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STUDIES ON LEVELS OF LUTEINIZING HORMONE AND PROLACTIN IN SERUM OF CYCLING AND ANOESTROUS COWS

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OPSOMMING: STUDIES OOR VLAKKE VAN LUTEINISERINGS HORMOON EN PROLAKTIEN IN SERUM VAN SIKLIESE EN ANESTRIESE KOEIE

Vlakke van luteiniserings hormoon (LH) in serum gedurende die estrus-siklus en die post partum periode is deur middel van radio-immunologiese bepaling in 14 Brangus en 5 Fries koeie bepaal. Met die uitsondering van die periodes tydens die pre-ovulasie golf, het die basale vlakke van LH tussen 0,2 en 5,0 ng LH/ml serum gevarieer. Hierdie basale vlakke van LH, het net soveel tussen diere as binne diere of tussen die rasse gevarieer. Gedurende die pre-ovulasie golf het die LH vlakke verhoog tot tussen 8,0 en 60,0 ng LH/ml serum. In 16 van die 21 gevalle waar ovulasie gepaardgegaan het met estrus, het die verhoging in LH serum met die dag van estrus saamgeval. In 4 gevalle het die toename in LH 1,2 of 3 dae na estrus plaasgevind en in een geval 2 dae voor estrus. 'n Hoë voorkoms van ovulasie sonder estrus is gevind. Géen standhoudende veranderings in die prolaktien vlakke van die serum is in twee van dié koeie gedurende die estrus siklus waargeneem nie.

SUMMARY

Blood levels of luteinizing hormone (LH) were measured, using a radioimmunoassay, during the oestrous cycle and during the post-partum period in 14 Brangus and 5 Friesland cows. Except during pre-ovulatory surges, basal blood levels of LH ranged between 0,2 and 5,0 ng LH/ml serum. These basal levels of LH varied more between animals than either within animals or between breeds. During the pre-ovulatory surge, LH levels increased to between 8,0 and 60,0 ng LH/ml serum. In 16 of the 21 cases when ovulation was associated with oestrus, the LH surge was detected on the days of oestrus. In 4 instances, the LH surge was detected 1,2 or 3 days after oestrus and in one instance, 2 days before oestrus. A high incidence of ovulation without oestrus was noted. No consistent changes were detected in levels of prolactin in the serum of two of these cows during the oestrous cycle.

Prolonged post-partum anoestrus and high incidence of silent heats (quiet ovulation or ovulation without associated oestrus) are two major factors which limit the reproductive performance of ranch cows in Rhodesia. Attempts to overcome these problems by relatively simple managerial procedures have met with only limited success (Symington & Hale, 1967; Symington, Gregor & Hale, 1967). These procedures included introduction of teaser bulls to anoestrous cows, manipulation of plane of nutrition and permanent or temporary early-weaning of the calf. Further progress with this line of research was hampered by the imprecise nature of the parameters which were available at that time to assess the effect of experimental regimes on the reproductive performance of the cows. Thus, cows show oestrus and ovulate only once in three weeks. Furthermore, oestrus and ovulation are "all-or-nothing" phenomena, that is. they either occur or they do not occur. Consequently, extremely large numbers of animals would have been required to assess, with any degree of accuracy, the value of treatments.

In recent years, development of sensitive and precise radioimmunoassay techniques has allowed accurate measurement of the polypeptide hormones of the anterior pituitary and gonadal steroid hormones (Kirkham & Hunter, 1971). Thus, it is now possible to evaluate definitively the effect of environmental factors such as plane of nutrition, photoperiod, temperature and managerial treatments on these reproductive hormones.

This paper presents results of a preliminary study of the levels of luteinizing hormone (LH) and prolactin in the blood of cows under typical Rhodesian farming conditions. Since changes in levels of reproductive hormones have been studied elsewhere in Frieslands (Hansel & Snook, 1970), levels of hormones were also studied in the blood of cows of this breed for comparative purposes. Data will be presented at a later stage on levels of oestrogen and progesterone in these animals.

Procedure

Five Friesland cows and 10 grade Brangus cows in various reproductive states were used. The Friesland cows were bled by jugular venipuncture three times each week. The Brangus cows were bled daily, eight of them from six weeks after calving until sexual activity resumed. Of the remaining two Brangus cows, one was a dry cow and the other a heifer.

Friesland cows were kept at the Veterinary Research Station of the Department of Veterinary Services, Ministry of Agriculture. Brangus cows were kept on a local farm (H.I. Hale (Pvt) Ltd) and were subjected to normal farm management. During the course of the study mean rate of increase in bodymass of Brangus cows was approximately 0,25 kg/day. A vasectomized bull was used to detect oestrus between 06h00 and 18h00 each day. Ovaries were palpated *per rectum* once each week.

