Indian Journal of Animal Sciences **96** (2): 145–149, February 2026/Article
https://doi.org/10.56093/ijans.v96i2.176491

Comparative efficacy of recombinant human follicle stimulating hormone and porcine follicle stimulating hormone on follicular response, oocyte recovery and *in vitro* embryo production in donor cows (*Bos indicus*)

M V BHALKE[1], S G DESHMUKH[2]✉, M S PATIL[3],C H PAWSHE[2], M V INGAWALE[2], D S RAGHUWANSHI[3], P S BANKAR[4], K Y DESHPANDE[5], K B KAYANDE[1] and J N ISOKAR[1]

College of Veterinary and Animal Sciences, Akola Maharashtra 444 104 India

Received: 25 February 2026; Accepted: 19 May 2026

ABSTRACT

The present study compared the efficacy of long-acting recombinant human follicle stimulating hormone (rhFSH) and pituitary-derived FSH (pFSH) on follicular response, oocyte recovery and *in vitro* embryo production in donor cows. Eight high-yielding multiparous cows were subjected to two ovarian superstimulation protocols in a crossover design. In the first trial, donors received 200 mg pFSH in tapering doses, while in the second trial a single dose of long-acting rhFSH (300 IU) was administered. Transvaginal ovum pick-up (OPU) was performed on Day 8, and follicular count, oocyte recovery rate, oocyte quality, cleavage rate and blastocyst rate were recorded. The mean total follicle count did not differ significantly between pFSH (27±1.83) and rhFSH (25±2.15) stimulated groups. The oocyte recovery rate was significantly higher in the rhFSH group (77%; 154/200) compared to the pFSH group (68.05%; 147/216). The mean total oocyte yield per OPU was comparable between rhFSH (19.25±2.03) and pFSH (18.37±1.53) treatments. Cleavage rate (66.23% vs 72.1%) and blastocyst rate (55.84% vs 62.58%) did not differ significantly between rhFSH and pFSH groups, respectively. It may be concluded that long-acting rhFSH was comparable to conventional pFSH in terms of follicular response, oocyte quality and embryo production, while providing significantly improved oocyte recovery efficiency.

Keywords: Blastocyst rate, Follicular response, *In vitro* embryo production, Oocyte recovery, Recombinant FSH, Superstimulation

Ovarian superstimulation followed by transvaginal ovum pick-up (OPU) and *in vitro* embryo production (IVEP) is an established tool for accelerating genetic gain in cattle (De Roover *et al.* 2005, Baruselli *et al.* 2018). The efficiency of this technique largely depends on the ovarian response to exogenous gonadotropins and the recovery of developmentally competent oocytes which are suitable for embryo production (Guerreiro *et al.* 2014, Hayden *et al.* 2023). Conventionally, pituitary-derived follicle stimulating hormone (pFSH) administered in multiple tapering doses has been widely used for follicular superstimulation prior to OPU (Goodhand *et al.* 2000, Carvalho *et al.* 2014) with an effective dose of 200mg of pFSH (Singhal *et al.* 2020). However, variability in hormone composition and the requirement of repeated handling may affect follicular dynamics and oocyte recovery efficiency (Khodadadi *et al.* 2022, Viana *et al.* 2024).

Recombinant DNA (rDNA) technology is a molecular technique in which a specific gene encoding a desired protein is inserted into the DNA of a host cell, enabling the host to produce the protein in large quantities. In the case of follicle-stimulating hormone (FSH), the FSH gene is introduced into the host cell to produce recombinant FSH (rFSH) (Johnson, 1983). Recent developments in recombinant biotechnology have enabled the production of long-acting recombinant FSH (rhFSH) formulations with extended biological activity, allowing sustained follicular stimulation following a single administration (Perlman *et al.* 2003, Fauser *et al.* 2009). Studies comparing recombinant and pituitary-derived FSH preparations have reported comparable follicular recruitment, oocyte yield, and embryo development, although differences in response have been observed depending on dose, breed and the physiological status of donor animals (Deshmukh *et al.*

