FIELD TRIALS WITH SUPEROVULATION, ARTIFICIAL INSEMINATION AND EMBRYO TRANSFER IN FALLOW DEER (Dama dama dama) IN WESTERN AUSTRALIA.

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ABSTRACT

A superovulation trial on four (4) European fallow does (Dama dama dama) was carried out in early April 1990 using natural means of fertilization to assess the ovulatory response, collection technique and embryo recovery and viability. Embryos were obtained.

The commercial trial was undertaken in late May-June 1990 replacing mating with artificial insemination using imported frozen semen from a pure Mesopotamian buck (Dama dama mesopotamica). Six (6) females were superovulated. Seven days after insemination these does were laparoscoped for response. All had multiple copora lutea and were flushed with 55% recovery rate. Fertilized eggs were recovered from have the donors. These were to various degrees, retarded or degenerate. They were transferred but failed to survive until pregnancy diagnosis (using ultrasonography) at 52 days.

INTRODUCTION

Mesopotamian fallow are known for their comparitively larger liveweights and increased venison potential. As there are no pure Mesopotamian animals in Australia, Australian fallow breeders have been importing frozen semen for insemination into Australian females, resulting in F1 hybrids. If some of these females should be successfully superovulated, inseminated and flushed and the embryos transferred, the genetic and financial gain is obvious.

AIM

To attempt to superovulate Australian fallow does, to artificially inseminate them with imported frozen semen from a pure Mesopotamian buck, and to collect and transfer the fresh embryos to Australian fallow recipients.

MATERIAL AND METHODS

Initially this programme was timed for late April-early May but to the unavailability of suitable semen the project took place in late May/early June 1990.

Semen was obtained as frozen 0.25ml straws from a pure Mesopotamian buck held at Ruakura Artificial Breeding Centre, Hamilton, New Zealand. The motility post-thaw was assessed at 60% and the concentration 30-40 million under field conditions.

DONOR AND RECIPIENT MANAGEMENT

The European fallow were owned by a syndicate of farmers located at Albany, Western Australia 35.025 South 117.53 East in a mediterranean-type climate - hot dry summers and cool moderately rainy winters.

The heavier and better does were selected as donors. All animals, donors and recipients, had had fawns the previous season. These animals were weaned 1 week prior to CIDR-G insertion, put onto new pasture (clover and rye grass), and fed lupins and oats to ensure a rising plane of nutrition. They were also made very familiar with the yards to reduce stress. Entire bucks were run in the next paddock and were introduced 8 days after embryo collection.

PROGRAMMING

Four (4) does for the pilot trial and six (6) does for the commercial trial were synchronized as donors. One CIDR-G was placed into the vagina for 14 days. On Day 12, 200IU of PMSG (Folligon(R)) were given intramuscularly and at the same time F.S.H. (Ovagen (R) 0.5IU) treatment was started, given twice daily for 4 days in a decreasing dose regime.

Recipients received a CIDR-G for 14 days which was removed 24 hours before the removal of donor CIDR's.

ARTIFICIAL INSEMINATION

Animals were anaesthetized with xylazine/ketamine (2mg./kg and 4mg./kg respectively IV) At 45 hours post-CIDR removal laparoscopic intrauterine insemination was performed. Half a straw of thawed semen was placed in each horn at the greater curvature. Each animal was given 500 IU HCG (Chorolon(R)) IM and the anaesthetic reversed with yohimbine (Reversine(R)) 0.4 mg/kg IV. CIDR's were replaced in donors 3 days after insemination to prevent luteal regression.

EMBRYO RECOVERY

Donors were anaesthetized with Zoletil (Bolus 2.2ml IV and 2.0ml IV top up). They were examined via a mid-line incision and flushed for embryos and/or ova. Recipient does were anaesthetized with xylazine/ketamine and reversed with yohimbine. They were examined via laproscope for the presence of a single copora lutea and the uterine horn exteriorized for embryo implantation if suitable.

Animals were examined for pregnancy at 52 days using a rectal probe with PIE DATA linear ultrasound scanner.

RESULTS

Only 50% of animals (4) in the April trial superovulated but embryos were obtained from one of these animals.See Table 1.

Of the commercial project, recovery rate of ova to corpora lutea was 55.3%. 16.1% of ova recovered were fertilized. 50% of superovulated animals had some fertilized eggs. About 50% of the ova recovered from these animals were fertilized. However, most of these were showing signs of degeneration or arrested growth. 4 of the 5 embryos recovered were transferred to recipients but all failed to hold. See Table 2

There were a large number of retained follicles but none were cystic as was the case for the early April trial.

The numbers of animals are too small to be significant.

RESULTS TABLE NO. 1 14TH APRIL 1990 ALBANY

DOE ID	OVARIAN RESPONSE				EGGS RECOVERED		
	COPOREA LUTEA		RETAINED FOLLICLES		UNFERTILIZED	FERTILIZED	COMMENT
	LEFT	RIGHT	LEFT	RIGHT			
G7	5	7	1	-	1	-	1 8 morula 1 tight morula
G12	-	1	-	-	not flushed		1 32 cell morula 1 early blast
G2	8	6	CYSTIC		0	5	1 degenerate
G19	-	-	-	-			not cycling
TOTAL	27		1		1	5	

NOTES
1. Irregular response to ovulatory regime
2. Some "problem" animals used
3. Very early in onset of oestrus - some animals may not have commenced true cyclicity.

RESULTS TABLE NO 2 12TH JUNE 1991 ALBANY

DOE ID	OVARIAN RESPONSE				EGGS RECOVERED		
	COPOREA LUTEA		RETAINED FOLLICLES		UNFERTILIZED	FERTILIZED	COMMENT
	LEFT	RIGHT	LEFT	RIGHT			
31	6	5	-	1	4	2	1 8 degenerate cell 1 16 cell
32	6	2	2	5	3	1	1 morula
27	6	4	2	4	4	-	
33	5	5	-	-	10	-	
28	4	2	3	3	2	2	1 blast collapsed 1 morula
30	5	6	-	-	3	-	
TOTAL	56		20		26	5	

NOTES
1. Each ovary responded
2. 4/6 had retained follicles

DISCUSSION

The results of this project wer disappointing but not totally unexpected. European fallow appear to be very sensitive to any gonadotrophins used for superovulation, and to the seasonal timing of the programme. The response in the early April produced a significantly greater number of CLs than the same regime in early June. This April response was erratic indicating an all or nothing effect as found by Asher in 1987. Perhaps use of the second overt oestrus would have yielded a more uniform with smaller and fewer retained follicles.

The fact that the weather was bleak and bitterly cold (relatively) in the 10 days prior to the June collection may have induced greater than expected stress. In this situation ACTH stimulation of the adrenals with resultant increase of progesterone may have halted or slowed ovulation and hence limited fertilization or whether blood glucose levels are significant (4).

We were also attempting to combine laparoscopic insemination of frozen semen with the latter part of the breeding season of fallow. It is difficult to determine whether HCG played any part in reducing the number of retained/large follicles and whether replacing CIDRs in donors after fertilization helped to reduce the effect of oestradiol secretion from those retained follicles which did occur. Perhaps anti-PMSG may play a part here.

Two points are significant. Firstly AI should not be attempted either too early or too late in the breeding season of fallow deer, and secondly stress would appear to play a significant role in reduced fertility. Yarding fallow just prior to insemination instead of overnight is undoubtedly important.

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