

INCREASED OVULATION RATE, BUT NOT FSH OR LH
CONCENTRATIONS, IN EWES SUPPLEMENTED WITH LUPIN GRAIN

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SUMMARY

The involvement of gonadotrophins in the control of ovulation rate was examined in ewes fed either oaten hay alone or supplemented with lupin grain for 10 days. The variability in plasma FSH concentration in ovariectomized ewes fitted with progesterone and oestradiol implants was greater for lupin-fed than control ewes ($P < 0.001$) but mean FSH values were similar. In progesterone - implanted entire ewes, supplementation increased the ovulation rate (1.19 vs 1.40) but had no effect on FSH. In ewes which subsequently produced twin ovulations, FSH levels were higher two days before implant removal and the decline in FSH two days after removal was greater than for single ovulating ewes. Live weights were also higher for ewes with two ovulations. The increase in ovulation rate produced by lupin feeding did not appear to be due to increased secretion of LH or FSH, but may have been due to increased ovarian sensitivity to gonadotrophins.

Keywords: Ovulation rate, lupins, FSH, LH

INTRODUCTION

The ovulation rate of ewes grazed on dry pasture can be increased after supplementation with lupin grain (Knight et al. 1975), even within six days of the start of feeding (Oldham and Lindsay 1984). Some studies show that lupin feeding may increase FSH secretion (Brien et al. 1976; Knight et al. 1981; Nottle et al. 1987), but Radford et al. (1980) reported no differences in LH or FSH associated with increased ovulation rates.

Lupin feeding is a useful model for investigating the effect of nutrition on ovulation rate mediated by gonadotrophins. However, in ewes with a higher ovulation rate there may be an altered feedback of ovarian hormones to the pituitary, making it difficult to interpret possible changes in circulating gonadotrophins. Accordingly, we examined the effect of a short period of lupin feeding on intact ewes, and on similar ovariectomized (ovex) ewes fitted with steroid implants to simulate ovarian feedback.

MATERIALS AND METHODS

Mature Merino ewes ($N = 120$; live weight 43.6 ± 0.6 kg) were stratified on live weight and allocated to either a control group (oaten hay fed ad lib.) or a lupin-fed group (additional 600 g/head of lupin grain/day for 10 days starting on Day 0). Live weights were taken before and after the experiment. Five days before lupin feeding commenced (Day -5), all ewes received a subcutaneous progesterone implant (Silestrus; Abbott Labs.). On the same day a subgroup ($n = 15$) from each treatment was ovex. and fitted with a 5 mm Silastic implant (601-265 tubing, Dow Corning Corp.) containing powdered oestradiol-17 Beta (release rate 8 $\mu\text{g/day}$). Progesterone implants were removed from the intact ewes on Day 6, and on Day 15 ovaries were examined by laparoscopy and corpora lutea counted.

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Blood samples were collected daily from all ewes, starting on Day -2. In addition ovex. ewes were sampled every 15 min. for 4 hr on Days -2, +3 and t8. Blood was centrifuged immediately and the plasma stored at -10°C until assay. All samples from each animal were examined in one assay. FSH concentrations in plasma were determined by the homologous ovine double antibody radioimmunoassay kit supplied by the Pituitary Hormone Distribution Programme of NIADDK and validated in our laboratory. The kit utilized highly purified ovine FSH (NIAMDD-oFSH-RP-1) and highly specific ovine FSH antiserum (NIAMMD-anti-oFSH-1). The coefficients of variation of three ovine plasma pools (mean \pm s.e.m.; 0.25 ± 0.02 , 0.74 ± 0.03 , 7.70 ± 0.60 ng FSH/ml) within assays were 13.9, 6.5 and 13.3% and between assays were 20.1, 8.1 and 15.6% respectively. The sensitivity of the assay (twice the standard deviation of the error in the zero standard) was always less than 0.1 ng FSH/ml. LH concentrations were measured by a double antibody radioimmunoassay (Martin et al. 1980).

FSH concentrations were analysed by analysis of covariance of a split plot design using the mean pre-feeding value as a covariate. In ovex. ewes it appeared that the variation of FSH was greater in ewes fed lupins than in controls and a rank analysis of covariance (Quade 1967) was used to test the hypothesis that standard deviations in the FSH concentrations of ovex. ewes in the two groups were equal. The two groups of intact ewes were also compared on individual days using the t-test. Pulses and basal (non-pulse) LH levels (see Goodman and Karsch 1980) were examined by analysis of variance.

RESULTS

Live weights of intact ewes fed the control ($n = 42$) and lupin ($n = 43$) diets were the same at the start of the experiment (43.4 ± 0.8 and 43.4 ± 0.9 kg) but were significantly different by the end of the feeding period (41.8 ± 0.9 and 44.7 ± 1.1 kg, $P < 0.001$). Ovulation rates for the lupin-fed animals was higher than for controls (1.40 vs. 1.19 ovulations/ewe, $P < 0.05$). At the end of the feeding period the live weights of ewes with a single ovulation were lower than those of ewes with two ovulations (41.4 ± 0.7 vs. 48.2 ± 1.2 kg, $P < 0.001$).

