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Evaluation of a Neck Mounted 2-Hourly Activity Meter System for Detecting Cows About to Ovulate in Two Paddock-Based Australian Dairy Herds

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Contents

Two studies were conducted to assess the performance of a commercially available neck-mounted activity meter to detect cows about to ovulate in two paddock-based Holstein-Friesian dairy herds. The activity monitoring system recorded cow activity count in 2-hourly periods. Study I investigated the ability of the system to detect cow ovulatory periods in dairy herds managed in two different Australian environments and breeding systems using five activity alert algorithms. Herd 1 consisted of approximately 130 milking cows calving year-round in a sub-tropical environment and kept in a single dry lot paddock. Herd 2 consisted of approximately 400 milking cows calving seasonally in a temperate climate and fed pasture by rotation through multiple grazing paddocks. Ovulatory periods and non-ovulatory days were identified using milk progesterone monitoring alone or in combination with ovarian ultrasonography; using these 'gold standards' 141 and 135 ovulatory periods were identified in 64 and 135 cows in Herds 1 and 2 respectively. Sensitivity of the activity monitoring system for detecting cow ovulatory periods ranged from 79.4% to 94.1%, specificity from 90.0% to 98.2% and positive predictive value from 35.8% to 75.8%. Study II investigated the ability of the activity meter system to predict the timing of ovulations in paddock-based pasture-fed dairy cattle (Herd 2). The time of ovulation was estimated by repeat trans-rectal ovarian ultrasonography at approximately 0, 12, 24 and 36 h after artificial insemination (AI). The mean times (±SD) from onset and end of increased activity to ovulation were 33.4 ± 12.4 and 17.3 ± 12.8 h respectively (n = 94). Fifty per cent of cows (n = 47) ovulated within the 8-h period between 30 to 38 hs after the onset of increased activity, 76.6% (n = 72) within the 16 h between 24 to 40 h, 85.1% (n = 80) within the 24 h between 18 and 42 h and 90.4% (n = 85) within the 32 h from 19 to 51 h after the onset of increased activity. Results from these studies show that in paddock-based dairy cows in two diverse management systems, this neck-mounted activity meter system detects high proportions of cows that are about to ovulate and provides a useful indication of when ovulation is likely to occur. However, the specificities and positive predictive values using the algorithms assessed may be lower than desirable.

Introduction

Managers of dairy herds using artificial insemination (AI) require practical methods to identify the optimal time to inseminate cows. To achieve acceptable herd reproductive performance, the AI submission method needs to detect a high proportion of cows that are about to ovulate, must have high accuracy (i.e. few false positives and so high specificity and positive predictive value) and must also predict when the detected cows are likely to ovulate (Nebel et al. 2000; Firk et al. 2002; Rorie et al. 2002). High proportions of ovulatory cows must be detected to maximize the number of cows submitted for AI soon after their first day of planned breeding and avoid unnecessary delays in time to pregnancy (Morton 2004). Accurate detection is necessary because excessive numbers of false positive inseminations will result in greater insemination costs, increased inseminations per pregnancy and reduced probability of pregnancy at each insemination. In addition, false positive inseminations in early pregnancy can cause iatrogenic embryonic death or abortion (Macmillan et al. 1977; Weaver et al. 1989; Sturman et al. 2000). An indication of the likely time of ovulation in ovulatory cows is needed so that herd managers can optimize the time of insemination to maximize the proportion of inseminations that result in pregnancy (Trimberger 1948; Dransfield et al. 1998; Van Eerdenburg et al. 2002; Roelofs et al. 2006).

Signs of standing oestrous have been the standard criteria for selecting cows for insemination. However, methods using these signs are becoming less practical with increasing herd sizes. These signs may also be less reliable than previously documented because of reductions in the frequency and duration of standing behaviour in the modern dairy cow (Kerbrat and Disenhaus 2004; Lopez et al. 2004). Thus, alternatives to traditional methods of selecting cows for AI are needed. Cow activity increases substantially in early oestrous (Farris 1954), and time of onset of this increase in activity has recently been related to time of ovulation (Nebel et al. 2000; Firk et al. 2002; Roelofs et al. 2005; Sakaguchi et al. 2007; Yoshioka et al. 2007). A number of completely automated activity meter systems are commercially available that monitor cow activity at frequent intervals. The full automation and relatively low cost of activity meter systems could make them a practical alternative to traditional methods of selecting cows for AI if their performance for detecting cows about to ovulate is adequate.

Ability to detect ovulatory cows for a range of activity meter systems has been reported (Firk et al. 2002). However, activity meter design and activity alert algorithms vary between many studies and this, as well as variation in cow behaviour as a result of differences in herd management and environmental conditions (Orihuela 2000; Stevenson 2001; Lopez-Gatius et al. 2005; Yaniz et al. 2006; Sakaguchi et al. 2007), is likely to affect the performance observed. For example, a study was conducted in a Dutch herd of approximately 70 lactating Holstein-Friesian cows housed in a free stall with slatted floor and cubicles and milked by an automated milking system (Roelofs et al. 2005). This system differs markedly from paddock-based dairy production systems and the ability of activity meter

systems to detect cows about to ovulate may differ between these. As few studies have observed cow activity in paddock-based dairy production systems, further research is needed to determine the performance of different activity meter systems and activity alert algorithms in paddock-based dairy cows.

The objectives of these studies were, for Study I, to determine the sensitivity, specificity and positive predictive value of an activity meter system for detection of ovulatory cows using five different activity alert algorithms in two paddock-based dairy herds with different animal environments and breeding systems, and for Study II, to determine the distribution of intervals from onset and end of increased activity to time of ovulation in a paddock-based pasture-fed dairy herd.

