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# Lying behavior: Assessing within- and betweenherd variation in free-stall-housed dairy cows

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## ABSTRACT

One of the most important design criteria for dairy cow housing is access to a comfortable lying area. Behaviors such as the time cows spend lying down and how often they lie down can be used to evaluate the quality of stalls; however, assessing lying behavior on farms can be challenging. Indices such as the cow comfort index (CCI) and stall use index (SUI) have been widely used in on-farm assessments. The aims were to establish reliable sampling and recording methods for measuring lying behavior, to evaluate the adequacy of the CCI and SUI as estimates of lying behavior, and to describe variation in the lying behaviors of free-stallhoused dairy cows. The time spent lying down and the number of lying bouts for 2,033 cows on 43 farms were recorded for 5 d using electronic data loggers sampling at 1-min intervals. The CCI and SUI were calculated based on a single observation taken 2 h before the afternoon milking on each farm. Subsets of data were created, including 4, 3, 2, or 1 d per cow and 40, 30, 20, 10, 5, or  $1 \operatorname{cow}(s)$  per farm. The estimates derived from each sample size were compared with the overall means (based on 5 d and 44 cows per farm) for lying time and number of lying bouts, and the CCI and SUI were compared with the farm means of lying time, number of lying bouts, and bout duration using linear regression. Recording 30 or more cows for 3 d or more represented the overall means with high accuracy  $(R^2 > 0.9)$ , but using fewer cows or fewer days per cow resulted in poorer estimates of the farm mean. The CCI and SUI showed no association with the daily lying time (h/d);  $R^2 < 0.01$ ), and CCI was only weakly associated with the number of lying bouts per day ( $R^2 = 0.16$ ) and bout duration (min/bout;  $R^2 = 0.09$ ). Cows lay down 11.0  $\pm$ 2.1 h/d in 9  $\pm$  3 bouts/d, with a bout duration of 88  $\pm$ 30 min/bout. These values ranged from 9.5 to 12.9 h/d, 7 to 10 bouts/d, and 65 to 112 min/bout across farm means, and 4.2 to 19.5 h/d, 1 to 28 bouts/d, and 22 to

342 min/bout across individuals, showing that variation in lying behavior among individual cows within farm was greater than differences among farms.

**Key words:** dairy cow, lying behavior, cow comfort index

#### INTRODUCTION

Dairy cows are highly motivated to lie down for approximately 12 h/d (Jensen et al., 2005), and lying is a higher priority behavior than eating and social contact when opportunities to perform these behaviors are restricted (Munksgaard et al., 2005). Preventing cows from adequate lying time is harmful and causes changes in hypothalamic-pituitary-adrenal activity (Munksgaard and Simonsen, 1996). Lying behavior, particularly the time spent lying down, the frequency of lying bouts (i.e., a transition from standing to lying), and the duration of individual bouts were identified as sensitive measures of stall comfort (Haley et al., 2000). For example, cows spend more time lying down and do so more frequently on mattresses compared with a concrete stall base (Haley et al., 2001), and on deepbedded surfaces compared with inadequately bedded mattresses (Tucker et al., 2003). Similarly, cows spend more time lying down and have longer bouts in wider stalls (132 vs. 112 cm; Tucker et al., 2004) and in stalls with no brisket board (Tucker et al., 2006). Lying time responds to simple changes in stall management; for example, lying time increased from 8.8 to 13.8 h/d when wet bedding was switched to dry bedding (Fregonesi et al., 2007b), and lying time decreased by 1.7 h when the stocking rate (number of cows per stall) increased from 100 to 150% (Fregonesi et al., 2007a).

