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LUTEINIZING HORMONE (LH) AND PROLACTIN LEVELS AT OESTRUS FOLLOWING SYNCHRONISATION WITH PROGESTOGENS IN THE EWE

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OPSOMMING: LUTEINISERENDE HORMOON EN PROLAKTIEN PEILE BY DIE OOI NA SINKRONISASIE VAN ESTRUS MET PROGESTOGENE

Die patroon van afskeiding van LH en prolaktien na sinkronisasie van estrus met progesteroon of SC-9880 is bestudeer. Drie van die 10 ooie wat met progesteroon behandel is, het geen bronstigheid getoon nie. Waar estrus met SC-9880 gesinkroniseer is, het die afskeiding van LH by of voor aanvang van bronstigheid in twee van vyf ooie plaasgevind. Met progesteroon is die afskeiding van LH slegs na aanvang van bronstigheid waargeneem. Die gemiddelde piek LH peil in die plasma van 223,2, 147,6 en 93,7 ng/cm^3 by die kontrole, SC-9880 en progesteroon behandelde ooie, respektiewelik. Die verskille was betekenisvol. 'n Duidelik gedefinieerde afskeiding van prolaktien is nie waargeneem nie. Die bydrae van lae piek LH peile tot verlaagde vrugbaarheid na progestogeen behandeling is bespreek.

SUMMARY

The pattern of luteinizing hormone (LH) and prolactin release following the synchronisation of oestrus with progesterone or SC-9880 was studied. Of the 10 ewes treated with progesterone three did not exhibit oestrus. Where oestrus was synchronised with SC-9880 the pre-ovulatory LH surge occurred at, or prior to, the onset of oestrus in two of five ewes. With progesterone the LH release occurred only after mating commenced. The average peak LH level in the plasma was 223,2, 147,6 and 93,7 ng/cm^3 for the control, SC-9880 and progesterone treated ewes respectively. The differences were significant. A clearly defined surge of prolactin was not observed. The contribution of low peak LH levels to reduced fertility following progesterone therapy is discussed.

The synchronisation of oestrus with progestogens commonly results in a reduced conception rate. Jöchle (1969) has described some of the abnormalities which may contribute to the reduced fertility. Generally, it would appear that the hormonal patterns which result in the controlled oestrus and ovulation deviate from those which occur during normal oestrus (Jöchle, 1969). The purpose of this investigation was to observe the patterns and levels of LH and prolactin in the blood of ewes at a synchronised oestrus.

Procedure

On April 27, 1973 (day 1), 20 Merino ewes which were exhibiting regular oestrous cycles were divided into four groups viz.,

Group 1 – Animals were selected in which oestrus was expected to occur spontaneously between days 11 and 14 (control group).

Group 2 – Polyurethane, intravaginal pessaries containing 40 mg SC-9880 (Synchro-mate, G.D. Searle) were inserted into each ewe on day 1 and removed on day 10.

Group 3 – Intramuscular injections of 10 mg progesterone in arachis oil were administered daily from day 1 to day 10.

Group 4 – Treatment was as for Group 3, but release of prolactin was inhibited by subcutaneous injection of 1,25 mg ergocornine hydrogene maleinate in ethanol-saline (EC). Injections were started at 08h00 on day 11 and repeated every eight hours until 24 hours after oestrus commenced (Louw, 1974).

Except for Group 1 the animals were randomly allocated to the treatment groups. At 10h00 on day 11 indwelling silastic cannulae were inserted in the jugular vein of all ewes. Immediately thereafter a sample of blood (5 cm^3) was collected into a heparinized syringe. The plasma was separated by centrifugation and stored at – 15°C. Further samples of blood were drawn at two-hour intervals until 30 hours after the onset of oestrus or 08h00 on day 14. Overt oestrus was detected by using active vasectomized rams which were joined with the ewes at intervals of not more than two hours.

The plasma samples were assayed for LH and prolactin by the double-antibody methods of Niswender, Reichert, Midgley & Nalbandov (1969) and Davis, Reichert & Niswender (1971), respectively. NIH-LH-S16 and NIH-P-S10 were used as standards. The sensitivity, specificity and repeatability of the LH and prolactin assays have been described by Lishman (1972) and Louw (1974), respectively. The significance of treatment differences was tested by analysis of variance.

