

Reproductive Management with Limited Hormonal Intervention

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■ Take Home Messages

- ▶ Detection of estrus is a key element in successful AI programs in dairy herds.
- ▶ Although timed AI programs are popular and pregnancy outcomes are generally good, automated activity monitors available to predict estrus have proven effective to increase heat-detection and AI-service rates.
- ▶ Automated activity monitors seem to closely identify onset of estrus compared with standing-to-be-mounted activity identified by HeatWatch. Time of ovulation relative to onset of estrus or activity is similar between methods.
- ▶ Within-herd comparisons described herein provide evidence that pregnancy rates may be increased (faster rate of achieving more pregnancies per unit of time) with activity monitors compared with applying timed AI programs.
- ▶ Rate of pregnancy establishment is greater when applying activity systems because inter-insemination intervals are reduced so AI service rate is greater.
- ▶ Surveys of activity system users show large rates (> 90%) of acceptance and satisfaction.

■ Introduction

As cows approach estrus, a profound increase in physical activity occurs including overt mounting and standing to be mounted. In addition to increased physical activity, a number of significant physiological changes occur in cows during the peri-estrous period (Lewis and Newman, 1984; Roelofs et al., 2010) enabling detection of estrual behavior and other correlated traits. Some of these changes include vaginal cytology and pH, electrical resistance of

vaginal mucus and genital tissues, body temperature, pulse and heart rates, blood flow, pheromones or odors, blood metabolites and hormones, milk yield, and feed intake.

Detection of estrus is a key element in a successful artificial insemination (AI) program. It can be the sole source of identifying cows in estrus or may be combined with fixed-time AI programs. Accurate prediction of ovulation is the goal of an estrus-detection program. A 12% improvement in AI-service rate was required for a 100% estrus-detection-based AI program to have the same economic value as a superior 100% timed AI program (Giordano et al., 2011). Furthermore, adding detection of estrus to a 100% timed AI program was only beneficial for a timed AI program with the lowest pregnancy rate. Thus, reducing costs associated with timed AI programs by improving efficiency and accuracy of detected estrus (increasing AI-service rate) should reduce the cost per pregnancy in most herds because inter-insemination intervals are reduced from 40 to 50 days to as little as 20 to 24 days when non-pregnant cows are identified at their first post-AI estrus.

Proper timing of AI has its source in the pioneering studies of George Trimberger. Cows were visually observed for estrus and then ovaries were palpated per rectum at frequent intervals to determine when a large follicle disappeared from the ovary. Based on his studies, Trimberger found that conception risk was maximal when cows were inseminated between 6 and 24 hours before ovulation. As a result, the AM-PM rule for AI was formulated—first detected estrus in the AM is followed by AI that PM or first detection of estrus in the PM is followed by AI the next AM. The fundamental basis for this rule requires a minimum of twice-daily visual observation of cows. Because appropriate timing of semen placement in the reproductive tract is critical to fertilization success, ovulation prediction rather than estrus detection is what is largely desired.

■ Detection of Estrus

For a variety of reasons, estrus-detection rates in North American dairy herds usually approximate 50% when based solely on visual observations. Poor estrus-detection rates have led to successful and profitable adoption of systematic timed AI programs (see recommended timed AI protocols at drcouncil.org). Despite the effectiveness of timed AI programs, many producers desire to breed cows based on estrus, and some voice their dislike of frequent hormone injections required with timed AI protocols and the public perception thereof.

Cows are monitored traditionally for visual signs of estrus such as “standing to be mounted” by a herd mate. In order to be effective in identifying when to inseminate, one must visually observe cows for estrual behavior at least twice daily. Common estrus-detection aids used on dairy operations assessed by a

2007 U.S. National Animal Health Monitoring System survey were: (1) visual observation (93%); (2) tail chalk or tail paint (35%); (3) bulls (40%); (4) heat mount patches (14%); and (5) other 14% (e.g., HeatWatch, pedometers).

