

Superovulatory response with once and twice daily injection of Follitropin-V in crossbred cows

S G DESHMUKH¹, C H PAWSHE², S K SAHATPURE³ and R K PATIL⁴

Post Graduate Institute of Veterinary and Animal Sciences,
Akola, Maharashtra 444 104 India

Received: 6 October 2009; Accepted: 31 May 2010

ABSTRACT

Cyclic cows (7) were divided into 2 different treatment groups for superovulation. Group 1 (n=4) cows were treated with follitropin-V (400 mg) subcutaneously diluted in 3.2% gelatin carrier vehicle in descending doses and group 2 (n=3) cows were treated with follitropin-V (400 mg) intramuscularly twice daily in descending doses. All the animals (100%) exhibited superovulatory estrus. The average time required for onset of estrus was 39.5 ± 6.65 h and 27.33 ± 10.53 h and the duration of superovulatory estrus was 27.5 ± 9.16 and 26.5 ± 8.81 h in group 1 and 2, respectively. The average number of corpora lutea and unovulatory follicles were 11 ± 7.58 and 6.33 ± 2.18 and 2.0 ± 0.70 and 2.66 ± 0.66 , in group 1 and 2, respectively. The average total number of embryos and transferable embryos in group 1 was 1.75 ± 1.75 and zero, respectively, while in group 2 it was 5 ± 4.51 and 4 ± 3.15 , respectively.

Key word: Cow, Embryo recovery, Superovulatory response

In the current practice of superovulation, highly purified FSH is being used. However these gonadotropins have much shorter half life in the circulation and are usually administered twice daily, requiring twice daily handling of the donor animals which may leads to addition stress further contributing to the variability in superovulatory response.

As an alternative to twice daily injection of FSH, once daily injection of FSH diluted in carrier is now being tried for superovulation. Attempts have been made to prolong the absorption of single FSH injection to reduce the handling of the donor cows during gonadotropin stimulation. Carboxymethyl cellulose diluents (1%) and 30% polyvinyl pyrrolidone are used to prolong the absorption of FSH. However the results were not very encouraging. In a subsequent study, Looney *et al.* (1981) got greater superovulatory response by using once daily injection of FSH diluted in a 3–2 per cent protein gelatin carrier vehicle as compare to that of twice daily treatment protocol.

In view of the earlier limited number of trials of single injection of FSH-P diluted in gelatin carrier vehicle in

crossbred cows under the semi arid geoclimatic condition of Vidarbha will be useful to bring about more specific understanding of this new approach to superovulation in cattle.

MATERIALS AND METHODS

Cows (7) selected as donors were synchronized by administering prostaglandin $F_2\alpha$ (lutalyse 25 mg) intramuscularly on day 0 and day 11. The cows were superovulated with two different protocols of follitropin-V on day 10 of the estrus cycle by randomly dividing into 2 different treatment groups.

In the group 1, cows (4) were superovulated with once daily injection of follitropin diluted in 3.2% gelatin carrier vehicle 130 mg (6.5 ml), 110 mg (5.5 ml), 90 mg (4.5 ml) and 70 mg (3.5 ml); total 400 mg follitropin-V was given for 4 day in descending doses. Group 2 total cows (3) were superovulated with 400 mg twice daily injection of follitropin-V (2×65 mg, 2×55 mg, 2×45 mg and 2×35 mg) intramuscular was given for 4 days in descending doses. The superovulatory treatment for both the groups was started from day 10th of estrus cycle. Lutalyse ($PGF_2\alpha$ 25 mg) was administered intramuscularly after 72 h of initiation of superovulatory treatment. The superovulated cows were artificially inseminated with frozen semen at 12 h interval for three times after onset of superovulatory estrus. The superovulatory response was estimated on the basis of

Present address: ¹Assistant Professor (e-mail: dr.deshmukh@rediffmail.com)

²Associate Professor and Head (e-mail: chpll@rediffmail.com)

³Associate Professor, Department of Animal Reproduction, NVC, Nagpur.

