# INSEMINATION STRATEGY BASED ON OVULATION PREDICTION IN DAIRY CATTLE

*Inseminatiestrategie gebaseerd op ovulatievoorspelling bij melkvee*

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## ABSTRACT

**Calving rates after first insemination are often less than 50% in practice. Part of this low percentage might be explained by wrongly timed inseminations. Our hypothesis is that it is better to time insemination according to ovulation instead of according to behavioral estrus, but up to now it has not been possible to predict the time of ovulation in practice. So, to better time inseminations in practice, there is a need for predictors of ovulation time. Therefore, the relationship between various estrus characteristics and time of ovulation was studied to investigate whether these characteristics could predict time of ovulation. First standing heat was displayed 26.4±5.2h before ovulation. The increase in number of steps during estrus predicted the time of ovulation (29.3±3.9h) best. The next question was: What is the best time for insemination relative to ovulation? When this time is known, an insemination strategy can be formulated. It was found that inseminations performed 24 to 12h before ovulation resulted in the highest number of good quality embryos at Day 7. This means that the optimal strategy is to inseminate 5 to 17h after the first increase in the number of steps. This strategy will result in optimal timed inseminations relative to time of ovulation.**

## SAMENVATTING

De afkalfpercentages na de eerste inseminatie zijn vaak minder dan 50%. Een deel van dit lage percentage zou verklaard kunnen worden door een suboptimaal inseminatietijdstip. Onze hypothese is dat het beter is om het tijdstip van inseminatie te bepalen aan de hand van de ovulatie in plaats van het bronstgedrag, maar tot nu toe was het niet mogelijk om het moment van ovulatie in de praktijk te voorspellen. Om het inseminatietijdstip nauwkeuriger in te schatten, is er behoefte aan een manier om de ovulatie te voorspellen. Daarom wordt de relatie tussen de verschillende tochtkenmerken en het ovulatietijdstip bestudeerd, om op die manier te onderzoeken of deze tochtkenmerken het ovulatietijdstip kunnen voorspellen. De eerste staande tocht werd vastgesteld op 26,4±5,2 uur vóór de ovulatie. De verhoging in het aantal stappen tijdens de tocht voorspelde het tijdstip van de ovulatie (29,3±3,9 uur later) het best. De volgende vraag is: wat is het optimale tijdstip van inseminatie ten opzichte van de ovulatie? Wanneer dit tijdstip bekend is, kan een algemene inseminatiestrategie geformuleerd worden. In een onderzoek werd het grootste aantal embryo's van goede kwaliteit gevonden bij inseminatie 24 tot 12 uur vóór de ovulatie. Alles bij elkaar genomen betekent dit dat inseminatie 5 tot 17 uur na de verhoging van het aantal stappen de optimale strategie is om koeien drachtig te krijgen.

## INTRODUCTION

For conception to occur, insemination must take place at the correct stage of the cow's estrous cycle. Successful fertilization greatly depends on the interval from insemination to ovulation. When cows are inseminated too early, the sperm are aged by the time ovulation occurs and they can no longer successfully fertilize an ovum (Hawk, 1987). When insemination takes place too late, fertilization and formation of a viable embryo may no longer be possible because of the ageing of the egg (Hunter and Greve, 1997). In practice, however, it is not possible to predict the time of ovulation.

Many experiments have studied the effects of the timing of insemination according to estrus (e.g Maatje *et al.*, 1997; Dalton *et al.*, 2001), but knowledge is scarce about the effects of time of insemination relative to the time of ovulation on final pregnancy results. So, to come up with an optimal insemination strategy that will achieve high calving rates for dairy cattle, parameters must be established that can predict the time of ovulation, and the optimal insemination-ovulation interval must be assessed. In several experiments (Roelofs *et al.*, 2004; Roelofs *et al.*, 2005bc; Roelofs *et al.*, 2006) a number of possible predictors of ovulation time were studied. In this overview, we discuss the applicability and quality of the various ovulation time predictors, namely visual observation, activity measurement, hormone profiles, vaginal mucus conductivity and body temperature. Subsequently, the optimal timing of insemination relative to ovulation is discussed. Lastly, the best predictors of ovulation time are combined with estimated calving rates as a function of the interval between insemination and ovulation to arrive at a practical insemination strategy that maximizes calving rates.