To reduce labor and improve efficiencies associated with detection of estrus, methods other than visual observation have been developed. Although technologically simple, such products as tail paint or tail chalk applied to the tail head is one of the most common forms of estrus-detection aids. Most large dairy operations use tail chalk or paint removal associated with once daily observation of cows, which is usually superior to visual observations, but still allow room for improvement with AI-service rates typically in the mid-60% range. Validation of accurate rubs by concentrations of progesterone in milk or subsequent pregnancy after insemination showed that accuracy of chalk or paint as estrus-detection devices varied widely from 33 to 90%. It seems from many studies that 5 to 30% of females inseminated are not really in estrus (Roelofs et al., 2010).

■ Automation of Estrus Detection

Automation of estrus-detection efforts in dairy cows is well documented (Firk et al., 2002). Technologies developed to take advantage of other physiological correlates of estrus include pedometry or activity monitors, pressure-sensitive, rump-mounted radiotelemetric devices, temperature sensors, and milking inline chemical sensors. Electronic mounting sensors are pressure-sensitive devices that are applied to the rump of the cow and activated by a mounting herd mate. Accuracy and efficiency of these systems are quite good because they are associated with specific sexual behavior and are functional 24 hours per day (Roelofs et al., 2010). The downside to these sensor systems is the labor associated with applying the sensor patches and the maintenance to keep sensor patches affixed to cows until pregnancy occurs.

A fully automated system of progesterone tests in milk using dry-stick technology has been marketed in Denmark since 2009 and is becoming available elsewhere. Since 2010 the Herd Navigator system was combined with a DeLaval milking robot or parlor and offers monitoring of reproduction by frequent measurements in milk of progesterone. For purposes of estrous cycle monitoring, the system collects several milliliters of milk during the milking process, measures progesterone, and provides a progesterone concentration profile. The actual test points are used to predict a smoothed curve prediction line. Milk progesterone concentrations follow the same pattern as blood progesterone, but the concentrations are greater, often by a factor of 2.

The software system adjusts the frequency of milk progesterone tests depending on the days in milk and the stage of the cycle, to an average of six

or seven progesterone tests per estrous cycle. From the smoothed progesterone curve, the algorithm developed in the system classifies the cows into categories: (1) postpartum anestrus; (2) estrous cycling; and (3) potentially pregnant. For estrous-cycling cows an alert is triggered by the software as soon as the progesterone concentration drops below 4 ng/mL. In case of an estrus alert, the algorithm also provides the probability of success of a prospective AI (between 0 and 100%) based on the duration of the previous luteal phase and the kinetics of the decrease in progesterone concentration. An average estrus-detection rate of 95% was later confirmed in commercial herds using this system in Denmark.

■ Activity Monitors

At least nine milking machine and AI companies are marketing activity monitor technology for use in dairy operations and will likely have a larger impact in dairy operations seeking low-cost methods of identifying cows in estrus. The latest version of electronic estrus-detection aids that appeared in the early part of this decade was the neck-mounted activity tags containing a microprocessor and a 2- or 3-dimensional activity sensor. Monitoring activity has formed the basis for many pedometer or neck-mounted monitor systems marketed to the dairy industry because increased activity (e.g., motion, movement, walking) is a correlated trait associated with estrus and it increases up to 400% in 93% of estrous periods (Kiddy, 1977). One challenge limiting any system is the lesser activity associated with estrus of cows maintained in tie stalls compared with free stalls or for cows in total confinement on concrete compared with dirt lots.

Increased walking activity that is associated with estrus (Roelofs et al., 2010) led to the development of pedometry as a means of detecting estrus as early as the 1970s. Pedometers affixed to the leg quantified cow movement or counted the number of steps taken by the cow. Increase in physical activity of the cow provided 70 to 80% accuracy of estrus detection. Cows housed in free stalls were approximately 2.75 times more active during estrus than when not in estrus. Most important to the success of this technology is the relative small amount of within-cow variation in day to day activity when cows are not in estrus. Therefore, activity monitors can be excellent predictors of behaviors associated with estrus (Roelofs et al., 2010).

Three types of activity monitors are currently available: (1) pedometers fixed on one leg that record the number of steps made by the cow per unit of time; (2) activity monitors attached to neck collars that record movement; and (3) activity monitors attached to the leg that comprehensively assess activity of dairy cows by measuring the number of steps and quantifying lying and standing behaviors (Saint-Dizier and Chastant-Maillard, 2012).

