BRUSHING ACTIVITY AS AN INDICATOR OF LAMENESS IN DAIRY COWS

שימוש במברשת כסממן לצליעות בבקר לחלב

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ABSTRACT

Lameness is a debilitating condition, which has significant economic and welfare implications on the dairy industry. Under- detection of lameness in the herd, leads to prolonged suffering for the cow until proper care is administered. Nowadays, common indicators for lameness are milk yield, rumination and activity levels. However, those measures are considered as core activities, and as such may not be sensitive enough to detect slight changes involved in an uncomfortable condition like lameness, especially in its early stages of development.

Brushing activity, is considered a "low resilient" behavior (i.e. behavior that typically decrease when energy resources are limited or when the cost involved in the behavior increases). As such, it is likely to be reduced earlier in cases of sickness or pain compared to core behavior which are more "resilient" by nature.

The aim of this study was to determine the association between different degrees of lameness and brush usage in dairy cows. Locomotion scores of 209 lactating Holstein dairy cows were collected once a week, for 14 weeks, for each cow individually, using a five point locomotion scoring system. The cows were housed in three sheds. In each shed, two rotating brush were installed, one installed next to the feed-bunk, and the second on the opposite side of the cowshed. Brushing activity data was collected automatically from each of the six brushes. Data on daily milk yield, rumination and activity was collected from the farm database. Statistical analysis was performed to evaluate the association between locomotion scores and daily measures of brush usage, milk yield, rumination and activity level.

We found that the daily proportion of cows using the brush at least once, as well as the daily duration of brush usage per cow were significantly lower in lame and severely lame cows

(locomotion score 4 and 5) compared to non-lame cows (locomotion score 1), only in brushes that were installed away from the feed bunk. However, mildly lame cows (locomotion score 3) and cows with uneven gait (locomotion score 2) did not differ significantly from non-lame cows (locomotion score 1) in both measures of brush usage. Daily milk yield of lame and severely lame cows as well as of cows with uneven gait was lower than that of non-lame cows. Daily rumination and daily activity of cows with uneven gait, mild lameness and lameness and severe lameness did not differ from that of non-lame cows.

The results of this study suggest that monitoring brush use when installed away from the feed bunk could be useful for detecting lame and severely lame cows, while detection of mild lameness or uneven gait using this method is, at this stage, less promising. Moreover, our results suggest, that milk yield is not a reliable measurement for detection of lameness, due to its inconsistent behavior in the different locomotion scores. However, monitoring of core behavior alongside "low resilient" behavior, such as brush use, may improve our ability to detect lameness even in its early stage.

תקציר

צליעה בבני בקר הינה מצב מגביל, בעל השלכות משמעותית מבחינת כלכליות המשק ורווחת בעלי חיים. תת-זיהוי של אירועי צליעה בעדר, יוביל לסבל ממושך ומיותר עבור הפרה. דרכי זיהוי צליעות בבקר המוזכרים באופן נפוץ בספרות, הינם תנובת חלב, העלאת גירה ורמת פעילות הפרה. אולם מדדים אלה נחשבים כמדדי ליבה, על כן לעיתים אינן רגישים מספיק לשינויים הקלים המעורבים במצבים כמו צליעות, בעיקר בדרגות חומרה קלות. שימוש במברשת גירוד, נחשבת כהתנהגות בעלת "קשיחות נמוכה" (פעולה שתדירות ביצועה תרד בתנאים בהם משאבי האנרגיה מוגבלים, או כשהמחיר הכרוך בביצוען עולה), על כן צפויה לרדת ברמת ביצועה מוקדם יותר במצבי מחלה וכאב בהשוואה להתנהגויות ליבה אשר "קשיחות" יותר מטבען. מטרת המחקר הנוכחי הינה, להעריך את הקשר בין דרגות הצליעה השונות לבין שימוש במברשת. דירוג צליעה מ209 פרות הולשטיין חולבות נאסף באופן פרטני לכל פרה, אחת לשבוע, במשך 14 שבועות, תוך שימוש במדרג תנועה בעל חמש רמות (1- פרה לא צולעת, 5- צליעה חמורה). נתוני שימוש במברשת נאספו באופן אוטומטי משש מברשות, שתי מברשות בכל אחת משלושת סככות החולבות, כאשר אחת המברשות הותקנה קרוב לאבוס ואילו השנייה רחוק מהאבוס. במקביל, נתוני תנובת החלב והעלאת גירה נאספו ממאגר הנתונים של הרפת. ניתוח סטטיסטי של התוצאות בוצע להערכת הקשר בין רמת הצליעה של הפרה לבין שני מדדים של שימוש יומי במברשת -משך השימוש במברשת ופרופורציית הפרות שהשתמשו במברשת לפחות פעם אחת ביום. כמו כן הוערך הקשר הסטטיסטי בין רמת הצליעה של הפרה ,לבין מדידות יומיות של תנובת החלב, העלאת גירה ורמת הפעילות. תוצאות המחקר הראו ירידה בשימוש במברשות שהותקנו רחוק מהאבוס בשני מדדי השימוש, בפרות בעלות צליעה (דירוג צליעה 4) וצליעה חמורה (דירוג צליעה 5) בהשוואה לפרות ללא צליעה (דירוג צליעה 1). לעומת זאת שימוש במברשת בפרות בעלות צליעה קלה או חוסר יציבות בהליכה (דירוג צליעה 3 ו-2 בהתאמה) לא נמצא שונה באופן מובהק בהשוואה לפרות ללא צליעה (דירוג 1). יתר על כן, נמצאה תנובת חלב נמוכה באופן מובהק בפרות בעלות צליעה קלה כמו גם בפרות בעלות צליעה וצליעה חמורה בהשוואה לפרות ללא צליעה, בעוד שלא נמצא שוני ברמת הפעילות והעלאת הגירה של הפרות בדרגות הצליעה השונות בהשוואה לפרות ללא צליעה. לאור התוצאות ניתן להסיק כי בשלב זה, שימוש במברשת כאשר היא ממוקמת רחוק מהאבוס, הינו כלי שימושי לזיהוי פרות בעלות צליעה וצליעה