## POSSIBLE PREDICTORS OF OVULATION TIME

For a parameter to be useful as a predictor of ovulation time, several prerequisites need to be met. The parameter should have a small variation in time to ovulation, and the measurements should be easy to carry out, repeatable and preferably automated. It should also be present in a high proportion of the animals. Various parameters were studied as possible predictors for time of ovulation and evaluated in terms of their practical applicability (Table 1).

One advantage of monitoring hormone concentrations to predict time of ovulation is that the changes in LH, estradiol and progesterone concentrations occur in all animals before ovulation (Table 1). However, monitoring of hormones to predict time of ovulation is not yet applicable in practice. Although the surge in LH shows a very good relationship with time of ovulation, it is not possible to monitor this hormone easily. Efforts have been made to monitor LH in the milk of dairy cattle and buffaloes, but up to now these efforts have not been successful (Batra and Pandey, 1983; Johnson and Reeves, 1988). Even when LH could be measured in milk, it would probably not be an appropriate tool to predict the time of ovulation because of the short duration of the LH surge in dairy cattle (on average 9.5±1.6h, Roelofs *et al.*, 2004).

Estradiol was measured in blood plasma, which is not feasible in practice, but estradiol concentrations can also be assessed in milk (Meisterling and Dailey, 1987). However, ovulation time can probably not be predicted by monitoring estradiol concentrations alone, because estradiol concentrations not only increase before ovulation

**Table 1. Overview of the practical applicability of various parameters to predict time of ovulation in dairy practice[1].**

| Parameter | Relation to ovulation time | | | Labor | Automation | Proportion |
|---|---|---|---|---|---|---|
| | Average | S.D. (h) | Range (h) | input | possible | of animals |
| Behavior (onset of showing) | | | | | | |
| All signs[2] | 30.6 | 5.1 | 20 | high | no | 100% |
| Standing heat | 26.4 | 5.2 | 27 | high/low | yes | 58% |
| Mounting | 28.7 | 5.3 | 30 | high | no | 90% |
| Walking activity increase | 29.3 | 3.9 | 17 | low | yes | 83% |
| Progesterone decline | 79.7 | 11.2 | 44 | high/low | no/yes | 100% |
| LH-peak | 25.3 | 2.0 | 6 | high | no | 100% |
| Oestradiol-decline | 23.9 | 3.9 | 16 | high/low | no/yes | 100% |

[1]Based on the results of the experiments described in Roelofs *et al.* (2004), Roelofs *et al.* (2005bc) and Roelofs *et al.* (2006); [2]All behavioral signs as described by van Eerdenburg *et al.* (1996), which include sniffing, flehmen, chin resting, mounting, being mounted, standing heat and restlessness.

but may also increase during the estrous cycle (Dieleman *et al.*, 1986), probably reflecting the growth of a dominant (but not preovulatory) follicle. Monitoring of the progesterone decrease before ovulation is not suitable because of the large variation in time of ovulation (Table 1). If estradiol and progesterone could be measured automatically during milking, then perhaps the monitoring of the two hormones simultaneously could be used to predict time of ovulation. This depends on the relationship between the peak or decrease of estradiol and time of ovulation. In our experiment (Roelofs *et al.*, 2004) the decrease in estradiol relative to time of ovulation was assessed in 12 animals, taking blood samples for estradiol at 3h intervals. The time from estradiol decrease to time of ovulation was 16h. Whether this time range would be similar if estradiol were assessed twice a day in milk, needs further investigation. If the decrease in milk estradiol could serve as a predictor for ovulation time, then simultaneous measurements of milk progesterone concentrations could be helpful for defining false positive alerts that occur due to increased estradiol during the estrous cycle. When progesterone concentrations are high, it is obvious that ovulation will not occur.

