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## The use of fullsibling pairs to measure plasma FSH concentrations in Booroola ewe and ram lambs

K.L. ISAACS, K.P. MCNATTY<sup>1</sup> AND R.P. LITTLEJOHN

MAF Technology, Invermay Agricultural Centre, Private Bag, Mosgiel, New Zealand.

### ABSTRACT

FSH concentrations were examined in 6 Fec<sup>B</sup>Fec<sup>B</sup> (BB) ewe, 3 BB ram and 5 Fec<sup>+</sup>Fec<sup>+</sup> (++) ram fullsibling pairs of Booroola Merino x Romney lambs between 2 and 20 weeks of age. The purpose of this study was to compare variation in FSH profiles within and between prepubertal fullsibling pairs of Booroola ewe and ram lambs, to evaluate their usefulness as control-treatment pairs in endocrinological studies.

In ewe lambs, FSH concentrations increased with liveweight and age to reach distinct peaks at 7-12 kg liveweight and at 3-9 weeks of age. Variation between pairs was significantly greater than within pairs for liveweight at all ages, FSH concentrations up to 9 weeks of age, and maximum FSH concentration, but not for age or liveweight at peak FSH.

In ram lambs, FSH concentrations were lower than in ewes but followed a similar pattern, with FSH peaks at 12-17 kg liveweight and 5-10 weeks of age. The geometric mean maximum FSH concentrations were 1.44 and 0.81 (LSR 1.78) ng/ml for BB and ++ groups respectively ( $P < 0.05$ ). Between-pair variation was significantly greater than within-pair variation for birth weight, liveweight to 7 weeks of age, peak FSH, and age at peak FSH.

Mean FSH concentration and liveweight showed no relationship within or between ram pairs, or between ewe pairs, but the heavier sibling within all six ewe pairs had the higher FSH concentration ( $P < 0.05$ ) most samples up to 7 weeks of age, but not thereafter. The reduced variation in gonadotrophin concentrations within compared with between fullsibling pairs suggests they have considerable value as control-treatment animals in endocrine studies on prepubertal development.

**Keywords** Booroola, Fec<sup>B</sup>, fullsibblings, dizygotic, ewe lamb, ram lamb, plasma FSH, liveweight.

### INTRODUCTION

Monozygotic twins used as control-treatment pairs in experimental studies have been shown to be superior to randomly selected animals for a number of characteristics in cattle (Hancock, 1950; 1951; 1952). In the ovine the use of dizygotic twins or fullsibblings is more appropriate as, in the absence of cloning, the incidence of identical twinning in sheep is very low (Willadsen, 1980).

Research aimed at understanding the physiology of reproduction relies in part on measuring gonadotrophin concentrations, but little is known of the usefulness of twins in reducing experimental variability in these situations. The aim of this study was to compare variation in follicle stimulating hormone (FSH) profiles within and between prepubertal fullsibling pairs of Booroola ewe and ram lambs.

### MATERIALS AND METHODS

Six Fec<sup>B</sup>Fec<sup>B</sup> (BB) ewe, 3 BB ram and 5 Fec<sup>+</sup>Fec<sup>+</sup> (++) ram fullsibling pairs were generated from an interbred Booroola Merino x Romney flock at the Invermay Agricultural Centre, Mosgiel, New Zealand. Two BB and two ++ rams were mated with ewes of like genotype to produce the lamb pairs. BB lambs were born in a range of litter sizes (2 pairs each of twin, triplet and quadruplet ewe lambs; 1 pair each of twin, triplet and quadruplet ram lambs), while ++ lambs were all twins. Within sex all BB fullsibling pairs had the same sire but four ++ ram pairs were sired by one ram and one ++ pair by the other. Sires were the sons of progeny tested rams classified as BB or ++ on the basis of four ovulation records from 31-36 daughters per sire, according to the criteria of Davis *et al.* (1982). Three ovulation rates

<sup>1</sup> MAF Technology, Wallaceville Animal Research Centre, P.O. Box 40063, Upper Hutt, New Zealand.

were used to classify the dams of the fullsibling pairs and dams of the sires used to generate the pairs (Davis *et al.*, 1982). The heritability of ovulation rate based on this classification has been confirmed by other research groups around the world (Piper *et al.*, 1991).

