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Review on Bovine Embryo Transfer

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Abstract: Over a period of approximately thirty years, commercial bovine embryo transfer has become a large international business. The technology is well established and more than 500,000 embryos are produced annually from super ovulated cows worldwide. Embryo transfer is the process by which an embryo is collected from the donor and transferred to another recipient to complete the gestation period. Bovine embryo transfer technology involves the selection and management, of donor and recipient animals and the collection and transfer of embryos within a narrow window of time following oestrus. The main use of embryo transfer in cattle has been to amplify reproductive rates of valuable females. Disease control, salvage of reproductive function, planned matting, increased farm income and researches are a few of the other benefits of embryo transfer. This technology is influencing the direction of cattle breeding industries; the numbers are small but the impact is high. However, embryo transfer is still not widely used despite its potential benefits. Embryo transfer technology is an important tool to improve livestock at faster rate as well as gives an opportunity to utilize the genetic contribution of both male and female at the same time. Commercial cattle breeders must recognize that they can benefit from well-designed embryo transfer programs providing selection criteria are appropriate for their environment and individual breeding objective.

Key words: *Economic importance · Embryo transfer · Superovulation · Synchronization*

INTRODUCTION

Bovine embryo transfer technology involves the selection and management, both physical and pharmacological, of donor and recipient animals and the collection and transfer of embryos within a narrow window of time following estrus. This technology has been incorporated into large dairy and beef cattle operations and often requires the participation of herd veterinarians [1]. An efficient reproduction in cattle herds is of great economic importance. During the past 30 years, embryo production has been a promising tool to enhance distribution of valuable genetics in different species of animals [2]. Initially much attention was placed on super ovulation, non-surgical embryo recovery from donor animals that are rapidly succeeded. Embryonic mortality in cattle is the main source of economic loss for livestock producers [3].

The first transfer of a bovine embryo was reported in 1949 and the first calf from embryo transfer in 1951 [4]. Milestones in the development of this technology have been evaluated from the point of view of their significance to our current knowledge of reproduction and to the improvement of animal agriculture [5]. Notably the development of procedures for non-surgical recovery and transfer and cryopreservation of embryos, with these improvements and a more realistic economic motivation, the industry now plays a useful role in the cattle industries of many countries [6].

By far the most common use of embryo transfer in animal production programs is the proliferation of so-called desirable phenotypes. Many breeders have identified individual females whose offspring are most saleable and used them exclusively in embryo transfer. As artificial insemination (AI) has led to the very valuable bull, embryo transfer has resulted in the very valuable female [5]. Embryo transfer has also been used to rapidly expand a limited gene pool. The production of AI bulls through embryo transfer is the most common application of planned mating [7].

In addition, new genomic techniques are being used increasingly to select embryo donors, especially for selection of dairy bull dams for super-stimulation, where a genomic analysis is becoming essential [8].

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Selection criteria for donor animals are very likely to differ depending on the reason for doing embryo transfer. Selection should be based on three criteria: genetic superiority, reproductive ability and market value of the progeny [9].The donor may be inseminated naturally or artificially and embryos will be collected non-surgically six to eight days after breeding. Following collection, embryos must be identified, evaluated and maintained in a suitable medium prior to transfer. At this point, they may also be subjected to manipulations, such as splitting and sexing and may be cooled or frozen for longer periods of storage [10].

Alternately, recipients must be synchronous with the stage of development of embryos that had been previously frozen. Initially, embryo transfers in the cow were done surgically, whereas most are done today using non-surgical methods [11]. This technology is influencing the direction of cattle breeding industries; the numbers are small but the impact is high. Commercial cattle breeders must recognize that they can benefit from well-designed embryo transfer programs providing selection criteria are appropriate for their environment and individual breeding objective [10].

Even though, embryo transfer has potential benefits, it's poorly applied throughout the world especially in developing countries which creates a pause of information.