Observation of behavioral estrous signs and changes in walking activity (measured by the number of steps in 2h periods) seem to be applicable predictors of ovulation time because of the good relationship with time of ovulation (Table 1). However, observation of behavioral signs every 3h for 30 min, as was done in our experiment (Roelofs *et al.*, 2005c), is too labor intensive to be carried out in practice. The effects of less frequent observations or limiting the number of behavioral estrous signs will be discussed in light of their usefulness as predictor of ovulation time. Measurements of walking activity can be automated and thus are easy to carry out. This parameter will be further discussed as a predictor of ovulation time. Changes in vaginal mucus conductivity and body temperature have been shown to have a close relationship with the LH peak (Schams *et al.*, 1977; Rajamahendran *et al.*, 1989; Mosher *et al.*, 1990), so these parameters are also discussed as possible predictors of ovulation time.

## Behavioral estrous signs

Ovulation occurred on average 30.6±5.1h after the onset of behavioral estrus when observations were done for 30 min every 3h and all behavioral estrous signs according to Van Eerdenburg *et al.* (1996) were included, i.e. sniffing, flehmen, chin resting, mounting, mounting headside, being mounted and standing heat (Roelofs *et al.*, 2005c). In practice, sniffing and chin resting may often not be noticed as estrous behavior. When only mounting behavior or standing heat is taken into account, ovulation occurred on average 28.7±5.3h and 26.4±5.2h after first display of these behavioral signs, respectively. Other studies that assessed the interval between onset of estrus based on standing heat (assessed with mounting detectors) and time of ovulation, found similar intervals (e.g. Walker *et al.*, 1996; Lopez *et al.*, 2002). However, the onset of standing heat is not practically suitable as a predictor for ovulation time, because of the low proportion of animals that show standing heat (Table 1). Moreover, observations of behavioral estrous signs as carried out in our experiments are very labor intensive. When observations are carried out less frequently, the variation in ovulation time, as presented in Table 1, is not much affected. However, only 30% and 19% of the cows in estrus would have been seen displaying standing heat if observations had been carried out three or two times per day for half an hour, respectively.

Mounting behavior (mounting, or trying to mount another cow) is displayed in a large number of estrous periods. It is observed in 89% of all estrous periods with observations every 3h. Unfortunately, this behavior cannot (yet) be recorded automatically. This makes mounting behavior a promising but not yet practical predictor of ovulation time.

In conclusion, the relationship between the display of behavioral estrous signs and time of ovulation is good (Table 1). However, observation of all the behavioral signs except for standing heat cannot be automated and requires high labor inputs. Decreasing observation frequencies result in lower detection percentages. Standing heat can be assessed automatically, but is displayed in a low proportion of the animals (Table 1). All together, prediction of ovulation time based on behavioral estrous signs is not feasible in dairy practice.

## Activity measurements

In contrast with behavioral estrous signs, prediction of ovulation time by an increase in walking activity (measured by the number of steps taken in 2h periods) can be automated and requires little time and effort (Table 1). The detection rates for activity measurements greatly depend on the algorithms and thresholds that are used to define an increase in activity. In our experiment, the best detection rate was 87% (Roelofs *et al.*, 2005b). The number of steps was stored in 2h periods. However, many pedometers used in practice store the number of steps in 12h periods. Liu and Spahr (1993) concluded from their experiment that activity counts that were stored in 12h periods identified the increased activity during estrus as well as the activity counts stored in 2h periods. When the data of our experiment are converted to number of steps in 12h periods (from 0.00h to 12.00h and from 12.00h to 0.00h) the percentage of correctly detected estrous periods become 92%. This detection rate is higher compared to storage of steps in 2h periods. However, the number of false estrus alerts increased (i.e. the increase in activity does not coincide with behavioral estrus and an ovulation): 1 to 2 false estrus alerts were found in 26% of the estrous periods compared to the 6% for the storage of steps in 2h periods using the same threshold. Although the detection percentage increased with the storage of steps in 12h periods (compared to 2h periods), the prediction of ovulation time became less accurate. The interval between onset of pedometer estrus based on 12h periods and time of ovulation is 33.3±5.9h with a range of 16 to 46h, while the interval between onset of pedometer estrus based on 2h periods and ovulation is 29.3±3.9h with a range of 22 to 39h (Roelofs *et al.*, 2005b). This larger variation could be a problem because AI has to be performed 24 to 12h before ovulation (Roelofs, unpublished results, see further), which implies that insemination should be performed 5 to 17h after the onset of increase in activity. When it takes at least 12h before the increase in activity is observed, this leaves almost no time for optimal timed insemination. More frequent reading of the pedometers seems therefore a logical approach to solving this problem.

