totey *et al.*, 1992). Long term superstimulatory effect of gonadotropin on ovarian function has not been reported. However, there are possibilities of production of antibodies against gonadotropins (Greve, 1982).

The number of total embryos flushed was also affected with repeated flushings. Mean numbers of total embryos recovered on repeated superovulations were 6.83 ± 0.70 , 4.83 ± 0.52 , 3.41 ± 0.35 , 3.08 ± 0.39 , 2.33 ± 0.87 respectively in I, II, III, IV and V attempts. Total number of embryo collected at the first attempt was significantly higher ($P < 0.05$) than that of II to IV attempts. Bastidas and Randel (1987) reported that repeated superovulations decreases the embryo production ranging from 7 embryo per donor in the first flush to 3.0 embryos per donor in sixth flush. Similarly, Totey *et al.*, (1992) also reported that mean number of embryo recovery was 5.85 ± 3.89 in the first and 2.5 ± 2.6 in the fifth flush.

The mean number of transferable embryos also decreased with subsequent flushings. The highest number was observed in first flush and the lowest in fourth and fifth attempt. Bastidas and Randel (1987) reported that the number of blastocyst and morulas recovered per donor cows was affected by repeated superovulations and flushings. Similar trend was also observed by Totey *et al.*, (1992) with the exception that mean number of transferable embryo increased during the 2nd flushing from the since in our study there was gradual decrease in the total transferable embryos after each flushing.

It may be concluded that Sahiwal cows also have detrimental effect on production of embryo after repeated flushings. However it is possible to flush these cows successfully upto fourth attempt in a year although fifth attempt is also possible only in 25% cows. The total number of transferable embryos appears to be 50% of the total number of ovulations irrespective of number of

attempts performed. A recovery of 13 to 15 transferable embryos is possible in 4-5 flushing in one year in this breed.

Acknowledgement

Indian council of Agricultural Research New Delhi is thankfully acknowledge for financial support as Adhoc research Project.

Table 1. Repeated superovulatory response in Sahiwal cows treated with FSH-P in one year.

| Attempts | Mean no. of CL±SE | Mean no. of total embryo±SE | Mean no. of transfe- embryo±SE |
|----------|----------------------|--------------------------------|-----------------------------------|
| I | 10.16±0.62 | 6.83±0.70 | 5.16±0.65 |
| II | 9.75±0.64 | 4.83±0.52 | 3.50±0.37 |
| III | 7.75±0.56 | 3.41±0.35 | 2.50±0.31 |
| IV | 5.91±0.37 | 3.08±0.39 | 2.00±0.27 |
| V | 4.66±0.87 | 2.33±0.87 | 2.00±0.57 |
| Total | | | 15.16±0.43 |

* Vth attempt was possible in only 25% of the cows.

REFERENCES

- Bastidas, P. and Randel, R.D. (1987): Effect of repeated superovulation and flushing on reproductive performance of *Bos indicus* cows. *Theriogenology* 28: 827-835.
- Donaldson, L.E. and Perry, B. (1983): Embryo production by repeated superovulation of commercial donor cows. *Theriogenology* 20: 163-168.
- Greve, T. (1982) : Embryo transplantation in dairy cattle. An attempt to analyze factor that may effect embryo number and quality. Proc. 2nd Internat Cong on Embryo transfer in mammals and in vitro fertilization. Anney. Sept. 20-22 1982 p.: 251-276.
- Munro, R.K. (1986). The superovulatory response of *B.taurus* and *B. indicus* cattle following treatment with follicle stimulating hormone and progesterone. *Animal Reproduction Science* 11: 91-97.
- Saumande, J. and Chupin, D. (1977): Superovulation: A limit to egg transfer in cattle. *Theriogenology* 7: 141-149.
- Totey, S.M., Singh Gurpreet, Taneja, M., Singh Gurcharan, Chillar, R.S. (1992). Effect of repeated superovulation and flushing on embryo recovery in crossbred cows. *Indian J. Anim. Reprod.* 13(2): 117-120