Materials and Methods

Overview of studies

Two prospective observational studies were conducted in two paddock-based Australian dairy herds between the months of May and August 2007 (late autumn to mid-winter). An outline of the methods is shown in Fig. 1. In study I, ovulatory periods and non-ovulatory days were identified by milk progesterone profile analysis alone in Herd 1, and in combination with ovarian ultrasound examinations in Herd 2. The method of identifying ovulatory cows differed for the two herds as a result of practical limitations associated with the daily prevalence of ovulatory cows in each herd. Activity alerts were determined using five different activity algorithms and the sensitivity, specificity and positive predictive value for detection of cow ovulatory periods using each algorithm was determined for each herd. In Study II, the time of ovulation was determined by repeat trans-rectal ovarian ultrasound in cows selected for routine insemination in Herd 2. The intervals from onset and end of increased activity to ovulation along with the durations of increased activity were calculated for these cows. Results of both studies were then reported with the results of Roelofs et al. (2005) to compare the performance observed in an environment and management system very different to either herd in the present studies. These comparisons were made because of a number of other common factors in the studies. The activity meters in both studies were manufactured by the same company (Nedap Agri B.V, Groenlo, The Netherlands), they monitored activity in 2-h time periods, and the algorithms in the Roelofs et al.(2005) study were based on a similar calculation to the DairyPlan® (Westfalia-Surge, Germany) algorithm used in study I. In addition, Roelofs et al. (2005) described the relationship between increases in cow activity and time of ovulation which was also an objective of study II.

The studies were approved by the University of Queensland Animal Ethics Committee (approval numbers SVS/292/07/DA and SVS/152/07/DA).

Herds and cows

Herd 1 was a university teaching and research herd located near Gatton (27°33′11S 152°19′60E), which has a subtropical climate and is approximately 100 km west of Brisbane, Qld, Australia. The herd consisted of approximately 130 milking cows and calved year-round. Approximately 80% of the herd were Holstein-Friesian, with the remainder being a mix of Ayrshire, Jersey and Milking Shorthorn. The cows were fed a mixture of silage, hay, grain and mineral supplements on a covered concrete feed pad located in the centre of a dirt loafing paddock (approximately 150 m by 150 m) within 500 m of the milking parlour. The cows were milked twice daily at approximately 0600 and 1600 h and were returned to this same paddock after each milking. The average 305-day milk yield per cow was approximately 6600 kg.

Herd 2 was a commercial herd located near Colac (38°20′09S 143°31′42E), which has a temperate climate and is approximately 150 km west of Melbourne, Victoria, Australia. The herd consisted of approximately 400 Holstein-Friesian milking cows that calved seasonally, with most cows calving from May to July (late autumn/early winter). The diet was predominantly ryegrass pasture with supplementary grain and minerals fed during milking and hay and silage fed on an open concrete feed pad following milking. The cows were milked twice daily at approximately 0500 and 1700 h and were grazed in a different paddock after each milking. The paddocks were open grazing areas (typical size approximately 300 m by 200 m) without any other

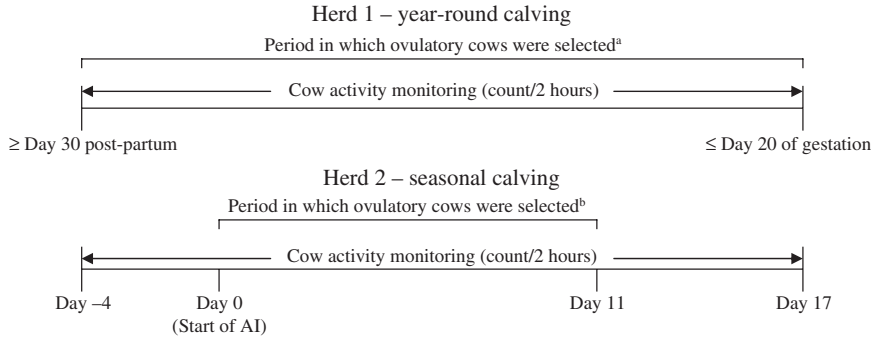

Fig. 1. Outline of methods used to evaluate a neck mounted 2-hourly activity meter system to detect cows about to ovulate and predict time of ovulation in two paddock-based dairy herds; [a]cow ovulatory periods identified by twice weekly milk sampling for P_4; [b]cow ovulatory periods (≥ 30 days post-partum) and time of ovulation identified by milk sampling for P_4 and/or repeat trans-rectal ovarian ultrasound (approximately 0, 12, 24 and 36 h from insemination)

housing structures; they were mostly flat and ranged in distance to the milking parlour from a few hundred meters up to approximately 3 km. The average 305-day milk yield per cow was approximately 8600 kg.

Activity monitoring

Activity was monitored in all lactating cows in both herds. Each cow was fitted with an activity meter (Rescounter II®, Westfalia-Surge, Germany) mounted on a neck collar. The activity meter counted each animal movement that had sufficient force to trigger the ball switch and stored total movement counts for sequential 2-h time periods. The activity data were automatically downloaded at each milking and stored in a database on a central computer. The data storage and retrieval were managed by a software program, DAIRYPLAN® (Westfalia-Surge, Germany); this software allows the user to graph the activity of individual cows and to create alert lists using a pre-determined algorithm with a number of user-definable inputs.

Milk sampling

In Herd 1, milk samples were collected from all lactating cows twice weekly during the evening milking for 10 weeks. The period between each sampling was always 3 or 4 days.

In Herd 2, milk samples were collected from all milking cows on the day prior to the start of AI and a subset of 113 cows were selected to have serial milk samples collected up until the 11th day of AI. These were selected by choosing as many cows that came into the milking parlour that morning that were identified as having calved at least 30 days previously and that were able to be sampled by a single technician without interrupting the routine milking. The cows chosen for serial milk sampling had a further four milk samples collected with intervals of 2–3 days between each sample. In addition to these, all cows selected by the farm manager for insemination during the first 11 days from start of AI had milk samples collected on the day of insemination. For cows inseminated in the first 6 days of AI, a second milk sample was collected 7–10 days after AI, and for those inseminated after the 6th day of AI, the milk sample collected on the day before start of AI (i.e. 7–10 days before insemination) was selected for milk progesterone assay.

Milk samples were collected by hand into 5 ml sterile plastic containers either immediately before or during milking, or at the time of AI. Samples were placed in a freezer (−20°C) following collection and remained frozen until the day of assay.