Present address: [1]M.V.Sc. Scholar, Department of Animal Reproduction, Gynaecology and Obstetrics, College of Veterinary and Animal Sciences, Akola 444 104, Maharashtra, India.[2] Professor, Department of Animal Reproduction, Gynaecology and Obstetrics, College of Veterinary and Animal Sciences, Akola 444 104, Maharashtra, India.[3]Professor, Department of Animal Reproduction, Gynaecology and Obstetrics, Nagpur Veterinary College, Nagpur 440 006, Maharashtra, India.[4]Associate Professor, Department of Animal Genetics and Breeding, College of Veterinary and Animal Sciences, Akola 444 104, Maharashtra, India. [5]Assistant Professor, Department of Animal Nutrition, College of Veterinary and Animal Sciences, Akola 444 104, Maharashtra, India. ✉Corresponding author email: shyamdeshmukh@mafsu.ac.in

2010, Rodrigues *et al.* 2023b, Patil *et al.* 2023).

Despite the encouraging result of recombinant gonadotropins in assisted reproductive technologies pertaining to OPU-IVEP program, limited literature is available under Indian field conditions. Therefore, the present study was undertaken to evaluate the comparative efficacy of long-acting recombinant FSH and pituitary-derived FSH on follicular response, oocyte recovery rate, oocyte quality and in vitro embryo production in donor cows. We hypothesized that the use of rFSH for superstimulation prior to OPU could match or improve the rate of embryonic production in donor cows.

MATERIALS AND METHODS

The present study was conducted from October 2025 to December 2025. Ovum pick-up (OPU) procedures were carried out at the Livestock Instructional Farm, and laboratory work related to in vitro embryo production was performed at the IVF–ET laboratory of the department.

Experimental animals: Eight high-yielding multiparous Sahiwal cows (2nd to 3rd parity : Age 4-5 yrs), each with a production potential exceeding 3500 litres per lactation and possessing phenotypically proven genetic traits, were selected as donor animals. The cows selected for the present experiment had an average body condition score of 3.0 to 3.5 on a 5.0 scale basis. The selected donor animals had been previously superstimulated with pFSH two months prior to inclusion in the present study. The animals were clinically examined to confirm the absence of reproductive abnormalities and the presence of normal ovarian cyclicity, as assessed through transrectal palpation. The experiment was conducted in a crossover design, with the same eight cows subjected to two ovarian superstimulation protocols in separate trials at an interval of one month duration between two trails.

Superstimulatory protocol: In both trials, cows at a random stage of the estrous cycle received a controlled internal drug release (CIDR) intravaginal device and estradiol benzoate (1 mg, IM) on Day 1. Prostaglandin $F_2\alpha$ (500µg cloprostenol sodium) was administered on Day 4. In the first trial, cows were superstimulated with 200 mg pituitary-derived FSH (Folltropin-V, Vetoquinol, Canada) administered in tapering doses from Day 5 to Day 7. In the second trial, cows received a single dose of long-acting recombinant human FSH (300 IU; Inceptova, Indian Immunologicals Ltd., India) on Day 6. The CIDR was removed on Day 8, and transvaginal OPU was performed on the same day in both protocols.

Before performing ovum pick-up (OPU), the donor cow was first properly restrained in an adjustable squeeze chute to ensure safety and minimize movement during the procedure. The rectum was carefully emptied by back raking, taking care not to allow aspiration of air. To reduce discomfort and facilitate smooth handling of the ovaries per rectum, caudal epidural anaesthesia was administered by injecting 3–5 ml of 2% Lignocaine Hydrochloride. This step effectively minimized defecation, abdominal straining and unwanted movement thereby ensuring better access to the reproductive tract. Prior to the insertion of the vaginal probe, thorough aseptic preparation of the external genitalia was carried out. The vulva and perineal region were first washed with clean water, then dried with a sterile napkin.