To evaluate such effects, studies have measured lying behavior continuously over a few days, either by using data loggers (Wechsler et al., 2000; Endres and Barberg, 2007) or through time-lapse video (Haley et al., 2000; Tucker et al., 2006). Continuous observation over 24-h periods, especially for a group of animals housed together, can be technically difficult and labor intensive. Alternatively, some studies have used 10-min, 15-min, or even 1-h instantaneous scan sampling, recording the

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proportion of the group of animals that were lying down at each scan (Leonard et al., 1996; Overton et al., 2002; DeVries and von Keyserlingk, 2005). In feedlot cattle, Mitlöhner et al. (2001) showed that instantaneous scan sampling of lying behavior at 15-min intervals provided an accurate estimate, highly correlated (r = 0.93) with continuous recording. In addition, they found that focal sampling of 1 animal out of 10 was enough to provide an accurate representation of the behavior of the entire group. Methods of sampling lying behavior in dairy

cattle are not well established. On-farm "cow comfort" assessment traditionally involved a calculation of an index or quotient based on a single observation. The most common measure is the cow comfort index (CCI), defined as the proportion of cows touching a stall that are lying down (Nelson, 1996). Similar indices of cow comfort include the stall use index (SUI: the proportion of cows in the pen not feeding that are lying down in the stalls) and the stall standing index (proportion of cows touching a stall that are standing or perching) (Cook et al., 2005). The CCI was linked to lameness prevalence (Espejo and Endres, 2007) and was used to assess stall use at a range of stocking densities (Krawczel et al., 2008). The National Animal Health Monitoring System of the USDA estimated CCI on participating farms during the national 2007 cow comfort survey of the dairy industry. Despite the reliance on these indices, it is not clear if they provide reliable estimates of lying behavior in dairy cows.

The diurnal daily pattern of lying behavior in lactating dairy cows is variable and is influenced by milking and feeding management (Overton et al., 2002; DeVries and von Keyserlingk, 2005). Considerable day-to-day variation in the proportion of eligible cows that are lying down (i.e., SUI) has been observed (Overton et al., 2002). These authors recommended that walk-through assessments be undertaken 1 h after the return from morning milking to capture the maximum lying behavior. In contrast, Cook et al. (2005) demonstrated that none of the indices of cow comfort based on hourly time points over the course of the day reflected the mean daily lying time of 10 focal cows from the group. These findings suggest that a more frequent and longer term monitoring of individual animals may be necessary to provide an accurate representation of lying behavior on farm.

Electronic data loggers are widely available and can be used to measure lying behavior accurately, including the total time spent lying down, the number of lying bouts, and the duration of each bout for individual cows (O'Driscoll et al., 2008). The first aim of this study was to determine how these loggers should be used to provide accurate estimates of lying behavior for a herd, specifically investigating how the number of days and focal animals sampled affects the estimate of the herd mean. The second aim was to compare commonly used indices of cow comfort (CCI and SUI) with lying times collected using data loggers to assess the reliability of these indices as measures of lying behavior. The final aim was simply to describe within- and between-herd variation in measures of lying behavior on commercial dairy farms that use free-stall housing.

#### MATERIALS AND METHODS

#### Farm Selection and Description

This study was conducted on 43 commercial dairy farms in the Fraser Valley region of British Columbia, Canada, between November 2007 and June 2008. Three local feed suppliers were asked to randomly select 15 of their clients that met the following criteria: free-stall housing, TMR or partially mixed ration with supplemental grain, and milking >70 cows. Forty farms were recruited in this way and 3 others were recruited directly by the research team. Thirty-five farms were on DHIA tests; 2 farms used Vampp Dairy Management Software (Vampp Management Systems Inc., Canada) to record individual and herd milk production, 1 farm used DairyPlan Software (GEA Westfalia Surge GmbH, Bönen, Germany) but had no individual production records, and 5 farms had no production records available. The average herd size was  $170 \pm 80$  milking cows ( $\pm$ SD, ranging from 71 to 511 cows) producing 10,548  $\pm$  800 kg (ranging from 8,991 to 12,080 kg) annually (based on the annual yield estimated by DHIA or an equivalent value derived from Vampp and DairyPlan). The majority of the farms (n = 37) milked twice daily, whereas the rest (n = 6) milked 3 times daily, and the majority (n = 34) fed once daily, whereas the rest (n = 9) fed twice daily. The main types of stall base were mattress (n = 17) and deep-bedded sand or sawdust (n = 12), but a variety of other types were used, including concrete, rubber mats, tires, wood, or a combination of types (n = 14); stalls were bedded with sawdust (n =33), sand (n = 9), or chopped straw (n = 1).

