DETECTION OF OESTRUS BY MILK PROGESTERONE ASSAY, VISUAL OBSERVATION AND CERVICAL MUCUS CONDUCTIVITY IN OESTRUS – SYNCHRONISED DAIRY COWS

G.J. SAWYER*, P.E. WILLIAMSON*, Adji DRADGET* and G. HOWELL*

Sixty cows were treated with Syncro-Mate B or prostaglandin and cyclical activity was monitored by frequent sampling for milk progesterone and measurement of the electrical resistance of cervical mucus. Visual observation of oestrus was practised four times daily. Oestrus and ovulation occurred in 80% of Syncro-Mate B and 87% of prostaglandin treated cows. The proportion of cows which could be inseminated at the correct time was highest if based on milk progesterone profiles, then visual observation of oestrus, and finally on measurement of the electrical resistance of cervical mucus. The large individual variation in resistance readings, labour requirement and irritation to the vagina caused by the probe lead us to conclude that this technique is not suitable for oestrus detection or the timing of insemination on commercial dairy farms.

INTRODUCTION

Strategic use of hormone treatments can reduce calving to conception intervals, concentrate calving patterns, reduce herd empty rates, and potentially increase the average lactation length and the period of peak milk production in dairy herds (Macmillan 1985). There have been numerous studies on synchronisation methods which either shorten the life span of the corpus luteum (prostaglandins), or which lengthen the cycle using progesterone treatments in intravaginal devices (PRID or CIDR) or subcutaneous implants with progestagen (Syncro-Mate B). In all cases conception rates are higher when insemination is based on detected oestrus rather than at a fixed post-treatment interval. Some encouraging results were obtained with fixed time insemination in programs using Syncro-Mate B (Pelot et al. 1984) but few experiments have been conducted in Australia with this product.

More information is required on factors influencing ovarian response to synchronising treatments, including stage of the oestrous cycle, follicular development and the level of endogenous hormones such as progesterone. Measurement of milk progesterone provides a useful aid in monitoring response to synchronising treatments and oestrous cycle activity. It may usefully be applied in evaluating methods of oestrus detection such as observation by the herdsman and measurement of the electrical resistance of cervical mucus by electronic probe (Foote et al. 1979; Cavestany and Foote 1985). The present study compared two treatments to synchronise oestrus and ovulation in dairy cattle, using milk progesterone levels to evaluate the response to treatment and the accuracy of oestrus detection based on visual observation and changes in electrical conductivity of cervical mucus.

MATERIALS AND METHODS

Sixty cows were paired at least 60 days after calving according to calving date and parity, and alternately allocated to one of two groups. Group 1 (n = 30) – injected with 5 mg oestradiol valerate, and 3 mg norgestomet on day 0 and given a norgestomet ear implant for 10 days (Syncro-Mate B treatment, Intervet, Sydney, Australia Pty Ltd); Group 2 (n = 30) – injected twice with prostaglandin (cloprostenol 0.5 mg, Estrumate, ICI Australia Ltd) 11 days

* Department of Agriculture, PO Box 1231, Bunbury, W.A. 6230.
Milk samples were collected according to the protocol illustrated in Fig. 1.

The concentration of progesterone in duplicate 10 μl milk samples was determined using a solid phase, microtitre plate progesterone enzyme-immunoassay (Munro and Stabenfeldt 1984). The sensitivity of the EIA standard curve was 0.4 μg/l and with three progesterone pools (4.9, 14.3 and 25.6 μg/l) the intraplate C.V. was 4.8% to 16.7%, the inter-assay C.V. measured 7.3% to 17.4% and the intra-assay C.V. was 3.9% to 8.2%

Three days after allocation to treatments the electrical resistance of cervical mucus was measured in 18 cows chosen at random from each group using the Animark Ovascan probe, sanitized and inserted into the anterior vagina against the cervix, until the displayed resistance units stabilised (30-40 sec). The protocol is described in Fig. 1. Cows were observed for oestrus four times daily at 0600, 1000, 1600 and 2200 h and were artificially inseminated using thawed frozen semen 12 h after first observed in oestrus.

Continuous data relating milk progesterone levels, the electrical resistance of cervical mucus and the timing of the onset of oestrus were analysed by analysis of variance and Student’s "t" test. Discrete data including synchrony response and detection of oestrus were analysed by Chi-square tests.