Lambs were born in September/October 1989. Blood samples were collected once weekly between two and 20 weeks of age by jugular venipuncture. The range of ages at first bleed was 14-21 days. Liveweights were recorded at two to three weekly intervals. FSH concentrations were measured by the method described by McNatty *et al.* (1989) using the same reference and iodination agents and antisera. The coefficient of variability was 8.6% between assays and 7.5% within assays. The detection limit was 0.2 ng/ml.

For individual plasma profiles the log FSH concentration, age and liveweight at peak FSH, birth weight, and liveweight at 7 weeks of age were analysed by analysis of variance for each sex, giving the variance ratio (VR) of between to within pairs. Ram liveweight and FSH parameters (Table 1) were adjusted for genotype to prevent possible differences from inflating the between-pair variance, but no birth rank adjustments were made for either sex.

The relationship between FSH concentration and liveweight, and FSH concentration and age were investigated within and between pairs. Sire effects could not be evaluated because only one ram was used to generate BB lambs, and there were insufficient numbers per sire of ++ lambs.

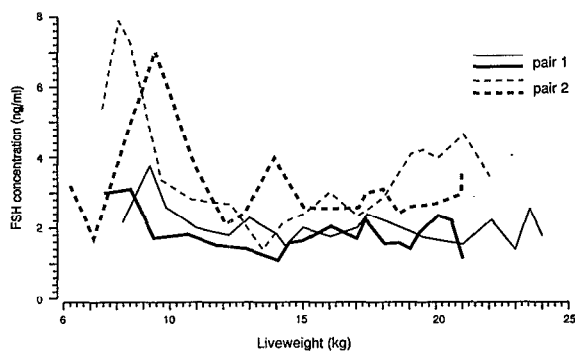
**TABLE 1** Variance ratios for FSH profiles and their associations with age and liveweight for Booroola ewe and ram lamb fullsibling pairs.

	Variance Ratio	
	BB ewe	BB and ++ ram
Log <sub>e</sub> Peak FSH	9.30 **	5.11*
Age at peak FSH	3.63	3.76*
Liveweight at peak FSH	3.20	3.11
Birth weight	12.13**	3.61*
Liveweight at 7 weeks of age	6.55*	3.94*
Critical value (5%)	4.39	3.58
(1%)	8.74	6.37

## RESULTS

### Ewe Lambs

Mean FSH concentrations increased with age and liveweight to a maximum of 3.86 (SE 0.70) ng/ml at 8 kg, and then declined. Of the 12 individual ewe profiles, 10 exhibited distinct peaks in FSH between 7 and 12 kg liveweight at ages varying from 3 to 9 weeks (Fig 1). There was significantly greater between-pair than within-pair variation for liveweight at all ages, FSH concentrations up to 9 weeks of age, and maximum FSH concentration between 3 and 9 weeks of age, but not for age or liveweight at maximum FSH concentration (Table 1). There was no evidence of a relationship between mean FSH concentration and liveweight between pairs at fixed ages. Within all six pairs, however, the heavier sibling had the higher FSH concentration ( $P < 0.05$ ) for most samples up to 7 weeks of age, but not thereafter.



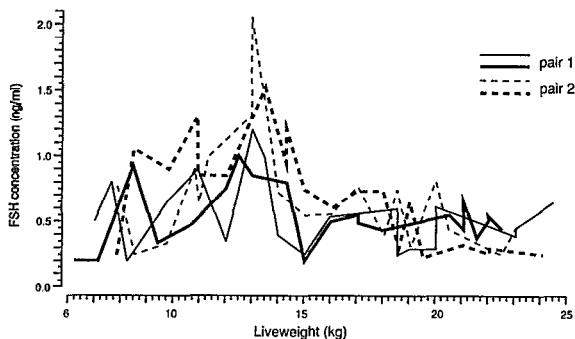
**FIG 1** Relationship between plasma FSH concentration and liveweight for two pairs of fullsibling BB ewes.