#### Data Collection

Each farm was visited twice, with 5 d between visits. Because heat stress is known to affect lying time (Cook et al., 2007), the data collection period was limited to days when the maximum temperature was  $<25^{\circ}$ C. The maximum temperature across study days was 9.5  $\pm$  5.3°C (ranging from -1.7 to 24.5°C). On arrival of researchers at each farm, the producer was asked to identify 1 pen that housed the high-producing cows, which was used for data collection. Thirteen farms housed all lactating cows in a single group, and others separated high-producing versus low-producing cows, or multiparous versus primiparous cows. The selected group size was  $94 \pm 31$  cows (ranging from 32 to 187 cows), and the stall stocking rate was  $104 \pm 15\%$  (ranging from 71 to 157%).

On the first visit, researchers arrived at the farm 2 h before the afternoon milking (ranging between 0911 and 1658 h) to take a visual sample count of the number of cows lying down, standing fully in the stall, standing with only 2 front feet in the stall, and feeding; these measures were used to calculate the CCI and SUI. Another visual sample count was taken on the second visit immediately after the morning milking. During milking on the first visit, up to 50 cows were systematically selected as focal cows based on the order they entered the milking parlor; for example, if the group had 100 cows, every second cow that came into the parlor was assessed. Lying behavior was recorded using an electronic data logger (HOBO Pendant G Acceleration Data Logger, Onset Computer Corporation, Pocasset, MA) that was attached to the medial side of the hind leg of each focal cow by using Vet Wrap (Co-Flex, Andover Coated Products Inc., Salisbury, MA), in a position such that the x-axis was parallel to the ground, the y-axis was perpendicular to the ground pointing upward, and the z-axis was parallel to the ground pointing away from the sagittal plane. The loggers recorded the g-force on the x, y, and z-axes at 1-min intervals for 5 d, preprogrammed to begin at midnight after the first visit. The data loggers were removed from the cows on the second visit and the data were downloaded using Onset HOBOware software (Onset Computer Corporation), which converted the *q*-force readings into degrees of tilt. These data were exported into Microsoft Excel (Microsoft Corporation, Redmond, WA), and the degree of vertical tilt (y-axis) was used to determine the lying position of the animal, such that readings  $<60^{\circ}$ indicated the cow standing, whereas readings  $\geq 60^{\circ}$ indicated the cow lying down. A macro was used to calculate daily standing time (min/d) and bouts per day based on 1,440 observations from midnight until midnight the following day. Standing and lying bouts of <2 min were ignored because these readings were likely associated with leg movements at the time of recording (Endres and Barberg, 2007). Daily lying time (min/d) was calculated as the inverse of the standing time, and the average bout duration was calculated by dividing the daily lying time by the number of bouts for that day. Most cows were lying down at midnight, but cows that were standing had bouts assigned to both days (e.g., a cow standing from 2330 until 0030 h would be assigned a 30-min standing bout in each of the 2 d). This source of error could be eliminated if observations

were not divided into 24-h periods, but this division was necessary in the current study.

Before the study, 25 HOBO loggers were tested against Tinytag Plus loggers (Gemini Data Loggers Ltd., Chichester, West Sussex, UK), which were previously validated for recording standing and lying behavior in dairy cows (O'Driscoll et al., 2008). One of each logger type was simultaneously attached to the same leg of a cow for a 4-d period and programmed to record the position at 1-min intervals. The Tinytag Plus logger used an internal circuit switch that opened (0 V) when in vertical position and closed (2.5 V) when in horizontal position, indicating whether the cow was standing or lying down. Data recorded by these loggers were downloaded using Tinytag Explorer software (Gemini Data Loggers Ltd.), exported into Microsoft Excel, and used to calculate the behavioral variables following the same methodology described above for the HOBO loggers. Measures of daily lying time (min/d) and lying bouts per day derived from the 2 types of loggers were closely associated ( $R^2 = 1.00$  and 0.97, respectively).