Ovarian follicles were examined on the day of OPU using a real-time B-mode ultrasound scanner (Exago, IMV, France) equipped with a 5 MHz transducer. Follicles were counted and categorized according to diameter as small (<6 mm), medium (6–10 mm) and large (>10 mm) (Rodrigues *et al.* 2023a, Hayden *et al.* 2022). The total number of follicles available for aspiration was recorded.

Oocyte recovery and grading: All visible follicles were aspirated using a portable ultrasound scanner (Exago, IMV, France) fitted with a 5.0 MHz convex transvaginal transducer and an 18 G stainless-steel needle guide (WTA, Brazil). The needle was connected to a vacuum system set to 70 mmHg negative pressure, and follicular contents were collected through 1.1 mm inner-diameter tubing into a 50 ml conical tube containing 15 ml OPU medium maintained at 37°C. After covering the transducer with a sterile vaginal sheath, it was inserted into the vaginal fornix, and the ovary was manipulated per rectum against the probe to align the aspiration line with the targeted follicle. Following assessment of follicle number and size, each follicle was punctured through the vaginal wall using a single-lumen 18-G sterile needle; aspiration was confirmed by follicle collapse on the monitor under constant vacuum pressure. The procedure was repeated for all visible follicles in both ovaries, withdrawing the needle under continuous suction and flushing it with medium to prevent blockage.

Follicular aspirates were filtered through a 75-µm embryo filter and examined under a stereomicroscope. Recovered cumulus-oocyte complexes (COCs) were morphologically evaluated at 8–20× magnification and graded into four categories (A–D) based on cumulus cell investment and cytoplasmic homogeneity. The parameters recorded included total follicular count, number of oocytes recovered, oocyte recovery rate (%), and grade-wise distribution of oocytes.

Maturation and fertilization of oocytes: Selected oocytes (Grade A, B and C) were subjected to *in vitro* maturation in 100 µl droplets of maturation medium under mineral oil and incubated at 38.5°C in a tri-gas incubator (5% CO_2, 5% O_2 and 90% N_2) for 22–24 h. After maturation, oocytes

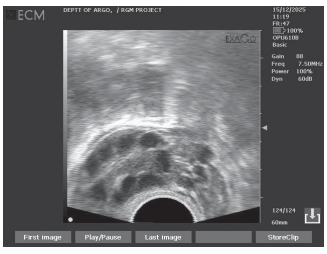

Fig. 1. Superstimulated ovary showing follicles of various size at the time of OPU

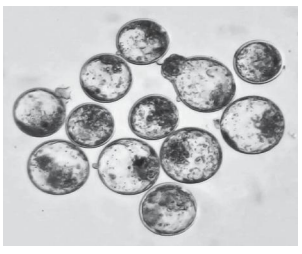

Fig. 2. Blastocyst stage of *in vitro* produced Embryo

were fertilized in vitro using frozen-thawed semen after density gradient centrifugation. Briefly, frozen-thawed semen exhibiting excellent initial motility (>80%) was layered over the density gradient prepared using upper and lower isolate in a 2.0 ml conical-bottom microcentrifuge tube. The composite mixture was centrifuged at 4700 rpm for 6 minutes, after which the supernatant was discarded and the pellet formed at the base of the tube was aspirated and transferred to pre-incubated IVF medium, followed by centrifugation at 1700 rpm for 3 minutes. After discarding the supernatant, the 100 µl sperm pellet was mixed thoroughly, and sperm concentration was estimated using a Neubauer's hemocytometer. Finally, the washed sperms were inoculated into the *in vitro* fertilization droplets containing oocytes at a final sperm concentration of 2×10^6/ml.

Presumptive zygotes were cultured *in vitro* for seven days under standard incubation conditions. Cleavage was assessed 24 h after fertilization, and blastocyst development was evaluated on the Day 7 of culture.