Blood samples were collected between 08h30 and 09h30 each day. Samples were placed in a refrigerator within an hour of collection and centrifuged (1000 X g) the following day in a refrigerated centrifuge. Sera were collected and stored (−20°C) until assay.

Concentrations of LH and prolactin were measured by use of radioimmunoassay techniques developed in this

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laboratory (Ellison, 1974). In both instances, assays used the "coated tube" technique which involves the solid phase principle of radioimmunoassay described by Catt & Tregear (1967).

Results and Discussion

General Sexual Activity

During the investigations, 3 of the 5 Friesland cows each showed one normal oestrus and two conceived when inseminated at this oestrus. The third cow showed one normal oestrus followed by two silent heats. The remaining two cows were approximately three weeks pregnant at start of observations.

Sexual activity in the Brangus cows was characterised by prolonged postpartum anoestrus (mean 169 days). During the period of observation, full oestrus was observed on 18 occasions, silent heat on 13 occasions and there were 6 instances of anovulatory oestrus. Three of these instances of anovulatory oestrus occurred in the heifer (Fig. 1). The remaining three anovulatory oestrous periods each occurred at the end of postpartum anoestrus and were followed approximately one week later by normal oestrus and ovulation (e.g. Fig. 2).

Concentration of Hormone in Serum

In general, levels of LH did not differ between Brangus and Friesland cows and were in agreement with overseas reports (Carr, 1972; Hansel, 1972; Schams & Karg, 1969; Nancarrow, Buckmaster, Chamley, Cox, Cumming, Cummins, Drinan, Findlay, Goding, Restall, Schneider & Thorburn, 1973). Blood levels of LH varied considerably between individual animals. Since differences between breeds were not detectable, for the purpose of this paper all data were pooled.

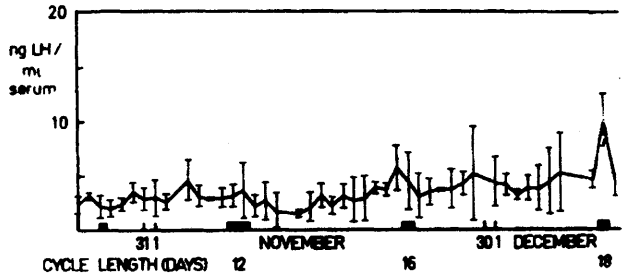

Fig. 1. *Levels of LH in the blood of a heifer during the onset of puberty. This animal experienced anovulatory oestrous periods on 26.10.72, 7–8.11.72, and on 23.11.72 followed by oestrus with ovulation on 11.12.72.*

Note: vertical bars represent 95% confidence limits of estimates

■ *Represents oestrus*

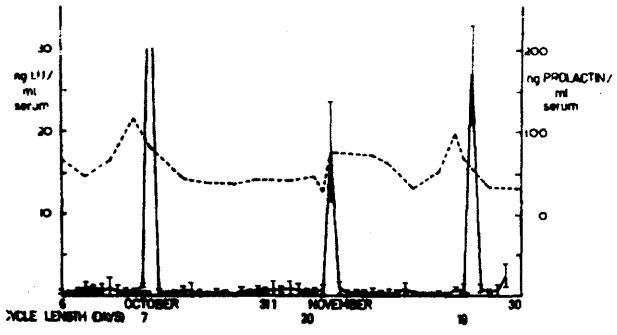

Fig. 2. *Levels of LH and prolactin in the blood of a lactating cow during resumption of post-partum sexual activity. This animal experienced an anovulatory oestrus on 9–10.10.72. A normal oestrus on 16–17.10.72 and delayed ovulation on 5–8.11.72 followed by delayed ovulation on 24–25.11.72*

•—• *LH*
•---• *Prolactin*

Note: vertical bars represent 95% confidence limits of estimates

■ *Represents oestrus*

With the exception of samples taken during the preovulatory surge of LH release, basal levels of LH in serum of mature cows varied little within animals. This was true, irrespective of sexual state of the cow – early pregnancy, postpartum anoestrus or at various stages of the oestrous cycle. However, LH levels varied greatly between animals – 0,2 to approximately 5,0 ng LH/ml serum(Figs. 1–4). Thus this "basal" concentration of LH in serum of cows gave no indication of reproductivity ability. By contrast, LH levels of the heifer increased during the course of the trial until oestrus with ovulation occurred (Fig. 1).

