EMBRYO TRANSFER IN DEER: AN UPDATE

M.J. Bringans
Central Southland Veterinary Services (CSVS)
Winton

Introduction

Embryo transfer in deer offers the industry an opportunity to increase numbers of elite stock at a faster rate than natural breeding within a herd; and gives a vehicle to transport genetic material within the country and overseas, as an export earner.

The main reasons for worldwide acceptance of frozen embryos as a means of importing livestock are:-

(i) Frozen embryos can be shipped at a fraction of the cost of shipping live animals;

(ii) embryos can be imported from the top females from several elite herds, offering a wider selection of genetic material than is possible by importing live animals;

(iii) there is a much reduced risk of disease importation;

(iv) the animal is born on site in the importing country and does not have to adapt to foreign climates, feeds and change in hemisphere effects.

The stage that research into embryo transfer in deer is at, presently, can only be considered early. Super-ovulation is achievable in red, wapiti, fallow and elk with varying degrees of success and consistency. Fertilized deer ova appear to have good conception rates when transferred fresh or frozen.

Superovulation of Donors

In the 1988 and 1989 seasons several different brands of drug have been used, in conjunction with CIDR progesterone priming. It is becoming obvious with red deer (Cervus elaphus) that the use of pregnant mare serum gonadotrophin (PMSG) and some less refined preparations of follicle stimulating hormone can give inconsistent superovulation, poor fertilization rates /or poor recoveries.

To quote P. Fennessy(1) "Both PMSG and FSHP have marked LH activity, plus have considerable batch variation in the LH/FSH ratios. There is evidence that the LH activity of these preparations may adversely affect ovulatory responses and embryo recovery rates in other species (2)(3)".

"Ovagen", "Follitrophin" and "FSHP" have been used predominantly in red deer recently. Some 1988 and 1989 results reflect the improved superovulation and quality of embryos with the more refined LH reduced preparations. Our group used mainly FSHP in 1988. We were flushing mid March in order to freeze embryos for one of our clients who was exporting to Australia.

Results, on the group of 12 red deer donors are as in the table below:-

Table One

Bringans/Wenkoff/CSVS 1988 - Red Deer

No. Donors	Av. CL count/ donor	Av. Embryo Recovery /donor	Av. Transferable /Embryos/donor	Av. Non Fertile Embryos	Av Degenerate Embryos
12	6.33	5.7	3.0	2.1	0.6

[recovery rate - 90%]

Approx. 30mg "FSHP" was delivered over 4 days in a miniosmotic pump device.

In 1989 we compared "FSHP" with "Ovagen" (delivered in mini-osmotic pumps). Fourteen red deer hinds were randomly selected and treated with 30 mg "FSHP". On the same property 14 red deer hinds were treated with "Ovagen"- one unit (mini-osmotic pumps).

Note: The "FSHP" group was flushed in the last 10 days of March and the Ovagen group flushed in last 5 days of March and first 3 days of April 1989.

On another property we treated 7 red deer hinds with "Ovagen" minipumps at the same time (Table 2).

Table Two

Bringans/Wenkoff/CSVS 1989 - Red Deer)

	No. Donors	Av. CL count	Av. Recovery	Av Transfer	Av. N.F.*	Av. Deg.
"FSHP"	14	12.0	7.49	2.8	4.26	0.43
	[Recovery Rate - 62.4%]					
"Ovagen"	14	5.6	4.9	4.2	0.64	0.07
	[Recovery Rate - 85%]					
"Ovagen"	7	12.0	11.0	6.0	5.0	0
	[Recovery Rate - 90%]					

N.F.* Non-fertile

"Ovagen" results summarized from both properties are summarised in Table 3.

Table 3

Total "Ovagen" Results - Red Deer
Bringans/Wenkoff CSVS - 1989

No. Donors	CL/Donor	Emb/Donor	Transfers/Donors	Av. NF	Av. Deg.
21	7.76	6.90	4.80	2.10	0.05

(i) Embryo recovery rate - 88%

(ii) Range of transferable embryos - 0-17 per donor.

Fennessy et al (1989) report the use of "Ovagen" in 1988 in red deer hinds and results are summarised in Table 4.

