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REPRODUCTIVE BIOTECHNOLOGIES IN FARM ANIMALS : A SYSTEMATIC LITERATURE REVIEW

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Abstract

It has been observed that reproductive inefficiency is a major constraint to productivity, profitability and genetic progress in livestock production systems in the world. It is a literature review methodology that will examine the quantitative and qualitative evidence on the effectiveness, adoption behavior and novelty in reproductive biotechnologies in large farm animals in literature published between 2010 and 2024. According to the conceptualization of PRISMA 2020 and a convergent mixed-method design, a total of 168 moderate to high-quality studies were included with respect to the initial 2,846 records. The findings reveal that the biotechnology that is mostly applied is artificial insemination (AI) and it has a conception rate of 48-62 percent in high dairy systems and a low rate in poor performance systems. Sex-sorted semen improves the performance of female offspring (85-92) at minimal cost in terms of fertility loss. Embryo transfer and most notably in vitro embryo production (IVP) technologies have increased by leaps and bounds and currently provide the largest portion of embryo manufacturing around the globe and enable genetic gain to be expedited. The invention of the cryopreservation or rather vitrification has enhanced the survival rates of post thaw embryos. The new gene-editing technology involving the CRISPR-Cas9 is exact and promising with a high level of efficiency (35-75) of resistance to diseases and improvement of characteristics, yet its extensive application in the corporate sector is controlled and ethically prohibited. The qualitative synthesis establishes the consistent obstacles which include infrastructure, cost, expertise shortage, environmental pressures, regulatory uncertainties. All in all, the reproductive biotechnologies are an immense influence on the genetic dispersion and the productivity of production in the intensive systems, yet the reasonable effect on the planet can only be achieved upon the context-specific adaption, greater accessibility and the homogeneity of regulatory laws.

Keywords: Reproductive Biotechnology; Artificial Insemination; Embryo Transfer; In Vitro Embryo Production; CRISPR-Cas9; Gene Editing; Livestock Reproduction; Cryopreservation; Sex-Sorted Semen; Genetic Improvement; Farm Animals; Systematic Review

INTRODUCTION

The livestock sector is among the agricultural production and food security pillars in the world since it provides essential nutrients and economic feed to billions of people throughout the world. According to the Food and Agriculture Organization (FAO) Statistical Yearbook 2024, the global quantity of meat production in 2022 totaled 361 million tonnes, which is 55 times more than it was in 2000 and livestock products are linked with an estimated 15% of the world caloric food intake and 31% of the protein intake (FAO, 2024). This enormous increase highlights the importance of sound animal production technologies in helping to meet the nutritional demands of a world population that is expected to exceed 9.7 billion by 2050 (United Nations, 2022). However, reproductive inefficiency is one of the most significant drawbacks to livestock productivity that has resulted in immense losses of an estimated billions of dollars annually in the animal agriculture sector as a whole all over the world (Rutledge, 2009; Diskin and Morris, 2008). One of the most widely used types of reproductive biotechnology is the artificial insemination (AI) that has changed the breeding paradigm and opened new perspectives on the genetic-improvement of animals in its commercially release in 1930s. Currently the estimates are that approximately 10 percent of the total cattle and buffalo population in the world is farmed by AI with advanced regions indicating more than 30-50 percent cover rates in the dairy cows with less developed regions showing an average coverage of less than 4 percent (Future Market Insights, 2025). The technology enables rapid inoculations of better genetics and minimization of threats of illnesses transmitted through natural mating. Over the years, approximately 81.9 million insemination procedures are performed annually in India, and this increases per capita supply of milk (Dhangada et al., 2024). However, the average conception rate varies widely among regions and systems of production with certain developing nations recording conception rates as low as 35 per cent and intensive dairy systems in Europe and North America registering conception rates of 50-60 per cent (Desta et al., 2024). These disparities describe the complex interplay between the technological availability, infrastructure development and technical capacity that determines the global trend in adopting reproductive biotechnology that can be adopted during livestock breeding systems. The development of embryo transfer (ET) technologies has further transformed the rate of genetic innovation that can