The activity monitors periodically download wirelessly to a base station or when the activity monitor is interrogated in the milking parlor, common feeding stations, or other high-traffic areas. The software operating on a personal computer downloads the activity data from the personal computer interface to the computer software for analysis. The activity analysis program algorithm examines within-cow activity to assist in detecting the amount of current activity as a function of the cow's most recent activity baseline. Once the current activity meets or exceeds a set threshold, the cow's identification is flagged by the software for further inspection and possible insemination. Results for detection rate, error rate, and specificity depend on the threshold value for investigations with known measures of estrus (Firk et al., 2002). Results are greatly influenced by the number of cows in estrus at one time because individual cow activity increases as more herd mates are simultaneously in estrus.

The newest generation of activity monitors employs an accelerometer device. Accelerometers are small (4 x 4 mm), reliable, and durable. Accelerometers now used in activity monitors were developed first for the military, aerospace, and automotive industries. They have the capacity to detect motion in all three spatial planes. Now they are more popularly used in industrial, medical, and consumer devices in any number of applications. The accelerometer allows accurate measurement of cow movement. The activity tag monitors specific estrus-related movement and its intensity resulting in estrus-detection accuracies up to 90% (Roelofs et al., 2010). By 2010, the bestselling activity system in the world with approximately 1 million estrus-detection tags sold, demonstrated that dairy farmers were willing to invest in technologies that provide a real solution to detection of estrus. A survey of 219 dairy farmers who had employed one automated activity system (Heatime, SCR Engineers Ltd, Netanya, Israel) revealed their nearly unanimous satisfaction (94.1%). More than 93% agreed that estrus-detection rates had increased; 92.3% believed reproduction had become more easily managed; and 94.5% said they would install the system again (Michaelis et al., 2013).

These activity systems are effective management tools in the AI program because their use will increase estrus-detection rates. Increasing estrus-detection rates result in increased AI service rates and more potential pregnancies. At least four patents have been issued describing some type of transponder system that is capable of detecting movement or motion that includes the ability to be interrogated in the parlor or send signals via wireless radiotelemetry.

Two studies documented accuracy of activity monitors to identify estrus. In one study (Aungier et. al., 2012), physical activity of 89 spring calving cows on pasture were monitored by activity monitors (SCR Engineers Ltd., Netanya, Israel). Based on twice weekly samples of progesterone, the activity monitors identified 72% of the preovulatory follicular phases (69 conceptions

resulted from 145 AI). One-third of the activity clusters were associated with elevated progesterone (i.e., false positives).

A second study (Valenza et al., 2012) examined dairy cows housed in free stalls that were treated with GnRH and prostaglandin $F_{2\alpha}$ (PGF) 7 days later. Cows enrolled in the study must have had a corpus luteum and a follicle > 10 mm in diameter, and subsequent luteolysis by 48 hours after PGF. Cows were fitted with activity monitors (SCR Engineers Ltd., Netanya, Israel) and heatmount tail patches to detect estrus. Activity monitors identified only 71% of the cows in estrus during 7 days after PGF injection; of which 95% ovulated. Of the 29% of cows not detected by the activity monitor, 35% ovulated.

■ Ovulation Prediction with Activity Monitors

The relationships between increased activity, time of ovulation, and fertility have been investigated with the help of activity monitors (Lopez-Gatius et al., 2005; Roelofs et al., 2005; Hockey et al., 2010). In general, ovulation takes place 29 to 33 hours after the onset of increased activity and 17 to 19 hours after the end of increased activity in lactating Holstein cows (Roelofs et al., 2005; Hockey et al., 2010).

One study estimated timing of ovulation by applying concurrently to each cow a pressure-sensitive, rump-mounted, radiotelemetric device and a foreleg attached pedometer (Gyuhō, Comtec, Miyazaki, Japan), which measured steps, to determine onset of estrus (Yoshioka et al., 2010). The pedometer was a real time radiotelemetric device consisting of a miniaturized radiowave transmitter linked to a pedometer enclosed in a hard plastic case. Ovaries in 20 cows were scanned transrectally every 2.5 hours beginning 20 hours after first standing estrus until the ovulatory follicle disappeared. Their findings are summarized (Table 1; Study 1).

