

## Lesson 2: Embryo Transfer Technologies

Today's science is tomorrow's applied technology. Embryo transfer is a good example of this trend. Once a complicated procedure requiring surgery, it has become a technology that some livestock producers are using in their own barns. This lesson will describe how embryo transfer is done, as well as some more advanced embryo manipulation methods, such as cloning.

### **The Process of Embryo Transfer**

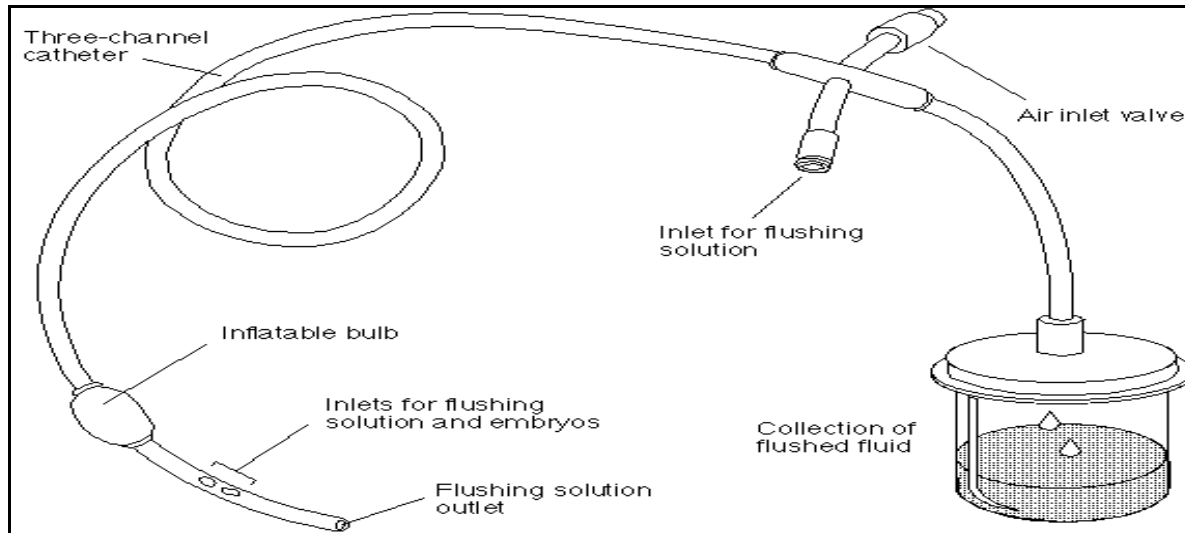
Embryo transfer (ET) is the process of transplanting embryos (fertilized eggs) from a donor female to a recipient female. Although ET is possible in several species of livestock, including sheep, goats, horses and swine, it is most common in cattle, which will be the focus of this lesson. The embryo transfer process has six steps. The first step involves the synchronization of estrous in the donor and recipient, which makes it possible for the collected embryos to be transferred to the recipient without being frozen. Next, the donor must be superovulated. Injecting the donor with a hormone like prostaglandin causes superovulation, or the release of multiple ova. The third step involves breeding the donor cow, either naturally or by artificial insemination. Next, the fertilized ova are collected from the donor through a process called embryo flushing in which fluid is used to wash the embryos out of the female reproductive tract. The fifth step in the embryo transfer process involves isolating and examining the embryos. Healthy embryos are transferred to recipients or frozen for later transfer.

### **Advantages of Embryo Transfer**

Embryo transfer has several distinct advantages over natural breeding. It can increase the reproductive potential of superior females by allowing the female to produce multiple offspring each year. In addition, embryo transfer increases the rate of genetic improvement in a herd. The average cow can produce four to five calves per year using embryo transfer technology. With a superior cow providing four to five calves per year, high quality herd replacement heifers and bulls accumulate faster. Another advantage of ET involves progeny testing, in which offspring are evaluated for growth characteristics to determine whether an animal produces quality offspring. Using ET, female animals can be more easily and accurately progeny tested since they produce offspring more quickly; in three years, a cow should produce the ten calves needed for progeny rating. Finally, since shipping live animals internationally is a difficult and expensive process, embryo transfer has been employed as a way to use breeding stock from other countries. ET has been used to import and export rare breeds and the offspring of genetically superior animals.

### **Embryo Transfer Equipment**

Embryo transfer requires the use of some specialized equipment. The equipment needed to flush the donor cow includes a special catheter that has three narrow tubes encased in one long tube (see Figure 2.1). One tube inflates the bulb found near the end of the catheter. Another tube injects the flushing solution into the uterine horn to flush out the embryos. The third tube collects the flushing solution and embryos from the donor's reproductive tract. The catheter is inserted into the vagina and through the cervix with a device called a stylet. A collection cylinder holds the flushing solution and embryos after they have been removed from the donor. The technician needs a shoulder-length glove and lubricant to palpate the donor and recipient. Syringes are needed to inflate the bulb in the catheter, inject hormones, and give a local anesthesia. The equipment needed to examine the embryos includes a microscope, a pipette embryo-handling tool, and a gridded search dish. Transferring the embryo to the recipient requires the use of a plastic embryo straw, which holds the embryo for transfer, and an embryo transfer gun, which is used to expel the embryo into the reproductive tract.