be acquired through livestock breeding regimes. The embryo business has experienced an astronomic expansion in the past three decades in the world with the current production exceeding two million cattle embryos annually (Viana, 2025). It is noteworthy that now such embryos are manufactured in vitro embryo production (IVP) at a rate of about 80 percent compared to the traditional in vivo methods of collecting such embryos, a paradigm shift in the industry practice (Stringfellow and Givens, 2000; Viana et al., 2022). Among them are increased efficiency, reduced dependence on the presence of the donor animals and the chance to be integrated with the new technologies including ovum pick-up (OPU) and in vitro maturation (IVM) (Galli et al., 2014). Using the statistics given by the International Embryo Technology Society (IETS) now, North America is the largest in the production of IVP embryos and it is the largest producer of the 2,160,000+ bovine embryos in 2024 (IETS Data Retrieval Committee, 2024). New technologies The advent of new technologies in the context of genetic engineering and genome editing has also been accompanied by drastic changes in the multiplication of genetically superior females, which actually reduces the breeding programs. The accuracy and effectiveness of genetic modification of livestock species have been transformed by the introduction of the clustered regularly interspaced short palindromic repeats (CRISPR)-Cas9 technology (Wang et al., 2025). Unlike the classical approaches of transgenic, where random insertion of genes occurred, CRISPR-mediated editing enables targeting of endogenous loci, which enables one to create animals with improved disease resistance, production attributes, and environmental footprint (Tan et al., 2013). The most recent applications include knockout mutation of the CD163 gene to produce pigs resistant to porcine reproductive and respiratory syndrome virus (PRRSV) (Whitworth et al., 2016), gene insertion in the NRAMP1 gene to increase the resistance of cattle to tuberculosis (Gao et al., 2017), and knockout of b-lactoglobulin gene to reduce allergenicity of goats to milk (Zhou et al., 2022). Reproductive technologies together with the precision genetic engineering is another significant development that can help transform the lives of animal production by addressing the old age problems in animal production. Sex-sorting technologies are also another major development with immense potential of transforming animal production. The potential of correcting the sex ratio of children through flow cytometric separation of X-

and Y-chromosome carrying sperm has been particularly utilized on dairy and beef production systems whereby there is gender predisposition (Garner & Seidel, 2008). Sexed semen technology gives dairy producers the opportunity to make replacement heifers using their best genetically endowed cows through using traditional semen in terminal crossbreeding to make the best genetic and economic gains (DeJarnette et al., 2009). However, technology has been linked to such issues as reduced fertility compared with traditional semen, high expenses and unavailability in some areas (Borchersen & Peacock, 2009). Existing research is geared towards improving the effectiveness of sorting, developing alternative sex-selection methods and apply the technology to other species besides cattle to preserve genetic material and genetic diversity. Sperm, embryo and more recent, oocytes and tissue of the ovary are now frozen and stored enabling the establishment of genetic resource banks, and the global exchange of livestock germ plasm to occur (Mapletoft and Hasler, 2005). To ensure the sanitary safety of the international embryo trade the International Embryo Technology Society has devised stringent measures as a way of cleaning up the embryo and treating it with trypsin in an attempt to curb the probability of disease transmissions (Stringfellow, 2011). Most critical is the popularization and optimization of reproductive biotechnologies despite the fact that new technologies have enhanced the viability of the methods of vitrification of embryos and oocytes and the new methods of preserving gonadal tissues (Saragusty and Arav, 2011). The procreation rate in modern high producing dairy cows has drastically declined over the past several decades due to the intensive modification of the genes in the production of milk to the extent of fertility traits (Lucy, 2001; Royal et al., 2000). This type of antagonistic genetic relation between the production and the reproduction is the fundamental issue, which requires concerted efforts in the form of genetic selection, nutritional regulation, and rationalization of the technology of reproduction (Walsh et al., 2011). In tropical and subtropical regions, especially heat stress has dire consequences on reproduction performance, reduced conception rates, elevated embryonic death rates, and poor general fertility (De Rensis & Scaramuzzi, 2003; Hansen, 2009). The future perspective of climate changes shows that the problems will not be less, and it is the need to develop genetically resistant properties in heat and cooling more effective schemes (Nardone et al.,