Milk progesterone assay

Milk samples were measured for progesterone concentration in singlet as whole thawed milk using a commercially available progesterone ELISA kit (Ridgeway Science Ltd., Gloucestershire, UK) described previously (Sauer et al. 1986; Groves et al. 1990). The intra-assay and inter-assay coefficients of variation observed in the present studies for milk progesterone concentrations between 2 and 20 ng/ml were 9.7% and 20.9% respectively.

Milk progesterone concentration cutpoints

It was necessary to identify milk samples from cows with and cows without an active corpus luteum with minimal classification errors. Thus two milk progesterone concentration cutpoints were determined and all milk samples were classified as having a milk progesterone concentration that was either low (below the low cutpoint indicating the sample is from a non-luteal cow), high (above the high cutpoint indicating the sample is from a luteal cow) or inconclusive (between the high and low cutpoints indicating that it could not be determined with great confidence if the sample was from a luteal or non-luteal cow). The milk progesterone cutpoints were determined based on milk progesterone concentrations from cows confirmed pregnant to an insemination during the studies. For this group of cows, distributions of milk progesterone concentrations on the day of insemination (a time when low concentrations are expected) and 7–10 days before or after insemination (a time when high concentrations are expected) were selected. Based on these results milk progesterone concentrations ≤ 2 ng/ml were classified as low (91.2% sensitive and 98.2% specific for detection of non-luteal cows, n = 57) and concentrations ≤ 6 ng/ml were classified as high (86.0% sensitive and 98.2% specific for detection of luteal cows, n = 57).

Study I

For Herd 1, calving, insemination and pregnancy detection data were used to identify those cows that were both > 30 days since previous calving and not pregnant for at least 20 days during the 10 week milk sampling period. Milk progesterone concentration profiles were then used to determine ovulatory periods and non-ovulatory days from these cows during the days they met the criteria. For cows that met these criteria and then subsequently became pregnant during the sampling period, only ovulatory periods and non-ovulatory days until and including 20 days following the day of successful insemination were analysed. In Herd 2, all cows monitored for ovulation were ≤ 30 days since previous calving and not-pregnant at the start of the monitoring period. Thus all data from cows that had an ovulatory period determined by milk progesterone concentration profile or repeat trans-rectal ovarian ultrasonography during the monitoring period were analysed.

Identifying ovulations and definition of ovulatory periods

For Herd 1, an ovulation (n = 141) was considered to have occurred during a period of low milk progesterone that was both preceded and followed by periods of high milk progesterone within 7–10 days of the start and end of the period of low milk progesterone. The ovulatory period was defined as the days between, and the 2 days before and after the first and last consecutive low milk progesterone sample days. The same method was used

to identify ovulatory periods (n = 28) amongst cows in Herd 2 that were selected for serial milk progesterone monitoring and not inseminated during the ovulation monitoring period (n = 59). In addition, for cows in Herd 2 that were inseminated during the data collection period (n = 111), an ovulation was considered to have occurred if either; an ovulation was observed by repeat ovarian ultrasonography (n = 100), or an ovulation was not observed but milk progesterone concentration on the day of insemination was low and this was either preceded or followed by a single high milk progesterone sample within 7–10 days (n = 7). In these cows, the ovulatory period was defined as the period from 2 days before to 2 days after the day of insemination.

Definition and selection of non-ovulatory days

Non-ovulatory days consisted of 24-h periods commencing at 4 AM each day. This time period approximately aligns with the decision-making process in a dairy herd where cows are selected for insemination once daily at the morning milking based on information collected prior to that milking. In Herd 1, all days (4 AM to 4 AM) on or between consecutive high milk progesterone sample days, and all days between high milk progesterone days and ovulatory period days, were classified as non-ovulatory days. In Herd 2, activity analysis was performed over a fixed 21-day period for all cows. These 21 days included the 11 days during which cows were observed for ovulatory periods for the two studies (the first 11 days of AI) plus 4 days before and 6 days after. Only cows that had an ovulatory period during the 11 observation days contributed to non-ovulatory days for analyses. For these cows, all days during the 21-day period other than ovulatory period days, were classified as non-ovulatory days.

Not all non-ovulatory days were used in the calculation of ovulation detection specificity and positive predictive value. For both herds, if a series of activity alerts occurred in consecutive non-ovulatory days, alerts on the second and subsequent days of these sequences were not classified as new false-positive alerts because they would generally be considered by herd managers as a continuation of the initial increased activity. Because by this definition any non-ovulatory day after an activity alert day could not be considered a false-positive alert, all non-ovulatory days immediately following an activity alert day were excluded from analysis regardless of whether an activity alert occurred or not. All other non-ovulatory days were used in calculating ovulation detection specificity and positive predictive value.

Definition of true-positive, true-negative, false-positive and false-negative activity alerts

Any ovulatory period with one or more activity alerts was considered a single true-positive period (TP). Ovulatory periods that had no activity alerts were considered a single false-negative period (FN). Non-ovulatory days that were preceded by an activity alert day were excluded from all cow ovulatory period detection performance calculations for reasons described above. All other non-ovulatory days with no activity alerts were each considered as true-negative days (TN), and those with one or more activity alerts were considered false-positive days (FP).

Calculation of sensitivity, specificity and positive predictive value

Sensitivity was defined as the proportion of ovulatory periods in which at least one alert occurred and was calculated by dividing the number of ovulatory periods with at least one activity alert (TP periods) by the total number of ovulatory periods (TP + FN periods). Specificity was defined as the proportion of non-ovulatory periods in which no alert occurred and was calculated by dividing the number of non-ovulatory days that did not have an activity alert (TN days), by the total number of non-ovulatory days (TN + FP days). Positive predictive value was calculated by dividing the number of ovulatory periods with at least one activity alert (TP periods) by the sum of the number of ovulatory periods with at least one activity alert and non-ovulatory days with activity alerts (TP periods + FP days). Because more than one activity alert occurred in some ovulatory periods, this will have resulted in lower estimates of positive predictive value relative to the alternative approach of including all activity alerts in ovulatory periods.