Table 1. Time of ovulation relative to estrus activity determined by automated activity monitors or by standing to be mounted.

Item	Study 1 ¹	Study 2 ¹
Cows, no.	20	61
Duration of increased activity, hours	15.8 ± 0.9 ³	11.4 ± 0.8
Duration of standing activity, hours	9.0 ± 1.3	7.1 ± 0.9
Standing events, no.	36.0 ± 7.6	5.9 ± 1.2
Ovulation after activity start, hours	30.2 ± 0.6	24.6 ± 0.7
Ovulation after first standing event, hours	29.0 ± 0.6	26.4 ± 0.7
Ovulation after activity end, hours	15.3 ± 0.9	13.2 ± 0.9

¹ Yoshioka et al. 2010 (Foreleg pedometer).

² Stevenson et al. Unpublished. (Collar-mounted activity monitor).

³ Means ± SE.

We recently conducted a study with the objective to determine the timing of ovulation in lactating dairy cows enrolled in an AI program exposed to Select Detect (SD; Select Detect™ advances estrus detection system; $n = 132$) and Heatwatch (HW; $n = 61$) heat-detection systems. Combining technologies allowed comparisons of the onset of estrus (first mount received per HW) with the timing of increased activity (first achieved threshold) as determined by an accelerometer activity system. When lactating dairy cows (60% primiparous and 40% multiparous cows) calved, they were fitted with SD accelerometer collars and HW transmitters to facilitate simultaneous collection of estrus and ovulation events. Cows enrolled in the study were setup for first AI beginning at 50 days in milk by receiving either 25 mg PGF or 100 μ g GnRH i.m. that preceded PGF injection by 7 days to induce estrus. Cows identified in estrus either before or after first AI were studied.

Once a test female was detected in estrus (at least 1 standing event), the hourly activity count reached threshold (based on SD software), or both, transrectal ovarian scans were initiated beginning 14.5 ± 0.5 hours later and continued every 3 hours until the ovulatory follicle(s) disappeared or until 36 hours. At the initial scan, all follicular structures were mapped and sized with electronic calipers. The largest two follicles were monitored until either or both disappeared. A blood sample was collected at the first ovarian scan to measure concentrations of progesterone (< 1 ng/mL, which was indicative of true estrus).

Of 132 cows enrolled in the ovulation study, 117 (89%) had concentrations of progesterone < 1 ng/mL (mean = 0.11 ng/mL) and 15 (11%) had elevated concentrations of progesterone (mean = 5.35 ng/mL) at the first ovarian scan. Eleven of the 15 high progesterone cows were identified falsely by the activity monitors (false positives). Of these 11 cows, three occurrences of 2 cows were detected together and one occurrence of 5 cows was detected together within 1 to 5 hours of each other. Of 117 low progesterone cows, 91% ovulated (85.5% ovulated < 36 hours after detection by the activity monitor). Of the total data collected with the activity monitor, 59 of 106 ovulating cows also had HW-transmitter data.

Figure 1 illustrates the relative proportion of cows that ovulated at various intervals after detection by either standing to be mounted or by reaching activity threshold. Average intervals to ovulation actually differed ($P < 0.05$) by only 1.8 hours (Table 1; Study 2). Mean interval to ovulation after the end of estrus or end of increased activity was greater ($P < 0.001$) for HW than for SD, respectively, whereas duration of activity was greater ($P < 0.001$) for SD compared with duration of estrus for HW (Table 1; Study 2).

