EMBRYO TRANSFER IN DEER - THE STATE OF THE ART

T E Dixon



INTRODUCTION

Embryo transfer is now a commonly used artificial breeding technique in most food-production animals. It has been beneficially applied in two situations. Firstly where superior animals have been identified in a breeding programme, ET can be used to apply increased selection pressure on the population by obtaining a greater number of offspring from a smaller number of selected animals. This is currently the situation with cattle. Secondly, ET has been successfuly used in simply speeding up the reproductive process as is being done in the Angora goat industry in New Zealand.

The development of the deer industry, and in particular the economic aspects of importing superior animals, has led to an interest in the application of embryo transfer technology in this species. Several attempts have been made to collect and transfer fertile embryos in deer (Cervus elaphus) and it is the intention of this paper to summarise results obtained so far.

There are several aspects peculiar to dealing with deer which should be noted.

- 1. There are still gaps in current knowledge of the reproductive physiology of deer. While work is continuing in this area, the ET programmes have relied mainly on the application, with a few modifications, of the protocols used for cattle and goats.
- 2. It has been well documented that stress adversely affects reproductive performance in animals, and this would be particularly noticeable during the hormonal interference involved in superovulating animals for embryo transfer. Deer, being a relatively recently domesticated species, would appear to be more susceptable to stress than other domestic species.
- 3. Most of the work undertaken so far has, for economic reasons, been under commercial conditions. Thus variations in results have been complicated by the effects of different management systems (eg: manpower available for observing oestrus) and different handling systems (stress?).

Further to this, only small number of deer have been involved in each programme, making it difficult to come to statistically meaningful conclusions.

Superovulation

Superovulation has been attempted using two hormonal compounds - FSH[a] and PMSG (Pregnecol [b] or Folligon [c]).

In most cases these drugs have been given to females primed with progesterone for 8-15 days, using either Controlled Internal Drug Releasing devices [d] or, in the case of elk and larger hybrids, Progesterone Releasing Intravaginal Devices.[e] When the progesterone was given for a shorter period, an injection of prostaglandin [f] was given 12 hours prior to removal of the progesterone source to achieve luteolysis.

Response to superovulation has been ascertained by laparotomy (at the time of surgical collection of the embryos) or by rectal palpation of the ovaries. However, in many successful attempts at non-surgical recoveries, the number of embryos or ova collected has greatly exceeded the ovulation rate as palpated per rectum (Degrofft pers. comm.). In a number of cases, laparotomy was performed following rectal palpation of the ovaries and the actual number of corpora lutea invariably exceeded the number estimated. This is possibly because of the very flaccid consistency of the CLs compared to those in cattle.

Responses to all methods of superovulation have been extremely variable (Table 1).

Synchronisation and Mating

Synchronisation of recipient animals has been achieved mainly using CIDRs or prostaglandin injections (Glover 1985). The biggest problem encountered has been the detection of oestrus, in both recipients and donors. For economic reasons, all animals synchronised for use as recipients have been red deer, which will show some degree of female/female mounting behaviour in a group of synchronised animals. Vasectomised stags with greased brisket have also been used to identify oestrus animals.

- [a] FSH-P, Burns-Biotec, Nebraska, USA
- [b] Commonwealth Serum Laboratories, Melbourne, Australia
- [c] Intervet, Australia
- [d] CIDR, AHI, Hamilton, New Zealand
- [e] PRID, Ceva, Paris, France
- [f] Lutalyse, Upjohn, Kalamazoo, USA

TABLE 1

| Source | Deer Type | Progesterone Source | Treatment | No Animals | Range of Ovulations | Ova or Embryos Recovered |
|-----------------------------|--------------|------------------------|-------------------------|---------------|---------------------------|--------------------------------|
| Dixon 198 | 4 E1k | MPA sponges | FSH 36mg | 3 | 1-4[g] | 5 |
| Fisher/ Fennessy 1984 | Red | CIDR | 500-1500iu Pregnecol | 17 | 0-5 | - |
| Fisher/ Fennessy 1985 | Red | CIDR | 400-1600iu Folligon | 18 | 0-3 | <u>-</u> |
| Fisher/ Fennessy 1985 | Red | CIDR | 4-60mg FSH | 25 | 0-28 | <u>-</u> |
| Dixon/ Hunter 1985 | Elk | PRID | 1500-2000iu Folligon | 8 | 0-6[g] | 26 |
| Dixon/ Hunter 1985 | Hybri ds | s CIDR | 2000-2500 | 7 | 0-16[h] |] 34 |
| Dixon/ Hunter 1986 | Hybri d | s CIDR | 48 mg FSH | 5 | 4-12 | 14 |
| Dixon/ Hunter 1986 | Red | CIDR | 42 mg. FSH | 3 | 2-8 | 7 |
| Bringans 1986 | Elk | CIDR | 1500iu Perganol | 2 | 2-4 | 6 |
| Bringans 1986 | Hybrid | s CIDR | 1100iu Perganol | 1 | 2 | 0 |

[[]g] As assessed by rectal palpation
[h] Unovulated follicles also present

Oestrus response has generally been better with CIDRs than with prostaglandin, as shown in Table 2.

TABLE 2

| Method of synchronisation | Time to oestrus (hours) | | | No. showing no oestrus |
|--|-------------------------|-------|-------|---------------------------|
| | 12-24 | 24-36 | 36-48 | |
| 15 days CIDRs | | 7 | 5 | 5 |
| 2 injections 3ml lutalyse 11 days apart | 1 | 1 | | 11 |

The achievement of successful donor matings has been one of the most variable factors in the ET programmes.

