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Automated and visual measurements of estrous behavior and their sources of variation in Holstein heifers. I: Walking activity and behavior frequency

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ABSTRACT

Holstein heifers (n = 57) were monitored using accelerometers and video observations with the objective of better understanding the behavioral expression of estrus, the variation within and between the heifers, and the possible sources of variation. IceTags recorded walking activity from 7 to 13 months of age. Activity peaks (n = 282) were obtained from a rolling sum of steps within 24-hour periods and validated to be estrus by ovarian ultrasonography. Behavior around activity peak of one estrus for each of 12 heifers was described in detail from video recordings. Baseline behavior was monitored in a corresponding interval 1 week before. Estrus and baseline total steps and steps per hour, estrus relative increase in activity, duration, and interval between episodes were analyzed by descriptive statistics and Spearman rank correlations. Effects of category of baseline walking activity, estrus order (pubertal vs. second and greater episodes), season, hour of estrus onset, and number of heifers simultaneously in estrus were evaluated with proc MIXED. Behavioral changes from baseline to estrus were evaluated by a signed-rank test. Estrus total steps varied greatly (4743 ± 1740; range: 837–10,070), as well as the relative increase in activity (290 ± 160%; range: 30%–1190%). Duration of estrus was 14 ± 4 hours, ranging from 4 to 26 hours. The interval between episodes was the trait that varied the least. Pubertal estrus was shorter and had a smaller relative increase in activity than second and greater episodes ($P < 0.05$). The number of steps during estrus was greater for heifers of high baseline activity ($P < 0.01$). Estrus episodes occurring in the winter and starting between 4 PM and 3 AM had the greatest relative increase in activity ($P < 0.05$). The number of heifers simultaneously in estrus did not influence estrus expression ($P > 0.05$). The behaviors with greatest change from baseline to estrus were chin rest, sniff, back mount, crossover, accept chin rest, and follow, but variation was large. Overall, estrus was apparent in behavioral changes with large variation within and between the heifers. Estrus order, onset hour, season, and baseline walking activity are important factors affecting estrus activity. Therefore, estrus detection tools should account for potential sources of variation. The visual and automated measurements of estrus expression reported in this study reveal possibilities for improved on-farm estrus detection technologies and potential genetic selection for estrus expression.

1. Introduction

Estrus detection is an essential component of reproductive programs in dairy cattle, but concerns about low rates of estrus detection are not recent (e.g., Helmer and

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Britt [1]). During the past 20 years, timed artificial insemination (AI) protocols have improved service and pregnancy rates with satisfactory results [2]. Recent interest in reproductive programs with minimal pharmacologic intervention presents a new opportunity for the use of automated estrus detection tools, thus the need for further research on behavior, detailed measurements from automatic monitors of activity, and variability between and within cows.

Automated systems for estrus detection identify preovulatory follicular phases with variable rates of success depending on predetermined thresholds [3]. There are relatively few studies with detailed measurements of estrus activity from sensors using sufficient numbers of observations. Furthermore, walking activity and behavioral measurements of estrus are subject to many sources of variation linked to lactation: social interactions, housing, age, genetics, and physiological aspects [4]. Studies of estrous behavior and walking activity of heifers provide assessment of individual and environmental sources of variation while controlling for some of the lactation-linked factors.

Further studies of automated and visually measured estrous behavior, their relationship, and inherent variation are essential for improvement of currently available automated technologies. For example, definition of time of estrus onset based on increased walking activity is one of the areas that needs refinement for improved accuracy and determination of AI timing. Measurements of baseline activity, time of the day of estrus onset, and its possible sources of variation among and within animals have not been properly reported in the literature and are key components to determine relative and absolute increase in activity and duration of estrus. Such measurements of estrus intensity have been shown to be possible indicators of fertility [5,6] and perhaps markers for phenotypic selection for this trait.

Acceptance of mount (i.e., stand to be mounted) is the classical and still the primary estrous behavior commonly observed. However, this behavior alone may not be as useful as believed for estrus detection [7]. Secondary behaviors that occur at greater frequencies could be as important and more likely to be detected.

In this study, estrous behavior of heifers was described in detail using automated measurements of walking activity and video observations of behavior. Our objectives were to (1) identify absolute and relative measurements from sensors, as well as behaviors from video observations during baseline and estrus periods, and (2) evaluate variation in estrus expression between and within heifers and possible sources of variation (e.g., number of heifers simultaneously in estrus, season, time of day at estrus onset), when measured by sensors.

2. Materials and methods

2.1. Animals and housing

This study was conducted at the University of British Columbia's Dairy Education and Research Centre (Agassiz, British Columbia, Canada), from March 2012 to July 2013. The experimental herd is closed and had an average 305-day mature equivalent milk yield of 12,236 ± 2219 kg/cow in 2013. The local Institutional Animal Care Committee, following the requirements of the Canadian Animal Care Council, approved all experimental procedures.

Holstein heifers (n = 57) were housed in a sand-bedded free stall barn with rubber flooring on the feed bunk alley from 6 to 13 months of age. Heifers were managed in groups of 7 to 12 animals/pen, where the maximum age difference was 3 months. Pens were 6.7 × 12 m and had 13 stalls each. Total mixed ration was offered once per day (9 AM) and pushed up three times per day (at approximately 11 AM, 6 PM, and 10 PM). Water was available *ad libitum* from one water bin per pen. All heifers were visually checked for signs of disease or injuries twice a week at the time of ultrasonography. No major occurrences of disease or injury were registered during the experimental period.

2.2. Estrus episodes and sensors

Ovarian dynamics were determined by twice-a-week ovarian scans with ultrasound equipped with a 7.5-MHz linear rectal transducer (Ibex Pro; EI Medical Imaging, Loveland, CO, USA). The existence of a preovulatory follicle before the activity peak followed by appearance of a CL in the same ovary in the subsequent scanning indicated ovulation and validated the activity peak as an estrus episode. Puberty was determined on the basis of presence of a CL at ovarian ultrasonography. Pubertal estrus was identified in 46 out of 57 heifers (aged 9.0 ± 1.0 months and 309.3 ± 34.3 kg body weight at puberty onset; mean ± standard deviation). The remaining 11 heifers had a CL at one or more of the first four ovarian scans (aged 8.6 ± 1.2 months and 295.1 ± 33.0 kg body weight); therefore, these could not be confirmed as the CL of the pubertal estrus.

Walking activity was measured automatically with accelerometers (IceTag sensors; IceRobotics Ltd., Edinburgh, Scotland) attached with a custom flexible plastic strap to the metatarsal region on one of the hind limbs during the whole period. The sensors recorded the number of steps per minute, lying and standing times, and frequency of lying bouts. The effects of estrus on lying and standing patterns are presented in a sister research article (Silper et al., [8]). Data were downloaded from these devices once a week and processed with the manufacturer's software analyzer (IceTag Analyzer 2011; IceRobotics Ltd.). Downloading and reactivation were performed without removal of the sensor from the animal resulting in a recording gap of approximately 10 minutes. IceTags have been validated for lying behavior [9], step counting by comparison with video recording [10], and estrus detection with research-developed algorithms [11]. IceTags accurately measure the number of steps, although false steps may occur [10]. In the present study, the accuracy of the sensors for step counting on growing heifers was confirmed by comparing those results to results obtained by two different observers counting steps from video recordings during two 24-hour periods (data not shown).

Estrus episodes were identified on the basis of the number of steps in relation to the baseline period. Files