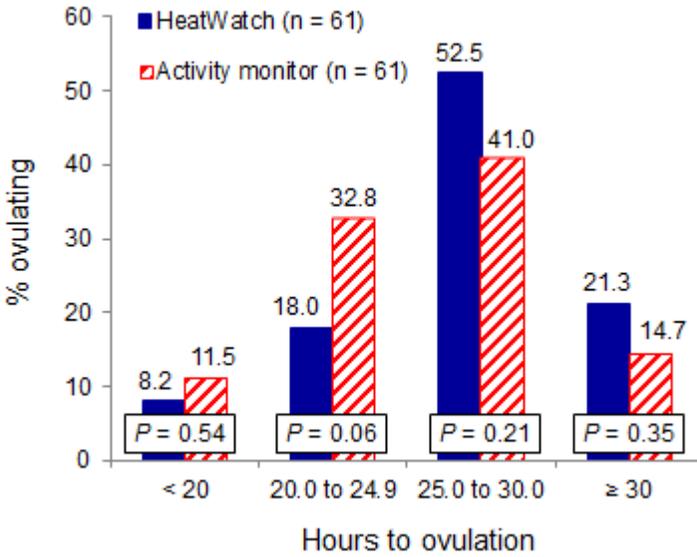


Figure 1. Hours to ovulation after first standing-to-be-mounted or achieved threshold activity in lactating dairy cows fitted with HeatWatch pressure-sensitive, rump-mounted estrus-detection devices and automated activity accelerometers.

In the two studies cited, predicted time of ovulation was in reasonable agreement with the gold standard study (Walker et al., 1996) where ovulation occurred at 27.6 ± 5.4 hours (mean \pm SD) after the onset of estrus (based on HeatWatch). From the literature cited and the relationship between ovulation and onset of estrus and ovulation and onset of achieved threshold activity in our study (Figure 2), it seems likely that time of ovulation is well correlated with the onset of standing estrus and increased activity in those studies where simultaneous measures were made.

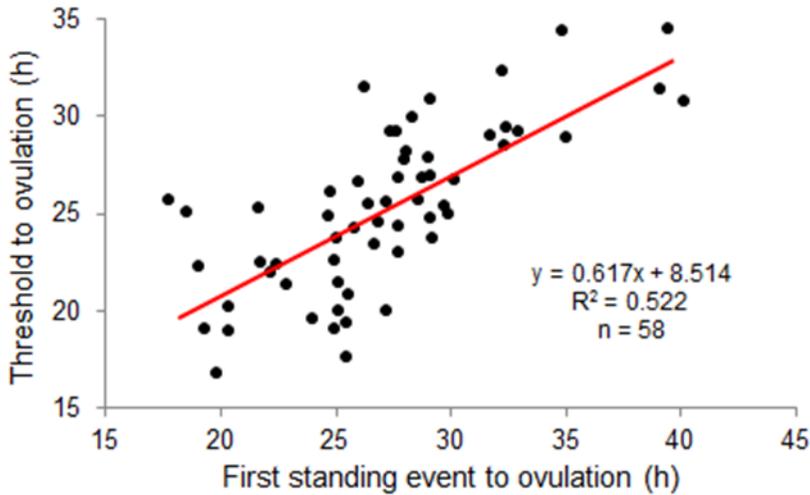


Figure 2. Linear relationships between time of first standing event and increased activity relative to time of ovulation.

■ Pregnancy Risk After Use of Activity Monitors

Insemination timing to maximize pregnancy outcome was predicted from SD systems installed in 19 free stall housed dairy herds located in 8 US states (Figure 3). Of greatest interest was the different conception risk observed between primiparous and multiparous cows (Figure 3). Among primiparous cows, a curvilinear relationship was apparent with optimum conception occurring at AI intervals of 13 to 16 hours after cows had achieved threshold activity and trended for lesser conception risk for both earlier and later AI intervals. Among multiparous cows, conception risk at intervals ≤ 12 hours was different than those > 16 hours with the 13 to 16 hour interval being intermediate. We interpreted these results to indicate that optimal conception risks were obtained when interval to AI was approximately 12 hours after detected activity threshold with shorter intervals less compromising to conception risk than greater intervals.

The objective of another study was to compare reproductive performance with management programs based on either a timed AI program or the SD activity system under field conditions in an 832-cow dairy located in central Pennsylvania. Cows were enrolled randomly at calving to either a timed AI program or AI according to the activity system for the first 3 services. Cows assigned for timed AI ($n = 413$) were enrolled in Presynch 14 x 11 Ovsynch 56, and received PG at 36 and 50 days in milk, followed 11 days later (day 61) with GnRH. On day 68 (7 days later) cows received PG followed by GnRH 56 hours later and timed AI 8 to 16 hours after GnRH. Cows found open at

pregnancy diagnosis were given PG at open diagnosis followed by GnRH 56 hours later and timed AI 8 to 16 hours after GnRH for up to 2 more services. If determined to be open after the third service, cows were fitted with an activity monitor collar.

