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Embryo Collection and Transfer Options for Beef Producers¹

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Introduction

Methods to accelerated genetic progress within a beef herd can include Artificial Insemination (AI) and Embryo Transfer (ET), but, when one considers the impact of reproductive technologies in the cattle industry, embryo collection, storage and transfer (ET) is probably not the first technology that comes to mind. AI has enabled dairy and beef producers to effectively utilize superior genetics of a single bull with little costs. Unlike AI, ET has not been used as extensively in beef industry, because of the extra cost associated with additional procedures (compared to AI) to collect and store embryos. However, compared to AI, ET offers seed stock producers a greater opportunity to propagate and disseminate superior maternal genetics. Under natural cover or an AI environment, cows and heifers are reproductively limited to one offspring per year. However, once a superior female has been identified, the process of super ovulation and ET potentiates multiple offspring from different sires, thus effectively exploiting

the genetics of a single female, which would have not otherwise occurred. Because ET has been perceived by producers as a rather difficult and expensive technology to use, the objective of this publication is to introduce beef producers to the procedures, equipment and relative costs associated with different ET programs with aims to expand this reproductive technology use in the beef industry.

Embryo Transfer Programs

There are five basic ET programs that beef producers can participate in.

1. Collect your own donor cow-freeze embryos and transfer at a later date.
2. Collect your own donor cow and transfer fresh embryos.
3. Purchase frozen direct transfer (DT) embryos and transfer into recipient cows (owned or purchased).
4. Buy embryos, and have an ET service thaw and transfer into recipient cows (owned or purchased) at their facility.
5. Buy females carrying ET derived calves.

Employment and program opportunities are offered to all people regardless of race, color, national origin, gender, age, or disability. North Carolina State University, North Carolina A & T State University, US Department of Agriculture and local governments cooperating.

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These programs are listed in a quantitative order of the time and average monetary investment when considering participation in all five programs. With the first three, heat detection and embryo-recipient synchrony are critical. Most embryos are flushed from cows about 6 or 7 days after ovulation induction, and thus, the transplantation of the embryos must take place into a recipient cow 6 or 7 days after the first signs of heat. The last two options can be the least expensive and easiest way to get embryo transfer calves on the ground. Unfortunately with the increasing popularity of ET, the final value of some recipient females is far surpassing the cost involved in conducting the first three programs listed.

ET Procedures

Superovulation, Breeding and Embryo Recovery

Programs one and two involve selecting a donor cow, watching her for heat and then superovulating her with a series of hormonal (gonadotropins) injections during mid portion of her estrous cycle. Following this series of injections, prostaglandins are also injected to cause the regression of the corpus luteum. This triggers estrus one and a half to two days later. The donor is then artificially inseminated 1 to 4 times 12 to 24 h after observed signs of standing heat. Variations to the breeding schedule will be used depending on the history of the donor, quality of semen used, and cost of semen. Factors important in superovulation include (1) the gonadotropin preparation and method of administration; (2) adjunct treatments aimed at controlling follicle growth and ovulation; (3) the genetics of the donor animal and the environment in which it is maintained; and (4) bull effects.

Non-surgical techniques are generally used to recover embryos seven or eight days after breeding. The donor is given an epidural block in the area of the tailhead to prevent straining. The vulvar area is scrubbed to prevent contamination. A flexible rubber tube catheter is gently threaded through the cervix and up into the uterine horns. A small rubber balloon

cuff is inflated with fluid to expand and fill the uterine lumen. This prevents any backflow of fluid. There are several holes near the tip of the catheter that precede the inflated cuff. Fluid enters the uterus by means of these holes after being injected through the catheter with a syringe. Once the fluid is in the uterus, the horn is gently massaged and the fluid (containing the embryos) is drawn back out through the catheter. This process is then repeated in the other uterine horn. The embryos are collected in a physiological saline. The embryos are searched for under a microscope and once located can be either frozen or loaded into an insemination straw and transferred into the recipient cows. In the case of fresh transfer, the receipts should be synchronized right along with the donor. Schedule recipient synchronization (see synchronization protocols) to coincide with the donor if fresh embryos are to be transferred. With frozen embryo transfer this event does not need to be synchronized.