2010). Climate changes will not only impact productivity of separate farms, but also agriculture development in the region and the entire world. Another significant component of the economy of the developed and developing nations is the livestock industry and one of the largest commodities in the international market is the animal products (Anderson et al., 2009). The benefits of the advanced reproductive technologies, however, are not evenly spread, and few people have access to them in the smallholder systems, where the technologies have a potential to cause a significant impact on livelihoods and food security (Ojango et al., 2016). Other complexities of applying reproductive biotechnologies, particularly genetic modification, are also presented by ethical aspects and regulatory provisions (Mrode et al., 2019). The perception of genetically engineered animals by the common people in cultural and geographical locations is somewhat different and this introspection impacts the means of controlling and market opportunities (Rollin, 1995). The distinction between transgenic and gene-edited animals in which the system has undergone any changes has prompted the review of regulatory systems in several jurisdictions (Van Eenennaam & Young, 2018). The balancing of the international regulations with the maintenance of the adequate degree of safety, at the same time, is also an issue in the international trade of animal products that are reared according to the principles of the most advanced reproductive technologies.

METHODOLOGY

The mixed-method research design adopted in this systematic literature review was convergent, as it involved the integration of the two methods of evidence synthesis (quantitative and qualitative) in providing a comprehensive approach to the reproductive biotechnologies in farm animals. The research design was grounded on the Preferred Reporting Items of Systematic Reviews and Meta-Analyses (PRISMA) 2020 (Page et al., 2021) and the Joanna Briggs Institute (JBI) model of the mixed-method systematic reviews. The study design was in terms of data based convergent synthesis where quantitative and qualitative data were transformed, analyzed and integrated to provide comprehensive information on the effectiveness, implementation and contextualization of the reproductive biotechnologies in the various farm animals species.

Search Strategy and Sources of Information.

A methodological literature search was conducted in a number of electronic databases to ensure that the area of reference was covered exhaustively. Web of science, Scopus, PubMed/MEDLINE, CAB abstracts and Google scholar were the databases searched. The search strategy was developed based on controlled vocabulary (MeSH terms) and free-text keywords that are related to the reproductive biotechnologies and farm animals. Search terms were combined using the Boolean operator AND and OR. The primary search query was made up of the following words: artificial insemination, embryo transfer, in vitro fertilization, cloning, transgenesis, sexed semen, oocyte retrieval, somatic cell nuclear transfer, gene editing, CRISPR, and cattle, buffalo, sheep, goat, pig, horse, poultry, and farm animals. The search concentrated on the peer-reviewed articles that have been published in English versions in the interval between January 2010 and December 2024 in order to determine the existing technological innovations and make the search manageable. Additionally, backward and forward citation (snowballing technique) search was conducted on the articles used in the research and on the systematic reviews as well that were relevant to the study to retrieve more articles that were not located during database search. To make sure all the literature was covered, manual screening of references lists of included studies and other articles of interest was carried out.

Recruitment and Suitability of the study.

The study was selected based on a rigorous multi-stage screening process by the screening by titles/abstracts and the screening of the entire text. All these inclusion criteria were chosen with a purpose to fulfill the need of the mixed-methods design: primary research (experimental, quasi-experimental, randomized controlled trials, cohort studies, case-control studies, cross-sectional studies), qualitative research (phenomenological, ethnographic, grounded theory, case studies), and mixed-method research without any limitation on

the topics of the research. There were artificial insemination, multiple ovulation and embryo transfer (MOET), in vitro embryo production (IVP), somatic cell nuclear transfer (SCNT), transgenic animal production, sex-sorting technologies, cryopreservation techniques, and new technologies of gene editing. Articles that were not eligible because they were not primary data, conference abstracts which could not be found via the full-text version were eliminated as exclusion criteria, and the articles on which laboratory rodents or companion animals were used as the sole study focus were eliminated as exclusion criteria. Two independent reviewers did the screening and in case of discrepancies, it was screened by a third reviewer after discussion or consultations with the third reviewer where there was no consensus. The inter-rater reliability was computed using Cohen kappa coefficient (k) and the results of above 0.80 were considered to be excellent agreement (McHugh, 2012). Figure 1 presents the broad selection process in the chronological details of the search of databases up to the final study inclusion.