It has to be noted that the activity measurements in our study were performed at only one farm. Although it is known that basal activity levels differ between cows and farms (Nebel *et al.*, 2000), the question is whether the relative increase in activity at estrus is also different between farms. In other words, the question is whether the high detection rate and the high correlation between increase in activity and ovulation time found in our experiment (Roelofs *et al.*, 2005b) will be similar for other farms. López-Gatius *et al.* (2005) studied the activity of two herds kept on different farms but under the same housing, management and milking conditions. They found no difference in increase in activity at estrus between the farms. However, activity levels were affected by parity, milk production and season. Nothing was mentioned about activity levels between farms outside estrous periods. Another study found similar increases in activity at estrus when cows were kept in a covered straw yard compared to cows kept in cubicles (Schofield *et al.*, 1991). These findings suggest that prediction of ovulation time by measurement of activity, as found in our study, should be applicable on other farms as long as the threshold for an increase in activity is based on the individual activity pattern of the cow.

In conclusion, the monitoring of the number of steps measured by pedometers meets all the prerequisites for ovulation time prediction: a good relationship exists between increase in activity and time of ovulation, the labor input for measurement of activity is low, the measurements can be automated and a high proportion of the animals show an increase in their activity before ovulation. Therefore, prediction of ovulation time based on an increase in the number of steps seems useable in dairy practice. However, the question as to whether this is true for all farms needs to be investigated.

### Vaginal mucus conductivity/resistance

Several studies have shown that vaginal mucus conductivity/resistance changes during estrus (reviewed by Rorie *et al.*, 2002). Electrical resistance is lowest during estrus because of changes in cell density, fluid volume and electrolyte content of the bovine vulva and vagina (Ezov *et al.*, 1990). Leidl and Stolla (1976) found evidence that the resistance of vaginal mucus is affected by estrogen. Several other studies have shown that the lowest resistance coincides with the LH surge, and the interval between lowest resistance and time of ovulation was between 32 and 24h (Schams and Butz, 1972; Leidl and Stolla, 1976; Schams *et al.*, 1977; Aboul Ela *et al.*, 1983). The resistance dropped from an average of 48 Ohm between estrous periods to an average of 30 Ohm during estrus (Schams and Butz, 1972). Various studies reported variation, both among and within cows, in conductivity measurements, resulting in undesirably high rates of false positives and false negatives (Elving *et al.*, 1983; Lehrer *et al.*, 1995). The labor-intensive nature of this approach and the hygiene risks it involves also greatly limit its practicality. Therefore, measurements of vaginal mucus conductivity/resistance do not seem useful for the prediction of time of ovulation.

### Body temperature

During the estrous cycle significant changes in body temperature occur (reviewed by Firk *et al.*, 2002). During estrus the temperature rises about 0.3°C (Nieuwenhuizen *et al.*, 1979; Mosher *et al.*, 1990). This may be caused by the higher activity during estrus, but the mechanism behind the rise is not clear. Although this temperature rise has been related to estrus characteristics, very little is known about the relationship of this parameter to time of ovulation. In heifers a rather variable interval between the peak in vaginal temperature and ovulation of 21.1 6.1h has been found (range: 16-33h; Mosher *et al.*, 1990). From this experiment, Mosher *et al.* (1990) concluded that since ovulation occurs within a consistent interval from the onset of a temperature spike, the onset of a temperature spike might be as good a predictor of ovulation as the LH peak. Rajamahendran *et al.* (1989) found that the peak in vaginal temperature occurred in heifers 22.0 3.5h and in cows 27.0 3.5h before ovulation. They found a high correlation between vaginal and rectal temperature, but the rise in temperature before ovulation was only significant in the vaginal temperature measurements. Their study demonstrated that the rise in vaginal temperature was a reliable measure of the time of ovulation and the time of the LH surge. However, several authors dispute the usefulness of body temperature measurements as an estrus detection method and therefore as a predictor of time of ovulation (Boyd *et al.*, 1969; Lewis and Newman, 1984; Firk *et al.*, 2002). The measurements could eventually be automated if one were to measure the milk temperature as a derivative of the body temperature. However, as already mentioned, the temperature rise during estrus is small, being only 0.3°C. Therefore, body temperature is not a specific indicator of the incidence of estrus, because a rise in temperature can also be caused by inflammatory reactions and variation in environmental temperature. In conclusion, temperature measurements do not seem to be a practical tool for predicting time of ovulation.

