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CHANGES IN PLASMA PROGESTERONE AND LH CONCENTRATIONS DURING THE PROGESTERONE SYNCHRONISED OESTRUS IN SHEEP

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OPSOMMING: VERANDERINGE IN DIE KONSENTRASIE VAN PROGESTEROON EN LUTEIENISERENDE HORMOON GEDURENDE DIE PROGESTEROON GESINKRONISEERDE BRONSTIGHEID BY SKAPE

Tien Merino ooie is binne die normale teelseisoen met 15 daaglikse inspuitings van 10 mg progesteroon gesinkroniseer, terwyl 'n verdere 10 ooie as kontrole gedien het. Daaglikse bloedmonsters, en vanaf die beëindiging van progesteroonbehandeling tot na die einde van bronstigheid, 6 uurlike bloedmonsters, is ontleed vir progesteroon en luteïeniserende hormoon (LH) konsentrasie. 'n Nou verband tussen die posisie van die LH-piek en die einde van bronstigheid is gevind in kontrole ooie. Hierdie verband is ontwrig in die gesinkroniseerde ooie. Geen verduideliking vir die afwyking in die LH-piek kon gevind word in die verandering in die progesteroonkonsentrasie by gesinkroniseerde ooie nie.

SUMMARY:

The effect of progesterone synchronisation on the plasma progesterone and luteinizing hormone (LH) concentrations in sheep were studied. During their normal breeding season 10 Merino ewes were injected with 10 mg progesterone daily for 15 days to synchronise their oestrous periods. Ten ewes served as controls. Blood samples were collected daily during the progesterone treatment period and then at 6 intervals up to the end of oestrus. The plasma was analysed for progesterone and LH. A close relationship was found between the LH peak and the end of oestrus in control sheep. This relationship was often disrupted by the progesterone treatment. No explanation for the shift in the LH peak could be found in the preceding progesterone changes in both treated and control animals.

Synchronisation of oestrus with progestagens commonly results in reduced fertility (Lamond, 1964; Robinsin, 1967; Lindsay, Moore, Robinson, Salamon & Shalton, 1967; Van der Westhuysen & Van Niekerk 1971; Le Roux, 1974).

Evidence of histochemical or morphological changes in the oviduct and uterus, possibly interfering with sperm transport, capacitation, motility and metabolism (Robinson, 1967; Jöchle, 1969) have been attributed to possible endocrine imbalances (Jöchle 1969). Although the oestrus-ovulation relationships were found to be normal (Robinson & Smith, 1967,; Van der Westhuysen, Van Niekerk & Hunter, 1971), the timing of the release of LH and the magnitude of the LH release is affected by progestagen treatment (Baumgartner, Lishman, Louw & Botha, 1974; Lishman, Botha & Louw, 1974). In part, these abnormalities following cessation of progestagen treatment may be due to the hormone imbalance caused by residual progestagens following the cessation of treatment (Quinlivan & Robinson, 1967). This experiment was therefore planned to study the relationships between peripheral progesterone concentrations, oestrus and the release of LH in control and progesterone synchronised ewes.

Materials and Methods

Twenty dry Merino ewes were included in this experiment which was conducted during April. Ten ewes received an intramuscular injection of 10 mg progesterone in propyleneglycol daily for 15 days. For the sake of convenience, the 10 control ewes were treated with methyl acetoxy progesterone intravaginal sponges (MAP) one cycle previously, so that a normal (control) cycle coincided with the progesterone synchronised cycle. All sheep were bled daily immediately prior to the injection of the progesterone treated group. The last (fifteenth) progesterone injection also contained approximately 25 μ Ci ^{3}H-progesterone. Following this injection, blood samples were collected by jugular venipuncture from all 20 ewes at 6 h intervals. At the same time the ewes were tested for overt oestrus using teaser rams. The collection of blood samples and teasing were ceased after the second negative test for oestrus following the commencement of oestrus. The onset of heat was then estimated as having occurred 3 h before the first positive test for oestrus while the end of heat was estimated as having occurred 3 h before the first subsequent negative test.

Plasma progesterone was determined by radioimmuno-assay using an antibody raised against Progesterone-succinate (Dr. J.C. Morgenthal Department of Human and Animal Physiology, University of Stellenbosch) and plasma LH by a modified method of Niswender, Reichert, Midgley & Nalbandov (1969). The decay of the injected ^{3}H progesterone was determined by extraction of 1 ml plasma with 10 volumes of ether. After evaporation of the ether, in counting vials, dioxane based scintillation coctail was added and the radioactivity determined. Although for the purpose of further determinations of peripheral progesterone these counts were neglegibly low, (Fig. 3) determinations were accordingly adjusted. Analysis of variance was used to test for treatment differences.

