# The use of milk progesterone measurements for genetic improvement of fertility traits in dairy cattle

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#### Abstract

Fertility measures based on calving and insemination dates are heavily affected by a farmers decision to cull or inseminate a cow. This might partially explain the low heritability for these fertility traits and the unfavourable association between yield and fertility. Progesterone measurements might provide an alternative. Heritability estimates ranged from 0.14 to 0.20 (s.e. = 0.08) for interval between calving and first luteal activity (CLA). However, measuring this trait in a progeny testing scheme is not feasible. Therefore, mean progesterone profiles (transformed to a binomial scale) were investigated for groups of 50 animals differing in CLA. Using splines clearly different curves were observed, even when only monthly progesterone measurements were available for each cow. Days between calving till 50% of the daughters is cycling (progesterone > 3 ng/ml) might provide a good trait for sire selection. Phenotypic association between CLA and milk yield were unfavourable. If milk yield increased with 1 kg per day over the first 100 days of lactation, CLA is expected to increase with 1.4 days. Similarly, the regression coefficient of CLA on energy balance was -0.33 (s.e. = 0.11). Hence, at cow level progesterone measurements provide a heritable trait which is useful for selection and for investigating the relationship between fertility and production traits. Indications from this study are that progesterone measures during monthly milk recording might be useful to establish a mean progesterone profile for a group of animals. Whether this measure of female fertility is heritable and useful for sire selection in a progeny testing scheme requires a larger data set for further investigation.

## 1. Introduction

Low fertility is of economic importance for dairy enterprises, because it results in higher levels of involuntary replacement, slippage in calving pattern, veterinary invention, hormonal treatment and reduced annual milk production (Esslemont and Peeler, 1993). Even when the definition of optimal fertility performance might change (for example the optimal length of the calving interval is still disputed productive for the highest cows) genetic improvement of fertility remains important: it gives the farmer control over its fertility management (i.e. which heat to inseminate, which month of the year does he wants most milk, what is the optimal calving interval). Hence, genetic improvement should give a the farmer the option to inseminate cows at the economically most optimal time period, whatever the time period is. However, genetic selection for improved fertility is hindered by the low heritability for fertility traits (Jansen, 1985; Pryce et al. 1997b, c).

One of the reasons for this low heritability might be that most fertility traits are based on calving and insemination dates. These are obviously affected by the farmers decision to inseminate or cull animals and many other factors. Error variances might be relatively large therefore, rather than that genetic variances for fertility traits are small. For example, compared with a genetic coefficient of variation of 9.5% for milk yield, this parameter for calving interval, days to first service and conception to first service are 1.8%, 4.4% and 10.3%, respectively (Price et al., 1997a). Further indications that errors due to management decisions might affect heritability estimates for fertility traits is that for the interval to first heat a relatively high heritability of 0.15 has been reported. Furthermore, heritability estimates coming from records collected on an experimental farm. under controlled fertility management, are also relatively high, i.e. 0.08 for calving interval, 0.13 for days open and days to first service (Pryce *et al.* 1997c). Hence, there appears to be sufficient genetic variation for fertility traits, but recording practises include too much noise with a negative effect on the heritability.

The second difficulty with using calving dates and insemination dates to measure a cows fertility objectively is that association between yield and fertility might be biased (Philipsson, 1981; Jansen, 1985). Farmers might cull low producing cows earlier or delay inseminating a high producing cow which both might have an affect on the observed correlation between yield and fertility.

To overcome some of these problems, Darwash et al. (1997a, b) suggested the use of progesterone profiles to measure the start of luteal activity. These authors demonstrated how early re-establishment of ovarian activity (CLA), measured using progesterone profiles, is an important prerequisite for high fertility; one day delay in the interval to first ovarian activity was associated with 0.24 and 0.41 days extra in the intervals to first service and conception, respectively. Each additional 21 days in CLA progressively reduced the probability of failure at a given insemination to 0.89 of its previous value Darwash et al. (1997a). Furthermore, Darwash et al. (1997b) reported a heritability of between 0.13 and 0.28 for CLA. Hence, using progesterone profiles, a measure of fertility is available which is unaffected by management decisions and seems to have a reasonable heritability.

Because only one estimate of the heritability for CLA is available yet in the literature, the first objective of this study was to estimate the heritability of days between calving and first ovarian activity measured using progesterone profiles.

