NEW A.I. TECHNOLOGY

Graham Bowen

1. SEMEN COLLECTION

Further development of semen collection techniques has been undertaken over the past six months to attempt to harvest larger quantities of semen from selected stags.

Present collection regimes using electroejaculation, as demonstrated and described in previous Deer Branch NZVA course proceedings, are yielding collections ranging from 0 to 200 doses. Equipment used has been modified somewhat to reduce the number of unsuccessful electroejaculation attempts made on anaesthetized stags. During the 1990 autumn (March-June) we made 88 collection attempts and were successful with a semen sample from 69 (78% success). Number of doses per ejaculate ranged from 20-247. While not having precise comparable data from previous years, these figures are substantially higher. The best period for collection was late May to late June.

Semen quality, which is naturally the key factor, has remained similar over the last three years and still varies from stag to stag and collection to collection. However, by virtue of the fact that we have had a high collection: non collection ratio, the end result has meant larger volumes of good quality semen in storage for subsequent usage. 49 of the 69 samples collected were of acceptable quality (71%). This gives an overall success rate of 55.6% of all attempts.

The single most limiting factor with the current collection regime is the necessity to anaesthetize stags. The procedure is high risk and also limits the frequency of collection attempts. To date we have spaced out our collections to a minimum of one week apart with no apparent effect on the individual stags. To avoid the necessity of anaesthetizing animals we have constructed a crush designed especially for semen collection. The crush allows stags to remain standing and limits their movement which in turn allows collection in a very quick time (on average three minutes). We have collected stags 2-3 times per week with this method without any increasing handling difficulties. The administration of 1/2ml 2% xylazine per 100kg bodyweight intramuscularly 5-10 minutes before collection assist with loading the animal in the crush and may even reduce any discomfort associated with the electro ejaculation collection. Semen volumes and sperm density remain similar to those of ejaculates from anaesthetized animals and although the risk of urination is still high it is not nearly as spontaneous.

To date we have collected semen from 13 individual stags and made 28 collections using this regime. Five collection attempts have not produced semen (82% success). A higher percentage of semen processed has subsequently passed quality control standards although this could be attributed to the stag being housed on the site, thus avoiding semen transportation and some on farm facility inadequacies.

23 of 28 samples collected were of acceptable quality (80.1%). This gives an overall success rate of 69.6% of all attempts.

2. CERVICAL INSEMINATION

This autumn we have inseminated a further 100 hinds cervically, and compared semen dose rates and laparoscopic vs cervical techniques.

The inseminations were carried out on two properties using semen from the same stag and the same technician. The programme included 12 days CIDR's, 200 I.U. PMSG. at withdrawal and insemination 48-50 hours later. Teaser stags were present with hinds at all times on both properties.

Results of these inseminations as determined by ultrasonography were:

Dose rate	40	million/spermatozoa	44.6%
Dose rate	30	million/spermatozoa	23%

The 30 million sperm/straw rate was also used laparoscopically and yielded a 61% conception rate.

All inseminations were on a fixed time basis as any attempt to identify oestrous hinds by using "greased" stags was relatively unsuccessful. Given that a fixed time insemination is not the most desirable regime we will probably not improve results significantly until heat detection is possible. An increase in sperm dose rate may lift conception rates but quickly disadvantages cervical A.I. by dramatically reducing the number of inseminations per ejaculate.

3. LAPAROSCOPIC TECHNIQUES

During the 1990 season two aspects of laparoscopic A.I. were investigated.

3.1 Unilateral vs bilateral insemination

It has always been the writer's objective when inseminating to disturb the reproductive tract as little as possible. On some occasions only one horn of the uterus is visible and accessible without "probing" and re-arranging the position of the tract. To avoid probing, inseminations were carried out placing all of the semen in one horn only. On these occasions semen was placed as close to the bifurcation as practicable. Results were as follows:

Insemination One Horn only	Both Horns
36/54 (66.6% conception)	360/573 (62.9% conception)

These inseminations were recorded on four properties using six different stags and a mix of fresh and frozen semen. Only one horn was available for insemination in 9.4% of hinds in this survey.

It appears that semen will be dispersed evenly through the uterus by natural reticulation. It may be that conception results are enhanced by avoiding disturbance and damage to the tract. Some damage is caused with every insemination and perhaps the damage is minimized when only one penetration is made.

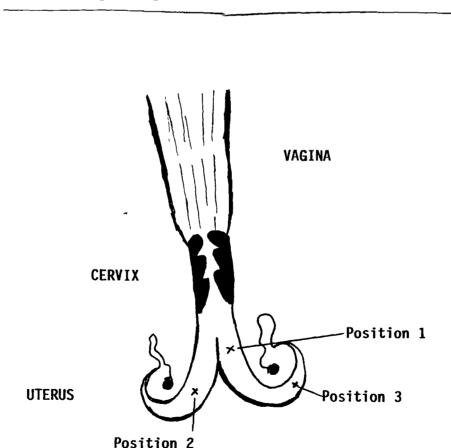
3.2 Semen delivery systems

Inseminations were carried out using a French produced sheath and needle (Aspic) connected to a small ruminant A.I. pistolette. A modified trochar and canula was developed to facilitate the system to keep air loss to a minimum. The reproductive tract can lay at varying angles and positions while the hind is elevated for laparoscopy.

Early techniques included the use of glass pipettes for semen delivery which while difficult to load and handle are very positive when it comes to semen being placed correctly in the lumen. Concerns with other delivery systems included the possibility of semen not being placed correctly and being injected into the wall or in fact completely through the horn, particularly if inseminating further down the horn than normal. Most operators aim to place the semen about 2.5cm from the uterine bifurcation, with half the semen dose in each horn. With glass pipettes the semen is sucked into the lumen when positioned accurately as there is insufficient pressure on the plunger to push the semen into the horn as there is with other semen delivery apparatus.

Since the uterine horn is of variable thickness we decided to examine the results of the semen being placed in three positions along the horn. The main reason for placing the semen in different positions is to ascertain whether it is necessary to inseminate in any one position to maximise results.

Figure 1 - Diagram of the reproductive tract of a hind showing three positions for laparascopic insemination



-170-

Conceptions/hinds mated (%)

Summary of Results:

	Placement 1	Placement 2	Placement 3	Totals
Property A	29/39	11/15	2/2	42/56 (75%)
B	18/22	31.51	1/2	50/75 (66.6%)
C	8/16	22/32	-	30/48 (62.5%)
D	4/8	13/28	0/2	17/38 (44.7%)
E	4/7	15/21		19/28 (67.8%)
Overall	63/92 (68.4%)	92/147 (62.5%)	3/6 (50%)	158/245 (64.4%)

A mix of fresh and frozen semen from seven different stags was used in this survey.

It would appear that there could be a slight advantage of insemination closer to the bifurcation than originally recommended with sheep and goats, but it would also be apparent that position 2 is presented in a higher percentage of deer. On the strength of the above data I would continue to recommend that semen should be placed in the uterine horn as close to the bifurcation as possible without disturbing or touching the horn. I believe the entry into the horn should be perpendicular to the horn and exactly in the middle of the horn. Operators should also take care not to move the pistolette about while it is inserted through the wall. For that reason I prefer to use a second technician to operate the pistolette plunger while the laparoscopic operator holds the equipment firmly in position.

4. ACKNOWLEDGEMENT

As with last years data special thanks must be directed to Noel Beatson for his commitment to the development of A.I. in the Deer species.

Peel Forest Estate, Mt. Hutt Station and Mt. Peel Station have been very supportive of this work.