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Results

The results of the changes in plasma progesterone concentration are presented in Table 1 and Fig. 1 and 2. The mean progesterone concentration in the plasma of control sheep in the luteal phase (diestrus period) differed grossly from that of the sheep receiving daily injections of progesterone. However, during the 48 hours preceding the onset of oestrus the progesterone concentrations of the plasma of both the control and experimental groups followed a similar pattern (Fig. 2) so that the mean plasma progesterone concentration did not differ significantly between groups at any point during this period. Although no significant differences were found between the duration of oestrus and the position of the LH peak relative to the onset and end of oestrus, the LH peak always occurred 24 h prior to the end of oestrus in control animals whereas its position varied greatly in progesterone treated animals (Fig. 3). The mean duration of the LH peak was 9,43 ± 2,45 and 8,76 + 3,02 h for progesterone treated and control groups respectively and the mean highest LH value of these groups were 122,8 ± 6,7 and 133,7 ± 7,5 ng/ml respectively. These differences were non-significant but since sampling only took place at 6 h intervals these values do not necessarily show the total time spread nor the highest LH value.

Table 1

The onset of oestrus following the cessation of progesterone injection, and the duration of oestrus, plasma progesterone concentration and position of the LH peak in progesterone synchronised and control ewes

	Control ewes	Synchronised ewes
Duration of estrus (h)	31,7 ± 2,7	28,0 ± 8,0
Final injection to onset of oestrus	–	55,0 ± 13,9
Plasma progesterone concentration (ng/ml):		
24 h before onset of oestrus	746,4 ± 83,8	1 687,9 ± 83,0
12 h before onset of oestrus	941,0 ± 54,4	1 266,7 ± 133,9
at onset of oestrus	826,1 ± 119,2	1 573,6 ± 137,8
LH peak value relative to:		
onset of oestrus (h after)	7,7 ± 2,7	4,0 ± 2,8
end of oestrus (h before)	24,0 ± 0,0	26,7 ± 7,5

Total plasma progesterone after cessation of the progesterone injection had a halflife of 14,4 h which is not significantly different from the halflife of the tritiated progesterone (18,0 h) included in the final injection. In the untreated group, the halflife of the following the peak which occurred prior to the onset

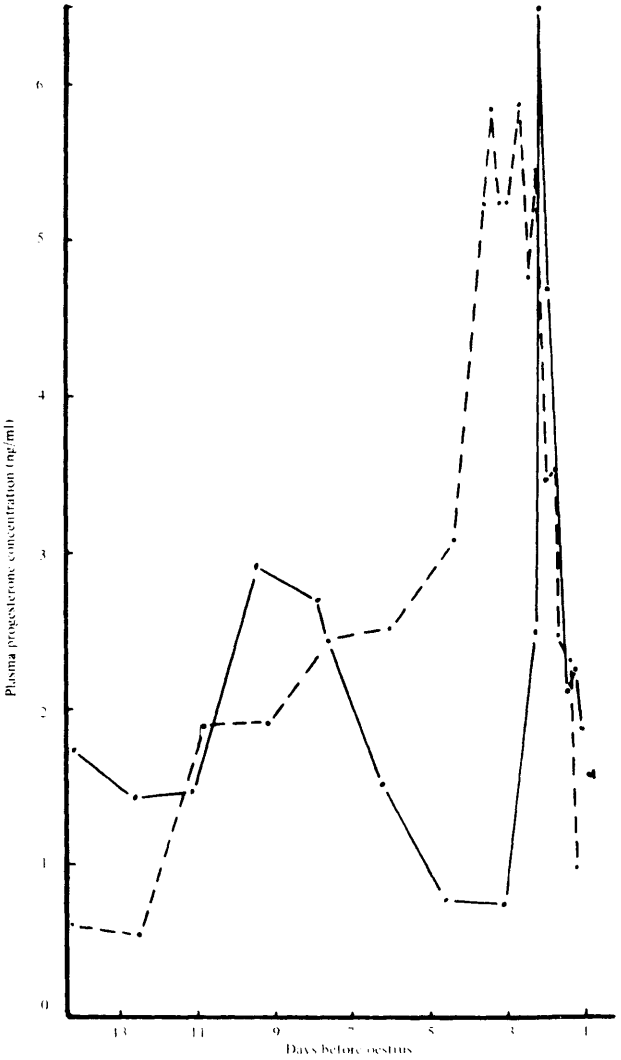

Fig. 1 *Changes in the plasma progesterone concentration of control ewes (---) and ewes receiving daily injections 10 mg progesterone intramuscularly (—) for 15 days prior to the onset of oestrus.*

