Fertility of anestrus ewes infused with gonadotropin releasing hormone or injected with pregnant mares' serum gonadotropin

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ABSTRACT

Two experiments were conducted to compare PMSG injection to GnRH infusion for induction of breeding in anoestrous ewes. In Experiment One, sixteen Suffolk ewes were pretreated for 14 days with 30 mg Cronolone sponges. At sponge removal half of the ewes received 0.125 µg/hr GnRH for 72 hrs via subcutaneous osmotic pumps. The other half received a single injection of 750 iu PMSG. Plasma LH levels gradually increased and were sustained for 24 hrs. Estrus, ovulation and *corpus luteum* function were expressed in half of the ewes treated with GnRH. All ewes treated with PMSG exhibited estrus, ovulated and formed functional *corpora lutea.*

In Experiment Two, 24 ewes were divided into two groups and the same treatments as in Experiment One were applied. Fertile rams were introduced to all the ewes at the time of sponge removal. Estrus was detected in 75% of the GnRH treated ewes vs 100% in PMSG treated ewes. Lambing rates were 44% and 56% in GnRH and PMSG groups respectively.

These results suggest that the induction of fertility using continuous infusion of GnRH is encouraging. However, other GnRH delivery systems should be investigated.

Keywords Ewes, seasonality, induced fertility, GnRH, PMSG

INTRODUCTION

Continuous infusion of anestrous ewes with low doses of GnRH through a cannula have been successful in inducing estrus, ovulation and normal luteal function (Mcleod *et al.*, 1983). This indicates that a sustained pulsatile delivery of LH to the ovary may not be an absolute requirement for follicular maturation (McNatty *et al.*, 1981).

Continuous GnRH infusion would be more practical from a developmental viewpoint if a more convenient method of GnRH administration could be substituted for the use of a cannula and external infusion pump. Two experiments were conducted to compare the effects of GnRH infusion and PMSG injection for the induction of fertility in ewes out-of-season.

MATERIAL AND METHODS

Experiment 1

Sixteen 3- to 4-year old Suffolk ewes weighing 71±2.5 kg (mean±SEM) were pretreated for 14 days with 30 mg intravaginal Cronolone sponges. At the time of sponge removal half of the ewes received osmotic pumps (Alzet 2001) which delivered 0.125 µg/hr GnRH for 72 hrs. The other half received a single injection of 750 iu PMSG. Ovaries of all ewes were examined using endoscopy on the day of sponge insertion and again on the day of pumps removal. Progesterone and LH concentrations were measured using RIA. The number of ewes that were observed in estrus and ovulated were recorded.

Experiment 2

Twenty-four 3-5 year old crossbred ewes weighing an average of 80.5±2.6 kg were pretreated for 12 days with 30 mg Cronolone sponges. On the day of sponge removal half of the ewes were infused for 72 hrs with 0.125 µg/hr GnRH via osmotic pumps. The other half received a single injection of 750 iu PMSG. The number of ewes detected in estrus, number of ewes lambed and number of lambs born were recorded.

Statistical analyses of both experiments were determined according to Ott (1984).

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RESULTS

Experiment 1

The first endoscopic examination revealed all ewes had "inactive" ovaries. *Corpora lutea* were observed in four of the eight ewes in the GnRH treatment group (Table 1). These ewes were detected in estrus. All ewes treated with PMSG were detected in estrus and ovulated. Differences between the two treatment groups were significant (P<0.05).

TABLE 1 Estrus and ovulation in ewes in each of the treatment groups in Experiment 1

Treatment	No. of Ewes in Estrus	No. of Ewes Ovulating	No. of Ewes
0.236 µg/hr GnRH	8	4[a]	4[a]
750 iu PMSG	8	8[b]	8[b]

[a,b] P<0.05

The mean plasma LH profile for GnRH treated ewes is shown in Figure 1. Plasma LH concentrations rose gradually and were sustained for 24 hrs. Levels started to decline and then increased to 6.6+1.8 ng/ml. Significant (P<0.05) differences in LH concentrations were obtained between times during GnRH infusion.

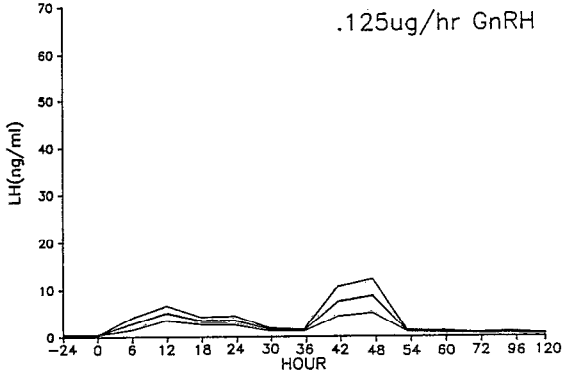

FIG 1 Plasma luteinizing hoemone concentrations (ng/ml, mean ± SEM) in ewes treated with 0.125 µg/hr gonadotrophin-releasing hormone before (6 through 72 hrs) and after (96 through 120 hrs) gonadotrophin-releasing hormone infusion.