Measuring progesterone profiles on individual cows is still costly and labour intensive as many milk samples (two or three times a week) need to be collected and analysed. This might only be feasible on a small scale in for example a nucleus herd. As an alternative, using the mean progesterone profile of many irregular sampled animals (for example monthly milk recording) might be sufficient for a progeny tested bulls. However, because not sufficient progesterone measurements were available to investigate average profiles for individual bulls, mean progesterone profiles for groups of around 50 animals differing in CLA were investigated herein.

Finally, because CLA is unaffected by management decisions, the phenotypic association between CLA and both milk yield and energy balance is investigated.

## 2. Material and Methods

## 2.1. Data

Data were available for Holstein Friesian heifers calving at the ID-DLO experimental farm 't Gen from 1994 to 1996. Part of the animals in this analysis are among the highest genetic merit animals, as these are part of the 'Delta' sib testing program. The other heifers originate from the ID-DLO farm and were on average about half a standard deviation below the Delta heifers for INET (the Dutch production index).

A data set was prepared containing two progesterone records per week for each animal (Monday afternoon and Friday morning). Animals with uterine infections or animals which had an abortion were excluded from the data set. For all other animals (n = 333) days to first luteal activity (CLA) was determined from the progesterone profile as follows: when two consecutive samples were above 3 ng/ml the date of the first sample was taken as start of luteal activity. Since progesterone samples were taken till approximately day 100 in lactation, animals not cycling received a CLA of approx. 100 days.

On a subset of these animals (n = 226), data on milk yield and energy balance during the first 100 days of lactation were available. More details of the recording and data handling are given by Oldenbroek *et al.* (1997).

## 2.2 Analysis

The heritabilities (plus s.e.) were estimated using VCE (Groenenveld, 1996). Additional to the random animal effect (including relationship matrix), year-season and age at calving were fitted as fixed effects. Because its skew distribution, not only CLA was analysed, but also two transformations of CLA: the natural log and 100 / CLA.

There were obviously not sufficient records to investigate average profiles for individual bulls. However, to investigate differences in mean progesterone profiles for groups of around 50 animals, data were grouped according to CLA. Then, average progesterone profiles were calculated for each group separately, whereby the actual progesterone records were transformed in a binomial trait: al records above 3 ng/ml received the value of 1 and all other records the value 0. This latter trait has the advantage that it is easily interpretable compared with actual progesterone levels, i.e. the average indicates the proportion of animals with progesterone > 3 ng/ml on a certain day (which might be indicative for the proportion of animals cycling at a certain date). Also the binary trait simulates a relatively simple 'yes or no' progesterone test. In practise, such a 'dipstick test' might have advantages when used on a large scale.

To investigate mean profiles, curves were created for each group separately. First, by simply averaging all the records per day of lactation, and secondly by fitting a smoothing spline in Genstat. The latter approach was applied on all records of individual cows (two a week; ca. 29 per animal), and on a subset of measurements for each cow (1 every month; ca. 3 per cow) simulating measurements during normal milk recording. Hereto, each animal was assigned a random start day of first test (uniform distribution between 5 and 35 days), and subsequently for each cow the records closest to four weekly intervals were selected. A simple linear regression was used to investigate the phenotypic association between CLA and energy balance or milk yield.

## 3. Results

# 3.1. Heritability for CLA

In this study mean CLA was 38 days and the standard deviation was 25 days. Heritabilities for CLA and its transformations ranged from 0.14 to 0.20 (Table 1). A genetic standard deviation of 11 days suggest that differences between animals are considerable for the interval between calving and first luteal activity.

*Table 1.* Mean, standard deviation (in days) and heritability (s.e.) for days till first luteal activity (CLA) based on progesterone profiles, and  $\log_e(CLA)$  and 100/CLA (n = 333).

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	Mean	s.d.	range	h²
CLA	38	25	11 - 105	0.20 (0.07)
log <sub>e</sub> (CLA)	3.5	0.6	2.4 - 4.7	0.17 (0.08)
100 / CLA	3.6	1.8	0.9 - 9.1	0.14 (0.08)

## 3.2. Average group curves

Mean progesterone profiles for groups of 50 animals with all twice weekly progesterone measurements animals are given in Figure 1a. Although curves are clearly distinguishable between the groups, between days there is a lot of noise. This is not unexpected, as, besides normal error variation, individual animals were only sampled twice a week and curves of individual animals are not synchronised. Whilst a normal cycling cows is expected to have one or two progesterone samples below 3 ng/ml every 21 days, and on average about 14 cows are measured each day, large sampling fluctuations in the mean progesterone curves are expected. For the same reason it is expected also that most curves asymptote around 0.7 - 0.8. Given that most of the animals in this study were not inseminated before day 100, it is expected of normally cycling cows that, on a given day, about 1/6 or 2/6 of the progesterone measurements is below 3.0 ng/ml, because these cows are in heat.

