USING MILK PROGESTERONE TO STUDY REPRODUCTIVE ACTIVITY IN DAIRY COWS CALVING IN DIFFERENT SEASONS

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SUMMARY

Milk progesterone profiles of 126 post-partum cows calving in August and September (n=41), January and February (n=42), or May and June (n=43) were determined from one week after calving through to four weeks after artificial insemination. Resumption of ovarian activity did not vary significantly with the different seasons, and the average length of the acyclic period after calving was 26.5 days. Sixty five percent of cows showed a transient rise in progesterone levels to approximately 4.0 ng/ml, lasting about 6.5 days, as their first indication of ovarian activity. The incidence of these short cycles did not vary with season or milk yield, but they extended the interval until oestrous cycles with a normal luteal phase accompanied by overt oestrus were established. Keywords: Milk progesterone, reproduction, dairy cows, season.

INTRODUCTION

In commercial dairy herds the optimal calving to conceptionintervalof 80-85 days is often extended due to problems including post-partum anoestrus, poor oestrus detection, inaccurate timing of insemination or asynchrony in the hormonal profile of cows (Bulman and Lamming 1978). The relative importance of these problems may be clarified by monitoring ovarian activity using milk progesterone profiles based on twice or thrice-weekly samples (Eulman and Lamming 1978; Peters and Riley 1982). Seasonal effects on fertility may also be studied this way. Studies by Bulman and Lamming (1978) and Hansen and Hauser (1983) showed intervals to first oestrus and ovulation were longer for cows calving in winter and spring than for cows calving in autumn or summer. However, most of these experiments used dairy cattle housed over winter under intensive feeding conditions, and seasonal effects may have been confounded by nutritional management very different from commercial conditions in Australia.

We investigated the return to reproductive activity in dairy cows calving in different seasons by monitoring milk progesterone levels, and examined some factors which could influence the acyclic period after calving in conditions typical of commercial dairying in Western Australia.

MATERIALS AND METHODS

Experimental animals and measurements

The study used 126 mature Friesian-Holstein cows aged between 3 and 10 years, and calving in August and September (n=41, Group 1), January and February (n=42, Group 2) and May and June (n=43, Group 3). Cows grazed annual pastures consisting of Trifolium subterraneum and Lolium spp. or irrigated permanent pastures comprising Kikuyu (Pennisetum clandestinum) white clover (Trifolium repens) and perennial ryegrass (Lolium perenne). Supplementary wheat or barley grain was provided to Groups 2 and 3 at an average of 4 kg/head/day. Milk samples were collected three times per week from one week after calving to four weeks after insemination, and assayed for progesterone. Milk production, liveweight

and condition score (Scale 1-8, Earle, 1976) were measured at monthly intervals. Pregnancy was diagnosed by rectal palpation 10 to 12 weeks after insemination and calving data were subsequently collected.

Ovarian activity and progesterone assay

Cows were observed for oestrus twice daily at 06.00 h and 16.00 h by experienced herdsmen, and were artificially inseminated using thawed frozen semen at the observed oestrus. Ovarian activity and timing of ovulationwere identified by the first progesterone measurement >1.3 ng/ml after calving, with the increase sustained for >14 days (Bulman and Lamming 1978). This signified the true commencement of cyclicity.

The concentration of progesterone in duplicate 10 ul milk samples was determined using a solid phase, microtitre plate progesterone enzyme-immunoassay (EIA) (Munro and Stabenfeldt 1984). The sensitivity of the EIA standard curve was 4 pg/l0 ul and with three progesterone pools (2.5, 10.0 and 40 ng/ml) the interplate coefficient of variation (CV) was 4.5 to 7.92, and the inter-assay CV measured 7.8 to 11.3%.

Statistical analysis

Continuous data, such as the length of the post-partum acyclic period, were **compared using** analysis of variance. Chi-square tests were used to examine the relationship between short cycles and the length of the acyclic period, milk production and occurrence of silent heat.



Fig.1. Average milk progesterone profiles of cows which: (a) did not show a transient rise after calving (n=45), and those which showed (b) one (n=63), (c) two (n=15) or (d) three (n=3) transient rises in milk progesterone. E/O: oestrus and ovulation, OEC: oestrous cycle, AP = post-partum acyclic period, TCC = true cycle commenced, TPR = post-partum transient progesterone rise.

Two distinct types of milk progesterone profiles were observed. The first pattern showed a period of low progesterone levels (< 1 ng/ml) followed by normal luteal activity (> 14 days) with progesterone levels rising from 1 ng/ml (day 3) to a peak of 18.6 ng/ml (days 13-15), (Fig. 1 a). The second pattern showed low progesterone levels, then a transient (< 9 days) rise no greater than 5 ng/ml and the commencement of normal cyclical activity (Fig. 1 b, c, d). Of 126 calving cows 35.7% had no transient rise in progesterone while 50%, 12% and 2.4% had one, two or three short cycles respectively (Fig. 1). Based on these profiles 76% of cows showed luteal activity and ovulation by 30 days after calving, but only 45% of these (43/96) were detected in oestrus. By 60 days after calving all cows had ovulated at least once and the incidence of no visible oestrus (NVO-failure to display/detect oestrus) decreased to 45%.