⁴Former Professor and Head, Department of Animal Reproduction.

number of corpora lutea and unovulatory follicles palpable per rectum on day seven after the superovulatory treatment and non surgical embryo recovery performed by using two ways Foley's catheter and Dulbecco's Phosphate Buffered Saline (DPBS) supplemented with 1.0% fetal calf serum (FCS) was used as a flushing medium. The recovered embryos were graded as transferable and non-transferable as per Linder and Wright (1983). The transferable embryos were of morula, early blastocyst and blastocyst stages. The non transferable embryos were either unfertilized ova or degenerated embryos. Data were statistically analyzed by using student "t" test (Snedecor and Cochran 1967).

Preparation of Gelatin Carrier Vehicle: It was prepared by dissolving 3.2 gm of protein gelatin to 96.8 ml of lukewarm sterile normal saline solution. Such gelatin saline carrier vehicle was then used to make a solution containing 20 mg of FSH-P in 1 ml of solution, i.e. 400 mg FSH-P in 20 ml gelatin solution.

RESULTS AND DISCUSSION

The effect of 2 different doses regimen of follitropin-V was observed for onset and duration of superovulatory oestrus (Table 1). All 7 (100%) animals in group 1 and 2 responded to follitropin -V treatment. The average time required for onset of superovulatory oestrus was more in group 1 (39.5±6.65 h) as compare to group 2 (27.33±10.53). Duration of superovulatory oestrus was slight more in group 1 (27.5±9.06 h) as compared to group 2 (26.5±8.81 h). However, there was no significant difference between 2 groups even at 5% level for onset and duration of oestrus. The time required for onset of oestrus and duration of oestrus confirmed the earlier observation of Loney *et al.* (1981).

The ovarian responses were graded on the basis of number

Table 1. Onset and duration of superovulatory estrus

Parameters	Treatment group	
	Group 1	Group 2
No. of cows treated	4	3
No. of cows responded (%)	4 (100%)	3 (100%)
Onset of superovulatory estrus (h)	39.5±6.5	27.33±10.53
Duration superovulatory estrus (h)	27.5±9.16	26.5±8.81

Table 2. Superovulatory response and embryo recovery

Parameters	Treatment group	
	Group 1	Group 2
No. of cows treated	4	3
No. of cows flushed	4	3
Average no. of corpora lutea	11±7.58	6.33±2.18
Average no. of unovulatory follicles	2.0±0.70	2.66±0.66
Average no. of embryos recovered	1.75±1.75	5±4.51
Average no. of transferable embryos	0	4±3.51

of corpora lutea and unovulatory follicles palpable per rectum on both the ovaries and presented in Table 2. The average number of corpora lutea was better in group 1 (11±7.58) with range from 0 to 33 as compare to group 2 (6.33±2.18) with range from 2 to 9. But the individual response of cow varied greatly. The student "t" test elicited non significant difference between the two groups. The average number of corpora lutea recorded in group 1 was more to those recorded by Looney *et al.* (1981) and Bo *et al.* (1994). Whereas, in group 2 numbers of corpora lutea recorded were similar to those recorded by Goulding *et al.* (1990) and Arora *et al.* (1996).

The average number of unovulatory follicles was less in group 1 (2±0.70) than in group 2 (2.66±0.66). The average number of unovulatory follicles recorded in group 1 was less with the finding of Walsh *et al.* (1993), who recorded an average number of unovulatory follicles to be 4.1 when normal saline was used as a carrier vehicle for the FSH P. In the present study the average number of corpora lutea in group 2 was similar with Pawshe *et al.* (1992) and Sarvaiya *et al.* (1992) who recorded (2.70±0.42 and 2.80±0.97), respectively. The total 7 and 15 with an average of 1.75±1.15 and 5±4.51 embryos recovered in group 1 were degenerated (ie) non-transferable quality embryos, while in group 2 out of 15 embryos recovered 12 with an average (4±3.51) were of transferable quality (morula stage embryo) and remaining 3 embryos were degenerated.