Therefore, the objectives of this paper are:

- To review on bovine embryo transfer
- To address the economic importance of bovine embryo transfer

Historical Frame of Embryo Transfer: The first successful transfer of mammalian embryos was performed by Walter Heape in 1890. Heape transferred two four-cell Angora rabbit embryos into an inseminated Belgian doe, which subsequently gave birth to four Belgian and two Angora young. Warwick and colleagues did considerable work on embryo transfer in sheep and goats in the 1930s and 1940s [12] but, it was reported on the first successful cattle embryo transfers in 1951and the first embryo transfer calf was born in Wisconsin following the surgical transfer of an abattoir-derived day-5 embryo [5].

The bovine embryo transfer industry as we know it today arose in North America in the early 1970's [13]. Continental breeds of cattle imported into Canada were very valuable and relatively scarce because of international health and trade restrictions. Embryo transfer offered a means by which their numbers could be multiplied rapidly. The use of embryo transfer technology in cattle breeding has continued to increase (Especially within the dairy industry) over the past 30 years with the movement toward genetic improvement as opposed to the production of desirable phenotypes [14].

Application of Embryo Transfer: The main use of embryo transfer in cattle has been to amplify reproductive rates of valuable females. Because of low reproductive rates and long generation intervals, embryo transfer is especially useful in this species. Cattle may be valuable for many reasons, including scarcity, proven genetic value, or having unique characteristics such as disease resistance [14].

It is possible to obtain offspring from genetically valuable cows that have become infertile due to injury, disease, or age by means of super ovulation and embryo transfer [15] although success rates are only about one-third of those achieved with healthy, fertile donors. Infertile heifers and cows with genetically caused sub fertility should not be propagated. Although success rates are low, it is possible to recover oocytes from genetically valuable, moribund cows, fertilize them in vitro, transfer them and obtain offspring [16].

The intercontinental transport of a live animal may cost several thousands of dollars, whereas an entire herd can be transported, in the form of frozen embryos, for less than the price of a single plane fare. However, the reduced risk of infectious disease transmission is the overwhelming benefit for using embryos in international trade [17].

General Procedures of Bovine Embryo Transfer: By collecting embryos from genetically elite females and transferring the harvested embryos into females of lesser genetic merit, it is possible to produce more calves from genetically superior females and fewer calves from genetically less valuable females [18]. The donor may be inseminated naturally or artificially and embryos will be collected non-surgically six to eight days after breeding. Following collection, embryos must be identified, evaluated and maintained in a suitable medium prior to transfer. At this point, they may also be subjected to manipulations, such as splitting and sexing and may be cooled or frozen for longer periods of storage [19].

Selection and Management of Donor Cow: Healthy, cycling cattle with a history of high fertility make the most successful donors. When there is a choice, animals without calving problems, such a s retained placenta, should be used. Donors at least two months post-partum produce more embryos than those closer to calving. Young cows seem to yield slightly more usable embryos than heifers under some conditions [19].

Super Ovulation: Once the donor cow is selected the first step is to super ovulate or produce multiple ova (Eggs) for simultaneous fertilization and subsequent collection. Initially, the donor female is treated with gonadotropin hormone called follicle stimulating hormone. This hormone is administered twice daily for four days in the range of eight to fourteen days following estrus. As the result of treatment multiple follicle should be develop on the ovaries of the donor. Multiple numbers of eggs will be released at estrus, one from each follicle [20].

Artificial Insemination: Following superovulatory treatment, the donor should be observed closely for a sign of oestrus.The time when the donor is first observed in standing oestrus is the reference point for insemination treatment. Because the multiple follicles ovulate over a period of time and transport of sperm and ova is altered by superovulatory treatment, it is wise to breed more often and use more semen than normal. Freshly collected liquid semen is slightly superior to high quality frozen semen since unfrozen spermatozoa probably remain viable in the female reproductive tract longer [21].

Flushing of Embryo: In the early days of commercial bovine embryo transfer, embryos were collected surgically from the cow around Day 4 after estrus. Non-surgical techniques are preferred as they are not damaging to the reproductive tract, are repeatable and can be performed on the farm. Non-surgical techniques involve the passage of a cuffed catheter through the cervix and into one of the uterine horns on Days 6 to 8 after estrus [22].