The following parameters were recorded and compared between the two superstimulation protocols: follicular size distribution, total follicle count, oocyte recovery rate, grade-wise oocyte distribution, number of cleaved oocytes, cleavage rate (%), number of blastocysts per OPU session and blastocyst rate (%).

Statistical analysis: Statistical analysis was performed using WASP 2.0 software (ICAR, Goa, India: https://ccari.res.in/waspnew.html). Data on follicular size distribution and oocyte grading were analysed using Factorial Complete Randomized Design (CRD). Total follicular count, number of oocytes recovered, number of cleaved embryos and number of blastocysts were analysed using Student's t-test. Oocyte recovery rate, cleavage rate and blastocyst rate were analysed using Chi-square test.

RESULTS AND DISCUSSION

Effect of superstimulation protocol on follicular response: The Effect of different superstimulation protocols on total follicular number and follicular size distribution is shown in Table 1. Both protocols were effective in stimulating the follicular response with non-Significant statistical difference in the variables of the number of follicles and follicular size distribution. These results agreed with Rodrigues *et al.* (2023b) and Gutierrez-Reinoso *et al.* (2022), who reported a non-significant difference in the number of follicles available for aspiration between donor superstimulated with rFSH and pFSH. Similarly, Carvalho *et al.* (2014) also reported that the number of follicles showed a non-significant difference between follitropin and rFSH groups. Patil *et al.* (2023) had reported significantly higher mean number of follicles available for aspiration in pFSH (200mg) superstimulated groups compared to rhFSH (@225 IU), which was in contrast with the present finding. The overall follicular size distribution between the superstimulated protocol using rhFSH and pFSH showed a non-significant difference, which is in accordance with Passamani *et al.* (2023) and Rodrigues *et al.* (2023b), who reported a non-significant difference in mean follicle size distribution at any moment between donors superstimulated with rhFSH and pFSH. The sustained biological activity of long-acting rFSH might have provided prolonged FSH exposure sufficient to support follicular growth comparable to that achieved with multiple pFSH injections (Perlman *et al.* 2003, Fauser *et al.* 2009, Hesser *et al.* 2011).

Effect of different superstimulation protocols on oocyte quality: The effect of different superstimulated protocols on the number of oocytes and grading protocols is depicted in Table 2.

In the present study, the mean total number of oocytes aspirated between the superstimulated protocol using rhFSH and pFSH is comparable with non-significant difference. The result was in accordance with Patil *et al.* (2023) who reported non-significant difference in mean number of oocytes aspirated between donor superstimulated with rhFSH and pFSH using total dose 225 IU and 200mg, respectively. While, the overall distribution of oocyte quality grades between the superstimulated protocol using rhFSH and pFSH showed non-significant difference but grade A oocyte were numerically higher in pFSH group, in contrast with Moura *et al.* (2025) who reported increased proportion of grade A cumulus oocyte complex (COC) with rhFSH than pFSH using total dose 10ug rhFSH in Nelore breed calves and prepubertal heifers. The comparable oocyte quality observed with rFSH stimulation might be attributed to its ability to promote a uniform follicular environment. Baruselli *et al.* (2023) stated that the FSH treatment prior to OPU promotes the growth of a homogeneous follicular population and facilitates the recovery of competent oocytes suitable for IVEP procedures. The absence of LH

Table 1. The follicular count (Mean ± SE) as per follicular size in donors stimulated with rhFSH and pFSH

Groups	Follicular count (Mean ± SE) as per follicular size			
	Small (<6mm)	Medium(6-10mm)	Large (>10mm)	Total
rhFSH	10.87b±0.78 (43.5%)	13.5b± 1.73 (54%)	0.62c± 0.18 (2.5%)	25a± 2.15
pFSH	11.87b± 0.63 (44%)	13.37b± 1.11 (49.53%)	1.75c± 0.25 (6.48%)	27a± 1.83