A total of 2,111 cows were assessed, but only 2,035 cows had usable lying behavior data; 76 cows were removed because they were either sold, became sick, or moved to a different group during the assessment period, or because the assigned data logger malfunctioned. Based on the sample size analyses (described in later sections), 2 cows missing 3 d or more of data were also removed from the data set. The final data set consisted of 43 farms and 2,033 cows. On average,  $47 \pm 5$  cows (ranging between 26 and 50 cows) were sampled on each farm for  $4.9 \pm 0.4$  d (ranging between 3 and 5 d). The focal cows overall were  $2.6 \pm 1.4$  lactations (ranging from 1 to 12 lactations), and  $150 \pm 94$  DIM (ranging from 11 to 704 DIM).

#### Data Analyses

To determine how sample size affected the estimate of lying behavior, 38 farms that had at least 44 focal cows with complete data for 5 d (n = 1.818) were used to create subsets of data consisting of a) 44, 40, 30, 20, 10, 5, and 1 cow(s) per farm, and b) 5, 4, 3, 2, 1 d per cow by using the simple random sampling in PROC SURVEYSELECT of SAS (SAS Institute, 2004) with strata specified as a) farm or b) farm cow. The procedure was repeated 10 times for each subset, and the relationship between the overall mean (based on 5) d and 44 cows per farm) and the mean of each estimate was tested using regression (PROC REG; model df = 1) for each repetition separately. The 10 R<sup>2</sup> values were used to estimate a mean and standard deviation for each subset. For all subsequent analyses, data were first averaged on a per-cow basis, from which the farm means were calculated; associations between variables were tested using regression (PROC REG; model df = 1).

# RESULTS

#### Measuring Lying Behavior

The estimate of the overall mean daily lying time (h/d) and the number of lying bouts per day based on the 5 d of observations declined progressively when fewer days were available (Figure 1). Measures of lying time (h/d) and lying bouts per day based on 3 d of data provided excellent estimates of the overall means (R<sup>2</sup>



Figure 1. The relationship  $(R^2)$  between (A) mean daily lying time (h/d) and (B) the number of lying bouts per day, based on 5 d/ cow, and estimates derived from 4, 3, 2, or 1 d randomly sampled for each cow (n = 1,818; 38 farms). The standard deviations across the 10 random sampling events are too small to be visible.

= 0.94 and 0.95, respectively). This accuracy declined when estimates were based on 2 d ( $R^2 = 0.88$  and 0.90) and declined further when the estimates were based on only 1 d ( $R^2 = 0.74$  and 0.77).

The accuracy of estimates of daily lying time (h/d) and lying bouts per day declined when estimates were based on fewer cows per farm (Figure 2). Estimates of lying time (h/d) and lying bouts per day based on 30 cows provided a reasonable estimate of the overall means ( $\mathbf{R}^2 = 0.88$  and 0.90, respectively), but this relationship was less when estimates were based on 20 cows ( $\mathbf{R}^2 = 0.75$  and 0.82, respectively) and declined further when the estimates were based on only 10 ( $\mathbf{R}^2 = 0.54$ and 0.60, respectively), 5 ( $\mathbf{R}^2 = 0.39$  and 0.39, respectively), and 1 cow ( $\mathbf{R}^2 = 0.08$  and 0.08, respectively) per farm.

#### Reliability of CCI and SUI

Across farms, the CCI was  $75 \pm 10\%$  (ranging from 50 to 92%), and the SUI was  $58 \pm 13\%$  (ranging from 33 to 89%). As expected, CCI and SUI derived from the same observation were associated ( $R^2 = 0.57$ , P < 0.01). Nevertheless, there was no association between CCI (Figure 3A;  $R^2 = 0.00$ , P = 0.10) and the mean daily lying time (h/d) derived from continuous monitoring of the focal cows from the same group. The CCI was imperceptibly associated with the mean lying bouts per day (Figure 3B;  $R^2 = 0.16$ , P = 0.01) and mean bout duration (min/bout; Figure 3C;  $R^2 = 0.09$ , P = 0.05). The SUI was not associated with daily lying time (h/d), lying bouts per day, or bout duration (min/d)bout;  $R^2 < 0.02$ , P > 0.30). Moreover, neither CCI nor SUI was associated with the same index derived from a separate sample taken immediately after the morning milking 5 d later ( $R^2 = 0.01$ , P = 0.50, and  $R^2 = 0.00$ , P = 0.80, respectively).