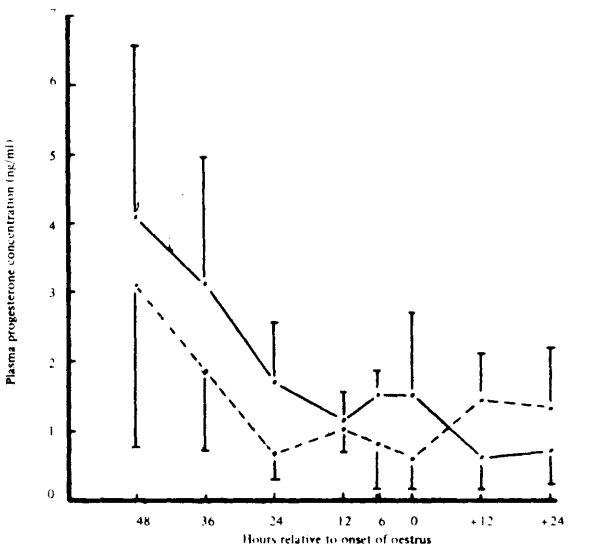

Fig. 2 *The mean plasma progesterone concentration and standard deviations in control (.---.) and progesterone synchronised (.—.) ewes for the period 48 h prior to, and until 24 h after the onset of oestrus.*

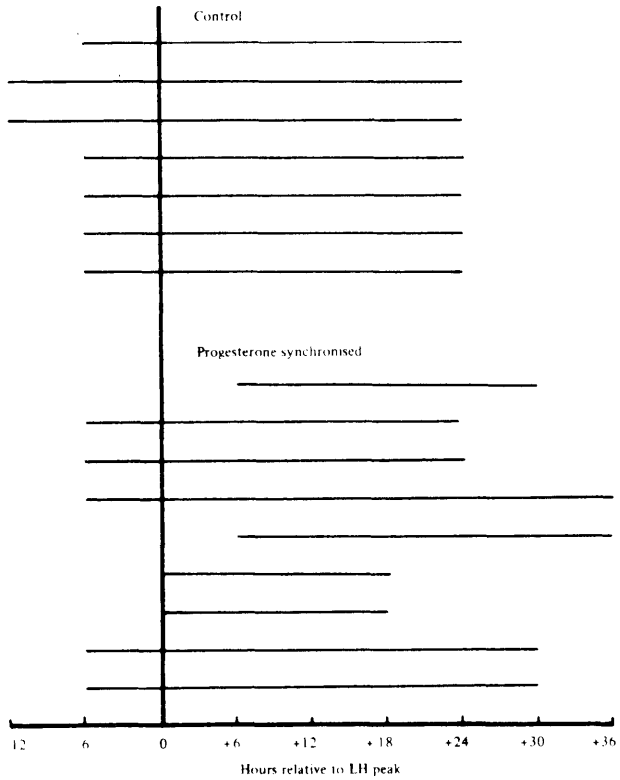

Fig. 3 *The duration and occurence of oestrus relative to the occurrence of the highest LH peak value.*

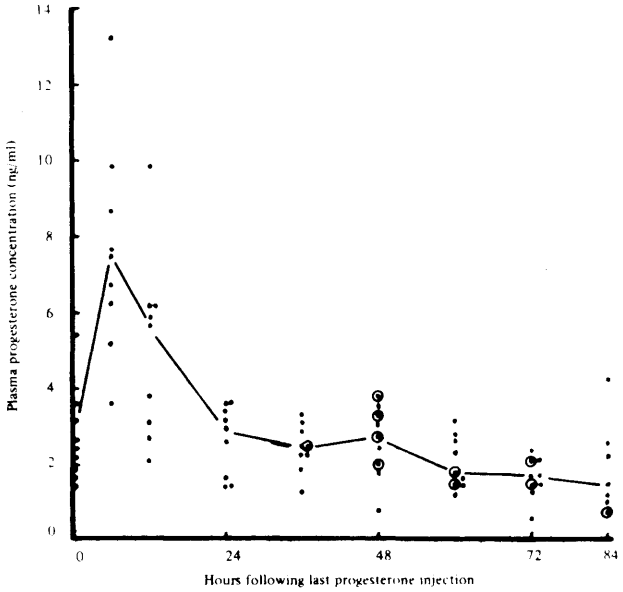

Fig. 4 *Distribution of plasma progesterone during the 84 hours following the final injection of progesterone. The graph represents the median of plotted values, and ewes in oestrus are indicated by a circle (⊙).*