חמורה, בעוד שאינו רגיש דיו לזיהוי פרות בעלות צליעה קלה וחוסר יציבות בהליכה. כמו כן, ניתן להסיק כי ניטור תנובת החלב, אינו מדד רגיש מספיק בפני עצמו, לזיהוי צליעה בפרות חולבות. זאת בשל התנהגותו הלא עקבית בדרגות הצליעה השונות. אולם יתכן כי שילוב של ניטור מדדים "קשיחים" כמו תנובת חלב ומדדים בעלי "קשיחות נמוכה" כמו שימוש במברשת, ישפר את היכולת שלנו לזיהוי צליעות בפרות גם בשלבים מוקדמים.

INTRODUCTION

Lameness is a common medical condition in the intensive dairy industry. In England and Wales a mean prevalence of 36.8% lame cows was estimated in 2010 (Barker et al., 2010), while a report from 2006 estimated a mean prevalence of 24.6% lame cows in Minnesota (Espejo et al., 2006). As for the Israeli dairy industry, a recent estimation revealed an annual mean prevalence of 12% lame cows. This seemingly low prevalence, may be a result of incorrect information (for example, reporting a diagnosed horn lesion as a lameness regardless if the cow is lame or not) and underestimation regarding lameness which reported by dairy producers in Israel (Department of herd medicine and Epidemiology "Hachaklait" 2017). From an economic stand, lameness is considered the third most important disease affecting the dairy herd (O'Callaghan, 2002), due to decreased milk yield, treatment costs, involuntary culling and reduced fertility (Green et al., 2002).

Apart from its economic implication, lameness has a major influence on animal welfare (O'Callaghan, 2002). Lameness is a debilitating condition, which usually involves tissue damage, pain and discomfort (Chapinal et al., 2009). The majority of lameness cases in cattle originates from lesions of the hoof. The lesion can be the results of an infection - for example: Dermatitis Interdigitalis, Dermatitis digitalis, Phlegmon Interdigitalis, or noninfectious - for example: Laminitis, sole ulcer, white line disease, double sole, tyloma etc (Newcomer and Chamorro, 2016). While the rest of the cases are caused by other disorders of the limb, such as diseases or injury of the bones or joints (Winckler and Willen, 2001), or from a systemic or metabolic disease (Greenough and Weaver, 1997). Lameness is a long-term developing disease. The actual onset of lameness can occur far before the diagnosis and treatment (Van Hertem et al., 2013). Cattle's natural instinct, as a survival strategy used by prey species, tends to mask any signs of pain and discomfort

(O'Callaghan, 2002). The little overt behavioral expression of pain during the early stages of lameness, makes the identification of lameness onset difficult, and prolongs the cow's suffering until proper care is administered (Anil et al., 2005). Studies have shown that dairy producers fail to detect more than two thirds of lame cows in the herd (Espejo et al., 2006), which leads to an underestimated number of lame cows reported by dairy producers (Borderas et al., 2008).

To improve the detection of lameness in the herd, especially in its early stages, several locomotion scoring methods have been proposed (Flower and Weary, 2006; Sprecher et al., 1997; Thomsen et al., 2008). Those methods focus on evaluating the degree of back arching and neck movement resulting from the cow's attempts to reduce weight on a particular limb (Flower and Weary, 2006). As the pain increases, the change in locomotion is more noticeable (Greenough and Weaver, 1997). Although proved to be helpful, these methods require training (Flower and Weary, 2006) and the scoring process itself is time consuming, especially when carried out on large dairy herds. Infrequent mobility scoring gives a snapshot of the prevalence of lameness in the herd, but has little value in continuous management of lameness on the present intensive farm routine (Reader et al., 2011). Therefore, there is a need to develop an objective and practical method for ongoing detection of lameness on the farm level.