## INSEMINATION-OVULATION INTERVAL

As previously mentioned, the optimal time for artificial insemination depends on the one hand on the lifespan of spermatozoa once released and on the other hand on the viable lifespan of the ovum after ovulation. The lifespan of spermatozoa in the cow's oviduct is reported to be 24 to 48h for fresh and 12 to 24h for frozen-thawed semen. The viable lifespan of the ovum is suggested to be around 6 to 12h (Gordon, 2003).

In pigs, the optimal time for insemination lies between 0 and 24h before ovulation (reviewed by Kemp and Soede, 1997). Not much is known concerning fertilization rates of cows at various insemination-ovulation intervals. Studies from the 1940's showed that conception rates in cattle differed when cows were bred during different stages of estrus (Trimberger and Davis, 1943; Barrett and Casida, 1946). The highest conception rates (varying from 73% to 86%) were obtained when inseminations were performed 24 to 6h before ovulation (Trimberger and Davis, 1943). As conception rate in their study was assessed by pregnancy diagnosis, it is not known whether the low conception rates were a result of fertilization failure, (early) embryonic death or both. Surprisingly, since then not much new information has appeared concerning the optimal time of insemination relative to ovulation in dairy cattle (reviewed by Hunter, 1994).

On the basis of research that was performed in the 1970's, it was assumed that in cattle, insemination should take place between 12 and 18h before ovulation to get good fertilization results (Hunter, 1994). However, Hunter assumed that ovulation takes place at a rather stable 12h after the end of estrus. From recent studies, it is known that the moment of ovulation relative to estrus is far more variable. In recent years, research has been done to study the effects of different insemination times relative to estrus characteristics on conception rate (Maatje *et al.*, 1997; Dransfield *et al.*, 1998) but time of ovulation was not assessed in these studies. These studies show variable results. Maatje *et al.* (1997) found conception rates (at 42 to 49 days after insemination) of around 80% for inseminations 0 to 24h after the onset of increase in walking activity and of 17.6% for inseminations more than 24h after the onset of increase in walking activity. Dransfield *et al.* (1998) found highest conception rates of 51% when inseminations were performed 4 to 12h after the first display of standing heat (as detected by mount detectors). In their studies, it is not known whether suboptimal insemination times resulted in higher fertilization failure or higher (early) embryonic death. This distinction was made in a study by Dalton *et al.* (2001), who assessed fertilization rate and embryo quality seven days after insemination when cows were inseminated 0, 12 or 24h after the onset of standing heat (using mount detectors). Inseminations performed 24h after the onset of standing heat resulted in the highest fertilization rate, whereas inseminations performed at the onset of standing heat resulted in the highest percentage of good quality embryos. They concluded that inseminations performed 12h after the onset of estrus provided a compromise between a potentially lower fertilization rate (insemination at the onset of standing heat) and lowered embryo quality (insemination at 24h after the onset of standing heat). In our experiment (unpublished data) cows were inseminated once between 36h before and 12h after ovulation. Seven days after ovulation the uterine horn was flushed and fertilization (yes or no) and quality of the embryo (good, fair, poor and degenerated) were assessed. In total, 122 embryos/ova were recovered. Fertilization rates were high when insemination was performed between 4 and 36h before ovulation. However, the quality of the embryos was influenced by the time of insemination. The highest number of good quality embryos was found when inseminations were performed between 12 and 24h before ovulation. The percentage of good quality embryos was 41% with inseminations performed 36 to 24h before ovulation, 67.7% with inseminations performed 24 to 12h before ovulation, 41.4% with inseminations 12 to 0h before ovulation and only 6.3% with inseminations performed from 0 to 12h after ovulation. The sex of the embryo was not influenced by time of insemination relative to ovulation (Roelofs *et al.*, 2005a)