of estrus, was estimated at 30,0 h. The non significant difference between the halflife of injected tritiated progesterone and that of the total plasma progesterone suggests that endogenous progesterone production at this stage of the synchronised cycle is of minor significance. In contrast, the slower decline of plasma progesterone in the control group is probably the result of a decrease in production of progesterone rather than an abrupt cessation of production.

Although plasma progesterone had to fall to a mean of approximately 1,0 ng/ml before oestrus could commence a great deal of variation occurred as regards the concentration of progesterone at which oestrus occurred and LH was released. So for instance no significant correlations could be found between plasma progesterone concentration, the decay of injected (tritiated) progesterone and either the onset of oestrus or the release of LH (Fig. 4 & 5). These findings suggest an individual variation in the sensitivity of animals to progesterone.

Discussion

Various workers have found that the synchronisation of oestrus with progesterone disturbs the normal time relationship between the release of LH and oestrus (Cumming, Blockey, Brown, Catt, Goding & Kaltenbach, 1970; Mauer, Revenal, Johnson, Moyer, Hirate & White, 1972; Linten & Lemming, 1973; Baumgartner *et al*, 1974; Lishman *et al*., 1974). It has been suggested that

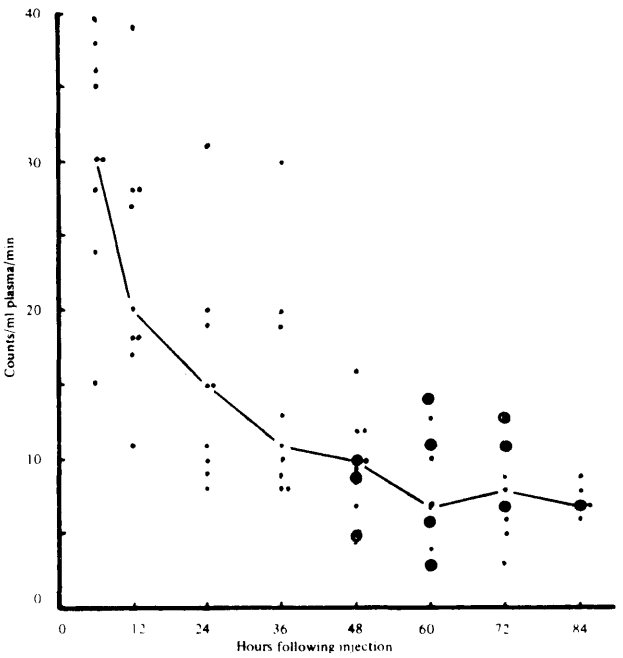

Fig. 5 *The decay of triteated progesterone in ewes over the 84 hours following the injection. (The graph represents the median of the plotted values and ewes in oestrus are indicated by a circle (⊙).*

this disruption is the result of an abnormal decline in progesterone following the cessation of treatment (Robinson, 1967).

The results of the present experiment substantiate the findings that progesterone synchronisation disturbs the normal release of LH (Fig. 2). On an individual basis the release of LH could not be related to the rate of decline in peripheral progesterone or to be actual plasma progesterone preceding LH release (Fig. 3 and 4).

Although the injected (exogenous) progesterone proved to have a shorter halflife than the decay of endogenous peripheral progesterone (14,4 cf. 30 h) changes during the 48 h preceding the onset of oestrus did not differ between the control and progesterone treated groups (Fig. 2). Considerable individual variation in the progesterone concentration at which ewes commence oestrus suggests individual differences in the sensitivity to progesterone and the subsequent events leading to oestrus and ovulation.

During the period of progesterone treatment, the plasma progesterone concentrations of the progesterone treated group differed significantly from that of the controls (Fig. 1). In addition, the ewes were at different stages of the oestrous cycle when progesterone treated commenced. It is therefore concluded that the disruption of the normal LH– oestrus relationship in this experiment cannot be related to the decline in circulating progesterone. It is postulated that the treatment and artificial alteration of the pattern and duration of the progesteronal phase may modify the hypothalamic – pituitary – ovarian interaction and thus the release of LH and the manifestation of overt oestrus.

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