Student "t" test elicited non significant difference between both the groups for average number of embryos recovered, but the transferable quality embryos were higher in group 2. The present finding for embryo recovery in group 1 is similar to Kharche *et al.* (1995) who recorded 2.33±0.26 embryo recoveries and also embryo recovery in group 2 was similar with that of Powlyshyn *et al.* (1987) and Ravindranath and Reddy (1999) who recorded the average number of embryo recovered to be 5.1 and 5.14±0.86, respectively.

It was concluded that the superovulatory response in terms of average number of corpora lutea, unovulatory follicles and total embryo recovery showed non-significant difference between both the groups. However, the total transferable quality embryos recovery in group 2 were better as compare to group 1.

ACKNOWLEDGMENT

This work was supported by the Indian Council of Agricultural Research, under "ICAR-AICRP Network programme on Embryo Transfer Technology for Animal Production"

REFERENCES

- Arora V K, Devanathan T G, Pattibiraman S R and John Edwin M. 1996. Studies on superovulatory response in cows treated with PMSG or FSH during luteal stage of the oestrus cycle. *Indian Journal of Animal Reproduction* 17: 9–11.

- Bo G A, Hockely D K, Nasser L F and Mapletoft R J. 1994. Superovulatory response to single subcutaneous injection of Follitropin-V in beef cattle. *Theriogenology* **42**: 963–75.
- Goulding D, Williams D H, Duffy P, Boland M P and Roche J F. 1990. Superovulation in heifers given FSH initiated either at day 2 or day 10 of the oestrus cycle. *Theriogenology* **34**: 767–78.
- Kharche S D, Dutt T K, Dutt T and Ansari M R. 1995. Efficiency of follicle stimulating hormone using different injection schedules for induction of superovulation in crossbred cows. Proceeding of XII National Convention of ISSAR and National symposium on “Modern trends in reproductive health care for enhancing livestock fertility and productivity” Jan. 13–15, 1995 held at University Department of Animal Reproduction, P.K.V., Akola: C–57.
- Linder G M and Write R W. 1983. Bovine embryo morphology and evaluation. *Theriogenology* **20**: 403–16.
- Looney C R, Boutte B W, Archbold L F and Godke R A. 1981. Comparison of once daily and twice daily FSH injection for superovulating beef cattle. *Theriogenology* **15**: 12–19.
- Pawshe C H, Kadu M S, Fasihuddin M and Totey S M. 1992. Superovulation with FSH-P and PGSG hormones in crossbred cows and heifers. *Indian Journal Animal of Reproduction* **13**: 18–20.
- Powlyshyn V, Lindsell C E, Braithwaite M and Mapletoft R J. 1986. Superovulation of beef cows with FSH-P: A dose response trial. *Theriogenology* **25**: 179.
- Ravindranath B M and Reddy S M. 1999. Superovulatory response in imported Holstein Friesian cows under field conditions. *Indian Journal of Animal Reproduction* **20**: 88–90.
- Sarvaiya N P, Chauhan F S and Mehta V M. 1992. Comparative efficiency of different gonadotropin preparations on superovulation and embryo recovery in Jersey×Kankrej cows. *Indian Journal of Reproduction* **13**: 120–23.
- Snedecor G W and Cochran W C. 1967. “Statistical Methods” 6th (Ed). Oxford and IBH publication Co, Calcutta, Bombay. Pp.: 64.
- Walsh J H, Mantovani R, Duby R T, Overstrom E W, Dobrinsky J R, Enright W J, Roche J F and Boland M P. 1993. The effect of once or twice daily injection of FSH-P on superovulatory response in heifer. *Theriogenology* **40**: 313–21.