## INSEMINATION BASED ON PREDICTION OF OVULATION TIME

Several studies have examined the optimal time for insemination relative to the onset of estrus (Table 2). Our results (unpublished data) have shown that the optimal time to inseminate is 24 to 12h before ovulation (based on the highest percentage of good quality embryos in that period). Combining this optimal interval with the interval between onset of behavioral estrus and ovulation time suggests that an insemination should be performed 3 to 15h after the observation of onset of behavioral estrus (assessing *all behavioral signs* for 30 min, three times daily). Using the same observation frequency, insemination should be performed 3 to 15h after the first observed *mount* or 0 to 12h after first observed *standing heat*. This is in the same range as suggested by other authors (Table 2). Inseminations based on *activity* should be performed 5 to 17h after the increase in activity. This optimal insemination interval is the same as reported by Maatje *et al.* (1997) (Table 2).

A survey done on 170 farms in The Netherlands seems to indicate that the average interval between detection and insemination on these farms varied from 3h to 24h. On almost half of the farms, the majority of inseminations were performed more than 15h after detection of estrus (Hensen *et al.*, 1992). Thus, it seems that insemination is performed too late on many farms. It has to be noted that only in 30% of all estrous periods is standing heat observed by three times daily observations (Roelofs, 2005). It is possible that when standing heat is observed, the timing of insemination is quite accurate, while in other estrous periods with less clear behavioral estrous signs, insemination is often wrongly timed. One reason for suboptimal timing of inseminations in the absence of standing heat could be that a farmer is not sure about the estrous status of a cow. Therefore, the decision to inseminate the cow is delayed, resulting in decreased calving rates because of inseminations performed too close or even after ovulation, especially when the cows are not inseminated by the farmer himself. According to the effects of the insemination time on em-

**Table 2. Overview of optimal insemination time relative to onset of oestrus based on various parameters.**

| Oestrus based on | Optimal insemination interval after onset of oestrus (h) | Conception rate (%) | Reference |
|---|---|---|---|
| Discharge, nervousness, interest in herd mates, mounting, standing heat | 7-12 | 55 | Hall *et al.*, 1959 |
| Walking activity | 6-17 | 83 | Maatje *et al.*, 1997 |
| Standing heat | 4-12 | 51 | Dransfield *et al.*, 1998 |
| Standing heat | 12-18 | 73 | Xu *et al.*, 1998 |
| Standing heat | 12 | - | Dalton *et al.*, 2001 |
| Behavioral, clinical and gynaecological symptoms | 8-18 | 66 | Martinez *et al.*, 2004 |
| Our results | | | |
| All behavioral signs | 3-15 | 62[1] | Roelofs *et al.*, 2005c |
| Mounting | 3-15 | 63[1] | Roelofs *et al.*, 2005c |
| Standing heat | 0-12 | 63[1] | Roelofs *et al.*, 2005c |
| Walking activity | 5-17 | 65[1] | Roelofs *et al.*, 2005b |

[1]Estimated calving rate when the survival rate of good, fair and poor quality embryos is 95, 68 and 53% respectively.

**Table 3. The percentage of inseminations for various insemination-ovulation intervals (IOI, 0=time of ovulation) when cows are inseminated based on first observed mount, first observed standing heat, observation of onset of behavioural oestrus (including all behavioural oestrous signs) or increase in activity.**

| IOI | Mounting[1] | Standing heat[2] | All signs[3] | Activity[4] |
|---|---|---|---|---|
| 36<AI=24 | 11 | 7 | 14 | 10 |
| 24<AI=12 | 78 | 81 | 72 | 84 |
| 12<AI=0 | 11 | 11 | 14 | 6 |
| 0<AI=-12 | 0 | 0 | 0 | 0 |

[1]insemination 9h after first observed mount; [2]insemination 6h after first observed standing heat; [3]insemination 9h after first observed behavioural sign; [4]insemination 11h after first increase of activity.

bryo quality, it seems better to inseminate (too) early than (too) late when the time of ovulation is not known.