Based on oestrus detection and progesterone levels of up to four successive oestrous cycles, the length of subsequent cycles did not differ in cows which had an initial short cycle, and those whose initial cycle was of normal length. There was no significant difference between seasonal calving groups in the number of cows showing short cycles post-partum, or the duration and peak levels of milk progesterone in the transient rise (Table 1).

The 81 cows with short cycles took approximately 10 days longer to commence normal cyclic activity than cows which did not exhibit short cycles (29.3 \pm 0.94 days vs 18.6 \pm 0.88 days; p < 0.01), and this did not differ with season.

Parameter	Group 1	Group 2	Group 3	Sig.
Number of cows	41	43	42	
Cows with transient rise in progesterone	28 (68.3%)	26 (60.5%)	27 (64.3%)	N.S.
Duration of transient prog. rise (days. mean ± SE)	7.4 0.42	6.0 0.32	6.3 0.38	N.S.
Peak level of milk prog. (ng/ml mean ± SEE	3.3 0.30	3.6 0.35	4.6 0.40	N.S.
Length of acyclic period (days. mean ± SE. all cows)	27.3 1.4	26.9 1.4	25.2 1.3	N.S.
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Table 1. Characteristics of the post-partum transient rise in progesterone level and the length of the acyclic period in the three calving groups.

Daily milk production ranged from 10.0 to 27.4 litres, but no relationship between milk yield and the incidence of short cycles was demonstrated. Similarly there was no significant correlation between milk yield andthelength of the acyclic period after calving. Seventy percent (57/81) of cows were pregnant to their first insemination after experiencing at least one short cycle, compared to 77% (35/45) of cows which did not have a short cycle (N.S.). The mean liveweight and condition score of all cows was 491 ± 7.1 kg and 4.0 ± 0.05 (Mean ± SE) and these did not differ significantly with season, the incidence of short cycles, or the length of the acyclic period after calving.

DISCUSSION

Combined information from milk progesterone profiles and observation of oestrus has proven especially useful in the study of longer-term serial changes in ovarian activity (Bloomfield et al. 1986). In the current study we found

there was no difference in the pattern of milk progesterone profiles from cows calving in spring, summer or autumn and winter and the average length of the post-partum acyclic period was 26.5 days. This interval is comparable to that measured by Bulman and Lamming (1978) and Van de Wiel et al. (1979). In contrast to our work, some studies have demonstrated a seasonal influence on the acyclic period post-partum with a negative correlation between this interval and photoperiod in late pregnancy (Bulman and Lamming 1978; Peters and Riley 1982; Hansen and Hauser 1983). It has been suggested that housing during winter, a common procedure in the northern hemisphere, could influence the proportion of cows in anoestrus or their ability to express heat symptoms, especially if animals were tied in stalls (Bulman and Lamming 1978). This may emphasise a tendancy towards seasonality in the length of ' post-partum anoestrus. Secondly. in the northern hemisphere, spring calving cows are more likely to calve in poorer condition than at other times of the year which could adversely influence subsequent ovarian activity; though Peters and Riley (1982) adjusted for the effect of bodyweight and still demonstrated seasonal differences in post-partum In our study the animal's condition and the high-plane of nutrition anoestrus. encountered in all seasons probably overrode any effect of season or milk it yield on reproductive activity. Under conditions of nutritional stress, or in highly productive animals, seasonality in reproductive activity may be observed, though this is yet to be tested.

The first indication of ovarian activity in 65% of cows was a transient rise in progesterone levels. The incidence of this did not vary with season, similar to the findings of Bloomfield et al. (1986). However, the transient rise in progesterone did extend the acyclic period after calving as normal cyclicity with a **luteal** phase of at least 14 days was delayed. The progesterone secreted during the short cycle may be from a luteinised follicle ((Lamming et al. 1981) or from a first corpus **luteum** which regresses early, resembling the situation described by Berardinelli et al. (1979) in pre-puberal beef heifers.

Three quarters of the cows in this study showed ovarian activity and ovulation before day 30 after calving, but less than half of these were seen in oestrus. These results confirm the prevalence of silent oestrus in dairy cows soon after **calving** recorded by Van'de **Wiel** et al. (1979) at 48%. It is likely the transient rise in progesterone is insufficient to prime cows to display oestrus coincident with the following ovulation similar to about 50% of "ram-induced" ovulations in ewes in spring (Oldham and Martin 1978).

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