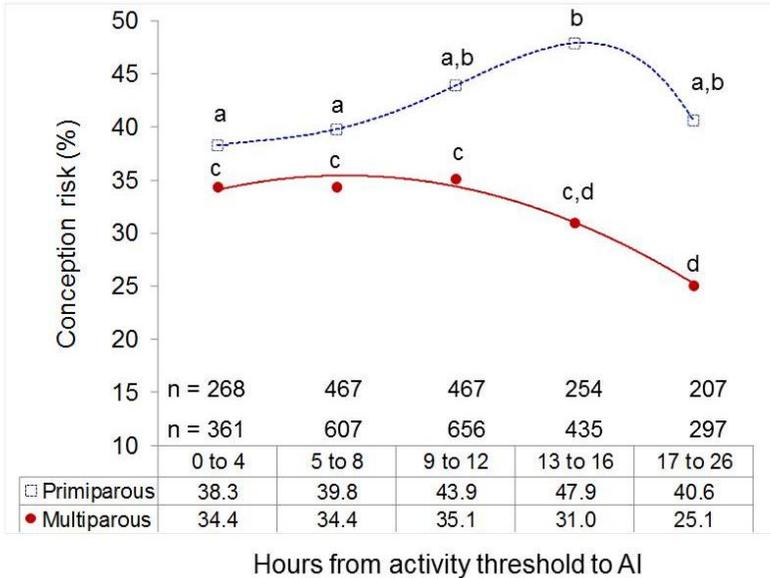


Figure 3. Conception risk of lactating cows in 19 herds in which inseminations were administered randomly at various intervals after achieved activity threshold by automated activity monitors. Within parity, data points with different letters differ ($P < 0.05$).

Cows assigned to the activity group (n = 394) were fitted with an accelerometer activity monitor (SD) the first Monday after 40 days in milk, and if not inseminated by 54 days in milk, PGF was administered. If not inseminated by 75 days in milk, a CIDR insert + Ovsynch-56 protocol was administered with timed AI between 85 and 91 days in milk. If cows in the activity group were not pregnant after third service, the resynchronization protocol used for timed AI cows was administered. Cows were enrolled in the study during an entire year.

Various reproductive characteristics are illustrated for this study (Table 3). Days to first AI service was 9 days less for the activity group, which included the 14% of the activity group cows receiving a timed AI for first service between 85 and 91 days in milk. Median days to pregnancy (days in milk in which 50% of the cows were pregnant) were 80 and 90 for the activity group and timed AI group, respectively (Figure 4). Mean days open were 88 ± 2 and 112 ± 3 for the activity and timed AI groups, respectively. The annual 21-day

pregnancy rate was 25 and 21% for the activity group and timed AI groups, respectively. The results demonstrate the activity system yielded a reproductive performance comparable with or superior to results using a timed AI-based program under field conditions.

Table 3. Reproductive characteristics of lactating cows submitted for insemination based on either timed artificial insemination or activity monitors in one herd

Item	System	
	Timed AI	Activity monitor
Cows, n	413	394
Days at first AI ¹		
Primiparous	76.1± 0.6	64.1± 0.6*
Multiparous	74.3± 0.5	67.6± 0.5*
Conception risk at first AI ² , %		
Primiparous	38.6	36.4
Multiparous	41.0	24.7*
Overall conception risk ³ , %	44	35
Estrus-detection rate ³ , %	42	74
Pregnant at 150 DIM ³ , %	52	68

*Differs ($P \leq 0.05$) from timed AI within parity
¹ Interaction ($P < 0.001$) between parity and system.
² Interaction ($P = 0.037$) between parity and system.
³ Statistical analyses could not be performed for these traits.