### Ram Lambs

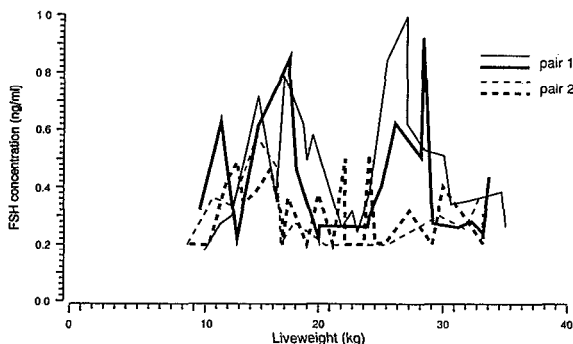
FSH concentrations in ram lambs were much lower than ewe lambs, but followed a similar pattern over age and liveweight, with 15 of 16 showing distinct peaks between 5 and 10 weeks of age and 12-17 kg liveweight (Figs 2 and 3). The geometric mean maximum FSH concentrations were 1.44 and 0.81 (LSR 1.78) ng/ml for the BB and ++ groups respectively, ( $P < 0.05$ ).

There was significantly greater between-pair than within-pair variation for birth weight, liveweight

to 7 weeks of age, peak FSH between 5 and 10 weeks of age, and age at peak FSH (Table 1). Variation in liveweight at peak FSH was similar within and between pairs (Table 1). No evidence of a relationship between mean FSH concentrations and liveweight was found within or between ram pairs at fixed ages.



**FIG 2** Relationship between plasma FSH concentration and liveweight for two pairs of full-sibling BB rams.



**FIG 3** Relationship between plasma FSH concentration and liveweight for two pairs of full-sibling ++ ewes.

Variance ratios were generally higher in ewe than ram lambs for the same parameters, and were highest for peak FSH, birth weight and liveweight at 7 weeks of age (Table 1).

### DISCUSSION

Plasma FSH profiles in BB ewe pairs followed a similar

pattern to that described by Bindon *et al.* (1985) and Montgomery *et al.* (1989). The lower FSH concentrations in ram compared to ewe lambs are also consistent with the findings of Montgomery *et al.* (1989), but the distinct peaks in FSH observed for individual ram profiles were not seen in the previous study. Maximum FSH concentrations occurred at approximately the same age range, but heavier liveweights in ram than ewe lambs.

The geometric mean maximum FSH concentration was higher for BB than ++ ram lambs, although the between genotype difference would have been greater but for two ++ ram lambs that had peak FSH values consistent with those observed for BB ram lambs. These two ++ rams were full-siblings sired by a different sire to the remaining ++ pairs, suggesting the difference observed may be the result of sire rather than genotype effects.

More data are needed before establishing with certainty whether it is genotype or sire which contributes to the apparent differences in FSH between Booroola genotypes. It is possible that the between-pair variance for liveweight parameters are inflated by an inability to adjust for birth rank. However, Bindon *et al.* (1985) and Montgomery *et al.* (1989) found no effect of birth rank on plasma FSH concentrations in Booroola ewe lambs. The greater variation in birth rank may also explain the greater between-pair than within-pair variation, for liveweight in ewe than ram lambs; 75% of ram pairs were born as twins but a maximum of 33% of ewe pairs were born to any one litter size.

Despite the possible effects of birth rank, variance ratios in the present study probably underestimate the value of dizygotic pairs for experimental studies. The majority of full-sibling pairs within sex and genotype groups were sired by the same ram, so that between-pair variation is largely a measure between half-siblings. The use of randomly selected pairs of animals for between-pair comparisons would have given greater between-pair variation.

That the heavier female full-sib within a pair had the higher FSH concentration is interesting. In adult ewes the ++ genotypes are generally heavier, but have lower FSH concentrations than the BB genotypes. The lack of evidence of a positive relationship between FSH and liveweight between pairs of ewe lambs suggests

that genetic variation in these traits may be masking such a relationship.

### CONCLUSION

Reduced variation in gonadotrophin concentrations within compared to between fullsibling pairs suggests they have considerable value as control-treatment animals in endocrine studies on prepubertal development.

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