When splines were fitted on all individual progesterone records, but for each group separately, mean profiles became more distinct (Figure 1b). Therefore it might be possible that from these smoothed curves, traits of interest for selection can be derived. For example, interval between calving and the time period that 0.5 of the samples is above 3.0 ng/ml seems to be an option to indicate fertility performance of a group of animals. The proportion

0.4 or 0.5 seem to be in the most distinctive part of the mean profiles.

The same conclusion can be drawn when only monthly progesterone samples were available for each cow: it was still possible to identify the curves for the six groups clearly (Figure 1c). Although the within group variation might be larger for sire progeny groups than for our six groups, similar mean differences in CLA are expected between progeny groups. This is because the genetic standard deviation for CLA is close to 11 days (Table 1). Therefore, it might be possible to estimate smoothed mean progesterone profiles for bulls using progesterone samples on daughters during normal milk recording.

## 3.3. Milk yield and energy balance

Another use of progesterone is that it measures fertility more objectively, compared with relying on insemination dates and recording of the farmer. For this reasons it might provide a tool that enables us to the management effect exclude (i.e. later insemination of high producing cows) on the relationship between yield and fertility. Figure 2 demonstrates that even when fertility is measured using progesterone, the phenotypic association between CLA and milk yield is unfavourable. When daily milk increased with 1 kg a day over the first 100 days, CLA is expected to increase with 1.4 days (s.e. = 0.3). Similarly, the phenotypic relationship between energy balance and CLA is negative: the regression coefficient is -0.33 (s.e. = 0.11).

# 4. Discussion

The use of progesterone measurements for the improvement of fertility records (as suggested by Darwash *et al.* 1997a and 1997b) is demonstrated in this study. Using twice weekly samples on individual cows, days till first luteal activity could be established. Heritabilities for CLA and its transformations were not significantly different from the values reported by these authors. Although, on the measured scale our heritability was slightly lower (although not significantly different from the much larger study).

The mean value of 38 days is higher than the value of 27 days reported by Darwash *et al.* (1997b) and the standard deviation was nearly twice the value reported by these authors. There are several possible explanation for this: i) in our data set progesterone was measured twice a week only and started at day 15 only. Darwash *et al.* (1997b) had at least three progesterone measures a week. Given that CLA was based on two consecutive samples above 3 ng/ml, in our study we might have missed some initial small increases in progesterone; ii) animals

included in the study reported here were part of a testing program and therefore hardly any animals were culled before day 100. In commercial practice, some animals might have been culled before they show heat, and in fact not showing heat might be among the culling reasons; iii) mean milk yield in our population was close to 30 kg/d, whereas the mean milk yield in the population reported by Darwash *et al.* (1997a, b) was about 18 kg/d. If the regression of CLA on yield demonstrated a causal relationship (Figure 2), then CLA was expected to be 17 days longer in our study.

However, the results in this study confirm that progesterone measures might provide a useful tool for genetic selection. In herds where intensive progesterone sampling is practised (i.e. for management reasons or in a nucleus herd) CLA can be measured for individual animals. In the situation where only monthly milk recording is practised, mean progesterone profile based on a simple 'dipstick test' might by sufficient to distinguish fertility performance of large progeny groups. Splines can be used to smooth mean progesterone profiles and based on these splines, the interval between calving and days till 0.5 of the samples are above 3.0 ng/ml provides a simple trait which could be used as trait to distinguish differences between groups. A larger data set with progesterone measured is required, however, to establish whether the heritability for this trait is as large as the heritability for CLA.

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*Figure 1.* Mean progesterone profiles (prog >3.0 ng/ml sample is 1 otherwise sample is 0) for cows grouped according to their CLA. Median CLA values for the six groups are 17, 19, 24, 31, 47 and 85 days, respectively, and each group contains around 50 records. The top graph is the average of the raw data based on two progesterone samples a week per cow. In the second graph smoothing splines are fitted through this data. In the bottom graph monthly milk recording was simulated by using only one sample every 28 days per cow.



*Figure 2.* Days till first luteal activity (CLA) measured using progesterone as a function of average a) milk yield and b) energy balance during the first 100 days of lactation