Freezing and Thawing Embryos

There are two types of procedures to freeze and thaw embryos for transfer: 1) Thawing and transferring or 2) direct transfer. Freezing needs to be done in a lab environment and requires some specialized equipment. Traditionally, embryos are frozen with a cryoprotectant called glycerol. Even though glycerol protects the embryo during the freezing process, it is lethal to the embryo if not removed before transfer. Once the embryos are frozen the thawing procedure requires 3 or 4 steps of rehydration (washing) to remove the glycerol before transferring. This procedure can take 15 to 20 minutes.

More recently, some ET services have switched to a procedure called direct transfer of embryos (DT). Collection procedures are similar to the traditional method; however embryos are frozen in ethylene glycol instead of glycerol. Ethylene glycol is safe enough to allow for the frozen egg to be directly transferred from liquid nitrogen to the recipient without going through a laboratory thawing and washing procedure. Because of

this advancement, some AI technicians are able to transfer frozen embryos since the procedure is much like AI with the exception that the embryo is placed much further up in the reproductive tract. For DT of embryos, skills in palpation are essential and the success can vary significantly depending on the experience of the technician because it is necessary to identify the ovary with the corpus luteum and place the embryo in the uterine horn on the same side as the CL. Although, DT eliminates the need for extensive thawing equipment, pregnancy rates may not be as good when compared to traditional transfer procedures. This must be considered at the time of freezing because embryos frozen with glycerol cannot be directly transferred with much success.

Transfer to Recipient Cows

For all types of ET, cows with proven reproductive performance should be used as recipients as to avoid complications of puberty, estrus synchronization and potential calving difficulties of heifers. Recipients should be healthy, and have received the appropriate vaccinations as advised by your veterinarian. Furthermore, recipient females must be on a gaining plane of nutrition and cycling regularly.

When transferring frozen embryos to one or two females, heat synchronization is usually not necessary. Briefly estrus detection is performed twice daily (AM and PM) in open females and following detection the time and date are recorded. On day 6 or 7 after estrus was detected, frozen embryos that were collected at a “synchronized” date and time (i.e. d 7 AM) are thawed and transferred to the recipient female.

Unless you put the embryos in yourself, and can afford the time to ET without prior scheduling, successful estrus synchronization procedures need to be established. In operations with large numbers of cows, synchronization can be avoided by using an electronic “heat watch” system. These types of estrus detection systems have been used with good success in some programs and especially with the transfer of frozen embryos. However, there are extra costs associated with this type

of program.

Transferring large numbers of embryos or fresh embryos requires an effective estrus synchronization program for recipient females. There are basically four estrus synchronization programs that could be used with estrus occurring 36-72 hours after the last procedure (stars indicate effectiveness and labor requirement with four stars being the best):

1. Single shot of Prostaglandin *
2. Two shots of Prostaglandin 12 days apart ***
3. Progesterone vaginal insert for 6-7 days followed by Prostaglandin **
4. GnRH followed by Prostaglandin on day 7 ****

Estrus synchronization is most critical when transferring fresh embryos to ensure that donor embryos are collected and transferred to the recipient female during the same physiological time of the estrous cycle (i.e. day 6 AM). If large numbers of embryos are to be transferred, synchronization of recipient females simply requires less handling, labor and if transfer service is used, a reduction in transfer costs.

Embryos to be transferred non-surgically are aspirated into 0.25-ml artificial insemination straws. Air bubbles are placed on either side of the fluid containing the embryo. The straw is fitted into a cassette gun, which is covered with a plastic breeding sheath. The recipient is placed in a squeeze chute and given an epidural. After being palpated for a corpus luteum, the cassette gun is inserted mid-way up the uterine horn (same side as the corpus luteum) of the recipient and the embryo is deposited there. Again, this technique takes considerable practice to achieve satisfactory pregnancy rates.

Data from a well managed ET program suggests that from the average donor, approximately eight to ten embryos can be collected. Several of these may be unfertilized or degenerate and approximately six to seven embryos are of transferable quality. On average, four or five pregnancies will result after a transfer. Approximately 65% of fresh and 60% of frozen embryos result in

pregnancies. However, there are many factors that can influence ET success rates, including: super ovulation response, embryo quality and handling, transfer synchrony between donor and recipient, recipient fertility and technician proficiency. Obviously, when purchasing recipient cows they should be guaranteed pregnant and although they may cost a little bit more, it ensures an embryo transplant calf.