Mean bearing same superscripts in column differ non-significantly

rhFSH, Recombinant Human Follicle Stimulating Hormone; pFSH, Porcine Follicle Stimulating Hormone

Table 2. The mean oocyte number of different grades in donors stimulated with rhFSH and pFSH

Groups	Oocyte Grading				
	Grade A	Grade B	Grade C	Grade D	Total
rhFSH	8.00b±1.03 (41.55%)	8.50b±0.86 (44.15%)	2.00c±0.18 (10.38%)	0.75c±0.16 (3.89%)	19.25a±2.03
pFSH	8.50b±0.82 (45.90%)	7.62b±0.59 (41.21%)	1.50c±0.18 (8.10%)	0.75c±0.25 (4.72%)	18.37a±1.53

Mean bearing same superscripts in column differ non-significantly
rhFSH, Recombinant Human Follicle Stimulating Hormone; pFSH, Porcine Follicle Stimulating Hormone

contamination in rhFSH preparations helps to maintain a stable intrafollicular environment, which is favourable for oocyte maturation and quality (Khodadadi *et al.* 2022).

Oocyte recovery rate and oocyte compatibility: The comparative effect of superstimulation protocol on oocyte recovery rate and oocyte compatibility is depicted in Table 3.

In the present study, rhFSH superstimulation resulted in a significantly higher (P<0.05) oocyte recovery rate and total oocyte yield compared with pFSH superstimulation. These findings were in accordance with Patil *et al.* (2023), who reported significantly higher oocyte recovery rates in rhFSH-stimulated donors compared to pFSH-treated donors. The finding are in contrast with that of Rodrigues *et al.* (2023b), who reported non-significant differences in oocyte recovery rate with rFSH and pFSH superstimulated cows. The significantly higher per cent of oocyte recovery rate observed in donor cows superstimulated with rhFSH might be due to rhFSH providing a more consistent and LH-free hormonal profile, resulting in a more homogeneous and synchronized follicular population that is favourable for efficient oocyte recovery (Baruselli *et al.* 2023, Lisi *et al.* 2001).

In the present study, the number of oocytes cleaved and the cleavage rate showed a non-significant difference in the donor cows superstimulated with pFSH and rhFSH, but resulted in a numerically higher number of cleaved oocytes and a higher cleavage rate in the pFSH superstimulated group as compared to the rhFSH superstimulation group indicating that both rhFSH and pFSH were capable of supporting early embryonic development up to the cleavage stage. The non-significant difference observed in the number of oocytes cleaved and cleavage rate in both pFSH and rhFSH superstimulation groups indicated that the source of FSH does not markedly influence post-fertilisation embryo development, once competent oocytes are obtained (Moura *et al.* 2023). In the literature searched, no suitable documentation on a comparative study of rFSH versus pFSH regarding the number of oocytes cleaved and cleavage rate in Sahiwal cattle could be found.

In the present study, the number of blastocysts and the blastocyst rate between the superstimulation protocols using rhFSH and pFSH showed no significant difference. These findings were in accordance with Patil *et al.* (2023), who reported that rhFSH-stimulated donors exhibited non-significantly higher blastocyst production rates and embryos per OPU compared to pFSH-stimulated donors. Similarly, Gutierrez-Reinoso *et al.* (2022) reported that early blastocyst and blastocyst parameters showed statistically nonsignificant differences when comparing bscrFSH (bovine single-chain recombinant follicle-stimulating hormone) with NIH-FSH-p. Further, Arreseigor *et al.* (2022) also detected no statistical difference in Cinna-f (recombinant human FSH) and Folltropin-V. However, in contrast, Rodrigues *et al.* (2023b) reported that the blastocyst rate and number of blastocysts were higher in donors stimulated with both 150ug brFSH and 100ug brFSH compared with the pFSH protocol. The present finding, with non-significant differences in rFSH and pFSH regarding the number of blastocysts and blastocyst rate suggested that the cows superstimulated with rFSH showed comparable embryonic development to those superstimulated with pFSH.

