Serum concentrations of insulin-like growth factor-I during the oestrous cycle in ewes selected for lamb production

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ABSTRACT

Concentrations of insulin-like growth factor-I (IGF-I) in serum are increased in cattle selected for twinning. To determine if a similar relationship exists in sheep, we examined changes in serum IGF-I concentrations during the oestrous cycle in Targhee ewes from lines with a documented differential in ovulation rate. Ewes were from a line selected for weight of lamb weaned (HI; N=24) or a control line (CON; N=25). Oestrous cycles were synchronized by insertion of vaginal pessaries containing 60 mg of medroxyprogesterone acetate for 12 days. Blood samples were collected daily for 19 days beginning on the day of pessary removal. Concentrations of IGF-I and progesterone in sera were determined by radioimmunoassay. Ovulation rates, determined by laparoscopy on day 7 after pessary removal, did not differ between HI and CON lines ($P>0.10$). Overall, concentrations of IGF-I did not differ between lines, averaging 127.4 and 135.7 ng/ml for HI and CON ($P>0.40$). Nor was there an interaction between line and day of oestrous cycle ($P>0.60$). The main effect of day of oestrous cycle on serum IGF-I concentrations was highly significant ($P<0.001$). Serum IGF-I concentrations peaked at 204.3 ng/ml on the day after pessary removal, declined thereafter, and returned to day 1 levels by day 19. IGF-I concentration varied inversely with progesterone in serum. In summary, serum IGF-I concentrations did not differ between selected and control lines, but changed markedly in association with stage of the oestrous cycle, peaking at the time of oestrus.

Keywords: Oestrous cycle, IGF-I, progesterone, sheep, genetics.

INTRODUCTION

The importance of the relationship between insulin-like growth factor-I (IGF-I) and ovarian function and activity is becoming increasingly clear. IGF-I is known to stimulate both follicular development and steroidogenesis in the ovary (Adashi *et al.*, 1985; Giudice, 1992). The observation that concentrations of IGF-I in blood and follicular fluid are higher in cows carrying twins than in single-bearing cows (Echternkamp *et al.*, 1990) implies a role for IGF-I in mediating genetic differences in ovulation rate. However, a recent study found no relationship between ovulation rate and serum IGF-I concentrations in ewes (Spicer and Zavy, 1992).

Other findings implicate the ovary as a potential regulator of IGF-I production. Carlsson *et al.* (1989), reported that levels of IGF-I mRNA and protein in the ovary vary with stage of the oestrous cycle in rats. Serum IGF-I concentrations declined after ovariectomy of cows but increased after estradiol treatment (Richards *et al.*, 1991). Others have shown that estrogen treatment increases IGF-I in circulation of cows (Bass *et al.*, 1989). Recently, Spicer and Zavy (1992) found that serum concentrations of IGF-I vary during the oestrous cycle in ewes, in conflict with an earlier report (Davis *et al.*, 1990). These reports highlight the need for further study of the interplay of IGF-I and ovarian function.

The objectives of this study were 1) to determine changes in serum IGF-I concentrations during the oestrous cycle in ewes and 2) to compare profiles of serum IGF-I in ewes from lines with a documented differential in ovulation rate.

MATERIALS AND METHODS

Targhee ewes were from a line selected for high weaning weight (HI; N = 24) or a randomly selected control line (CON; N = 25). Lines were established at the U.S. Sheep Research Station in Dubois, Idaho, USA, and differed significantly in lamb weaning weight and ovulation rate (Stellflug *et al.*, 1990). The animals were managed under range conditions at the USDA Sheep Station. To synchronize oestrous cycles, vaginal pessaries containing 60 mg medroxyprogesterone acetate were inserted in all ewes. Pessaries remained in place for twelve days. Blood samples were collected daily for 19 days beginning on the day of pessary removal. Sera were harvested and frozen pending assay. Seven days after pessary removal, laparoscopy was performed to determine ovulation rate.

Concentrations of IGF-I in daily serum samples were determined by radioimmunoassay after methanol:formic acid (9:1) extraction of sera as described previously (Herring and McFadden, 1990). Concentrations of progesterone in sera were determined in samples collected on alternate days of the trial using a commercial radioimmunoassay kit (Coat-a-count, Diagnostic Products Corp.).

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Statistical analyses were done using the general linear models procedure of SAS (1985). Serum IGF-I and progesterone data were analyzed as a split-plot design in which effects of genetic line, sample-day, and the line by sample-day interaction were tested. Ovulation frequency data were compared by Chi-square analysis.

RESULTS AND DISCUSSION

Despite previously-established differences (Stellflug *et al.*, 1990), ovulation rate was similar in both genetic lines (P>.10) in the present experiment. The explanation for this discrepancy is unclear, but may be related to the oestrus-synchronization procedure used. Although in previous studies ovulation rate was higher in the selected line both before and after oestrus-synchronization, a possible effect of synchronization was observed in both lines (Stellflug *et al.*, 1990).