Factors which may affect this include:-

- Programming the animals too early in the roaring season before they have cycled.
- 2. Management factors causing stress over the mating period. These factors would include excessive handling of the animals prior to the expected oestrus. For this reason, the use of a single injection of PMS-G may be preferable, in some circumstances, to the twice-daily injections of FSH.
- 3. The presence of a sexually active male.

Embryo collection

Parous elk and larger hybrids can be flushed non-surgically. The technique involves palpation of the reproductive tract per rectum, and the placement of a 2-way catheter (Rusch, Germany) through the cervix and into the uterus. The cuff is inflated either in the body of the uterus, and both cornua flushed simultaneously, or into each cornua so that they are flushed separately. Phosphate buffered saline is infused into the uterus and the fluid recovered and searched for the embryos. The collection procedure can be carried out with the animal standing, with or without sedation, so long as it is adequately restrained.

The flush is carried out on day 6 (day of oestrus = 0).

In smaller or non-parous animals, and in red deer, the collection is surgical. The technique is similar to that in goats and sheep (Tervit 1976), using a 12G or 14G catheter. As with other species care must be taken to minimise trauma to the tract which may cause the formation of adhesions which could affect later fertility.

In one case, where poor recovery was obtained from the uterine flush, the oviducts were flushed and the ova recovered, despite this animal being 6 days post-oestrus.

The fertile embryos were of variable quality, though being morphologically similar to bovine embryos.

Table 3 summarises the results of successful embryo collections.

TABLE 3

| Source | e Deer Type | Superovulation | Collection | Embryos Recovered |
|--------------------------|----------------|------------------------|--------------|--|
| Dixon 198 | 4 E1k i | Folligon/Prostaglandin | Non-Surgical | 1xmorula 1xNF[a] |
| Fisher/ Fennessy | Red 1985 | Folligon/CIDR | Surgical | 1xgranular morula |
| u | Red(x2) | FSH/CIDR | п | 7 x 2-4 cell 1 x 2 cell 7 x degenerating |
| Dixon/ Hunter 1985 | Elk | Folligon/PRID | Non-Surgical | 6 x blastocysts |
| | 85 | и | 11 | 8 x degenerate |
| 11 | н | 11 | 11 | <pre>11 x NF 6 x expanded blastocysts[b]</pre> |
| Bringans 1986 | Elk | Perganol/CIDR | Non-Surgical | 2 x blastocysts |
| | u | Perganol/CIDR | и | 4 x morulae |
| Dixon/ Hunter 1986 | Red (x3) | FSH/CIDR | Surgical | 4 x blastocysts 1 x morula 2xNF |
| Dixon/ Hunter 1986 | Hybrids (x3 |) FSH/CIDR | Surgical | 3 x 8 cell 3 x morulae |
| | | | | |

Embryo Implantation

Embryos were in all cases transferred surgically into red deer. The occurrence of a synchronous heat does not ensure that a recipient will have an adequate corpus luteum on the ovary (M Bringans, pers. comm.). For this reason, a laparoscope can be used on recipients to ascertain the presence of a CL before performing a laparotomy.

[[]a] Non-fertile

[[]b] Embryos frozen

The embryo is injected into the U-T lunction of the ipse-lateral horn using either a glass pipette or a tom-cat catheter (after making a puncture with a blunt instrument).

When a laparoscope was used to visualise the CL, the ipsilateral horn was exteriorised, using a pair of modified bowel clamps, through a slightly enlarged midline puncture. This avoids the necessity of exteriorising both horns of the uterus.

Pregnancy results from embryos implanted up to 1985 are shown in Table 4.

TABLE 4

| Source | Embryo | Recipient Synchronisation | Number of Recipients | Number of Pregnancies |
|-----------------------|-----------------------------|------------------------------|-------------------------|--------------------------|
| Dixon 1984 | Blastocyst | MPA sponge | 0ne | None |
| Fennessy 1985 | 1x granular m | orula CIDR | 0ne | One |
| | 7 x degeneration 1 x 2-cell | ng CIDR | Five | None |
| Dixon/ Hunter 1985 | 6 x blastocyst | s Prostaglandin | Six | Five |

DISCUSSION

As can be seen from the data given, the embryo transfer technique in deer is still in the early stages of development. Although pregnancies have been achieved, the number of pregnancies per donor programmed is very low, and is a long way from being commercially viable.

The seasonal nature of reproductive activity in deer, and the economic pressures previously mentioned, limit the rate at which development can take place. However, the introduction of, for example, artificial insemination, would remove some of the variables being encountered at present.

REFERENCES

- Fennessy P.F.; Fisher M.W. (1985): Manipulation of Reproduction in Red Deer. Proc. of the ANZAAS Deer Farming Seminar, ed. R C Couchman, Agric. Notes Series No. 159.
- 2. Fisher M.W.; Fennessy P.F. (1985): Reproductive Physiology of Female Red Deer and Wapiti. Proceedings, NZVA Deer Branch, Course No. 2.
- 3. Glover G.J. (1985): Aspects of Reproductive Physiology of Female Wapiti. PhD Thesis, Univ. of Saskatchewan.
- 4. Tervit H.R.; Havik Pamela G. (1976): A modified Technique for flushing ova from the sheep uterus. NZ Vet. J. 24: 138-140.