Activity alert algorithms

Activity alerts were calculated using five different algorithms. The first algorithm was that used by DAIRYPLAN® and the last four algorithms were the same as described by Roelofs et al. (2005). All of the algorithms were based on comparisons of the activity in each 2-h period with the mean and standard deviation of the activity for the corresponding periods at the same times of day in the previous 10 days for the same cow. An example of activity data from an individual cow is shown in Fig. 2.

For the Westfalia-Surge DairyPlan® algorithm (W-DP), an alert was recorded if; (i) the activity in each of three consecutive 2-h periods was equal to or greater than two standard deviations above the mean of their corresponding periods in the preceding 10 days, and (ii) the mean of the deviations in total activity count for each of the three periods (expressed as ratios of the standard deviation for the corresponding periods in the preceding 10 days) was at least three. An alert was recorded for each of the three 2-h periods.

The remaining four algorithms were as described by Roelofs et al. (2005); an activity alert was recorded if deviations in total activity count from the mean for the corresponding periods in the preceding 10 days for two consecutive 2-h periods were both at least 2 (2SD), 2.5 (2.5SD), 3 (3SD), or 3.5 (3.5SD). An alert was recorded for each of the two 2-h periods.

Study II

Detection of time of ovulation by ovarian ultrasound

Cows from Herd 2 that were inseminated during the first 11 days from the start of AI had their ovaries examined

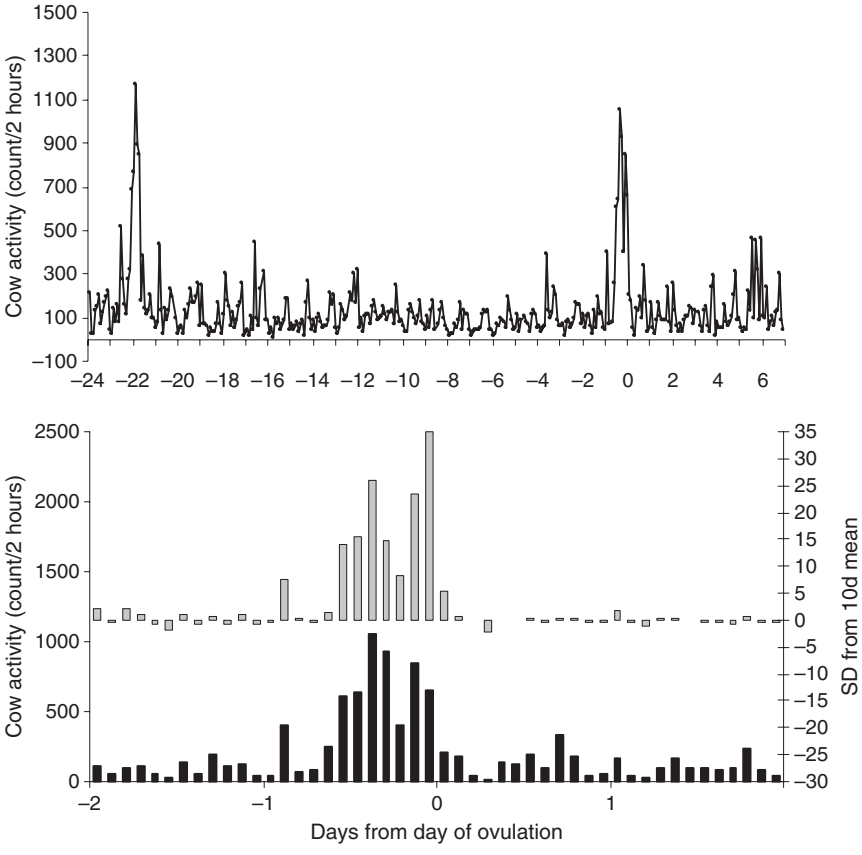

Fig. 2. Activity of an individual cow from Herd 2 as measured by a neck-mounted activity meter system (Rescounter II®, Westfalia-Surge, Germany). The top section shows the activity over a 30-day period. The black line represents the activity count for each 2-h period. The lower section shows a detailed insert of the cow's activity around the time of the observed ovulation. The day of ovulation (determined by ultrasound examination) was chosen as day 0. The black bars represent the activity in each 2-h period (left *Y*-axis) and the grey bars express this same activity as the number of standard deviations from the mean of the corresponding periods during the previous 10 days (right *Y*-axis)

by trans-rectal ultrasound (Easi-Scan, 4.5–8.5 MHz, BCF Ultrasound Australasia Pty Ltd, Melbourne, Australia) up to four times. The first examination was performed at the time of insemination with subsequent examinations performed just prior to or after each subsequent milking (approximately every 12 h) until the disappearance of the largest follicle (≥10 mm) or up until four ultrasound observations were performed, whichever occurred first. Ovulation was assumed to have occurred half way between the time of the last observation where the largest follicle was present and the time of the subsequent observation. To prevent bias in the distribution of interval from onset of increased activity to time of ovulation; for cows where an ovulation was not observed but was likely to have occurred before or after the ultrasound observations (n = 10), a time of ovulation was estimated so their data could be included in this distribution. An ovulation was considered likely to have occurred if the milk progesterone profile indicated an ovulatory period as described for Study I (n = 7), or if the milk progesterone profile was inconclusive but the cow was later confirmed pregnant from that insemination by palpation per rectum by an experienced veterinarian between 35 and 56 days following insemination (n = 3). For those cows that had no large follicle present at the time of the first ultrasound scan (four of 100 ovulations where time of ovulation was determined), ovulation was assumed to have occurred 6 h prior to the first ultrasound examination. Similarly, for those cows which had a large follicle still present after the last ultrasound examination (six of 100 ovulations), ovulation was assumed to have occurred 6 h later. These assumptions were based on the previous studies where few ovulations occurred earlier than 12 h or later than 48 h after the onset of oestrous (Trimberger 1948; Hall et al. 1959; Hernandez-Ceron et al. 1993; Walker et al. 1996; Van Eerdenburg et al. 2002; Bloch et al. 2006).