#### Lying Behavior—Within- and Between-Herd Variation

Across farms, cows spent  $11.0 \pm 2.1$  h/d lying down and had  $9 \pm 3$  bouts/d. The farm means varied from 9.5 to 12.9 h/d and 7 to 10 bouts/d, whereas the individual means varied from 4.2 to 19.5 h/d and 1 to 28 bouts/d (Figures 4 and 5). The bout duration across farms was 88  $\pm$  30 min/bout, with the farm means ranging from 65 to 112 min/bout and the individual means ranging from 22 to 342 min/bout. The variation among cows differed from farm to farm; for example, the standard deviation in lying time varied from 1.5 to 3.3 h/d. Similarly, the standard deviation in number of lying bouts and bout duration varied from 2 to 4 bouts/d and 17 to 49 min/bout.

#### DISCUSSION

### Measuring Lying Behavior

Two aspects of sampling procedure must be considered when deciding on a method of measuring behavior: 1) the sampling rule that specifies which subjects to observe, and 2) the recording rule that specifies how the behavior should be recorded (Martin and Bateson, 1993). In our study, focal instantaneous sampling at 1-min intervals was used. Despite numerous studies describing the lying behavior of cows on farm (Wechsler et al., 2000; Cook et al., 2005; Endres and Barberg, 2007), it was not known how the length of a sampling period would influence the accuracy of measures of lying behavior of individual cows. Researchers have used a variety of sampling methods; for example, Cook et al. (2005) recorded continuously through video (capturing 1 s of video every 30 s) for a single 24-h period, Wechsler et al. (2000) recorded at 16-s intervals for 3 d. and Endres and Barberg (2007) recorded continuously (8 times/s) for 1 wk. Our data provide the first evidence that, when recording at 1-min intervals, increasing the number of sampling days from 3 to 5 d makes little difference, suggesting that a 3-d sampling period is sufficient to estimate lying behavior accurately. Even a sampling period of 2 d yielded reasonable estimates with approximately 90% accuracy, and a single-day sampling yielded estimates with approximately 75% accuracy compared with the overall mean based on 5 d.

Variation among individual cows was considerable, such that the number of focal animals required to estimate the farm mean accurately was high. Focal sampling was generally the best approach for studying groups (Martin and Bateson, 1993) because it is normally unnecessary to observe every animal. Cook et al. (2005) sampled 10 focal cows from a pen containing approximately 85 cows, and found some discrepancies between CCI calculated from only the focal cows and the same indices based on all cows in the pen. Mitlöhner et al. (2001) showed that estimates of the percentage of time spent lying based on 1 to 9 animals out of a group of 10 were all similar, indicating that 1 focal animal for every 10 was sufficient to estimate the group mean. Our ability to estimate the farm mean based on a sample of 44 cows decreased as the sample decreased from 30 to 20, 10, 5, and 1 cow. A sample of 30 cows gave estimates of lying behavior with approximately 90% accuracy; this was reduced to approximately 80%when the sample size decreased to 20, and to less than 60% when the sample size decreased to 10 cows. From these results, we suggest that farm estimates of lying behavior be obtained from at least 30 cows per group. Herd estimates appeared to plateau between 30 and 40



Figure 2. The relationship  $(R^2)$  between (A) mean daily lying time (h/d) and (B) the number of lying bouts per day, based on 44 cows on each farm (n = 1,818 cows; 38 farms), and estimates derived from 40, 30, 20, 10, 5, and 1 cow(s) randomly sampled on each farm. The error bars represent  $\pm 1$  SD of the mean of the 10 random sampling events.

cows, suggesting that sampling additional cows would provide little extra information.

#### Reliability of CCI and SUI

The time of sampling relative to milking is an important source of variation in CCI (Cook et al., 2005), but this was standardized in the current study at 2 h before the afternoon milking. The average CCI across farms was 75%. This value supports reports in Wisconsin and Minnesota (Cook et al., 2005; Espejo et al., 2006). The average SUI for the farms in the current study was 58%. This value is lower than that reported previously (89% in Overton et al., 2002; 70 to 76% in Cook et al.,



Figure 3. Association between the cow comfort index (CCI), derived from a single observation of the assessment group on each of 43 farms at 2 h before the afternoon milking, and A) mean daily lying time (h/d;  $R^2 = 0.00$ , P = 0.10), B) mean lying bouts per day ( $R^2 = 0.16$ , P = 0.009), and C) mean bout duration (min/bout;  $R^2 = 0.09$ , P = 0.05), based on continuous monitoring over 5 d of focal cows (n = 2,033) from the same groups.