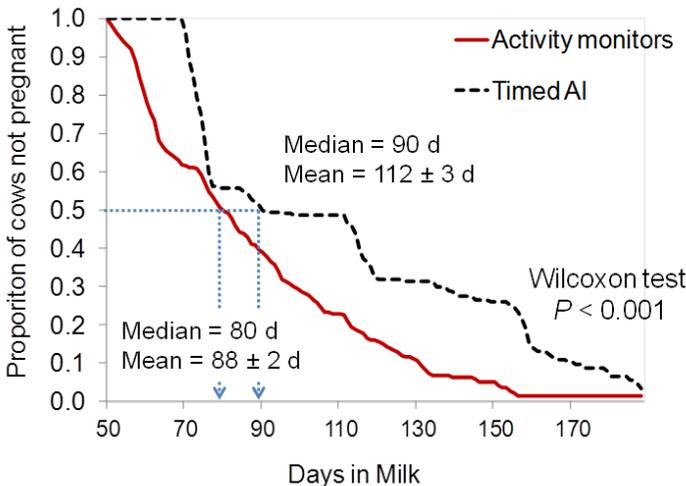


Figure 4. Kaplan-Meier survival curves for proportion of non-pregnant cows according to whether cows were enrolled either a timed AI program through 3 inseminations or inseminated after achieved activity threshold by automated activity monitors.

Another multi-herd study compared reproductive performance with management programs based on automated activity monitoring or a timed AI program (Neves et al, 2012). Holstein cows ($n = 1,429$) in 3 commercial herds were enrolled during 1 year in a randomized controlled experiment. The two systems produced different outcomes among herds to the automated activity monitoring system (Heatime, SCR Engineers Ltd., Netanya, Israel) and timed-AI based program. Median time to first AI service and to pregnancy did not differ in two herds, but were less in the third herd in which 1,985 cow 6-month periods were examined. The results were confounded by the fact that 19 to 32% of all inseminations in both treatments were made after visual estrus detection (not by according to the timed AI or activity system). Considering only 924 cow 6-month periods in which inseminations were made only according to the assigned management program, time to pregnancy was reduced in two of the three herds when cows were fitted with activity monitoring compared with using timed AI. Furthermore, individual conception risk at each AI did not differ between programs (31% for activity and 30% for timed AI).

A collection of non-experimental before-and-after results from 5 dairy farms that employed activity monitors illustrates their added value. Regardless of 21-day pregnancy rates that ranged from 13 to 23% before employing the activity monitors, 21-day pregnancy rates increased significantly to range from 18 to 26% after installation of the activity monitors. This increase in 21-day pregnancy rate occurred while estrus-detection rates ranging from 51 to 65% before installation increased to 56 to 75% after installation. All other factors increased (milk per day, first and overall conception risk), whereas days to first service decreased in all but 1 herd and days in milk decreased in all herds.

Another non-experimental before-and-after testimony reveals a similar story. The Benthem Brothers Dairy of Michigan installed an activity system in June 2010. They immediately ceased using timed AI, but continued to inject PG beginning 3 days before the end of the voluntary waiting period and continued every 14 days until cows were inseminated for their first AI. Cows not responding to PG by 100 days in milk were checked by the veterinary practitioner to determine normality before applying progesterone (CIDR insert) to promote better estrus expression. Currently, only 5% of their cows reach 100 days in milk not yet inseminated. Cows diagnosed not pregnant are given PG if they have a corpus luteum. Otherwise, they are rechecked 1 week later.

■ Conclusions

Automated activity monitors identify the onset of estrus compared with standing-to-be-mounted activity identified by pressure-sensitive, rump-mounted transmitters (HeatWatch). Average time to ovulation relative to onset of estrus or activity threshold differed by less than 2 hours. Within-herd

comparisons provide evidence that conception risk resulting from AI services based on automated activity monitors were similar or less than that achieved by timed AI, but more cows became pregnant earlier in lactation (faster rate of achieving more pregnancies per unit of time). It is inferred from these studies that because of greater estrus-detection rates and earlier establishment of pregnancy, rate of pregnancy establishment was greater when applying activity systems because inter-insemination intervals were reduced so insemination or service rates were increased. Activity monitor results are consistent with similar studies based on observed mounting activity in which optimal conception risks were obtained when interval to AI was approximately 12 hours after detected activity threshold, with shorter intervals less compromising to conception risk than greater intervals. Activity monitors described can effectively predict ovulation and the best time to AI to maximize conception risk.

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