RESULTS

Control of oestrus and ovulation was similar in both treatment groups at 80% within three days for Group 1 and 87% within five days for Group 2, but fewer cows treated with Syncro-Mate B were detected in oestrus (n.s., Table 1). Cows injected with prostaglandin displayed oestrus 27.7 hours later, on average, than those treated with Syncro-Mate B (P <0.001, Table 1). Examination of progesterone profiles showed that six cows in Group 1 and four in Group 2 were not synchronised. Half of these Group 1 cows had low concentrations of progesterone (<2 μg/l) at implant insertion, which stayed low for 3-5 days then gradually increased, signifying the presence of a new, active corpus luteum. Progesterone did not decline at implant removal. The other cows not synchronised had luteal concentrations of progesterone which declined, then a new ovulation occurred. Four cows did not respond to prostaglandin. One had uniformly low (<1 μg/l) progesterone and was judged to be not cycling, and in the others progesterone concentrations fell in response to prostaglandin, but a new corpus luteum was not initiated and hence, cows could not respond to the second injection of prostaglandin. Based on milk progesterone profiles all cows which were not detected in oestrus were apparently synchronised. Undetected oestrus could not be related to the progesterone concentration or stage of the oestrous cycle when treatment commenced. Only 37.5% (9/24) cows of Group 1 and 50% (13/26) cows of Group 2 conceived to AI at the synchronised oestrus,
All inseminations were made when milk progesterone was below 0.5 ug/l.

Table 1 Cows responding to synchronising treatments assessed by milk progesterone profiles, visual detection of oestrus and vaginal probe readings

<table>
<thead>
<tr>
<th>Measurement/</th>
<th>Number of cows</th>
<th>Number synchronised according to MP profiles (%)</th>
<th>Number detected by visual observation (%)</th>
<th>Number detected by vaginal probe (%)</th>
<th>Standing oestrus</th>
<th>Low probe resistance</th>
<th>Hours (+ s.e.) from end of treatment:</th>
</tr>
</thead>
<tbody>
<tr>
<td>Syncro-Mate B treatment</td>
<td>30</td>
<td>24* (50%)</td>
<td>12 (79%)</td>
<td>2* (6%)</td>
<td>47 ± 2.9</td>
<td>23.1 ± 3.4</td>
<td></td>
</tr>
<tr>
<td>Prostaglandin treatment</td>
<td>30</td>
<td>26** (97%)</td>
<td>24 (92%)</td>
<td>13** (76%)</td>
<td>74.7 ± 5.0</td>
<td>69.9 ± 20.3</td>
<td></td>
</tr>
</tbody>
</table>

* Within 3 days. ** Within 5 days. #Significantly different, P<0.001. Cows not included because they were not synchronised or a lack of readings due to irritation of vaginal mucosa.

Successful detection of oestrus based on retrospective analysis of vaginal probe data was worse than that using visual detection (Table 1). Average electrical resistance readings are presented in Fig. 2 and show that the lowest resistance values are generally associated with the peri-oestrus period (± 2 days), but the large standard errors (Table 1) indicate that this measurement is much less reliable than visual observation of oestrus. The drop in electrical resistance from luteal to oestral phases ranged between 20-30% for individual cows (Fig. 2) whereas cyclical differences in profiles of milk progesterone at similar stages of the oestrous cycle range between 8 and 15 fold.

DISCUSSION

Concentrations of milk progesterone proved useful in interpreting the response to both synchronising regimes used in this experiment. The work highlights the value of this technology when studying reproductive activity in dairy cattle. Profiles during and after Syncro-Mate B treatment generally confirmed the luteolytic action of oestradiol valerate on corpora lutea at least six days old. When treatment began just after oestrus and ovulation the secretory activity of the corpus luteum was sometimes modified and cows showed a synchronised oestrus, but in three cases oestrus and subsequent ovulation...
was delayed due to a functional corpus luteum established in the presence of Syncro-Mate B, similar to observations by Thimonier et al. (1975). This may be inherent biological variation in response to oestradiol valerate early in the oestrous cycle, and precludes the possibility of good results to fixed time insemination with this treatment. In those animals which responded to treatment with Syncro-Mate B the time until oestrus was one day shorter than the response to prostaglandin and was within five hours from the timing of oestrus after treatment published by Whittier et al. (1986). Failure to synchronise oestrus and ovulation in response to prostaglandin was due to the lack of establishment of a new corpus luteum ready for a second luteolytic dose of prostaglandin.

In this experiment, concentrations of milk progesterone served as a yardstick against which other methods of oestrus detection were measured. Though a useful research tool, successive measurement of milk progesterone to form a profile for each cow and establish the timing of oestrus and ovulation, is neither practical nor economic on commercial farms. The next most reliable method was visual observation of oestrus. The proportions of cows detected were similar to other work where oestrus was looked for frequently (Foote 1975). Measurement of the electrical resistance of cervical mucus by vaginal probe proved the least reliable method for detection of oestrus. The large individual variation and sometimes erratic readings meant that although the resistance measured tended to follow cyclical patterns, results were too variable to accurately predict oestrus or time insemination appropriately. This result is in general agreement with other studies (Gartland et al. 1976; Cavestany and Foote 1985) and at variance with other work by Foote et al. (1979) who inseminated cows on the basis of probe readings. We took care not to let air in the vagina influence probe readings. The time taken to perform the task. This time requirement, and the fact that frequent use of the probe around the expected time of oestrus led to irritation of the vulvo-vestibular mucosa and some discomfort to cows on a few occasions, mitigates against widespread use in commercial dairy herds run at pasture.

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REFERENCES