Definition of onset, end and duration of increased cow activity

Duration of increased cow activity was defined as the time between onset and end of increased activity for ovulatory periods based on alerts using the 2SD algorithm. Times of onset and end of activity were defined respectively as the start times of the first and last of each sequence of 2-h periods where activity was increased. Periods were considered to have increased activity where this was at least 2 standard deviations above the mean activity for the corresponding periods over the preceding 10 days. Activity was considered to be continuously increased across sequential 2-h periods even if up to two consecutive 2-h periods did not have increased activity, provided these periods were both immediately preceded and followed by at least one 2-h period with increased activity.

Statistical analyses

Data were analysed using the Stata statistical software package (Release 10, 2007; StataCorp, College Station, TX, USA). Sensitivity, specificity and positive predictive value was calculated for each algorithm in each herd.

Ninety-five per cent confidence intervals (95% CIs) were calculated for each estimate of sensitivity, specificity and positive predictive value within each herd using robust standard errors that accounted for clustering by cow. Intra-class correlation coefficients (ICCs) were calculated to estimate the extent of clustering; an ICC of 1 indicates complete clustering and an ICC of 0 indicates no clustering. For example, if complete clustering of outcomes for ovulatory periods was present within cows, each cow would fall into one of two categories; within cows, either all ovulatory periods would have at least one alert or none would have alerts. Similarly, if complete clustering of outcomes for non-ovulatory days was present within cows, either all non-ovulatory days for any particular cow would have at least one alert or none would have alerts. ICCs were calculated as the cow-level (i.e. the random intercept) variance divided by the sum of the cow-level variance and $\pi^2/3$ using Stata's -xtlogit- command. The p value for testing the null hypothesis (the hypothesis that the actual ICC is 0 and that there is no clustering by cow) was calculated using the likelihood ratio test for testing whether the cow-level variance is 0. Mean number of ovulations per day during the study periods were calculated for each herd by dividing the total number of ovulations identified for each herd by the total number of days in which the herd was monitored for ovulations.

Results

For Herd 1, using milk progesterone profiles, 141 ovulatory periods and approximately 2000 non-ovulatory days (range = 1952–2143, depending on algorithm being assessed) were identified from 64 cows and used to calculate the performance of activity monitoring for detecting cow ovulatory periods. This herd had a mean of 2.0 ovulations observed per day during the study. The mean intervals from calving to the beginning and the end of the activity observation period for each cow used in the final analyses were 94.6 and 154.9 days respectively. For Herd 2, using a combination of milk progesterone concentration and repeat trans-rectal ovarian ultrasound, 135 ovulatory periods and approximately 2000 non-ovulatory days (range = 1941–2118, depending on algorithm being assessed) were identified from 135 cows and used to calculate the performance of activity monitoring for detecting cow ovulatory periods in Study I. From these 135 cows, 111 were selected for insemination by the farm manger and so were also enrolled in Study II and examined by repeat trans-rectal ovarian ultrasound to determine the time of ovulation. There was a mean of 12.2 ovulations observed per day for this herd during both studies. The mean intervals from calving to the beginning and end of the activity analysis period were 61.2 and 82.2 days respectively.

Study I – Sensitivity, specificity and positive predictive value for detection of cow ovulatory periods

The performance of the activity meter system for detecting cow ovulatory periods using each activity algorithm in Herds 1 and 2 is shown in Table 1. These are compared with the performance reported from a

Table 1. Performance of the activity meter system using five activity alert algorithms for detecting cow ovulatory periods in two different herds

Herd	Activity alert algorithm	Ovulatory periods	Ovulatory periods with at least one activity alert	Sensitivity		Non-ovulatory days in analysis	False-positive activity alert days	Specificity		Positive predictive value	
				% (95% CI)	ICC ±SE, *P*			% (95% CI)	ICC ±SE, *P*	% (95% CI)	ICC ±SE, *P*
1, Cows = 64	W-DP	141	114	80.9 (73.4, 88.3)	0.350 ± 0.206, 0.045	2143	41	98.1 (97.4, 98.8)	0.165 ± 0.089, 0.014	73.5 (66.0, 81.1)	0.043 ± 0.101, 0.325
	2SD	141	128	90.8 (85.4, 96.1)	0.373 ± 0.262, 0.100	1952	195	90.0 (88.6, 91.4)	0.005 ± 0.020, 0.398	39.6 (35.8, 43.4)	0.000 ± 0.000, 1.000
	2.5SD	141	123	87.2 (80.8, 93.6)	0.415 ± 0.220, 0.039	2012	156	92.2 (90.9, 93.6)	0.026 ± 0.026, 0.131	44.1 (39.9, 48.3)	0.000 ± 0.000, 1.000
	3SD	141	116	82.3 (75.0, 89.5)	0.364 ± 0.207, 0.041	2061	112	94.6 (93.5, 95.6)	0.006 ± 0.031, 0.427	50.9 (46.2, 55.6)	0.000 ± 0.000, 1.000
	3.5SD	141	112	79.4 (71.9, 87.0)	0.303 ± 0.202, 0.065	2086	92	95.6 (94.7, 96.5)	0.000 ± 0.000, 0.498	54.9 (50.0, 59.8)	0.000 ± 0.000, 1.000
2, Cows = 135	W-DP	135	122	90.4 (85.3, 95.4)	N/A	2118	39	98.2 (97.6, 98.7)	0.000 ± 0.000, 0.497	75.8 (70.2, 81.4)	0.000 ± 0.000, 1.000
	2SD	135	127	94.1 (90.0, 98.1)	N/A	1941	186	90.4 (89.1, 91.8)	0.003 ± 0.033, 0.460	40.6 (37.4, 43.8)	0.000 ± 0.000, 1.000
	2.5SD	135	125	92.6 (88.1, 97.1)	N/A	2009	138	93.1 (92.0, 94.2)	0.000 ± 0.002, 0.496	47.5 (43.7, 51.4)	0.000 ± 0.000, 1.000
	3SD	135	119	88.1 (82.6, 93.7)	N/A	2067	92	95.5 (94.8, 96.3)	0.000 ± 0.000, 0.498	56.4 (52.1, 60.7)	0.000 ± 0.000, 1.000
	3.5SD	135	117	86.7 (80.9, 92.5)	N/A	2104	62	97.1 (96.4, 97.7)	0.000 ± 0.000, 0.497	65.4 (60.2, 70.5)	0.000 ± 0.000, 1.000