Costs

You can expect the total cost per calf born to vary considerably between the described programs. Much of the costs will depend on professional fees and services for the described procedures. One of the ways producers can reduce costs in ET programs is to contract ET services on a per live calf born basis. Most services, however, will charge a separate fee for each service they provide; i.e. cost per embryo collected, stored or transferred. Contact our extension office for a complete list of individuals or embryo transfer companies. Purchasing individual embryos or recipient females carrying ET calves can be more or less expensive than collecting your own, depending the genetic value and demand of the proposed offspring from a planned mating. It is also important to consider that some embryos may or may not carry a pregnancy guarantee (i.e. purchase 5 guaranteed pregnancies vs. 5 embryos). If a pregnancy guarantee is associated with the embryos, a premium will likely be charged for the guarantee. Finally, if you plan on registering ET derived calves (i.e. purebred), you will have to do blood typing on the donor and receipts to identify the parentage of the calf. These are additional costs and your breed association generally provides these services indirectly if you submit blood samples.

Suggested Readings

- May, P. Bovine Embryo Transfer. *Vet Rec.* 1996 Mar 16; 138(11): 262.
- Vanderbloom, R. J., T. C. McCauley, R. Tappan, and R. L. Ax. 1997. Bovine Reproductive Biotechnology. In: *Current Therapy in Large Animal Theriogenology*. R. S. Youngquist, Ed. W.B., Saunders Company, Philadelphia.

Summary table of embryo transfer programs options for beef producers.

Program	Resources^a	Procedures Involved	Cost Factors^b	Specific Factors Influencing Success
1. Collect your own donor cow-freeze embryos and transfer at a later date to recipient cows.	- Cattle working facilities - Super ovulation drugs and injection equipment - AI equipment and Liquid Nitrogen storage tank - ET Services - Recipient Cows	- Super ovulation - Artificial Insemination - Embryo Collection - Embryo Freezing	- Service fees per flush (\$100-300) - Freeze fee (\$50-100/embryo) - Transfer fee (\$50-150/embryo) - Semen (\$4-100) - Ovulation Drugs (\$15-30)	- Donor Fertility - Recipient Fertility - Embryo stage/recipient female synchrony. - Estrus detection skills - Embryo quality
2. Collect your own donor cow and transfer fresh embryos	Same as Program 1	- Super ovulation - Artificial Insemination - Embryo Collection - Recipient estrus sync.	- Service fees (\$100-300/flush) - Transfer fee (\$50-150/embryo) - Semen (\$4-100) - Ovulation Drugs (\$15-30) - Sync. Drugs (\$2-12/cow)	- Donor and recipient female estrus synchrony. - Estrus detection skills - Same as program 1
3. Purchase frozen DT embryos and transfer into recipient cows (owed or purchased).	- Cattle working facilities - AI and Storage equipment - Recipient Cows	- Recipient estrus Sync. - Embryo Transfer	- Embryos (\$50-1000+) - Transfer fee (\$50-150/embryo) - Sync. Drugs (\$2-12/cow)	- Estrus Detection and Synchronization - Embryo Quality - Technician experience
4. Buy frozen embryos/ET service thaw and transfer into recipient cows (owned or purchased) at their facility	- ET Services - Recipient Cows	N/A	- Embryos (\$50-1000+) - Transfer fee (\$50-150/embryo) - Sync. Drugs (\$2-12/cow) - Recipient Cows (\$500-?)	- Estrus Detection and Synchronization - Embryo Quality - Transfer procedures
5. Buy females carrying ET derived calves.	Not Applicable (N/A)	N/A	- Pregnant Recipient female (\$500- ?)	- Same as above plus - Successful pregnancy (health) - Dystocia

^a Programs can be conducted on the producers farm or donor cows may be brought to ET service facilities where a daily boarding fee will be charged in addition to the service fees.

^b Cost ranges are based on available advertised rates and could be considerable more or less. ET services may also charge travel expenses if procedures will be conducted on the farm. Transfer fees may be eliminated in DT program if a producer transfers embryos (extensive training recommended).