Evaluation of Embryo: Embryos were searched under stereomicroscope with approximately 10x magnification and transferred to holding media and then they were identified, evaluated and graded according to the morphological criteria of quality and viability determined based on the International Embryo Transfer Society Manual. The cows with more than 5 transferable embryos were categorized as Rank A and those with less than 5 transferable embryos were categorized as Rank B [23].

Preparation of Recipient Cows: Proper recipient herd management is critical to embryo transfer success. Cows that are reproductively sound, that exhibit calving ease and that have good milking and mothering ability are recipient prospects. They must be on a proper plane of nutrition (Body condition score 6 for beef cows and dairy body condition score 3 to 4 for dairy breed recipients.) These cows also must be on a sound herd health program. Acceptable pregnancy rates in embryo transfer are partially dependent upon the onset of estrus in the recipient being within 24 hours of synchrony with that of the embryo donor [19].

Recipients can be selected for an embryo transfer program by detection of natural estrus in untreated animals or by detection after drug-induced estrus synchronization. Regardless of the method of synchronization used, timing and critical attention to estrus detection are important. Recipients synchronized with PGF must be treated 12 to 24 hours before donor cows because PGF-induced estrus will occur in recipients in 60 to 72hours and in super ovulated donors in 36 to 48 hours [23].

Embryo Transfer: Both surgical and non-surgical methods of embryo transfer can be made to work well. Under most circumstances, non-surgical transfer is greatly preferred, although surgical transfer can be done quite rapidly, even in rather primitive circumstances. Surgically embryos can be transferred via mid-line abdominal incision to cows under general anaesthesia; in most circumstances a flank incision is far more practical. Recipients are placed in squeeze chutes that give access to either flank [21].

Non-surgical embryos are to be transferred fresh and loaded directly into 0.25 cc plastic straws and transferred using a rod similar to an artificial insemination rod. Most AI rods are designed to use 0.5 cc straws, but those used to inseminate with sex-sorted semen are designed for 0.25 cc straws [24].

Economic Importance of Bovine Embryo Transfer: In approximately 40 years, commercial embryo transfer in cattle has become a well-established industry with more than 500,000 embryos being transferred on an annual basis throughout the world [25]. Although this results in a very small number of offspring on an annual basis, its impact is large because of the quality of animals being produced. Embryo transfer is now being used for real genetic improvement, especially in the dairy industry and most semen used today comes from bulls produced by embryo transfer [25].

Faster Genetic Improvement: The most important factor in increasing the rate of genetic improvement is shortening the generation interval. From 1980 to 2000 pregnancy rates declined 6% in dairy cattle which is equal to a 24 days increase from calving to conception.Genetic progress has generally been considered to be slower with embryo transfer than with conventional artificial insemination, especially on a national herd basis. However, with increased selection intensity and shortened generation intervals, i.e., transferring female offspring, genetic gain can be made on a within-herd basis [26].

Increased Farm Income: Reproduction can have a multitude of impacts`on a farm, from altering culling policies, increasing retention of better replacements, moving primiparous cows into a more productive the second lactation and improving milk production. Because production accounts for more than 88% of the gross income of a dairy farm [27] it is no surprise that most attention paid to improvements in reproduction evolve around altering milk production during the productive life of cows. In most cases, altering milk production has to be considered per day of calving interval, as improvements in reproduction increase the time a cow spends in the dry period, which is considered a nonproductive stage of the lactation cycle. Improving reproduction oftentimes results in greater availability of replacement animals, which increases herd turnover [28].

Planned Mating: As AI has led to the very valuable bull, embryo transfer has resulted in the very valuable female. Many breeders have identified individual females whose offspring are most saleable and used them exclusively in embryo transfer. Embryo transfer has also been used to rapidly expand a limited gene pool. The dramatic rise of the embryo transfer industry in Canada in the early 1970's was a direct result of the introduction of European breeds of cattle, which were then in short supply. The production of AI bulls through embryo transfer is the most common application of planned mating [1].