Results

When observations ceased on day 14 two ewes in Group 3 and one ewe in Group 4 had not exhibited oestrus. Although the delay to oestrus following termination of

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progestational therapy was shorter in Groups 3 and 4 than for Group 2 the variation in time of onset was greater (Table 1).

The timing and duration of the pre-ovulatory release of LH is illustrated in Fig. 1. The progestogen treatments did not significantly influence the duration of the LH surge. The maximum level of LH in the plasma was significantly lower (P=0,01) following pre-treatment with SC-9880 than with progesterone (Table 1). Both progestogens resulted in a significantly lower (P=0,01) maximum LH level than that of the untreated ewes. One ewe viz. no. 168 accepted service without exhibiting an LH surge.

Table 1

Onset of oestrus following synchronisation and the release of LH and prolactin

Treatment	Group	Ewe No.	Latency to oestrus (h)*	Maximum concentration (ng/cm^3 plasma) LH	Prolactin
Control	1	316	–	237,0	532,0
		71–99	–	276,0	110,0
		69–292	–	223,2	810,0
		66–49	–	232,2	900,0
		72–86	–	148,2	1375,0
		Mean		223,2	945,4
		SE		±20,9	±141,9
SC-9880	2	67–26	53,8	160,0	1316,5
		68–223	57,3	146,0	1865,7
		66–39	57,0	120,0	729,3
		67–49	54,0	132,0	448,2
		67–10	41,8	180,0	734,7
		Mean	52,8	147,6	1018,9
		SE	±2,8	±10,5	±254,5
Progesterone	3	66–58	84,0	31,0	688,2
		71–100	57,3	128,0	361,6
		68–173	57,0	124,0	665,0
		70–147	–	–	552,0
		U3	–	–	1324,7
		Mean			718,3
		SE			± 162,2
Progesterone + E.C.	4	66–97	77,9	138,0	–
		70–95	64,0	112,0	–
		68–294	58,2	29,0	–
		66–136	–	–	–
		168	89,0	–	–
		Mean	69,6x	93,7 x	–
		SE	± 5,1	± 9,1	–

* Measured from time of last progesterone injection or removal of intravaginal pessaries.

x Mean and SE after pooling results for Groups 3 and 4.