For Herd 1, 141 ovulatory periods were identified by serial milk progesterone concentration profiles; for Herd 2, 135 ovulatory periods were identified by either; (i) repeat trans-rectal ovarian ultrasound observations (n = 100), (ii) serial milk progesterone concentration profiles (n = 28), or (iii) two milk progesterone concentrations 7–10 days apart (n = 7); 95% CI = 95% confidence interval; ICC = Intra-class correlation coefficient (an indication of clustering of results by cow); SE = the standard error of ICC; N/A = ICC could not be estimated because each cow contributed only one ovulatory period.

Table 2. Summary of activity meter performance for detecting cows that are about to ovulate in three different management systems using a range of activity alert algorithms

Measure of performance for detecting cows about to ovulate	Herd	Activity alert algorithm				
		W-DP	2SD	2.5SD	3SD	3.5SD
Sensitivity (%)	1	80.9	90.8	87.2	82.3	79.4
	2	90.4	94.1	92.6	88.1	86.7
	Roelofs et al. (2005)		87.3	87.3	82.5	79.4
Specificity (%)	1	98.1	90.0	92.2	94.6	95.6
	2	98.2	90.4	93.1	95.5	97.1
	Roelofs et al. (2005)		97.2	98.8	99.6	99.7
Positive predictive value (%)	1	73.5	39.6	44.1	50.9	54.9
	2	75.8	40.6	47.5	56.4	65.4
	Roelofs et al. (2005)		64.7	80.9	92.9	94.3

Specificities were not quoted in Roelofs et al. (2005) but have been calculated using the reported numbers of correct and false alerts and total analysis period of approximately 18 days per ovulatory period. It was assumed that each ovulatory period was only one day. Therefore, the number of non-ovulatory days = 17 days × number of ovulations.

similar study in a research herd in the Netherlands in Table 2. (Roelofs et al. 2005). Estimates of sensitivity for detecting cow ovulatory periods in Herd 1 using the 2SD, 2.5SD, 3SD and 3.5SD algorithms were very similar to those reported by Roelofs et al. (2005). Sensitivity for the corresponding algorithms was slightly higher in Herd 2. Herds 1 and 2 had similar specificity and positive predictive value for corresponding algorithms and these were markedly lower than estimates calculated using the findings by Roelofs et al. (2005). Within each herd, specificity and positive predictive value estimates were lower with algorithms where sensitivity was higher. In both Herds 1 and 2, the W-DP algorithm gave the best specificity and positive predictive value of all the algorithms. Sensitivity of this algorithm was only slightly lower than that for other algorithms.

For ovulatory periods in Herd 1, the occurrence of at least one alert was clustered by cow (Table 1); estimated ICCs were between 0.303 and 0.415 indicating modest clustering by cow and demonstrating that some cows were at repeatedly greater risk than other cows of having or not having an alert within ovulatory periods. Actual ICC is 0 when no clustering is present and 1 when complete clustering is present. For non-ovulatory days, there was some evidence of clustering of occurrences of false-positive activity alerts by cow with the W-DP algorithm in Herd 1 (ICC = 0.165), but not in Herd 2, or for other algorithms in either herd. In Herd 1 where the ICC for the W-DP algorithm was 0.165, 41 false-positive activity alerts occurred in only 26 of the 64 cows that contributed non-ovulatory periods (an average of 1.6 false positives in these 26 cows). By contrast, in Herd 2 where the ICC for the W-DP algorithm was 0, all 39 false-positive activity alerts occurred in 35 of 135 cows (an average of 1.1 false positives for these 35 cows which is just above the minimum possible of 1).

Study II – Interval between onset and end of increased activity and time of ovulation

Of the 111 inseminations in Herd 2, the milk progesterone profile was inconsistent with a normal ovulation for one insemination and one or more ultrasound observations were missed for 10 inseminations. These were excluded from the analysis of intervals from onset and end of increased activity to ovulation. The time of ovulation was recorded for the remaining 100 ovulations. Of these ovulations, 90 were observed between successive ultrasound examinations, four were assumed to have occurred before the first ultrasound examination and six after the last examination. From these 100 ovulations with estimated ovulation times, 94 also had an associated increase in activity. The distributions of intervals from onset and end of increased activity to time of ovulation for these 94 ovulations are shown in Fig. 3. The mean times (±SD) from onset and end of increased activity to ovulation were 33.4 ± 12.4 and 17.3 ± 12.8 h respectively. Fifty per cent (n = 47) of ovulations occurred within the 8-h period between 30 to 38 h after the onset of increased activity, 76.6% (n = 72) within the 16 h between 24 to 40 h, 85.1% (n = 80) within the 24 h between 18 and 42 h and 90.4% (n = 85) within the 32 h from 19 to 51 h after the onset of increased activity.

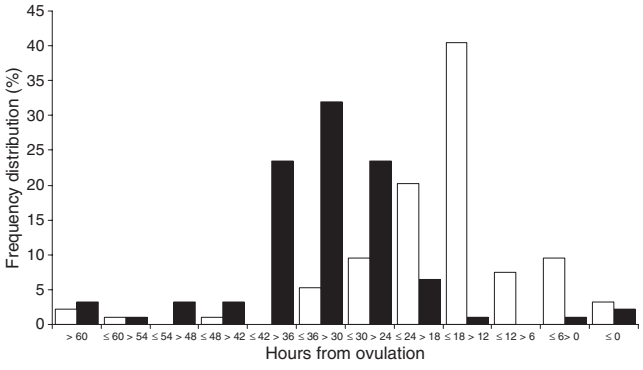

Fig. 3. Distribution of intervals from start (black bars) and end (white bars) of increased cow activity to time of ovulation (all data is from Herd 2, n = 94 ovulations)

From the 135 ovulatory periods selected for calculating cow ovulatory period detection performance in Herd 2, 127 had an associated increase in activity. The mean duration (±SD) of increased activity for these periods was 16.1 ± 7.0 h.