Ovarian superstimulation using long-acting recombinant follicle stimulating hormone produced follicular response, oocyte quality, cleavage rate and blastocyst production

Table 3. Oocyte recovery rate and oocyte Competence using recombinant FSH and pFSH in donor cows.

Sr. No.	Parameter	rhFSH	pFSH
1	Oocyte recovery rate	77%^{a} (154/200)	68.05%^{b} (147/216)
2	No. of oocyte cleaved	12.75^{c}±1.17	13.25^{c}±1.08
3	Cleavage rate	66.23%^{c} (102/154)	72.1%^{c} (106/147)
4	No. of blastocyst	10.75^{c}±1.17	11.5^{c}±0.98
5	Blastocyst rate	55.84%^{c} (86/154)	62.58%^{c} (92/147)

Mean and Percent bearing different superscript (a,b) in a row differ significantly (p<0.05)
rhFSH, Recombinant Human Follicle Stimulating Hormone; pFSH, Porcine Follicle Stimulating Hormone

comparable to conventional pituitary-derived follicle stimulating hormone in donor cows. However, recombinant follicle stimulating hormone resulted in significantly higher oocyte recovery efficiency. The findings indicated that long-acting recombinant follicle-stimulating hormone can serve as an effective alternative to conventional protocols while reducing repeated hormone administration, animal handling, and treatment cost. Due to the small sample size in the present study, further studies required to confirm the findings.

ACKNOWLEDGEMENT

The authors are highly thankful to ETT-IVF laboratory Department of Animal Reproduction, Gynaecology and Obstetrics, Nagpur Veterinary College, Nagpur and Maharashtra Animal and Fishery Sciences University, Nagpur.

REFERENCES

Arreseigor C J, Gutierrez-Reinoso M A, Driedger B, Cabezas I, Hugues F I, Parra N C, Sanchez O, Toledo J R and Garcia-Herreros M. 2022. Superovulation efficiency and embryo production by using recombinant FSH (bscrFSH) vs. pituitary-derived FSH (FSH-p) in Brangus heifers. *Animal Reproduction* **19**(2): p.e22180.

Baruselli P S, Abreu L Â D, Catussi B L C, Oliveira A C D S, Rebeis L M, Gricio E A and Rodrigues C A. 2023. Use of new recombinant proteins for ovarian stimulation in ruminants. *Animal Reproduction* **20**(2): e20230092.

Baruselli P S, Batista E O S, Vieira L M, Ferreira R M, Guerreiro B G, Bayeux B M and Gimenes L U. 2018. Factors that interfere with oocyte quality for *in vitro* production of cattle embryos: Effects of different developmental and reproductive stages. *Animal Reproduction* **13**(3): 264–272.

Carvalho P D, Hackbart K S, Bender R W, Baez G M, Dresch A R, Guenther J N and Fricke P M. 2014. Use of a single injection of long-acting recombinant bovine FSH to superovulate Holstein heifers: a preliminary study. *Theriogenology* **82**(3): 481–489.

De Roover R, Genicot G, Leonard S, Bols P and Dessy F. 2005. Ovum pick up and in vitro embryo production in cows superstimulated with an individually adapted superstimulation protocol. *Animal Reproduction Science* **86**(1–2): 13–25.

Deshmukh S G, Pawshe C H, Sahatpure S K and Patil R K. 2010. Superovulatory response with once and twice daily injection of follitropin-V in crossbred cows. *Indian Journal of Animal Sciences* **80**(9): 851.

Fauser B C J M, Mannaerts B M J L, Devroey P, Leader A, Boime I and Baird D T. 2009. Advances in recombinant DNA technology: corifollitropin alfa, a hybrid molecule with sustained follicle-stimulating activity and reduced injection frequency. *Human Reproduction Update* **15**(3): 309–321.