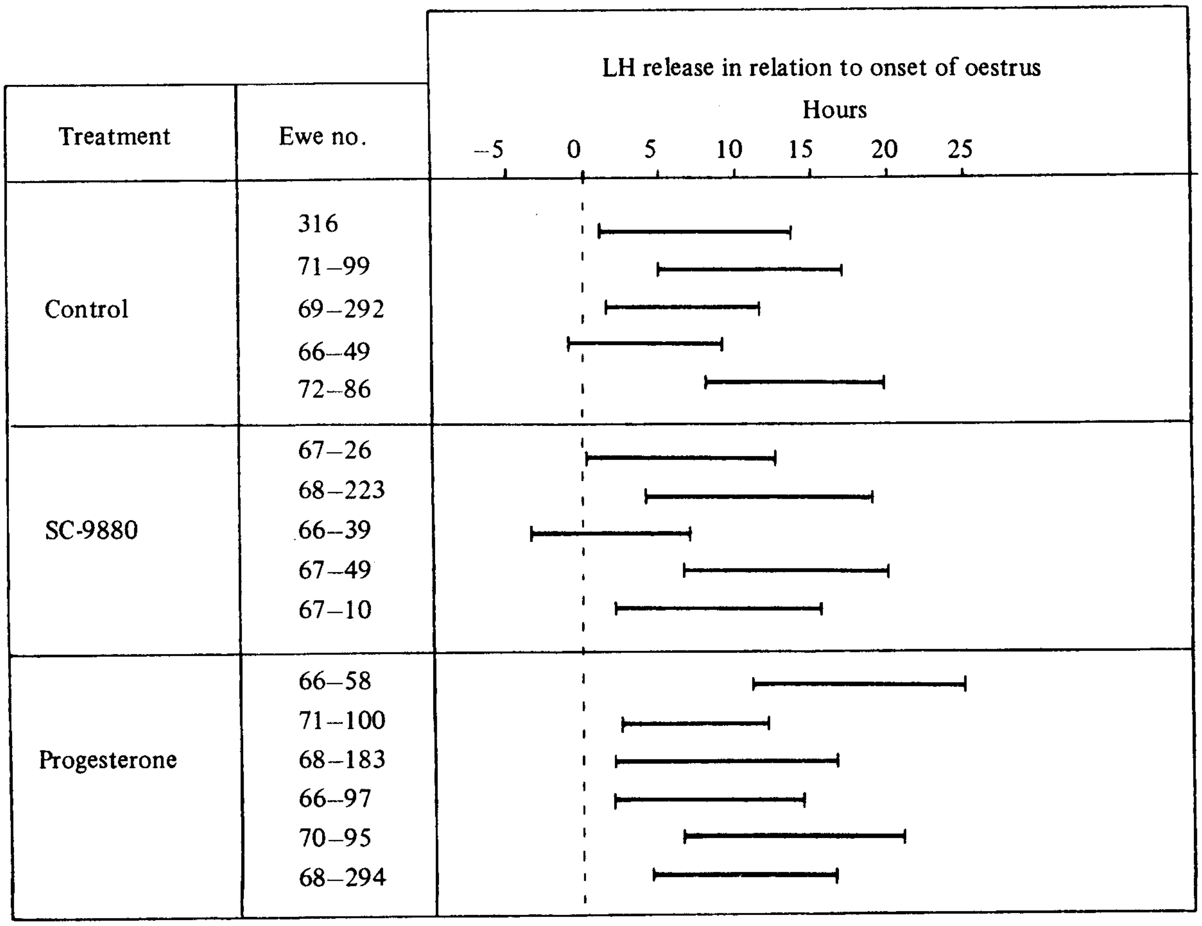

Fig. 1 *Timing and duration of LH surge in relation to onset of oestrus following spontaneous oestrus or synchronisation of oestrus with progestogens*

The patterns of LH and prolactin secretion in individual ewes, selected at random from Groups 1,2 and 3, are depicted in Fig. 2. Levels of prolactin varied markedly between samplings and a clearly defined release, such as that recorded for LH, was not observed. No treatment differences in the blood levels of prolactin in Groups 1,2 and 3 could be demonstrated. Furthermore, the LH surge and onset of oestrus in the ewes in which the release of prolactin was blocked (Group 4) was not different from that in the animals not treated with EC.

Discussion

The present results confirm the observation that synchronisation of oestrus with SC-9880 (Cumming, Blockey, Brown, Catt, Goding & Kaltenbach, 1970), progesterone implants (Mauer, Revenal, Johnson, Moyer, Hirata & White, 1972) or fluorogestone acetate (Lintin, & Lamming, 1973) induces an LH release which may even occur prior to oestrus and thereby make fertilisation improbable. However, the conclusion that a premature surge of LH is abnormal (Cumming *et al.*, 1970) is not entirely correct since Lishman (1972) showed that in cycling ewes the LH surge commonly commences prior to the onset of oestrus.

The present results show that the concentrations of LH in the plasma were lower during LH surges which occurred after administration of progesterone than during unsynchronised LH surges. Grieg & Weisz (1973) concluded that ovulation could be stimulated by blood levels of LH which were only 14% of the levels actually measured in rats during normal pre-ovulatory surges of LH. However, Pelletier & Thimonier (1973) suggested that subnormal levels of LH were associated with poor fertility. Consequently, the present results indicate that the amounts of LH which are released after progesterone pre-treatment may be inadequate for optimum fertility.

