

## IN VITRO FERTILIZATION: A POWERFUL TOOL

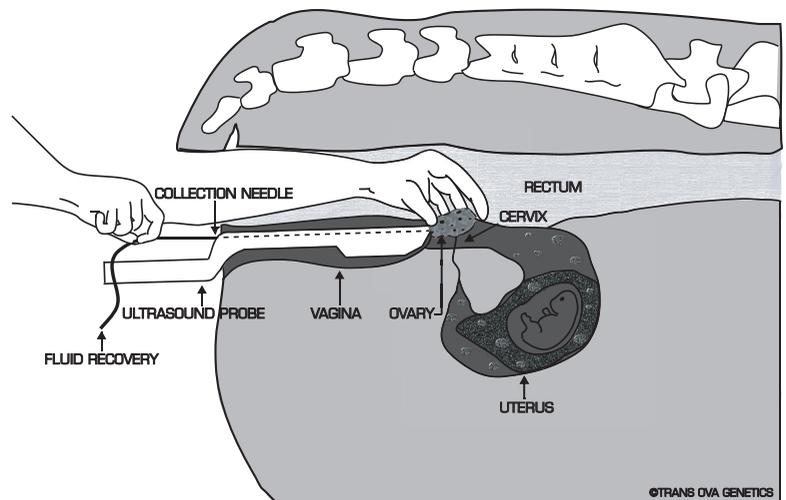
During the past 50 years technological advances in the field of bovine reproduction have led to some dramatic changes in the way cattle look, reproduce, perform, and even taste. Artificial Insemination (AI) and conventional Embryo Transfer (ET) are two of the main tools used by progressive producers to accelerate genetic improvement within their herds. However, recently many producers have introduced In-Vitro Fertilization (IVF) into their reproductive programs. Early on, IVF struggled to establish itself as a legitimate technology because embryo production and pregnancy rates were low, which made the cost of a live calf unacceptable to most. At that point IVF was used as a last chance option for donor cows that could not produce pregnancies through other means – but now the technology has improved to such a level that it has become a very practical and competitive reproductive tool with numerous applications for seedstock and commercial operations alike.

### HOW DOES IVF WORK?

In-Vitro Fertilization resembles conventional ET by allowing cows to produce multiple calves per year. This is accomplished by generating embryos from elite cows called donors and transferring them into cows with less genetic merit called recipients. After this, there are many differences in the way the procedure works.

In IVF, oocytes (unfertilized eggs) are collected using a specially designed probe, fitted with an ultrasound transducer which allows the ovary to be visualized during the aspiration procedure. The technician locates and stabilizes the ovary via rectal palpation of the reproductive tract. The aspiration probe is inserted vaginally where the transducer and ovary meet with only the vaginal wall separating the two. The probe has an attached needle guide that houses a 55cm long needle. This needle will transverse the vaginal membrane and is inserted into fluid filled follicles, containing oocytes, located on the ovary. The contents of the follicles are aspirated out and captured in a searchable filter. The filter is then taken into the lab where it is rinsed and searched using a microscope; the oocytes are then retrieved, counted, and graded.

#### TRANSVAGINAL OOCYTE RECOVERY



Once the oocytes have been processed they are moved into dishes with special media designed to mature them. The dishes are placed in an incubator for 18 to 24 hours where the maturation process takes place. The following day, semen is added to the dishes containing the matured oocytes and fertilization takes place. The media and incubator are designed to mimic the cow's uterine environment as pertains to temperature, pH, etc. Following fertilization, the oocytes are left in the incubators for the next 7 days as a percentage of them develop into embryos. They can then be evaluated under a microscope and quality graded just as you would conventional ET embryos. The grade one and two embryos are now ready to be loaded into transfer straws and implanted into recipient cows that were in standing heat 7-8 days prior to the transfer date or frozen for transfer at a later date.

## WHAT ARE THE POTENTIAL APPLICATIONS?

**1. Embryo production from pregnant donors** — Because the cervix and uterus are not penetrated during the process of aspiration, oocytes can still be collected without disturbing the fetus. Donors can be safely aspirated from about 45-100 days of gestation. On rare occasions donors can be aspirated up to 6 months of gestation, as long as the ovaries are accessible to the technician. This application is a good alternative for operations wanting to get a jump on the next generation. In years past, breeders were forced to decide whether to risk future productivity of young donors by flushing them as virgins or just postponing embryo production until after their first calf. By getting the heifers pregnant first, they are already on their way to calving in a normal window with their counter parts. It also works well for those operations that want to keep donors on a 365-day calving interval, and can help reduce reproductive failures such as cysts in donors that remain open for long periods of time.



**2. Production from donors that do not make embryos in ET** — Various reasons can cause donors to be non-productive in ET. Blockages, scarring, uterine infections, and un-passable cervixes are some of the more common reproductive tract abnormalities that can be overcome through IVF. Also, donors prone to overstimulation or those that produce a high percentage of unfertilized eggs, generally perform well in IVF. Donors that typically fail to stimulate and ovulate little to no ova will not necessarily benefit from IVF.

