Benefits of embryo transfer

Embryo transfer (ET) technology gained commercial prominence in the international movement of cattle genetics in the 1970's and the increase in the North American population of European breeds of cattle. Similar, though less dramatic, events have occurred in sheep and goat breeds with the recent importation to Canada of Boer goat and Texel, East Friesian, Dorper and Charollais sheep.

Small ruminant ET is a well described and yet underexploited animal breeding technology. The size of sheep and goats, aspects of their anatomy and seasonal reproductive behaviour, present challenges not common to cattle. Those considerations have not deterred serious breeders and ET practitioners in sheep and goat producing countries.

Transrectal palpation and manipulation of the reproductive tract is not possible. The cervix of sheep and goats is difficult to penetrate, other than during estrus. It is possible to assess ovarian and follicular status with ultrasound but this may not always be logistically practical.

Non-surgical or transcervical embryo recovery is possible in goats, and embryo recovery rates can be comparable to, and generally lower than, surgical embryo recovery attempts. Non-surgical embryo transfer remains under-developed commercially. While it is possible to identify the presence of a CL by transrectal ultrasonography, transcervical embryo transfer into the appropriate uterine horn is challenging.

Most practitioners rely upon laparoscope-assisted surgical embryo recovery. Surgery of the reproductive is invasive and limits the number of times a donor can be flushed in a season. Laparoscope-assisted transfer of embryos into recipient sheep and goats minimizes potentially traumatic handling of the reproductive tract and may be performed rapidly. The investment in developing technique, performing anesthesia and surgical procedures, and the requisite equipment, all contribute to the overall cost of the procedure.

Superovulation

Superovulation increases the efficiency of ET by increasing the potential number of embryos available for recovery at the time of embryo collection. As with cattle, super-ovulation can be brought about by the administration of hormones. More reliable results are obtained with FSH (porcine follicle stimulating hormone) obtained from sheep or swine than with eCG equine chorionic gonadotrophin – formerly known as PMSG).

Super stimulation generally occurs while donors are treated with a source of progesterone, usually delivered in a device called a CIDR-G placed in the vagina of the doe or ewe. Injections of FSH are given twice daily, close to 12 hours apart, for 3-4 days. Donors are usually in standing heat (estrus) 24-36 hours after the CIDR-G is removed. We recommend that breeding should begin no earlier than 24 hours after progesterone withdrawal (assuming the donor is in standing estrus), and continue every 8 hours until the donor is out of heat.



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Laparoscopic AI can be performed if only a limited number of high quality males is available. We have experienced good results with trans-cervical AI in sheep (fresh extended semen) and goats with frozen semen suing laparoscopic AI (LAI). Embryo recovery occurs 8 days after the withdrawal of the CIDR.

Embryo recovery

Success in non-surgical embryo recovery is limited to goats. We recently participated in a commercial trial of non-surgical embryo recovery.

Sedation and local anesthesia of the donor may provide adequate restraint but we prefer general anesthesia for optimal control and safety. The anesthetised donor is placed in dorsal recumbency (on her back) and the underside of the abdomen, in front of the udder, is prepared routinely for surgery. The donor's response to super-stimulation is assessed laparoscopically. Poor responders (<=3 CL) are usually not flushed unless at the request of the owner. Typically 15-20% of sheep and 10% of goats fail to stimulate adequately to warrant being flushed.

The uterus and uterine horns are exteriorised through an abdominal incision. A Foley catheter is introduced into the uterine horn, threaded up the horn and the cuff of the catheter inflated. Embryo flush fluid is introduced through a blunted 20g needle and directed down the uterine horn, towards the Foley and collected in a gridded search dish. The procedure is repeated with the opposite horn. The uterine incisions are closed and the surfaces of the reproductive tract are rinsed free of blood clots or foreign material with saline. Abdominal closure is routine.

Careful, delicate surgical technique is required to minimise the formation of adhesions which can impair future fertility. While donors may be flushed 2-3 times in a season, allowing them to become pregnant after a few flushes lets the reproductive tract become stretched, and breaks down some of those adhesions.

The number of embryos recovered from donors is highly variable but well fed and managed, dry dairy goats often will produce 12 transferrable embryos although 7 is more common.

Identification and classification of embryos

Examination of the recovered flush fluid is routinely performed under magnification of 20-80X using a microscope. The size, morphology and developmental stages of small ruminant embryos are similar to those of bovine embryos. Small ruminant embryos may be held in embryo holding solutions formulated for bovine embryos.

Embryo cryopreservation (freezing)

Sheep and goat embryos are generally frozen in 0.25 mL plastic straws, each straw containing 2-4 embryos of a similar developmental stage from the same flush. Conventional cryopreservation in 1.5M EG is effective for morulae and blastocysts. It is reported that blastocysts have greater post-thaw survival than morulae.



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Vitrification may offer a cost effective alternative to conventional freezing of small ruminant embryos by circumventing the need for controlled rate freezers. Post-thaw survival of vitrified sheep and goat embryos compare favourably with conventionally frozen embryos of the same developmental stage.

Donor and recipient cycle synchrony

As with other species, donor and recipient estrus and ovulations should be synchronised as closely as possible. Asynchrony of 12-24 hours results in acceptable pregnancy rates. Synchronisation usually involves CIDR-G devices inserted concurrently with the donors. In our practice, eCG is given at CIDR-G withdrawal.

We prefer prolific breeds as recipients. Most will have multiple CLs at the time of ET and have a greater potential for milk production.

Despite hormonal manipulation, some potential recipients fail to synchronise or are otherwise found to be unfit as embryo recipients at the time of ET.

In our practice, final selection of recipients is made at the time of ET. Assessment of CL number and quality can be performed rapidly by laparoscopy. A detailed examination of the entire reproductive tract is impractical but obvious pathology (extensive abdominal adhesions, pelvic inflammatory disease, abscesses, distended uterine horns) disqualifies potential recipients. A single CL will support pregnancy and if the recipient is deemed satisfactory, ET can be performed immediately.

Transfer of embryos

ET can be performed quickly so heavy sedation, with or without local analgesia, provides adequate restraint. Once the animal is placed on the tilt table, the ventral abdomen is prepared routinely for surgery.

The location of laparoscope ports and incisions for exteriorization of the uterine horn are largely a matter of personal preference. The tip of the uterine horn corresponding to the ovary where ovulation, and therefore one or more CL(s) is found, is gently exteriorized and embryos transferred though a fine catheter at the tip of the uterine horn. The uterine horn is allowed to return to the abdomen and the small incisions are closed routinely. Recovery is normally rapid with animals standing and eating within an hour of ETs

Typical responses	<u>Sheep</u>	<u>Goats</u>
Super-ovulatory response	12 CL (0-30)	15 CL (0-45)
Efficiency of embryo collection	75% (60-100%)	75% (60-100%)
Transferable embryos / donor	6 (0-30)	9 (0-45)
Embryos surviving to birth	70% (0-100%)	70% (0-100%)
Recipients pregnant	65% (0-100%)	65% (0-100%)



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Important points

- Donors and recipients should be dry. Lactating donors tend not to respond as predictably.
- Sick donors with a history of reproductive disorders are liable to disappoint.
- Nutrition must be excellent with attention paid to energy, mineral and vitamin levels.
- All animals being considered for ET should be in good health.
- All management procedures foot trimming, vaccinating and deworming should be performed at least a month before programming any animal.
- Embryo collection can occur in season, as well as out of season.
- Embryos may be frozen for transfer at a later date or time.
- Please contact us well in advance of considering your ET program



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