The high levels of prolactin which were observed in this study are in accord with the results of Reeves, Arimura & Schally (1970), Bryant, Greenwood, Kann, Martinet & Denamur (1971), Kann (1971) and Cumming, Brown, Goding, Bryant & Greenwood (1972). The role of prolactin in the oestrous cycle of the ewe remains obscure (Louw, Lishman, Botha & Baumgartner, 1974; Niswender, 1974) and the present results support the contention (Fall, Beck, Brown, Catt, Cumming & Goding, 1972) that the

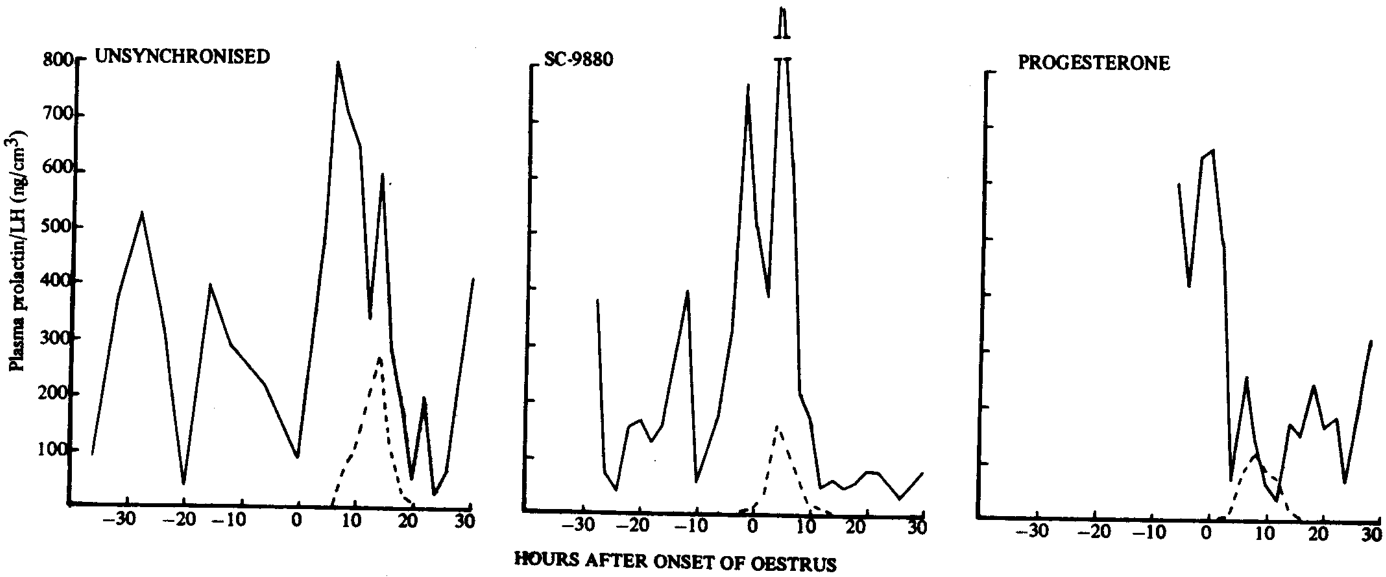

Fig. 2 *Plasma prolactin (——) and LH (– – –) levels in individual ewes following synchronised or unsynchronised oestrus*

simultaneous occurrence of prolactin and LH peaks are merely incidental.

One of the principal aims of oestrous synchronisation in beef cows and ewes is to facilitate the use of artificial insemination. Robinson & Moore (1967), Colas & Cognie (1968) and van Niekerk & Belonje (1970) recommend that following synchronisation of oestrus insemination should be conducted at a time relative to the termination of progestogen therapy rather than to the onset of oestrus. This recommendation appears to be sound, but in view of the limited survival time of dilute semen, means should be sought whereby synchronisation of the release of LH and the resultant ovulation can be improved.

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References

BRYANT, G.D., GREENWOOD, F.C., KANN, G., MARTINET, J. & DENAMUR, R., 1971. Plasma prolactin in the oestrous cycle of the ewe: Effect of pituitary stalk section. *J. Endocr.* 51, 405.

COLAS, G. & COGNIE, Y., 1968. Artificial insemination, with or without oestrus detection, following progestogen treatment in the ewe. *Proc. VIth int. Congr. Anim. Reprod. Paris,* 2, 1407.