Disease Control: Several large studies have now shown that the bovine embryo does not transmit infectious diseases should be noted that none of the infectious diseases studied has been transmitted by *in vivo*-produced bovine embryo provided embryo handling procedures were followed correctly. Consequently, it has been suggested that embryo transfer be used to salvage genetic material in the event of a disease outbreak, which could be a useful alternative in establishing disease-free herd [29].

Import and Export of Embryo: The intercontinental transport of live animals costs several thousands of dollars, whereas an entire herd can be transported, in the form of frozen embryos, for less than the price of a single plane fare [25].

Research: Embryo transfer techniques have proven to be a very useful research tool. In fact, many technical developments in embryo transfer before 1970 were directed toward research purposes rather than for the propagation of superior livestock [11]. These studies included natural limitations to twin pregnancies, uterine capacity, endocrine control of uterine environment, maternal recognition of pregnancy, embryo-endometrium interactions and the endocrinology of pregnancy. Studies that were originally planned to answer basic physiological questions are now being used to improve and increase the utilization of embryo transfer. Newer techniques have added an entirely new perspective to the utilization of embryo transfer for research purposes. The production of identical twins, clones and chimeras will certainly advance many of these sciences. The IVF techniques are being used to study the fertilizing capacity of semen and IVF techniques are of immense value in the study of oocyte competence and embryo metabolism [1].

Application of Bovine Embryo Transfer in Developing Countries: After artificial insemination and oestrus synchronization, embryo transfer is the third most commonly used biotechnology [30]. Embryo transfer from one mother to a surrogate mother makes it possible to produce several livestock progenies from a superior female. However, embryo transfer is still not widely used despite its potential benefits. In developing countries this is mainly due to absence of the necessary facilities and infrastructure. An evaluation of country reports [31] shows that only five of the African countries providing information (Cote d?Ivoire, Kenya, Madagascar, Zambia and Zimbabwe) use ET technology, all on a very limited scale. The use of ET also been independently reported in South Africa [32]. The first successful embryo transfer in Ethiopia, resulted in the birth of a Holstein-jersey calf at the Adami Tulu Animal Research Center in the beginning of May, 2010 and five more calves had born. In April 2010, eighty frozen embryos that were imported the previous August were implanted in the native cows [33].

Embryo transfer increases reproductive rate of selected females, reduces disease transfer and facilitates the development of rare and economically important genetic stocks as well as the production of several closely related and genetically similar individuals that are important in livestock breeding research [34].

Challenges of Embryo Transfer in Developing Countries: The constraints and limitation of biotechnology in animal production in developing countries are due to factors such as the poor conditions of the human population in such countries that include poverty, malnutrition, disease, poor hygiene and unemployment [35].

The major constraints of animal biotechnology includes: insufficient access to land and other productive resources, unfavourable terms of trade for food products, especially for animal products, lack of database on livestock and animal owners in most of the developing world, uniqueness of animal breeds in developing world, lack of trained scientists, technicians and field-workers, absence of coordination between industry, universities and institutions for technology transfer, expensive technology to be purchased from developed world, high cost of technological inputs, poor bio-safety measures of biotechnology developed in developing countries, negligible investment in animal biotechnology in Africa, lack of clear policy and commitment from the government, disregards for indigenous knowledge and local agricultural resources management [36].

CONCLUSION

During the past 30 years, embryo production has been a promising tool to enhance distribution of valuable genetics in different species of animals. Although embryo transfer is generally costly in both developed and developing countries it is frequently still profitable. However, embryo transfer is still not widely used despite its potential benefits. In developing countries such as Ethiopia, this is mainly due to absence of the necessary facilities and infrastructure.

Therefore, on the basis of this conclusive remark, the following recommendations are forwarded:

- There should be well trained technician for embryo transfer technology.
- There should be of clear policy and commitment from the government about embryo transfer technology.
- Awareness should be created in the community and companies about the profitability of embryo transfer.
- Coordination should exist between industry, universities and institutions for technology transfer.
- Investors should be encouraged to invest on embryo transfer technology.

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