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Effects of Ovarian Hormones on Ovarian Capillary Blood Flow in Anoestrous Ewes

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Abstract

The indicator fractionation technique with [^{86}Rb]rubidium chloride as the indicator was used to determine the relative blood flow (RBF) as a measure of capillary blood flow in the ovaries of conscious, hormonally treated, anoestrous ewes. Treatment of ewes with either progesterone only or oestradiol only had no effect on ovarian RBF, but treatment with oestradiol subsequent to progesterone caused a significant increase ($P < 0{\cdot}001$).

Consequently, it appears that progesterone-induced sensitivity of the ovarian vasculature to the vasodilatory effects of oestradiol may be responsible for increased ovarian blood flow around oestrus in cyclic ewes.

Introduction

In ewes, capillary blood flow in the non-luteal tissue of the ovary (stroma plus follicles) is greater around oestrus than at the mid-luteal stage of the oestrous cycle (Brown *et al.* 1974*a*). In rats (Wurtman 1964) and rabbits (Janson 1975) luteinizing hormone (LH) causes an increase in ovarian blood flow. However, it has been reported that, in sheep, LH treatment produces only a small increase in ovarian blood flow (Cook *et al.* 1969; Niswender *et al.* 1976) or has no significant effect (Hixon and Clegg 1969). On the other hand, a considerable amount of oestradiol-17β is secreted by the ovaries at oestrus and this hormone is known to induce marked and prolonged increases in capillary blood flow within organs of the female genital tract (Brown *et al.* 1974*b*; Brown and Mattner 1977). Further, in a study on uterine blood flow in sheep, Greiss and Anderson (1970) found that progesterone sensitizes the vascular bed of the uterus to the effects of oestradiol. It is possible therefore that these two hormones might also stimulate ovarian capillary blood flow. Accordingly, in the present study, anoestrous ewes were used so that the effects would not be confounded by endogenous ovarian hormones.

Materials and Methods

During the normal anoestrous period, 16 mature Merino ewes that had not exhibited oestrus for 2 months were used. The ewes were randomly allocated to four groups of four animals and were treated with various regimens of oestradiol-17β and progesterone as shown in Table 1. The hormones were dissolved in peanut oil and injected intramuscularly; blood flow measurements were obtained in each ewe 24 h after the last hormone injection.

The indicator fractionation technique of Sapirstein (1958) as modified by Brown *et al.* (1974*a*) and Brown and Mattner (1977) for the purpose of determining ovarian capillary blood flow was used. The sheep were given a local anaesthetic and then a catheter (1·4 mm i.d.; 1·9 mm o.d.) was inserted into the posterior vena cava via a recurrent tarsal vein so that the catheter tip lay near the entrance

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to the right atrium. Two hours later, with the ewe conscious and standing, 5·6 MBq [^{86}Rb]rubidium chloride (Australian Atomic Energy Commission, Lucas Heights, N.S.W.) in 4 ml of saline was injected rapidly through the catheter and the heart was stopped 30–40 s later by the injection, through the same catheter, of 40 ml of saturated potassium chloride containing 10 g sodium pentobarbitone. The ovaries were removed within 1 min, weighed and their radioactivity measured in an autogamma spectrometer (Packard Instruments; 400–2000 KeV).

As cardiac output was not measured in these experiments, relative blood flow (RBF) [amount of radioactivity in the tissue expressed as a fraction of the dose—an expression devised by Waites and Setchell (1966)] was calculated for each ovary.

Data were analysed by one-way analysis of variance followed by a modified least significant difference (l.s.d.) test (Shirley 1979) using the variation between sheep within treatments to determine the l.s.d.

Results

At autopsy of the ewes, none of the ovaries contained current or recently regressed corpora lutea or obvious corpora albicantia. Furthermore, none of the treated or control ewes had ovulated at the time of blood flow measurement (*c.* 24 h after the last hormone injection).

As there was no significant difference between RBF for the right and left ovaries, the mean of the two values in each sheep was used. The grand mean (±s.e.m.) RBF values and the grand mean weights of the mean ovarian weight per ewe are shown in Table 1.

Table 1. Grand mean ± s.e.m. relative blood flow (RBF) values and grand mean weights of the ovaries of anoestrous ewes following treatment with oestrogen and progesterone
Treatments: 1, control (oil only); 2, 10 mg/day of progesterone for 6 days; 3, 50 μg oestradiol-17β as single injection; 4, 10 mg/day progesterone for 6 days followed 48 h later by 50 μg oestradiol-17β

	Treatment groups (four ewes per group)			
	1	2	3	4
Ovarian RBF	0·27 ± 0·11	0·25 ± 0·13	0·27 ± 0·12	1·20 ± 0·12*
Ovarian wt (g)	0·85 ± 0·09	0·73 ± 0·06	0·77 ± 0·07	0·83 ± 0·16

*Value differs from all other RBF values by one-way analysis of variance and l.s.d. test ($F_{3,12} = 14·919$, l.s.d. = 0·7418, $P < 0·001$). Standard error of mean from one-way analysis of variance was 0·12 and was calculated by using $\sqrt{(s^2/r)}$.

The RBF values for ovaries from control ewes, from ewes treated with progesterone only and from those treated with oestrogen only did not differ significantly. However, the grand mean RBF value for ewes which received progesterone prior to treatment with oestrogen (group 4) was significantly greater than that for each of the other three groups.

Discussion

Since a significant increase in ovarian capillary blood flow occurred only in those ewes in which progesterone was given prior to oestradiol, it appears that in the ewe, progesterone sensitization of the ovarian vasculature may be necessary to permit a stimulatory influence of oestradiol on ovarian capillary blood flow. Such an effect by progesterone might be responsible for the rise in ovarian capillary blood flow which occurs in ewes around the oestrous period (Brown *et al.* 1974*a*). It also appears that LH release did not contribute to the above effect of progesterone, as an oestrous-type LH release occurs in anoestrous ewes given treatments similar to those received by groups 3 (oestradiol alone) and 4

(progesterone, then oestradiol) (Goding *et al.* 1969; Radford *et al.* 1970). Furthermore, Hixon and Clegg (1969) showed that LH has little or no effect on ovarian blood flow in ewes.

In anovular ewes, progesterone treatment prior to either the introduction of rams or treatment with GnRH, results in normal functioning of the corpora lutea that develop following the first ovulation (Hunter *et al.* 1971; Oldham *et al.* 1980; McLeod *et al.* 1982; Martin and Scaramuzzi 1983). It has been suggested that this effect may be due to progesterone delaying the onset of the induced LH surge, thereby allowing adequate follicular development to take place before the induction of ovulation (Haresign and Lamming 1978; McLeod *et al.* 1982; Pearce *et al.* 1982). Alternatively, the effect might arise as a result of the progesterone treatment facilitating a response by the ovarian vasculature to endogenous oestrogen. A pronounced increase in ovarian capillary blood flow, similar to that induced in the present study and comparable with the rise which occurs around the ovulatory period in cyclic ewes (Brown *et al.* 1974*a*), might be critical for the establishment of an adequate nutrient supply for the developing corpus luteum.

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