RESULTS

As Table 1 shows, studies incorporated were distributed unequally according to species, study design, and geographical contribution and more publications on cattle-based research were published in the developed areas. Table 2 is a pooled measure of reproductive performance of conventional artificial insemination and sex-sorted semen that comprises of conception rate, calving rate, services/conception, and female calf output. Table 3 shows the relative parameters of technical efficiency of multiple ovulation and embryo transfer (MOET) and in vitro embryo production (IVP) that have differences in the embryo yield, pregnancy rate, collection frequency and genetic gain potential. Table 4 shows the ranges of efficiencies, most important applications and major obstacles to the implementation of advanced reproductive technologies: SCNT, transgenesis, CRISPR-Cas9 and sex-sorting systems.

Table 1. Distribution of Included Studies by Species, Design, and Region

Category	Subcategory	Number of Studies (n)	Percentage (%)
Species	Cattle	104	62
Species	Pigs	24	14
Species	Sheep & Goats	22	13
Species	Buffalo	10	6
Species	Other Species	8	5

Caption: Table 1 shows the distribution of the 168 included studies according to species focus, study design, and geographical contribution, highlighting the dominance of cattle-based research and concentration in developed regions.

Table 2. Reproductive Performance Indicators: Conventional AI vs Sex-Sorted Semen

Parameter	Conventional AI	Sex-Sorted Semen	Difference (%)
Conception Rate (%)	55	48	-7
Calving Rate (%)	52	45	-7
Services per Conception	1.8	2.1	+0.3
Female Calf Output (%)	50	90	+40

Caption: Table 2 shows pooled reproductive performance indicators comparing conventional artificial insemination with sex-sorted semen, demonstrating slightly reduced fertility but substantially higher female calf output in sexed semen programs.

Table 3. Comparative Technical Efficiency: MOET vs IVP

Parameter	MOET (In Vivo)	IVP (In Vitro)	Relative Advantage
Embryos per Donor	5.2–8.6	2.8–4.5 per OPU	MOET higher per flush
Collection Frequency	Low	High (Repeated OPU)	IVP higher frequency
Pregnancy Rate (%)	55–65	50–60	Comparable
Genetic Gain Speed	Moderate	High	IVP advantage

Caption: Table 3 shows comparative technical and biological efficiency indicators between multiple ovulation and embryo transfer (MOET) and in vitro embryo production (IVP), emphasizing the increased genetic dissemination potential of IVP systems.

Table 4. Advanced Reproductive Technologies: Efficiency, Applications, and Barriers

Technology	Efficiency Rate (%)	Primary Applications	Major Barriers
SCNT (Cloning)	10–25	Elite genotype replication	Low efficiency, high cost
Transgenesis	5–20	Trait enhancement	Regulatory restrictions
CRISPR-Cas9	35–75	Disease resistance, trait editing	Regulatory & ethical concerns
Sex-Sorting Tech	85–92 accuracy	Gender selection	Reduced fertility

Caption: Table 4 shows the reported efficiency ranges, principal applications, and major implementation barriers associated with advanced reproductive technologies including SCNT, transgenesis, CRISPR-Cas9, and sex-sorting systems.

The PRISMA 2020 flow diagram presented in Figure 1 illustrates the manner in which the studies have been searched in a systematic manner starting with the search and identification of the first database, all the way to the point where 168 researches are identified. Figure 2 indicates the relative pregnancy rates in the usual artificial insemination and that of sex-sorted semen with the intensive and extensive production systems and indicates that there are some minor losses in fertility with sexed semen but more efficient gender

targeting. Figure 3 demonstrates how the world has transformed the production technologies of the production of embryos in vivo MOET to the in vitro embryo production (IVP) in the same period of 2010-2024 wherein the adoption of IVP will rise exponentially with the years. Figure 4 is the post-thaw survival rate of the various modes of the vitrification as the most successful mode of survival of the embryos and the slow freezing mode as the least predictable mode of preserving the oocyte.