Table 4

Fennessy - "Ovagen" 1988 - Red Deer

Trial 1:	No. Donors	Av. CL/Donor	Av. Transfers	Deg/NF
(March)	3 @ .036 units/day	1	.3	
	3 @ .071 units/day	2	.66	
	3 @ .11 units/day	4.3	2.3	
	3 @ .14 units/day	15.3	5	
	[Recovery Rate - 38%]			
Trial 2:	**No. Donors**			
(June)	(i) 4 @ .14 units day in osmotic pump	3	1.25	0.5
	(ii) 4 @ .14 units/day as injections	11	3.25	2.25
	[Recovery rate - 72%]			

Trial one showed an increasing response with the larger "Ovagen" dose. Trial two suggested a better response by injection than pumps. However "Ovagen" was delivered by pump in trial 1. P. Fennessy expresses caution with analysing trial (ii). This was a subfertile group of red deer hinds, which may have led to the range of embryo stages that were found in the fertilized ova.

J. Hunter reports (pers. comm.) the use of "Follitrophin" in 1989. The results are summarized in the table below.

Table 5

Total "Follitrophin" Results - Red Deer
Dixon/Hunter (Beatson) - 1989

No. Donors	CL/Donor	Emb/Donor	Transfers/Donor	Av. NF	Av. Deg
61	8.5	6.80	5.06	1.7	0.1
(i)	Embryo recovery - 80%				
(ii)	Range of transferable embryos - 0-16 per donor				

Recent superovulation trials in other species of deer:-

(i) **Fallow Deer** - G. Asher(4) superovulated 36 mature fallow does in May 1987. They were in 3 treatment groups and all CIDR progesterone primed.

1st Group were given 100 I.U. PMSG 48 hours before CIDR withdrawal

2nd Group were given a total of 20 mg "FSH" ("Follitrophin") I/M b.i.d. for 4 days, stopping at CIDR withdrawal

3rd Group were given 750 I.U. PMSG 48 hours before CIDR withdrawal and 14 mg FSH the same way as in the second group.

Table 6

	No. Donors	Av. CL	Total ova	% Recovery	% Fert.	Response range
Group 1	12	9.2	3.7	40	70	
Group 2	12	6.3	1.1	17	84	8 no resp. [0-30]
Group 3	12	11.2	1.9	17	52	

A wide range of embryo development stages was seen in the fertilized ova. This, combined with the low recovery, shows the fallow deer is very easy to overstimulate.

Wapiti: In 1988 (March) our group did a trial on 18 mixed-age Wapiti hinds (average body weight 150 Kg).

The hinds were randomly divided into 2 mobs, and 3 elk bulls mated some from each mob.

Programme 1

Nine hinds were given a total of 30 mg "FSHP" (osmotic minipump) over 4 days prior to CIDR withdrawal and 300 I.U. of PMSG ("Pregnecol") on CIDR and pump withdrawal.

Programme 2

Nine hinds were given a total of 30 mg "FSHP" (osmotic minipump over 4 days prior to CIDR withdrawal and 300 I.U. of PMSG ("Pregnecol") at pump insertion.

Table 7

Wapiti 1988 - CSVS

Donor	Programme	NF Recovered	Transferable Recovered
1	1		4
2	1		10
3	1		3
4	1		5
5	1		5
6	1	5*	
7	1		6
8	1		6
9	1	4*	
10	2	2	
11	2	10	
12	2	N.R.	
13	2	17	
14	2	14	
15	2	N.R.	
16	2	7	
17	2	17	
18	2	N.R.	

* Bull problem

Programme 1 gave us 39 fertilized embryos (average of 4.33 transferable embryos per donor or taking the mating problem into account an average of 5.57 T. embryos per donor). Programme 2 gave us 0 fertilized embryos (Table 7).

Elk

Programmes have been reported in previous Deer Branch NZVA proceedings (1987(5)).

Recipient Programme

Insertion of CIDR for 12 days and 200 I.U. PMSG on withdrawal is now an accepted procedure. With the hold rates currently being gained, devising methods of heat detection are a low priority J. Hunter (pers. comm.) reports a rejection rate of 10-45% depending on the property and the timing of implantation.

Our group found in NZ in the 1987 and 1988 seasons, a 10% rejection rate of recipients with more than 100 recipients being involved. However, in 1988 in Australia we found more than a 30% rejection rate. This was due to a possible stress factor from lack of routine handling, and nutritional problems. In 1989 in Australia the stress factors were still obvious but the rejection rate was 10%. The hormone levels in the treatment programme were increased.

Expected Results

(i) **Fresh Implantation**

Average results taken over several seasons are in the range of 60% to 80%. As in all species quality of embryos can be ascertained and is proportional to the hold rate.

J. Hunter (pers. comm.) gives an example using fresh embryo implantation):

15 grade 1 Morulae (E. Dixon's Grading) resulting in 8 pregnancies
27 grade 4 Morulae resulting in 26 pregnancies.