In our experiment, the embryos were flushed seven days after ovulation, so it is impossible to know which embryos would have survived and would have resulted in the birth of a healthy calf. Average pregnancy rates of various studies using embryo transfer were 60, 45 and 30% for good, fair and poor quality embryos, respectively (Schneider *et al.*, 1980; Wright, 1981; Hoogenkamp, 1984; Hasler, 1998). It seems likely that the survival rate of the embryos is higher for non-transferred embryos compared to transferred embryos. Therefore survival rates of good, fair and poor quality embryos of none-transferred embryos are assumed to be 95, 68 and 53%, respectively. Based on the occurrence of good, fair and poor quality embryos in the different I-O intervals, these assumed embryo survival rates result in the expected calving rates of 55, 67, 43 and 19% for inseminations performed 36 to 24, 24 to 12, 12 to 0h before and 0 to 12h after ovulation, respectively. Even when the cows are inseminated in the optimal period after the first display of mounting, standing heat, behavioral estrus or increase in activity, still not all inseminations will be performed in the optimal interval of 24 to 12h before ovulation, because of the variation in the interval between the first display of these parameters and time of ovulation of individual cows (Table 3). Combining the number of cows detected in estrus with the insemination times shown in Table 3 and with the expected calving rates based on embryo quality it would result in the fact that 38% (for first mount), 19% (for first standing heat), 55% (for first display of any behavioral estrous sign) and 54% (for increase in activity) of the cows in estrus ultimately calf (Figure 1).

In conclusion, the expected number of calves born per 100 cows in estrus are similar when a cow is inseminated

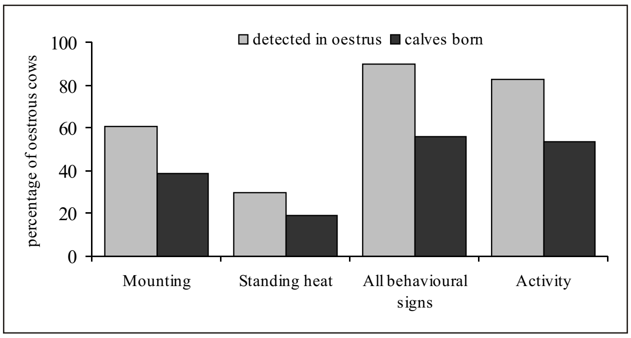


**Figure 1. Percentage of cows detected in oestrus and expected percentage of calves born of cows in oestrus, using different oestrus detection strategies. Oestrus detection strategies consist of visual observations three times daily of only mounting behaviour, only standing heat or all behavioural oestrous signs or is based on an increase in activity (measured by pedometers). Inseminations are per- formed on average 9h after the first observed mount, 6h after the first observed standing heat, 9h after the onset of behavioural oestrus (all signs) or 11h after the increase in activity. Giving the known variation in ovulation time, a known expected percentage of good, fair and poor quality embryos per I-O interval and an expected chance of survival of these embryos (see text), an expected calving rate is calculated for these strategies.**

either 3 to 15h after the onset of behavioral estrus or 5 to 17h after the first increase in activity. However, insemination based on an increase in activity seems the best strategy, because it requires low labor input and is easily implemented in dairy practice.

## PRACTICAL IMPLICATIONS

It was found that pedometer readings best predict time of ovulation: ovulation takes place 22 to 39 h after the first increase in activity. However, pedometer readings may also give false positive alerts. Therefore, to optimize insemination time (and thus calving rates), in practice the strategy should be to combine activity measurements with visual observations. When the pedometer readings show an increase in activity for a certain cow, that cow should be observed for the display of estrous behavior (including sniffing and chin resting). When she shows any of these behavioral signs, insemination should be performed. Since it was found that the optimal insemination time is 24 to 12h before ovulation, the cow should be inseminated between 3 and 15h after the first increase in activity. On the other hand, when a cow shows behavioral estrus signs, pedometer readings should be evaluated to decide on the right time for insemination.

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