Goodhand K L, Staines M E, Hutchinson J S M and Broadbent P J. 2000. *In vivo* oocyte recovery and *in vitro* embryo production from bovine oocyte donors treated with progestagen, oestradiol and FSH. *Animal Reproduction Science* **63**(3–4): 145–158.

Guerreiro B M, Batista E O S, Vieira L M, Sá Filho M F D, Rodrigues C A, Netto A C and Baruselli P S. 2014. Plasma anti-Müllerian hormone: An endocrine marker for *in vitro* embryo production from *Bos taurus* and *Bos indicus* donors. *Domestic Animal Endocrinology* **49**: 96–104.

Gutierrez-Reinoso M A, Aguilera C J, Navarrete F, Cabezas J, Castro F O, Cabezas I and Rodríguez-Alvarez L. 2022. Effects of extra-long-acting recombinant bovine FSH (bscrFSH) on cattle superovulation. *Animal Reproduction* **12**(2): 153.

Hayden C B, Sala R V, Absalón-Medina V A, Motta J C, Pereira D, Moreno J F and García-Guerra A. 2022. Synchronization of follicle wave emergence before ovarian superstimulation with FSH and ovum pick-up improves *in vitro* embryo production in pregnant heifers. *Theriogenology* **188**: 71–78.

Hayden C B, Sala R V, Pereira D C, Moreno J F and García-Guerra A. 2023. Effect of use and dosage of porcine follicle-stimulating hormone for ovarian superstimulation before ovum pick-up and *in vitro* embryo production in pregnant Holstein heifers. *Journal of Dairy Science* **106**(11): 8110–8121.

Johnson I S. 1983. Human insulin from recombinant DNA technology. *Science* **219**(4585): .632–637.

Khodadadi A, Niasari-Naslaji A, Nikjou D and Mohammadi B. 2022. Superovulation of high-producing Holstein lactating dairy cows with human recombinant FSH and hMG. *Theriogenology* **191**: 239–244.

Moura R M, Martins L P, Passamani G, Fernandes C A C, Siqueira L G B, Figueiredo R A and Viana J H M. 2023. Using a long-acting recombinant human follicle-stimulating hormone in cattle: superovulatory response. *Reproduction, Fertility and Development* **36**(2): 269–269.

Patil S P, Layek S S, Karuppanasamy K K, Doultani S, Raj S, Raval K B and Gorani S. 2023. The use of recombinant human follicle-stimulating hormone in comparison to pituitary-derived follicle-stimulating hormone prior to ovum pick up *in vitro* embryo production in cattle. *Reproduction, Fertility and Development* **36**(2): 271–271.

Perlman S, van den Hazel B, Christiansen J, Gram-Nielsen S, Jeppesen C B, Andersen K V and Schambye H T. 2003. Glycosylation of an N-terminal extension prolongs the half-life and increases the *in vivo* activity of follicle stimulating hormone. *The Journal of Clinical Endocrinology and Metabolism* **88**(7): 3227–3235.

Rodrigues C A, Netto A C, Catussi B L C, Rebeis L M, Pinho P, Randi F and Baruselli P S. 2023a. Superstimulation prior to the ovum pick up with a single dose of recombinant FSH improves *in vitro* embryo production in Holstein heifers. *Animal Science Proceedings* **14**(3): 483–484.

Singhal S, Singh N, Singh H, Malik V S and Brar P S. 2020. Curtailing the conventional dose of Folltropin-V for superstimulation and embryo recovery in Sahiwal cattle (*Bos indicus*). *Indian Journal of Animal Sciences* **90**(11): 1471–1475.

Viana J H M, Moura R M, Martins L P, Figueiredo R A, Siqueira L G B and Fernandes C A C. 2024. Superovulating cattle with corifollitropin-alpha, a long-acting recombinant human FSH: dose-response, half-life, effects on the ovaries and embryo outcomes. *Theriogenology* **226**: 302–307.