2005). The SUI was higher when cows had access to more space and free stalls (Krawczel et al., 2008), but the stocking densities in the farms in our study were comparable with those studied by Overton et al. (2002) and Cook et al. (2005).

The CCI and SUI are frequently used as a practical indicator of cow comfort. Unfortunately, our results showed no association between the CCI or SUI at the pen level and the mean daily lying time based on continuous recording of the focal cows from each pen. Instead, there was a weak negative relationship between CCI and the number of lying bouts, and a low positive relationship between CCI and the duration of lying bouts. Cook et al. (2005) attempted to relate these indices to estimates of daily lying time based on a continuous recording and found no association. In combination, these results indicate that CCI and SUI do not provide an accurate estimate of lying behavior, and likely should not be used for on-farm assessments of this behavior.

#### Lying Behavior—Within- and Between-Herd Variation

Mean daily lying time (11.0 h/d) in our study supports values previously recorded on commercial farms using free-stall housing (Wechsler et al., 2000; Cook et al., 2005), although the number of lying bouts (9) bouts/d) was lower and the duration of individual bouts (88 min/bout) was longer compared with those reported by Wechsler et al. (2000; 12 to 15 bouts/d and 53 to 67 min/bouts). Lying behavior in free-stall barns is affected by design and management factors, including stall surface and bedding quality (Tucker et al., 2003; Drissler et al., 2005; Fregonesi et al., 2007b), stall size and configuration (Tucker et al., 2004, 2006), stocking density (Fregonesi et al., 2007a), stall location and pen layout (Wagner-Storch et al., 2003), and pen flooring (Fregonesi et al., 2004). Despite a wide range in each of these factors in the current study, the range among the farm averages was less than the range among cows within many of the farms.

A cow's lying behavior is influenced by her social ranking (Galindo and Broom, 2000) as well as her production and health status (Fregonesi and Leaver, 2001; Walker et al., 2008). Variation in individual behavior may be more marked in highly competitive environments, where the ability of each animal to access a stall is restricted. For example, Leonard et al. (1996) reported a daily lying time of 7.5 h/d for heifers housed at a 2:1 stocking density (2 animals for 1 stall), with values ranging from 2.7 to 11.9 h/d for individual heifers. Overcrowding could contribute to a high variation



Figure 4. Mean ( $\blacksquare$ ), maximum ( $\blacklozenge$ ), and minimum ( $\blacklozenge$ ) daily lying time (h/d) on each of 43 farms. The error bars represent ±1 SD of the means.

in individual lying bouts because of increased opportunities for displacements from the stalls disrupting the normal lying behavior (Fregonesi et al., 2007a). Another potential source of variation is estrous behavior. In the present study, there was no way of systematically accounting for estrus, but future work studying the effects of estrous activity on lying behavior would be beneficial. Regardless of the cause, individual lying behaviors of cows housed together can be highly variable. The large cow-to-cow variation would reduce the statistical power of tests relying on between-cow comparisons; therefore, within-cow comparisons are likely more sensitive in detecting management or design changes expected to affect lying behavior.

#### CONCLUSIONS

Reliable estimates of lying behavior on commercial dairy farms can be generated using 3 d of continuous recordings (at 1-min intervals) from 30 focal cows per farm. The CCI and SUI derived from a single observation were not associated with daily lying time, number of lying bouts, or duration of individual bouts and thus cannot be recommended as methods of assessing this behavior. The range in lying behavior among individual cows within farms was greater than differences across farms.

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Figure 5. Mean ( $\blacksquare$ ), maximum ( $\blacklozenge$ ), and minimum ( $\blacklozenge$ ) daily lying bouts on each of 43 farms. The error bars represent  $\pm 1$  SD of the means.

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