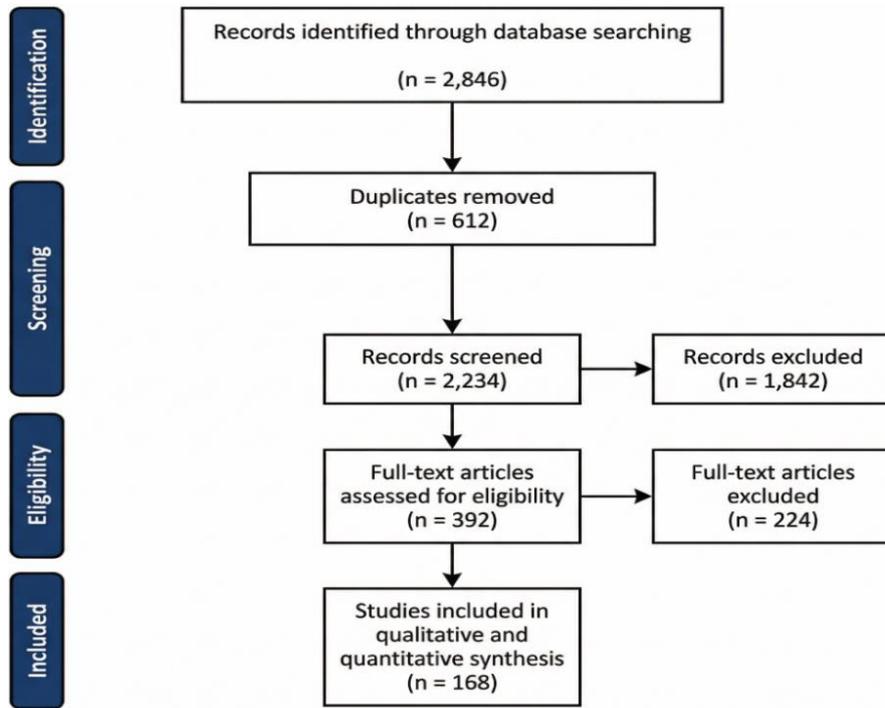


Figure 1 – PRISMA 2020 Flow Diagram

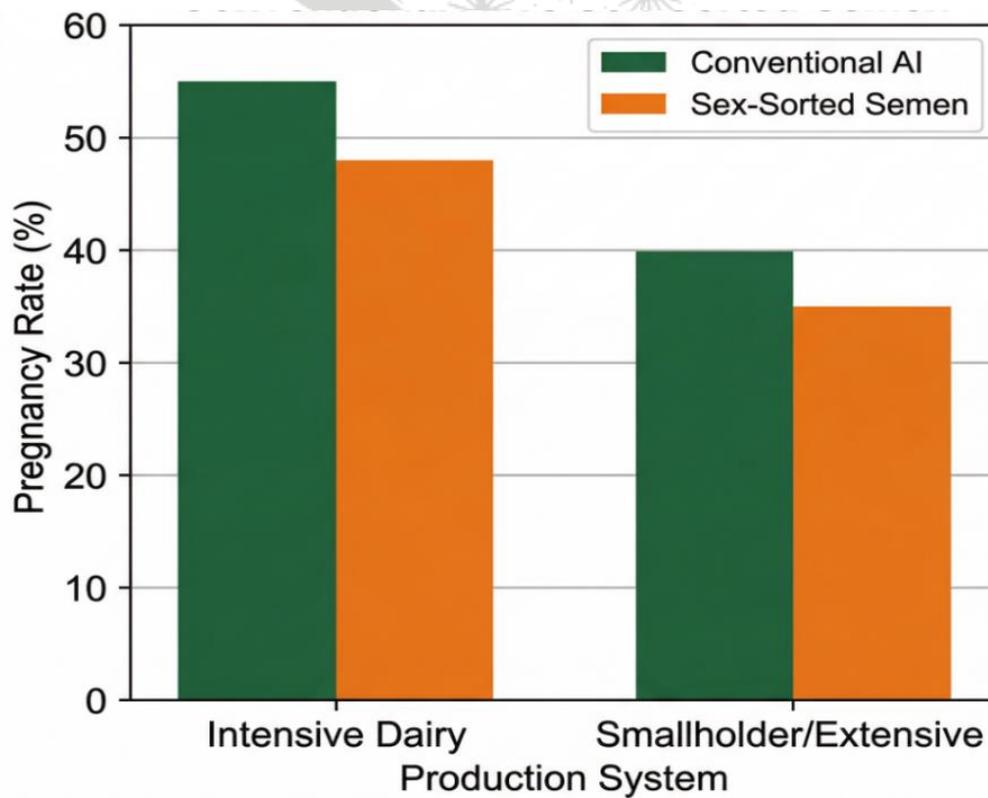


Figure 2 – AI vs Sex-Sorted Semen Pregnancy Rates

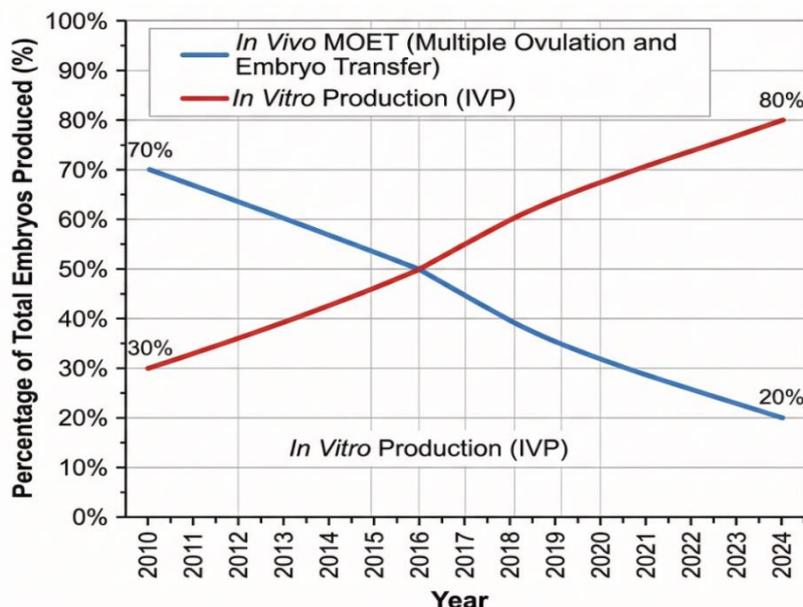


Figure 3 – Global Shift from MOET to IVP (2010–2024)

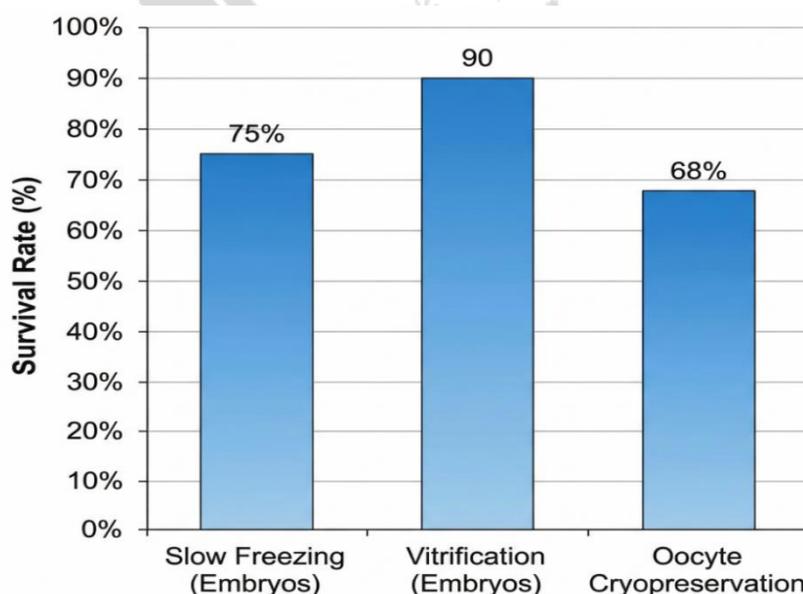


Figure 4 – Cryopreservation Survival Rates

DISCUSSION

The findings of such a systematic review illuminated the phenomenal breakthroughs and the unaddressed issues that have characterized the scenery of reproductive biotechnologies in farm animals. Studies on cattle conducted in Table 1 are reflective of the economic significance of the dairy and beef industry across the entire globe yet simultaneously are reflective of a severe shortage of diversification of the species that may constrain the generality of such technologies to the rest of the agricultural field. This species bias suggests that regardless of the fact that cattle is the principal paradigm of technological