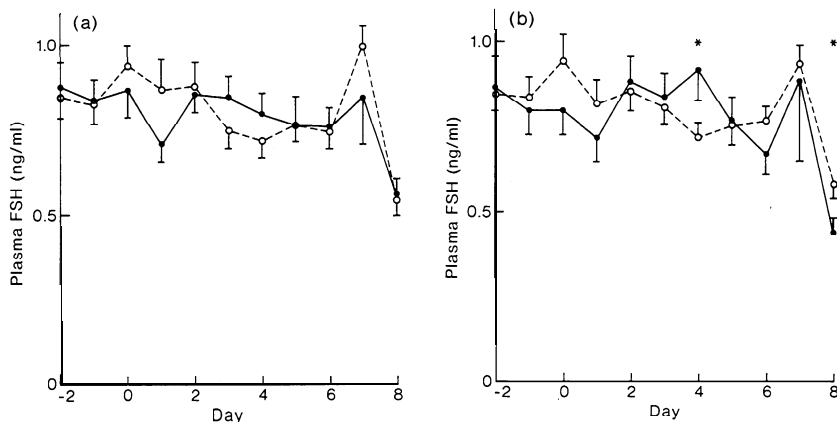


Fig. 1 Mean (\pm s.e.m.) plasma FSH concentrations in intact ewes (a) fed hay (0---0) or hay supplemented with lupins (●---●) and (b) which subse quently had single (0---0) or twin (●---●) ovulations. * - $P < 0.05$

FSH decreased markedly in intact ewes on Day 8, two days after removal of progesterone implants (Fig. 1). The mean daily FSH concentrations during the experimental period did not differ between control and lupin-fed groups. When

ewes were grouped according to ovulation rate, significant differences were found on Days 4 and 8 (i.e. 5 and 1 days before the expected ovulation); ewes which subsequently had twin ovulations had higher FSH on Day 4 (two days before implant removal), and lower FSH on Day 8 than single ovulating ewes.

FSH in ovex. ewes increased during the experiment (Fig. 2), but there was no difference in the overall mean concentrations between control and lupin-fed animals (4.54 ± 0.12 vs. 4.90 ± 0.15 ng FSH/ml plasma respectively). Rank analysis of covariance showed that the ranks of standard deviations of lupin-fed ewes were higher ($F_{1, 28} = 14.66$, $P < 0.001$) than for control ewes. This enhanced variability was not detected in the lupin group before the feeding period commenced, and was also not seen in intact ewes.

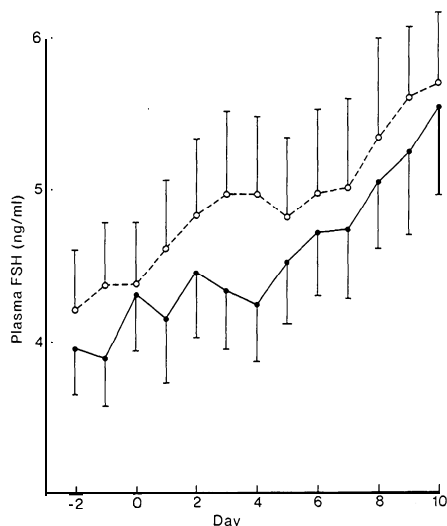


Fig. 2 Plasma FSH concentration in ovex. control (●—●) and lupin-fed (○—○) ewes. Lupin feeding commenced on treatment Day 0.

Plasma LH concentrations in ovex. ewes increased between Day 3 and Day 8 regardless of diet (Table 1). However, there was no difference in LH between control and lupin-fed animals; whether expressed as the mean number of pulses, basal concentration or the overall mean concentration of LH.

Table 1 Mean plasma LH in ovex. ewes with oestradiol and progesterone implants and fed hay alone (C-control) or supplemented with lupins (L)

	No. pulses/ 4 h/ewe		Basal LH (ng/ml)		Mean LH (ng/ml)	
	C	L	C	L	C	L
Pre-feeding	2.67	2.87	1.72	2.06	2.69	3.39
3 days feeding	2.73	2.47	1.52	1.43	2.69	2.37
8 days feeding	3.67	3.47	3.20	2.96	4.92	5.08

DISCUSSION

The ovulation rate was higher in intact ewes fed lupins than in controls, but we found no difference between these groups in the circulating FSH levels. Lupin feeding also had no effect on LH or mean FSH in ovex. ewes. However, there was an increase in day-to-day variability in FSH after lupin feeding of ovex. ewes and this may be worthy of further study. The steroid implants used in

this study were small, which permitted a slight increase in LH and FSH in ovex. ewes throughout the experiment (see Fig. 2). Thus, the failure to detect a gonadotrophic response to lupin feeding in ovex. ewes was not because pituitary function was completely suppressed or because of maximal pituitary secretion.' Interestingly, Nottle et al. (1987) recently found that FSH in oestradiol-implanted ovex. ewes increased significantly one day after the start of lupin feeding but the FSH levels were about 10 times greater in that study suggesting that our assay may have higher specificity for FSH.

FSH levels were greater two days before progesterone implant removal in ewes which were going to have twin ovulations, which is similar to previous reports (Davis et al. 1981; McNatty et al. 1985). Higher FSH concentrations before luteolysis have been reported previously in lupin-fed ewes (Brien et al. 1976; Knight et al. 1981), but in those studies the ewes were fed lupin grain for several weeks before examination, so that the observed differences in FSH may have reflected longer term altered ovarian feedback (steroid, inhibin) from the increase in twinning. The decrease in FSH two days after implant removal was greater ($P < 0.05$) in ewes with twin ovulations, and may have been due to the enhanced secretion of oestradiol and inhibin from the two preovulatory follicles. By contrast, McNatty et al. (1985) did not observe a difference between single and twin ovulating Romney ewes in preovulatory concentration of FSH after luteolysis.

The present study was larger and more thorough than previous studies, and was carried out over a very short period so that ewes were examined before the new equilibrium between ovary and pituitary had been established. We conclude that lupin feeding did not cause an increase in the secretion of FSH. The difference in ovulation rate may therefore be due to the increased ovarian sensitivity to gonadotrophins.

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