Discussion

Results from these studies show that monitoring paddock-based dairy cow activity in 2-h time periods can result in detection of a high proportion of cows about to ovulate under different management systems, and can give a good indication of when ovulation is likely to occur. However, the specificities and positive predictive values using the algorithms assessed may be lower than desirable.

Our estimates of sensitivity, specificity and positive predictive value of algorithms for detection of cow ovulatory periods were within range of those reported from the previous studies using cow activity monitoring (Lehrer et al. 1992; Koelsch et al. 1994; de Mol et al. 1997; Nebel et al. 2000; Firk et al. 2002, 2003; Cavalieri et al. 2003; Roelofs et al. 2005). However, estimates of

these variables will vary depending on methods of calculation and definitions of ovulatory periods and non-ovulatory days. In addition, while some authors have reported the specificity (Koelsch et al. 1994; de Mol et al. 1997; Firk et al. 2003), others have reported only the positive predictive value (often indicated by 'accuracy' or 'error rate'). However, positive predictive value is not only dependant on specificity; it is also affected by the prevalence of ovulation and the sensitivity. So comparisons of estimates of positive predictive value may not be valid in circumstances where prevalence of ovulation and the sensitivity differ. Accordingly, it is preferable that specificity is reported along with positive predictive value and sensitivity when evaluating systems for detecting ovulatory cows.

The performance of the activity meter system to detect cow ovulatory periods was generally similar between Herds 1 and 2 for each algorithm despite substantial differences in animal environments and calving and reproductive management systems; the only differences of note were modestly higher sensitivity estimates in Herd 2. These higher sensitivities may have been because of greater numbers of cows ovulating on the same day compared with Herd 1. The difference in number of ovulations occurring concurrently was because of both the differences in the size of the herds (130 and 400) and calving systems (year-round versus seasonal calving) in Herds 1 and 2 respectively. Roelofs et al. (2005) found the proportion of ovulatory cows detected by activity meters increased when more than one cow was in oestrous at the same time (proportion of ovulatory cows detected was 67, 85 and 95% when one, two, and three or more cows were in oestrus at the same time respectively). Other researchers have also shown that intensity of oestrous behaviour and frequency of activity is positively correlated with the number of animals in oestrous at the same time (Hurnik et al. 1975; VanVliet and VanEerdenburg 1996; Yaniz et al. 2006).

Sensitivity estimates for both herds were generally similar to those found by Roelofs et al. (2005). This suggests that relative changes in ovulatory cow activity were similar across all three herds despite substantial differences in the environments and management systems of these herds. However, estimates for specificity and positive predictive value for both herds 1 and 2 were substantially lower compared with Roelofs et al. (2005). This may have been because of different positions of the activity meter on the cow (neck in the current study vs leg in Roelofs et al.'s study). Eradus et al. (1992) observed significantly higher false positive peak activity values in neck-mounted activity meters relative to foreleg activity meters and this was confirmed by Sakaguchi et al. (2007). Koelsch et al. (1994) reported that although leg activity meters gave readings at or above frequencies recorded by neck-mounted activity meters, they consistently followed the same pattern. In addition, cows in Herd 1 and 2 were milked twice daily in contrast to those in the study by Roelofs et al. (2005) where cows were milked using an automated milking system. It is possible that twice daily movement of the lactating cow herd to the dairy and holding cows in the yard until milked disrupts normal behaviour patterns and so results in increased false positive activity alerts.

Our ICC estimates show that cow-level factors contribute to the sensitivity of detecting cow ovulatory periods. Differences in sensitivity between individual cows may be because of a range of factors including social hierarchy, dioestrous activity levels, lameness, milk production, endocrine abnormalities or potentially the positioning of the collar that holds the activity meter on the neck (Orihuela 2000; Firk et al. 2002; Wiltbank et al. 2006). In addition, ranges of days from the previous calving varied between cows in the study and this may explain some of the clustering within cows. This is because time from calving affects the intensity and duration of standing oestrous behaviour (Shipka 2000; Isobe et al. 2004), and the sensitivity for detection of ovulatory cows using activity meters (Peter and Bosu 1986).

Poor specificity is thought likely to be a major limitation in the uptake of technology to select cows for AI by dairy farmers because of low positive predictive values and consequently low probability of pregnancy to each insemination (Firk et al. 2003). Based on the results of Study I, modifications to the algorithms used to define activity meter alerts are required to improve specificity. Such changes must not result in reduced sensitivity. Although the 2SD algorithm resulted in the highest sensitivity for both herds, it also resulted in the poorest specificity. This is because the 2SD algorithm requires smaller increases in intensity and duration of activity to produce an alert. The W-DP algorithm, however, gave the highest specificity for both Herd 1 and Herd 2 whilst maintaining modest levels of sensitivity. The higher specificity of the W-DP algorithm compared with the SD algorithms is likely to be because of the requirement for three rather than two consecutive time periods to be above a set threshold. Koelsch et al. (1994) reported a substantial increase in specificity with only a minor reduction in sensitivity by requiring multiple time periods to exceed a statistical threshold. The highest combination of sensitivity and specificity was found when activity alerts required both a statistically unique increase in the rate of activity and in the duration of that increased rate of activity (Koelsch et al. 1994). The improved specificity and high sensitivity achieved using the W-DP algorithm compared with the SD algorithms demonstrates the importance of requiring prolonged periods of high activity when designing algorithms to calculate activity alerts. It may be possible to improve specificity without a marked reduction in sensitivity by further increasing the duration of increased activity required to create an alert. As more than 95% of cows who demonstrated an increase in activity around the time of ovulation had a duration of increased activity of at least 8 h, the duration of increased activity required for an alert could be increased to four consecutive 2-h activity periods or even four within a larger number of given 2-h activity periods (e.g. an increased activity in any four 2-h periods within six consecutive periods), without a substantial reduction in sensitivity. Further study of the pattern and causes of false-positive alerts could identify if this approach is useful and may help to find other ways to improve activity alert algorithms.