Progesterone patterns were normal in all ewes ovulating in the GnRH treatment group (Figure 2). Levels rose within 4 days of GnRH infusion to a level of 1.5+0.1 ng/ml. This was maintained for 6 days then increased to a level of 2.4+0.4 ng/ml for the next 6 days. Plasma levels in PMSG treated ewes increased to 2.0+0.3 ng/ml within 4 days of sponge removal and injection of PMSG. They then increased rapidly to 9.9+1.2 ng/ml and remained at this concentration for 9 days. Differences in progesterone concentration in ewes that ovulated in response to GnRH and PMSG treatment groups were significant (P<0.05).

Experiment 2

Estrus was detected in 9 of the 12 ewes in the GnRH treatment group (Table 2).

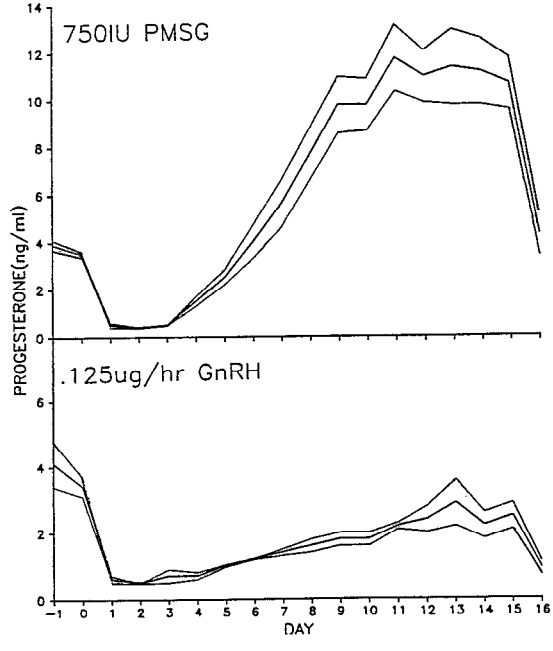

FIG 2 Plasma progesterone concentrations (ng/ml, mean ± SEM) in ewes that ovulated in gonadotrophin-releasing hormone (GnRH) and pregnant mare serum gonadotrophin (PMSG) treatment groups. In GnRH treated ewes prosgesterone-releasing devices were removed on day 0. GnRH osmostic pumps were inserted in day 0 and left in place for 3 days. In PMSG treated ewes progesterone-releasing devices were removed and PMSG was injected on day 0.

Four of the 9 ewes that showed estrus lambed. Five lambs were born in this treatment group. All ewes treated with PMSG showed estrus. Five of the 12 ewes lambed and a total of 12 lambs were born. These treatment differences were not significant (P>0.05).

TABLE 2 Estrus and lambing data from ewes treated with gonadotropin-releasing hormone and pregnant mare serum gonadotropin in Experiment 2.

Treatment	No. ewes treated	No. ewes in estrus	No. ewes lambed	No. of lambs
0.125 μg/hr GnRH	12	9	4	5
750 iu PMSG	12	12	5	12

DISCUSSION

Continuous infusion of GnRH resulted in estrus, ovulation and *corpus luteum* function in only half of the ewes. Infusion of 0.125 μg/hr GnRH did not induce spike release of LH. Plasma levels gradually increased and were then sustained for a period of time. This was expected to improve the response to GnRH infusion. Factors underlying the 50% success rate are unknown. Results demonstrated that mean plasma LH levels and LH peaks were significantly higher in ewes that ovulated than in ewes that did not. These results suggest that there may be some aspect of the LH profile that is important and only half of the ewes treated were able to achieve it.

Another factor that may affect responses to GnRH treatment is the stage of follicular development at the onset of GnRH administration. It has been shown that during seasonal anestrus there are alternating waves of follicular development and regression (Matton *et al.*, 1977). It is possible that ewes that showed estrus, ovulated and formed a functional *corpus luteum* were those that had ovarian follicles at a responsive stage.

Infusion of GnRH is not as effective as PMSG in stimulating estrus and ovulation. This may be related to the FSH-like and LH-like activity of PMSG. It has been reported that PMSG rapidly changes growing follicles into antral follicles (Mariana and Machada, 1976), stimulates multiplication of granulosa cells and prevents follicular atersia (Mauleon and Mariana, 1977). The mode of action of GnRH is different from that of PMSG. The low dose of GnRH used in this study is below the threshold needed to cause FSH release (Wheaton *et al.*, 1984). The lower response to GnRH treatment may be related to lack of FSH activity and inability to stimulate early follicular development.

The overall lambing rate in Experiment 2 was lower (38%) than usual (60 to 70%) for out-of-season breeding trials. Reasons for lower fertility, especially in the PMSG treatment group, are not known. The absence of a second PMSG injection in the PMSG treatment group may explain part of the lower fertility. Two PMSG injections 16 days apart produce about 20% higher fertility than one injection (Gordon, 1963; Hulet and Foote, 1969).

In conclusion, continuous infusion of GnRH via osmotic pumps cannot be considered of practical value. Other GnRH delivery systems need to be developed and evaluated.

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