



OPU AND IVF: AN OPPORTUNITY FOR BOVINE PRACTITIONERS

Evolving technologies and processes make *in vitro* fertilization a viable option for dairy and beef clients.

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Though the non-invasive technology that currently allows veterinarians to perform ovum pick up (OPU) was first developed more than 25 years ago, only recently has this process become commercialized, resulting in over 206,000 *in vitro* fertilization (IVF) embryos produced in North America in 2014; 93,000 of these embryos were either transferred or frozen. Numerous advancements in the OPU and IVF process have allowed the technology to evolve from only being performed in research, on very valuable donors or in terminal situations, to a technology that is both efficient and affordable for producers.

It is a straightforward procedure for clients. They simply select the donors and super-stimulate them, with no need to breed or heat detect. Following follicle stimulating hormone stimulation, a veterinarian performs transvaginal ultrasound-guided OPU, collecting oocytes in a vial. The contents of the vial are then filtered and searched, similar to a conventional embryo flush. Once recovered, oocytes are rinsed and placed into vials containing media and shipped in an incubator overnight to an IVF lab. The oocytes mature during shipment, and maturation is completed after arrival at the lab (22 to 24 hours, total), followed by fertilization.

After the gametes are allowed to co-incubate overnight, the dead cumulus and sperm cells are removed and presumptive zygotes are moved to a different type of medium to mature, known as culture medium. The culture medium is designed to mimic the chemical make-up of the uterus, and it is in this medium that embryo development occurs. One week after aspiration, the resulting embryos can either be shipped back to the client for fresh embryo transfer or frozen for transfer at a later date. Transferring of embryos is another simple way for veterinarians to generate revenue as part of an OPU/IVF service.

One of the factors limiting OPU/IVF program adoption is



Veterinarians need training and considerable practice to master ovum pick up (OPU) skills, along with investment in equipment and facilities.

that a highly skilled veterinarian must perform the aspiration. Mastering OPU requires a highly technical skill set, as well as an investment in training, equipment and facilities. Therefore, a limited number of practitioners perform the OPU procedure; however, as the demand for this service grows, more veterinarians are being trained and incorporating OPU/IVF services into their business plans and generating new revenue for their practices.

WHAT DOES IT TAKE?

The basic equipment required to perform OPU includes an ultrasound machine equipped with an endovaginal probe to visualize the ovaries and a needle guide (also referred to as a probe holder) through which a needle/tubing apparatus is guided to puncture the follicles and collect their contents. Additionally, an aspiration pump is needed to create suction, which causes the follicular fluid and oocytes to flow through the tubing and into a collection vial. Numerous heating devices including a test tube warmer, filter warmer, warming table and stage warmer are necessary to keep the oocytes at a consistent temperature, between 35° C and 37° C. Cold shock of oocytes can cause dissociation of the meiotic spindle, resulting in arrest prior to embryo development, so keeping oocytes warm throughout the entire process is critical.

In addition to equipment, an interested veterinarian would also need to look into training to master the aspiration technique. Training can be completed through different avenues, but upon completion of the initial course, weeks of practice must follow. While veterinarians who have experience in ovarian ultrasound and embryo transfer (ET) make great candidates for offering OPU services, there are certainly technical aspects that differ between the two techniques, and many long-time ET practitioners have described learning the OPU process as humbling. It is a standard recommendation

that a minimum of 100 aspirations in the first three months following training are performed on practice animals prior to offering the service commercially. If recovery rate (i.e., the number of oocytes recovered divided by the number of follicles aspirated) is still sub-par, additional practice sessions are required. Recovery rate increases with experience and should exceed 75 percent to make the procedure economically effective for clients. The length of time from the initial training to offering the service commercially is dependent on a number of factors: Preparation of equipment and facilities prior to the initial training, palpation and ultrasound skills, availability of practice animals, frequency of practice sessions, technical support and initiative all play a role. This process usually takes between four and eight months.

The final investment required is in adequate facilities in which to collect oocytes. Collection centers are often referred to as satellite centers, because they are typically located a fair distance from a centralized IVF lab. One of the most important aspects of facilities design is the ability to efficiently control temperature. For best embryo development, both the aspiration and oocyte searching area should be maintained between 27° C and 32° C, and having a small but workable area allows for more cost-effective heating/cooling. Like all handling facilities, one designed for OPU should allow for easy movement of cattle, and a staging area for donors is beneficial for time management. Lastly, a squeeze chute that can adequately restrain donors is recommended, especially with certain breeds of cattle.

In an effort to accommodate those clients concerned with biosecurity or who would prefer not to haul cattle, some veterinarians have responded by creating mobile labs. These mobile labs allow practitioners to aspirate anywhere there is a heated collection room, followed by processing the oocytes in the mobile lab. This convenience makes OPU/IVF services available to more producers but does require a bit more travel and scheduling coordination by the veterinarian. Both mobile and stationary satellite OPU centers have allowed greater access to OPU/IVF technologies for producers throughout the United States and Canada, while the regional IVF lab offers benefits of concentration of resources and skilled personnel as well as enough throughput to justify equipment and labor costs.

BENEFITS OF OPU/IVF PROGRAMS

Given the additional investment in training, equipment and facilities required, some may question why one would chose OPU/IVF over conventional embryo recovery and transfer. The reason is simple: Generating embryos using an OPU/IVF system offers many benefits. In comparison to conventional flush, donors can be collected every two weeks for OPU versus every six to eight weeks with conventional flushing. Also, due to the fact that the follicles are aspirated rather than entering the uterus, OPU can be done on donors as young as 7 months of age as well as during the first three months of pregnancy, so there is no need to take a donor out of production to get multiple



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offspring from her. Furthermore, if a donor does not perform well with conventional flush due to an oviduct issue or corpus luteum/progesterone failure, this can be by-passed by using OPU/IVF techniques.

Because more problem donors are able to make embryos in an IVF program, the failure rate (producing zero embryos) is considerably lower compared to conventional embryo recovery. Another benefit of OPU/IVF over conventional flushing is that fertilization rates using sexed semen are much higher than *in vivo* and often comparable to results with conventional semen. Lastly, IVF is a more efficient use of rare or expensive semen as multiple donors' oocytes can be fertilized with a single unit of semen versus two to three straws used in conventional flushing; reverse sorting semen (sorting frozen semen) allows for offspring of the desired sex from nearly any bull. About the only down side of OPU/IVF technology for reproducing cattle is that, currently, the total costs to the client per calf produced are higher than with conventional super-stimulation, flushing and ET.

CONCLUSION

The technologies associated with OPU/IVF have evolved considerably in the last five years, resulting in an *in vitro* embryo that is beginning to resemble an *in vivo* embryo much more closely than it had just a few short years earlier. More advanced media that closely mirror the uterine environment are leading to better quality embryos, and in turn, these better quality embryos are more likely to survive freezing and result in pregnancy. These things combined have led to more confident clients who are seeking out these advanced reproductive technologies, a great opportunity for practitioners to offer value-added services. 🐾