Detection of cow ovulatory periods was based only on activity algorithms and no other information about the cow was considered. Performance may be improved by taking into account other information about the cow, such as intervals since previous activity alerts, visual observations of the cow and intensity and duration of the current alert. Firk et al. (2003) showed that by taking into account times since previous oestrous events in a multivariable fuzzy logic model, the frequency of false positive alerts was reduced without large reductions in oestrous sensitivity. Others have also improved specificity in a similar way (de Mol et al. 1997; de Mol and Woldt 2001). In addition, manual rectal palpation and visual assessment for oestrous signs by experienced AI technicians can allow exclusion of non-ovulating cows without eliminating excessive numbers of ovulating cows (Sturman et al. 2000). Performance may also be improved by combining activity monitoring with other automatically sensed variables. The previous studies have shown that combined interpretation of multiple detection methods can improve overall performance for detecting cows about to ovulate (Lehrer et al. 1992; Senger 1994; Firk et al. 2002). The clustering of sensitivity by cow observed in the current studies also suggests that a combined approach might be effective. If reasons for this clustering can be identified and these factors included in combination with activity data, detection performance may be improved.

Few previous studies have reported the temporal relationship between increase in cow activity and time of ovulation (Roelofs et al. 2005; Sakaguchi et al. 2007; Yoshioka et al. 2007). The mean of intervals from onset of increased cow activity to ovulation in our study was within close range of those reported previously. Roelofs et al. (2005) observed 63 ovulations from 43 cows; the mean and standard deviation of intervals from onset of increased activity to ovulation were 29.3 ± 3.9 h. In a study of 62 ovulations by Yoshioka et al. (2007), mean and standard deviation were 29.8 ± 4.7 h. Sakaguchi et al. (2007) observed 15 heifers with both neck and leg-mounted activity meters under paddock and tie-stall conditions, mean intervals from onset of increased activity to ovulation ranged from 24 to 32 h depending on the activity meter position and environment.

Duration of increased activity was approximately 6 h longer in our study than reported by Roelofs et al. (2005), (16.1 ± 7.0 h vs 10.0 ± 4.2 h respectively). This longer duration was because of both an earlier onset and slightly later end of increased activity relative to time of ovulation than in the study by Roelofs et al. (2005). The longer duration and earlier onset of increased activity may be related to each other as Yoshioka et al. (2007), reported a modest correlation between duration of activity and the interval to ovulation ($r = 0.54$; $p < 0.0001$). Mean time from activity to ovulation for herds may also differ between cows in paddocks rather than free stall barns, and between neck and leg-mounted activity meters (Sakaguchi et al. 2007).

The standard deviation of the interval from onset and end of increased activity to time of ovulation in our study is markedly greater than that reported by Roelofs et al. (2005). This is important because the smaller the variation in this interval, the greater the accuracy of cow activity for predicting the time of ovulation. The higher variation may partly be explained by the reduced frequency of ovarian ultrasounds in this study compared with Roelofs et al. (2005), (approximately every 12 h vs every 3 h). However, this does not explain the higher variation in the duration of activity observed in our study (±7.0 h vs ±4.2 h) because activity was monitored in 2-h time periods in both studies. It is possible that this variation is because of the differences in the housing and milking systems or the positioning of the activity meters on the cows. In a number of studies by Sakaguchi et al. (2007) comparing neck and leg-mounted activity meters under paddock and tie-stall barn conditions, standard deviations for duration and interval to ovulation were generally higher for data from neck-mounted activity meters compared with leg-mounted activity meters. Time from activity to ovulation may also be affected by interactions between cow genetics, nutrition and metabolism (Arney et al. 1994) and prevalence of ovulation (Yaniz et al. 2006).

Despite the larger variability in intervals from activity to ovulation compared with Roelofs et al. (2005), over 75% of the animals ovulated during a 16-h time period relative to onset of increased activity and approximately 90% within a period of 32 h. Based on the previous studies assessing the effect of time of AI on pregnancy rate (Trimberger 1948; Hall et al. 1959; Nebel et al. 1994, 2000; Maatje et al. 1997; Dransfield et al. 1998; Pursley et al. 1998; Van Eerdenburg et al. 2002; Martinez et al. 2004), timing inseminations based on the onset of increased cow activity using this activity meter system is likely to result in an acceptable proportion of inseminations becoming pregnant. In addition, the close association between time of onset of increased activity and time of ovulation suggests that most of the true-positive alerts used in the calculation of sensitivity did not occur by chance alone during a period of low milk progesterone, but were actually temporally associated with ovulation.

Conclusions

These results show that monitoring cow activity with neck-mounted activity meters in 2-h time periods in paddock-based dairy cows in two diverse management systems can detect a high proportion of cows about to ovulate and provide a good indication of when ovulation is likely to occur. However, the specificities and positive predictive values using the algorithms assessed may be lower than desirable. Of the algorithms assessed, the W-DP algorithm gave the best specificity and positive predictive value and the sensitivity was only slightly lower than that for the other algorithms. If specificity and positive predictive value can be improved without substantially reducing sensitivity by taking into account other information about the cow or by studying the pattern and causes of false-positive activity alerts and developing better algorithms, this automated activity meter system could be an accurate, practical and efficient method for selecting cows for AI in paddock-based dairy herds.

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Author contributions

CD Hockey was responsible for the study design, data collection, data analysis, and drafting and editing all parts of the manuscript. JM Morton contributed to the study design data analysis and discussion of results and editing of the manuscript. ST Norman contributed to the discussion of results and editing of manuscript. MR McGowan contributed to the study design discussion of results and editing of the manuscript.

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