Hunter reports overall in 1989 that of 269 recipients, 218 scanned positive for pregnancy (81%). Our own experience gives a similar result. 21 embryos which were classified as unsuitable for freezing because of poor quality (Grade 3 Morulae - Wenkoff classification) were put in recipients as fresh transplants rather than destroying them, resulting in 10 pregnancies. Twenty Grade 1 Morulae resulted in 18 pregnancies during fresh transplanting.

(ii) **Frozen Embryo Transplantation**

Bringans/CSVS/Wenkoff

	No. implanted	Results	%
1987	6 reds	4 calves born	66%
1988	60 elk + reds	36 calves born	60%
1989	72 elk + reds	(30/37 scanned to date are pregnant - 81%)	

Dixon/Hunter

	No. implanted	Results	%
1988	38	21 scan +	55%
1989	78	45 scan +	58%

COMPLICATIONS AT EMBRYO TRANSFER

(1) Donors

Infertility, as in all species, can result from ET procedures. Adhesions following surgery have the potential to cause problems. We re-operated on 12 donors in the 1989 season that had surgical transplantation in 1988 and noticed only one of these had adhesions of any consequence.

Subsequent calving - Approximately 3% of donors actually calved to the transplant date (i.e. they failed to abort any embryos that had not been recovered despite all donors being injected with prostaglandins 9 days post surgery). These have all been births of singles.

Some donors will conceive to the next cycle following surgery, some will conceive at a later cycle. A small percentage remain dry for the season, especially if surgery is done mid to late April rather than in mid March.

J. Hunter (pers. comm.) states that 35 donors have been presented for repeat surgery. Four have shown adhesions of a minor degree to body and horns of the uterus which has not interfered with flushing. One had a major consequence of a blocked utero tubal junction on one side.

(2) Recipients

Deer used as recipients suffer no ill effects. Implantation is usually done early in the season and those not conceiving to the ET usually fall pregnant at their next mating. Because better quality hinds are usually chosen as recipients, we have never experienced a recipient that failed to conceive during the season that an implant had been performed.

(3) Anaesthetic Deaths

For several hundred donors and recipients operated on in the past two seasons, we have never had an anaesthetic death. However, it must always be considered a possibility.

Hunter reports only the loss of one recipient plus one donor in 4 years over approximately 1,000 procedures. Anaesthetic used is Rompun/Fentaz i/v followed by intubation and maintenance on fluothane. Our group is yet to experience a death and use Fentaz/Rompun i/m for all procedures.

(4) Post Surgery Deaths

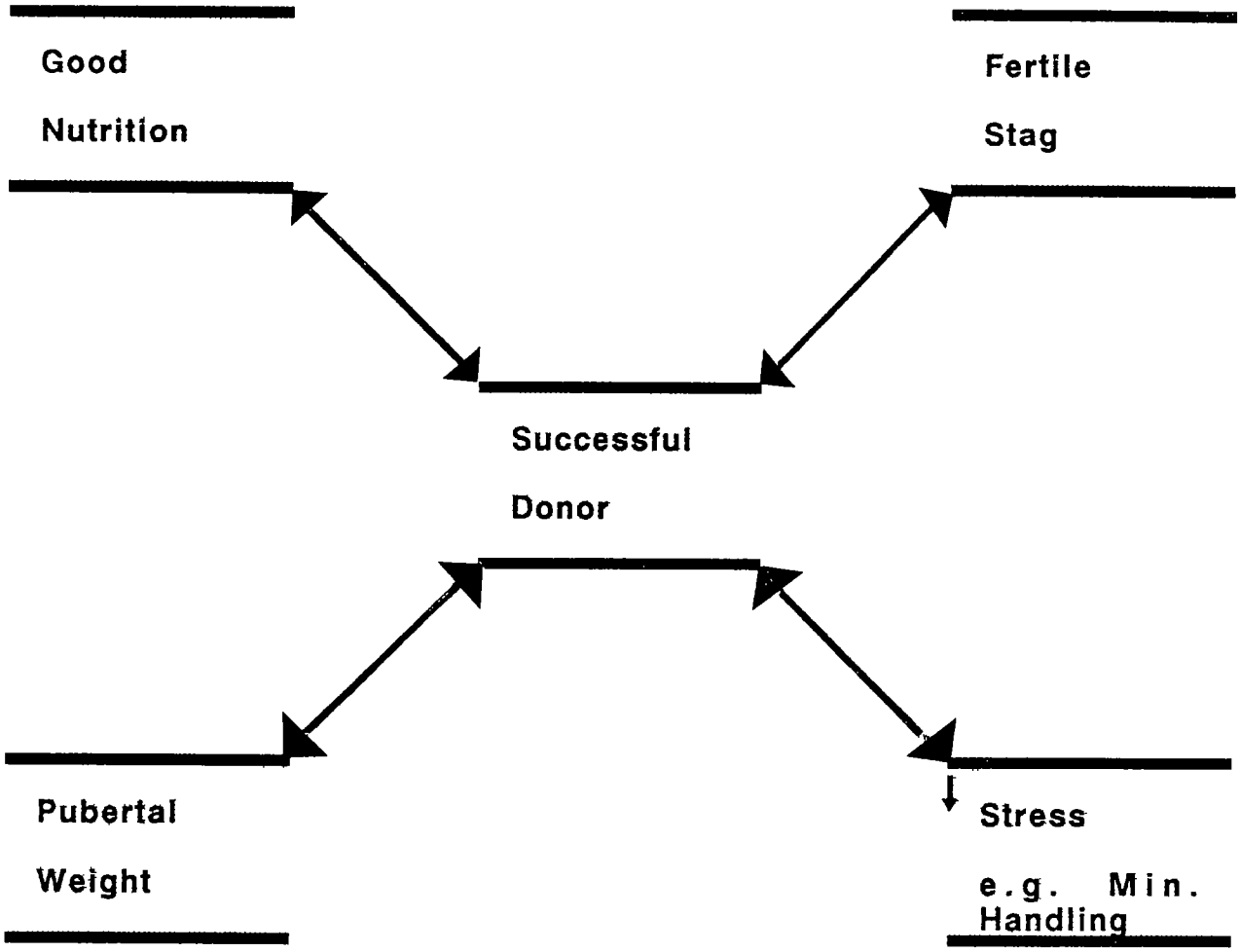
Likewise we have never seen any post surgical deaths or illnesses subsequent to ET in either donors or recipients.