### Management of Superovulation

When prostaglandin is used to manipulate the heat cycle, preparation of the donor cow begins ten days after she is in standing heat with an injection of the hormone FSH, or follicle stimulating hormone. The injection of large amounts of FSH causes the ovaries of the donor to release multiple ova. These injections are given once in the morning and once in the evening, until a total of seven injections have been given. On the third day of the procedure, prostaglandin is injected into the donor in the morning and evening. These two injections will cause the donor to come into estrus in 48 hours, and she can then be bred either naturally or through artificial insemination. The management of the process of superovulation is slightly different when other hormones are used.

### The Embryo Flush Process

The embryo flush process is done seven days after breeding. The technician injects an epidural anesthesia into the space between two cervical vertebrae. The epidural causes the rectal muscles to relax, which aids in the insertion of the technician's hand into the rectal tract to guide the catheter. The technician uses the stylet to insert the special catheter into the vagina, through the cervix, and into the right uterine horn. He or she must palpate the donor carefully to guide the catheter into the proper location. The bulb near the end of the catheter is inflated to block off the uterine horn while it is being flushed.

A sterile flushing solution is allowed to flow into the uterine horn under the force of gravity until 500 milliliters of the solution is in the horn. The technician then starts to massage it to loosen the embryos. When the uterine horn is filled with flushing fluid, the technician opens the outlet tube of the catheter and collects the fluid and embryos in the collection cylinder. This process is repeated with the left uterine horn.

Because of their weight, the embryos settle to the bottom of the collection cylinder. The fluid above the embryos is carefully siphoned off. The embryos are ready to be counted and characterized, or examined for quality. They must be normal in appearance and of the correct size to be usable.

### Transferring the Embryo into the Recipient

After the collected embryos have been washed and examined, technicians load embryos that are to be transferred to recipients into embryo straws. They are then prepared to be either transferred into recipient cows or frozen in a container of liquid nitrogen at -320 degrees Fahrenheit. The recipients have already been prepared to receive the embryos through estrous synchronization. A technician loads an embryo transfer gun

with a straw and inserts it into the recipient cow's vagina. He or she guides it through the cervix and into the uterus, where the embryo is expelled.

### **Cloning**

Cloning is the asexual reproduction of an organism in which the resulting organisms are identical. The livestock industry uses two basic methods of cloning. In the first method, the researcher physically splits the embryo into two halves as it is dividing. Each half is transferred to a recipient and develops normally. Embryo splitting doubles the number of embryos available for transfer.

Nuclear transfer is a second method of cloning. Nuclear transfer involves removing the nucleus of an unfertilized ovum. A cell is then extracted from a parent organism and fused into the ovum without a nucleus using an electrical pulse. The new cell has a diploid number of chromosomes and will develop as if it were a natural embryo. However, it must be stimulated to act like a fertilized ova and begin dividing. Nuclear transfer technology can multiply the number of embryos by 16, 32 or even 64, depending on the number of cells available in the parent embryo.

### **Benefits of Cloning**

Cloning has several advantages. It can increase the number of highly prized animals produced because it multiplies the number of collected embryos. Cloned animals are valuable to researchers doing live animal experiments. Fewer animals can be used in these tests because all of the animals--control animals and experimental animals--are identical. Animals genetically altered to produce pharmaceuticals could be cloned as well, which would reduce the cost of producing the animals.

### **In Vitro Fertilization**

In vitro literally means "in glass." In vitro fertilization (IVF) is a process in which immature follicles (ova) are collected from the ovaries of a female animal, stimulated to mature, and fertilized outside the female reproductive tract in a test tube. The fertilized embryos can be transferred to recipient animals.

The process of IVF begins a few days before ovulation. A technician inserts a special probe containing an ultrasound sensor into the vagina and moves one of the ovaries to a position directly above the vaginal wall. The ultrasound equipment is then used to locate the follicles. The technician inserts a needle through the vaginal wall and into the ovary. The needle is attached to a vacuum device that sucks the follicles into a collection bottle. The follicles that are collected are stimulated to mature in the laboratory and are then fertilized.

### **Summary**

Embryo transfer, which has only been in use since the early 1980s, is a relatively new technology in the livestock industry, but the number of dairy and beef cattle producers using it is growing rapidly. Embryo transfer is the process of transplanting embryos from a donor female to a recipient. The procedure involves superovulating a donor, flushing the embryos out of the donor, and transferring the collected embryos to a recipient. More advanced techniques such as embryo splitting, nuclear transfer, and in vitro fertilization are also beginning to be used in the livestock industry.

### **Credits**

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