Unfortunately, in the present observations, detection of precise time of ovulation was not practical. Consequently, time relationships between the surge of LH release and ovulation were not studied. However, in every instance, ovulation, as indicated by subsequent development of a new corpus luteum, was associated with an elevated level of LH in the daily blood samples. In these samples, levels of LH were at least five times greater than the "basal" level of LH. Thus, levels at this time rose to between 8 and 60 ng LH/ml serum. In 16 of the 21 cases where ovulation was associated with oestrus, the elevated levels of LH were detected in blood samples taken on the day of oestrus (Fig. 1). In 4 cases, the LH surge was detected 1, 2 or 3 days after oestrus (Figs. 2 & 3).

Ovulation takes place 25 hours after the time when serum LH levels are maximal during the preovulatory surge of release of LH (Dobson, Hopkinson & Ward, 1973) or 15–22 hours after the end of the LH surge (Schams & Karg, 1969). Thus, where the LH peak occurred one or

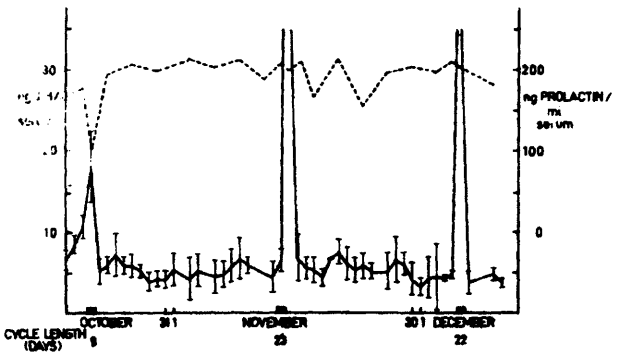

Fig. 3. *Levels of LH and Prolactin in the blood of a lactating cow during three oestrous periods, two normal (20.10.72 and 6.12.72) and one with delayed ovulation (14–15.11.72)*

LH
Prolactin

Note: vertical bars represent 95% confidence limits of estimates

■ *Represents oestrus*

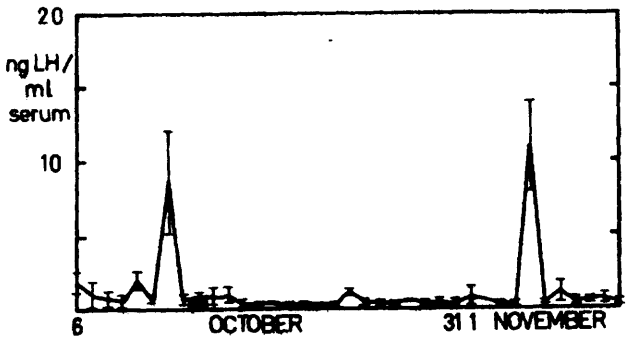

Fig. 4. *Levels of LH in the blood of a lactating cow during apparently premature ovulation (12–14.10.72) and silent heat (5.11.72)*

Note: vertical bars represent 95% confidence limits of estimates

■ *Represents oestrus*

more days after oestrus, it is likely that ovulation was delayed to such an extent that mating at oestrus would have been unsuccessful (van Rensburg & de Vos, 1962). By contrast, in one instance, elevated levels of LH were detected 2 days before manifestation of oestrus (Fig. 4). These instances of apparent lack of synchronisation of oestrus and ovulation all occurred in Brangus cows.

During the preovulatory surge of LH release associated with silent heat, levels of LH in the daily blood samples were lower than those recorded during the preovulatory surges of LH release associated with normal oestrus (9,9 ng/LH/ml *cf* 25,4 ng LH/ml respectively). This difference was significant statistically ($P < 0,001$). However, the half life of LH in circulating blood is short and rates of LH release change rapidly. Thus, levels of hormone as measured once daily may only partially reflect amounts of hormone released. Consequently, until data are available from more frequent samples and until levels of ovarian steroids are measured, the physiological significance of the difference in hormone levels in single daily samples between silent heat and normal oestrus cannot be evaluated.

To date, levels of prolactin have been measured in only two animals. Levels varied between 150 ng prolactin/ml and 220 ng prolactin/ml serum. No definite pattern of prolactin secretion is yet apparent from these data. Thus, in general terms, levels of prolactin remained constant throughout the period of observation. No tendency was detectable for prolactin levels to decline with the progression of lactation or with the onset of sexual activity after postpartum anoestrus. Further, prolactin levels were not consistently greater during oestrus than at other times of the oestrus cycle. This finding is at variance with the observations of Hansel & Snook (1970). However, it must be emphasised that data in the present study refer to only two animals.

Results of this study show that levels of LH and prolactin vary widely between individual animals. This great variability implies that radioimmunoassay techniques must be used to examine the effect of experimental treatments on individual animals, rather than to assess differences between groups of experimental animals. In other words, where the effects on reproduction of treatments such as different planes of nutrition are assessed by use of radioimmunoassays to measure hormone levels, each animal must be used as its own control.

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