TRANSLATIONAL RESEARCH IN VETERINARY SCIENCE

SUPEROVULATION NEW MODELS INCLUDING FSH DOSE REDUCTION AND NEW PREPARATIONS

Translational Research in Veterinary Science Vol. 5 No. 1 (2023) p. 9 – 10

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It was estimated that every year around the world, approximately half a million bovine embryos are produced (Cremonesi, 2020). The classic superovulation program involves administering of a series of FSH injections to embryo donor cows starting on day 9-12 of the oestrus cycle. In Europe, Pluset and Folltropin preparations are commercially used for this purpose, both available on the market in Poland (Kulus, 2019). This is the mechanism underlying superovulatory protocols that were developed in the 1980s, which were further improved by refining hormonal preparations like human menopausal gonadotropin (HMG), equine chorionic gonadotropin (eCG), or follicle-stimulating hormone (FSH) from porcine or ovine pituitaries (Cremonesi 2020). Superovulation protocols vary widely based on the FSH source, the diluent used, the number and timing of FSH injections and the timing and utilisation of various prostaglandins, controlled internal progesterone releasing devices, gonadotrophin-releasing hormone, and other means of controlling follicular development and ovulation (Mikkola, 2019). These may include nutritional status, reproductive history, age, season, breed, effects of repeated superovulations and ovarian status at the time of treatment (Cremonesi 2020). An approach that has shown promise is to initiate FSH treatments at the time of the emergence of the first follicular wave following GnRH-induced ovulation. Using of aspiration of dominant follicle is possible to induce new follicular wave. This simple biotechnical treatment has a final impact on the number of in vivo produced embryos. Its seems, that of the interval from follicle aspiration to initiation of lengthened FSH treatment (Cirit, 2019). However, it has been shown that it may be possible to ignore follicular wave status, and by extending the treatment protocol induce smaller follicles to grow and reach maturity and superovulate. Finally, the short halflife of pituitary FSH necessitates twice daily treatments which are time-consuming, stressful and subject to error. Recent treatment protocols have permitted superstimulation with a single or alternatively, two FSH treatments, reducing the need for animal handling during FSH treatment (Mapletoft, 2013). A single dose protocol of FSH for superstimulation in cattle may improve compliance and superovulatory response. A single subcutaneous (sc) administration of pFSH was

efficacious, but response depended on body condition and injection site; the adipose tissue pad behind the shoulder was most efficacious. Inconsistent results in Holsteins were partially overcome by sc administration of 75% of the total pFSH dose behind the shoulder on the first day followed by 25% 48 h later (Bo, 2018). The split-single injection given ischiorectal fossa (split-single IRF administration had a comparable superovulatory response to the traditional twice-daily protocol. Moreover, the ovulation rate, ovarian follicle responses, and embryo quality were affected by heat stress (Chumchai, 2021, Ratsiri, 2022). In the other study the ovarian responses in the split-single IRF group were similar to those of the control group ($p > .05$) but higher compared with the split-single IM group. Regardless of the route of administration of FSH. The high THI affected ovulation rate as well as the numbers of transferable embryos and degenerated embryos (Thanaporn, 2021). A recombinant long-acting ovine follicle stimulating hormone (roFSH) has been devised and its biological effectiveness following a single dose has been assessed in several experiments under field conditions, in pasture-based beef and dairy farming in New Zealand. Owing to the molecular structure of this long-acting roFSH, which includes additional N-glycosylation sites, a single dose combined with a simple CIDR-based superovulatory regime elicits successful ovarian stimulation with averages of 11.8 corpora lutea and of 6.1 good quality embryos collected in cattle. Solid performance of this novel FSH was demonstrated in several beef and dairy breeds which included yearling heifers and mixed age cows, with embryo production results in the same range as those observed nowadays with eight doses of commercial pituitary FSH (pFSH). Viable embryos produced from these collections, when implanted either fresh or frozen and thawed, gave pregnancy rates in recipients similar to those collected from cows and heifers superstimulated with pFSH. Repeated superovulatory treatment of the same cows was not associated with a decrease in ovarian response or embryo yield. The single administration of this long-acting roFSH when combined with a modified simple superovulatory regime has the advantage of reducing animal welfare concerns, lowering labour resource requirements and giving similar results to other commercially prepared pituitary FSH extracts (Sanderson, 2020, Gutierrez-Rreinoso, 2022). It is well known that 70% of embryos are produced by 30% of donors. This very high variability was tried to be reduced, with very good results by administrating platelet rich plasma (PRP) inside the ovary before the superovulation protocol. This hemocomponent is rich in growth factors and cytokines known for their regenerative properties in human and veterinary medicine (Cremonesi, 2020, Cirit, 2020).

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Key words: superovulation, results improvement, cows