**3. Adding value to semen** —With IVF, less semen is needed than in a typical AI or ET procedure because oocytes are contained in small dishes during the fertilization period. This allows for tremendous opportunities. Depending on quality, one straw of conventional semen can fertilize oocytes from as many as 15 donors. This allows breeders to maximize the effectiveness of rare or expensive semen. If sex selected pregnancies are desired the IVF system offers distinct advantages over conventional ET. Quality sexed frozen semen tends to be more effective in IVF because less sperm cells are necessary. Generally 1-2 straws are sufficient per donor, depending on how many oocytes she has produced. Semen does not have to be sexed prior to freezing in order to get sexed pregnancies. Conventionally frozen semen can be sorted for the desired sex prior to fertilization in the IVF process – this is termed reverse sorting. It should be noted that not all bulls will work after reverse sorting. Also, oocytes from high producing donors can be placed in separate dishes making it possible to use more than one sire and still be able to identify the matings of the resulting embryos.

**4. Short interval between procedures** —The IVF procedure can be repeated more often than conventional ET. Donors are routinely placed on bi-weekly schedules. During a 90 day period a donor could be aspirated up to 6 times. Within a given time frame, more total pregnancies can be created through an IVF program when compared to ET.

**5. Other applications** — Because donors do not have to be cycling in order to perform the IVF procedure, embryos can be created from cows that have not yet returned to estrus post-partum and heifers that have not reached puberty. Oocytes can also be recovered from the ovaries of slaughtered females or donors prior to a death event.

## WHAT ARE THE DISADVANTAGES?

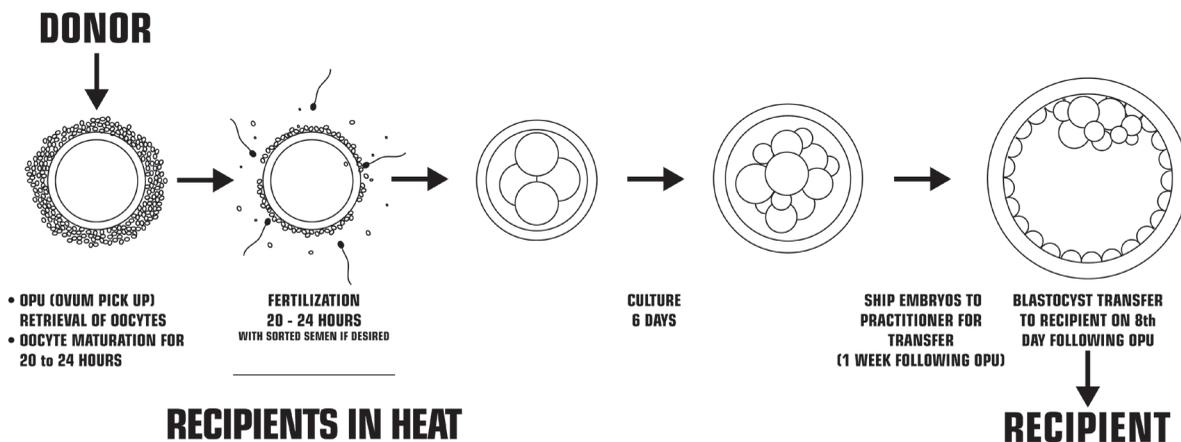


No doubt, the best environment to develop bovine embryos is within the uterus of the cow. Therefore, embryos developed in a lab are not quite as hearty as those from ET. Lower pregnancy rates are to be expected and the freezeability of the embryos is compromised. The industry has not developed a standard method to cryopreserve IVF embryos that will produce acceptable results and most companies will advocate fresh transfers only. However, Trans Ova Genetics has developed methods for freezing IVF embryos and has achieved favorable pregnancy rates. For more information about IVF freezing methods consult with a Trans Ova Genetics' client service representative. Some instances of Large Offspring Syndrome (calves born abnormally large) can occasionally occur. These instances are very rare and generally correlate with matings where extreme birth weights are expected.

## WHAT SHOULD YOU EXPECT FOR RESULTS?

Oocyte recovery should range from 12-30 oocytes per procedure and will vary by breed, age, health, nutritional status, and cow-to-cow variation. Most high-quality conventional semen has a development rate of approximately 30%. For example, if a cow has 20 oocytes you could expect around 6 transferable embryos. IVF embryos transferred fresh should yield on average a 50% pregnancy rate in well-managed recipients. You can expect a slight decrease in pregnancy rate from frozen IVF embryos compared to embryos transferred fresh. Ten to 15% of IVF procedures will result in zero useable embryos. These results represent averages; individual results can be higher or lower than the average.

## IVF (IN VITRO FERTILIZATION) TIMELINE



## DESIGNING YOUR OWN PROGRAM?

Some breeders are using IVF to maximize the impact of their very best cow. One donor currently in the clinic has been worked 10 times in approximately a 5-month period and has made 80 pregnancies. This also happens to be a donor that came in because she was no longer making embryos in ET. Another client wanted to rapidly increase his herd size. In a 2 ½ month period he aspirated 10 heifers 5 times each and used the reverse sorting technology to produce females. He ended up with 103 female pregnancies. Another set of clients wanted to maximize the use of a rare straw of semen. They aspirated 9 total cows and fertilized the oocytes with one straw of semen, resulting in 252 fresh and frozen embryos. These are not typical results but are used as testimony of how powerful a tool IVF can be in a reproductive program.

Depending on the specific needs of your program, various approaches can be made to accomplish your goals. The main thing is to understand exactly how the technology can be used to benefit your operation. In-Vitro technology is not for everyone or every cow, but more and more cattlemen and women are realizing that IVF has the potential to do things never before possible through other reproductive technologies.



## QUESTIONS?

Contact a Trans Ova Genetics client service representative:

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