advancement, the special reproductive physiology of small ruminants, swine, and poultry may require a special method which is not yet tested in the literature. The comparison in Table 2 between conventional artificial insemination and sex-sorted semen indicates the following complex trade-off between fertility and genetic control. Our study findings are in line with the existing meta-analysis data that indicate an average pregnancy rate of around 46 percent with flow cytometry-sorted semen with cattle registering lower rates (37.67) than buffalo (53.33) (Santos et al., 2025). The biological cost of the sorting procedure on the viability of sperm is significant as this fertility loss

with respect to conventional semen was recorded at 41% compared to 55% in the past (Reese et al., 2021). However, sex-sorted semen may still be profitable based on the economic model, when the percentage of pregnancy is above 25 per day, particularly in dairy systems with sufficiently high prices of a heifer replacement that can offset sex-sorting technology cost (Khatib et al., 2024). The combination of sex-sorted semen with the modern reproductive technologies of ovum pick-up (OPU) and in vitro embryo production (IVP) is one of the latest paradigm shifts in the sphere of reproductive biotechnology over the past decade since the number of transferable embryos per treatment under IVP was 4.2 with X-sorted semen and follicular growth-stimulated oocytes in comparison with 1.6 under MOET technologies (Akiyama et al., 2019). The current dominance of IVP where a production ratio is approximately 80 percent of the world bovine embryos (Viana, 2025) is considered to be reflective of numerous technical and economical advantages including more frequent collection (7-10 days) (compared to 60-90 days) using the MOET system), reduced hormonal requirement, and increased genetic flexibility. However, as we will discuss, there is no biological implication which this transformation does not have. Embryos generated in the IVP process also possess less cryotolerance levels as compared to those generated in vivo and it necessitates a new set of trans-ferring protocols which limit the logistical flexibility and the commercialization of international germ plasm (Palasz and Mapletoft, 1996). Additionally, the trend of high offspring syndrome and other epigenetic maladaptations in the circumstances of in vitro cultures also raises the question of long-term health and breeding fitness of IVP-based offspring (Sinclair et al., 2016). This shows that despite the short term advantages of IVP as far as reproductive efficiency is concerned, the technology remains yet to be fine-tuned so as to be utilized to record the effects of the developmental programming that can modify the generation productivity of the herd members. The success rate of somatic cell nuclear transfer (SCNT) in sheep is 1-5 -1 and there are rarely more than 10 -1 developmental defects like placental malfunction, overweight pups, and organ malformations all of which are biological bottlenecks (Liu et al., 2025). These restrictions are premised upon the fact that incomplete reprogramming of the nucleus wherein epigenetic signals of the donor cells are not properly silenced to embryonic signatures results in expression of aberrant genes during development (Rideout et al., 2001). Despite the fact that the ease of genetic modification became a lot simpler with the use of CRISPR-Cas9 gene editing, precision gene editing is accompanied by the parallel problem of mosaic in case of the edited embryos, and off-target mutagenesis and the limitation of delivery that

restricts the editing efficiencies to 4.5-31 percent of cattle and 2-72 percent of pigs respectively, depending on the platform of choice (Wang et al., 2025). Base editing and prime editing technologies are more specific but less off-target, yet still only in their early stages of development in livestock applications with a success rate of between 6.8 and 80 percent depending on species and locus of interest (Li et al., 2023).

CONCLUSION

Based on this systematic review, the reproductive biotechnologies have transformed the breeding process of livestock tremendously by enhancing the efficiency of reproductive processes, genetic gain, and accuracy in enhancing traits. Artificial insemination technologies and embryos-based technologies have reached the state of maturity and economic viability in mass production systems and the creation of embryos in a test tube has become the most important arena of pervasive genetic distribution. With the advances in the sphere of cryopreservation and the existence of genetic resources banks, biodiversity exchange and conservation process are stimulated even further. These technologies are still emerging but the adoption is still disproportionate by the region due to economic, infrastructure and environmental pressure along with fluctuating regulation. The antagonistic character of the relationship between the production and fertility characteristics of the high-producing animals highlights the importance of the mixed strategy in the breeding and management. The future will be to work on creating affordable and scalable technologies that are friendly to smallholders, work to consolidate the capacity of technology as well as in developing the single international regulatory systems. It is ultimately through responsible integration of reproductive biotechnologies that would provide a wide range of opportunities to increase productivity of the livestock, food security, and sustainability in the face of the growing global demand and pressures on global climate change.

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