CUMMING, I.A., BLOCKEY, M.A de B., BROWN, J.M., CATT, K.J., GODING, J.R. & KALTENBACH, C.C., 1970. The release of luteinizing hormone in ewes following the withdrawal of intravaginal sponges containing progestagen. *Proc. Aust. Soc. Anim. Prod.* 8, 383.

CUMMING, I.A., BROWN, J.M., GODING, J.R., BRYANT, G.D. & GREENWOOD, F.C., 1972. Secretion of prolactin and luteinizing hormone at oestrus in the ewe. *J. Endocr.* 54, 207.

DAVIS, S.L., REICHERT, L.E. & NISWENDER, G.D., 1971. Serum levels of prolactin in sheep as measured by radioimmunoassay. *Biol. Reprod.* 4, 145.

FELL, L.R., BECK, C., BROWN, J.M., CATT, K.J., CUMMING, I.A. & GODING, J.R., 1972. Solid-phase radioimmunoassay of ovine prolactin in antibody-coated tubes. Prolactin secretion during oestradiol treatment, at parturition, and during milking. *Endocrinology,* 91, 1329.

GREIG, F. & WEISZ, J., 1973. Preovulatory levels of luteinizing hormone, the critical period and ovulation in rats. *J. Endocr.* 57, 235.

JÖCHLE, W., 1969. Latest trends and practical problems arising during oestrus synchronisation. *Proc. S. Afr. Soc. Anim. Prod.* 8, 23.

KANN, G., 1971. Variations des concentrations plasmatiques de l'hormone luteinisant et de la prolactin au cours du cycle oestrien de la brebis. *C.R. Acad. Sci. Paris,* 272, 2934.

LINTIN, K. & LAMMING, G.E., 1973. The effects of progestagens and PMSG on peripheral plasma LH levels in the Clun Forest ewe. *J. Reprod. Fert.* 35, 608.

LISHMAN, A.W., 1972. *The influence of variations in the level of nutrition on reproduction in the ewe.* Ph.D. Thesis, University of Natal.

LOUW, B.P., 1974. *The secretion and function of prolactin during the oestrous cycle of the ewe.* M.Sc. Thesis, University of Natal.

LOUW, B.P., LISHMAN, A.W., BOTHA, W.A. & BAUMGARTNER, J.P., 1974. Failure to demonstrate a role for the acute release of prolactin at oestrus in the ewe. *J. Reprod. Fert.* (In the press).

MAUER, R.E., REVENAL, P., JOHNSON, E.S., MOYER, R.H., HIRATA, A. & WHITE, W.F., 1972. Levels of luteinizing hormone in sera of ewes near the time of oestrus as determined by radioimmunoassay. *J. Anim. Sci.* 34, 88.

NISWENDER, G.D., 1974. Influence of 2–br-α-ergocryptine on serum levels of prolactin and the oestrous cycle in sheep. *Endocrinology,* 94, 612.

NISWENDER, G.D., REICHERT, L.E., MIDGLEY, A.R. & NALBANDOV, A.V., 1969. Radioimmunoassay for bovine and ovine luteinizing hormone. *Endocrinology,* 84, 1166.

PELLETIER, J. & THIMONIER, J., 1973. Comparison of the induced preovulatory LH discharge in lactating and dry sheep during seasonal anoestrus. *J. Reprod. Fert.* 33, 311.

REEVES, J.J., ARIMURA, A. & SCHALLY, A.V., 1970. Serum levels of prolactin and luteinizing hormone (LH) in the ewe at various stages of the oestrous cycle. *Proc. Soc. Exp. Biol. Med.* 134, 938.

ROBINSON, T.J. & MOORE, N.W., 1967. The evaluation of progesterone and Sc-9880 – impregnated intravaginal sponges for the synchronisation of oestrus for large scale artificial insemination of Merino ewes in Spring. In, *Control of the ovarian cycle in sheep,* ed. T.J. Robinson, Sydney University Press.

VAN NIEKERK, C.H. & BELONJE, P.C., 1970. The times of ovulation after the synchronisation of oestrus in Merino ewes with medroxy-progesterone acetate. *Proc. S. Afr. Soc. Anim. Prod.* 9, 177.