MANAGEMENT

Management is as important to the success of all aspects of an ET programme as any other factor.

Deer are an easily stressed animal. Nutritional and emotional stress must be kept to a minimum to ensure a successful ET programme.

For example in 1987 we used only a CIDR, with no PMSG on withdrawal in red deer recipients Only 50% of these recipients cycled. The PMSG seems to overcome this stress inhibition of ovulation, with only 5-10% of recipients being rejected.



Range 0-17 transferable embryos/donor

(1) **Recipients**

In my opinion, for best results when preparing recipients, they should be weaned at least two weeks before CIDR's are inserted and kept in their own settled mob at this stage. It is important to put them on a rising plane of nutrition. By the time CIDR's go in the hinds they should be showing signs of "lifting" in body condition such as beginning to lose their roughness of coat. Handling should be kept to a minimum. They should be kept near stags at this stage. We have not bothered with heat detection in our recipients. It may be possible to increase conception rates by doing this, but until an easy method is developed for heat detection, I feel the conception rate currently being experienced negates its importance.

After implantation, the hinds are kept on good nutrition for a few weeks. As far as the introduction of a stag for back-up reasons, it is difficult to advise. Many farmers keep the stag away for 20 days so that a distinct split in fawning patterns can be seen. Some of our clients have introduced stags immediately post-surgery and have suffered no drop in conception rates to the ET. Pregnancy scanning will easily distinguish between the ET pregnancy and the back-up stag pregnancy if done before 65 days post-surgery. Second and third calvers with good histories are usually chosen as recipients, as they are in their optimum reproductive state.

(2) **Donors**

Donors must be treated as for recipients i.e. wean early. (Note: donors lactating but in good condition will still flush satisfactorily if superovulated, but in most situations the easiest way to achieve condition is to wean). We have superovulated a number of 14 month old red deer. They responded well to superovulation, but mating seems to be a problem with about 50% of these animals producing unfertilised ova. Generally speaking we prefer adult deer as donors.

Time is important to flushing success. The flushing average improved as we neared the end of the season. We were flushing a number of deer for freezing embryos for Australia in mid March but found flush results erratic until the last few days in March.

We introduce the stag to the donors when the CIDR goes in. It is important to use a proven sire. Don't budget on using a stag over more than three donors per day for any extended period of time. A programmed hind unmated is expensive.

References

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(4) Thompson, J.G.E. and Asher, G.W. "Superovulation and Ova Recovery in Farmed Fallow Deer". Proc. Aust. Soc reprod Biol 1988 20: 4.

(5) Bringans, "ET in Deer" - Proc. Deer Course for Veterinarians, Deer Branch NZVA No. 3, 1987.

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