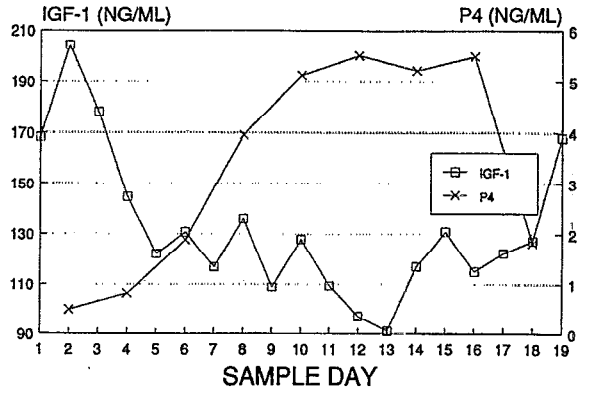
Because ovulation rate did not differ in this trial, inferences relative to this parameter cannot be based on observed differences, but only on the established genetic difference between lines. In this regard, IGF-I concentrations did not differ between lines, averaging, over the entire trial, 127.4 and 135.7 ng/ml for HI and CON lines, respectively (P>.40). Furthermore, there was no significant interaction between genetic line and sample-day (P>.60), thus data were pooled for determination of mean IGF-I concentrations for each day of the oestrous cycle. The absence of an interaction between genetic line and day of the oestrous cycle indicates that changes in endocrine profiles, as well as overall means, were similar in both genetic lines.

Although effects of genotype on concentrations of IGF-I in blood have been reported in several species, Medrano and Bradford (1991) found only small differences due to genetic line in plasma IGF-I of lambs from lines of Targhee sheep selected or controlled for high weaning weight, similar to the lines used in the present study. As for genetic influence on reproduction, Echternkamp *et al.* (1990) reported concentrations of IGF-I in serum were higher in twinning than single-bearing cattle. In sheep however, Spicer and Zavy (1992) found no difference in serum IGF-I between genotypes of cross-bred sheep differing widely in ovulation rate, but did observe age-related differences. Our findings confirm those of Spicer and Zavy (1992) in cross-bred sheep and extend the comparison to divergent lines within a single breed.

Profiles of IGF-I and progesterone in serum during the oestrous cycle are depicted in Figure 1. IGF-I concentrations varied significantly during the sampling period (P<.001), increasing markedly to peak at oestrus (day 2; 204.3 ng/ml; P<.01), then declining rapidly to 122.8 ng/ml on day 5 (P<.01). Low levels continued during the luteal phase, before increasing again in association with the ensuing predicted oestrus. Minimum concentrations of IGF-I (90.1 ng/ml on day 13) were approximately one-half of peak levels (P<.01). As expected, progesterone concentrations also varied significantly (P<.001) during the cycle, and serve to confirm the relative normalcy of the cycle as well as revealing the timing of changes in follicular function.

These results, based on daily sampling of a large number of ewes (N=49), confirm the report of Spicer and Zavy (1992)

FIGURE 1: Changes in serum concentrations of IGF-I and progesterone during the ovine oestrous cycle. Pooled SE's were 6.8 ng/ml for IGF-I and 0.2 ng/ml for progesterone.



who found a similar pattern of serum IGF-I concentrations during the oestrous cycle in ewes sampled every other day. In contrast, Davis *et al.* (1990) failed to detect such changes in IGF-I in ewes sampled twice-weekly during the oestrous cycle. In addition, our data show that IGF-I concentrations vary inversely with progesterone concentrations in blood. Whether this is a direct effect of progesterone or simply a reflection of changes in estrogens is uncertain, however the latter seems more likely based on the stimulatory effect of estrogen on hepatic IGF-I production (Bass *et al.*, 1989; Richards *et al.*, 1991). Nevertheless, it is not clear that the liver is the sole source of the oestrus-associated rise in serum IGF-I since IGF-I production by ovary (Carlsson *et al.*, 1989) and uterus (Murphy *et al.*, 1988) is also increased by estrogen.

We conclude that 1) serum IGF-I concentrations in ewes change markedly in concert with the oestrous cycle and peak at oestrus, but 2) do not differ based on genetic difference in ovulation rate/lamb production potential. The observed changes in IGF-I concentrations during the oestrous cycle are significant in several respects. First, the importance of the association between ovarian function and circulating IGF-I concentration is further emphasized. Second, concomitant changes in ovarian steroids and IGF-I potentially complicate interpretation of endocrine regulation of events associated with oestrous cyclicity. And third, variation in serum IGF-I during the oestrous cycle clearly must be considered in designing and interpreting experiments involving cycling females.

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