Automated ways to detect pain and discomfort are used widely in both routine animal husbandry and in research - For example in detection of metritis (Fogsgaard et al., 2012; Mandel et al., 2017), pneumonia (Toaff-rosenstein, 2016) and for lameness (Borderas et al., 2008; Kocak and Ekiz, 2006; Reader et al., 2011; Thorup et al., 2016; Van Hertem et al., 2013). The behavioral indicators used for detecting lameness involve mostly production parameters, such as milk yield (Kocak and Ekiz, 2006; Van Hertem et al., 2013; Warnick et al., 2001), visits to the automatic milking system (Borderas et al., 2008), eating related behaviors such as rumination (Thorup et al., 2016; Van Hertem et al., 2013) ,visiting the feed bank (Thorup et al., 2016) and level of activity (Reader et al., 2011; Van Hertem et al., 2013). However conflicting results were found regarding these indicators and their association with lameness.

Animals adjust their behavior according to the "costs" of each activity in terms of time and energy (Aubert, 1999; Dawkins, 1990). The sick animal changes its behavioral priorities. The animal is willing to invest more or spend more time on behaviors with a primary function of promoting survival, over behaviors that promote other aspects of fitness (McFarland, 1999). At a time of illness, the animal recruits resources (by means of time or energy) to perform activities of critical short-term fitness, while activities that offer long-term fitness are likely to decrease (Weary et al., 2009). Activities which promote long-term fitness, are usually characterized as luxury or low- resilience behavior (i.e. an activity that is expected to decline when time and energy are limited) for example maintenance (e.g., grooming) (Dawkins, 1990; Weary et al., 2009). On the other hand, core behavior, for example, feeding or related behaviors such as rumination (Dawkins, 1990), is usually characterized as short-term fitness behavior. As such, it is more resilient by nature and expected to decrease only at a relatively later stage of disease.

The mechanical brush is an example of an environmental enrichment device that allows the cow to perform grooming behavior (Wilson et al., 2002). Research has shown that when given the opportunity, cows groom by mechanical brush rather than by inanimate objects in the pen (DeVries et al., 2007). Studies have suggested that as an expression of grooming, brushing activity falls under the category of a "low resilience" activity. As such, brush use was reduced when time and energy were limited (e.g. heat load; Mandel et al., 2013). Moreover, brush utilization was shown to be influenced by its distance from the food resource (Mandel et al., 2013). The farther the brush is located from the feed bank, the higher the "cost" involved in its utilization. Thus, the proportion of cows using the brush and daily average number of brushing events were reduced when the food is served farther from the brush (Mandel et al., 2013). Therefore brushing activity might fit the criteria of being a good objective indicator and could evolve to be a valid, reliable and feasible automated measure of a lameness (Rushen et al., 2012).

The aim of the current study was to investigate the association between locomotion scores and brush usage. We hypothesized that the daily proportion of cows using a brush, and the daily duration of brush usage would be inversely related to the severity of lameness. Daily measures of brush usage are expected to decrease even in case of mild lameness, while measures of core behaviors (rumination, milk yield, and general activity) are expected to decrease only in more severe cases of lameness, due to their high resilience. In order to test this hypothesis, we also analyzed the effect of lameness on daily milk yield, rumination, and general activity. Furthermore, we expected that changes in our measures of brush usage would be more pronounced in brushes located away from the feed bunk, compared with brushes located next to the feed bunk, because the cost involved in utilizing the former is higher.

MATERIALS AND METHODS

Cows and Management

The study was carried out at Shomria, A commercial dairy farm located in the northern Negev of Israel, between September and December of 2015. The herd consisted of Holstein dairy cows, which were kept in 3 groups divided according to their lactation status (1st, 2nd and 3rd+ lactation). Each group consisted of 70-80 cows (the number of cows in each of the groups changed depending on parturition in the herd). The groups were housed year-round in loose-housing cowsheds, 6.6x90.0m, bedded with dried manure which was cultivated on a daily basis. Each shed was ventilated by five overhead ventilators in order to facilitate the drying of the bedding. The cows were fed a TMR diet twice daily on a concrete slab (minimum 38m long for every 33 cows) at 08:00 h and 16:00 h. The food was pushed closer six times a day. Water was available ad-libitum from six self-filling water troughs (approximately 6m trough for every 33 cows). Cows were milked three times a day, at 04:00-06:00, 11:00-13:00 and 19:00-21:00. The average milk yield at that time was 36.8±8.7 L/day per cow. Due to warm climate conditions, during the first month of observations (September), all lactating cows were cooled down using water showers installed at the entrance to the milking parlor. Cow's hooves were trimmed by a trained staff member twice in lactation - once at 120-150 DIM and the second time before drying. Routine care of the animals was done by the farm's staff. Farm veterinary care was provided by a veterinary surgeon from Hachaklait Veterinary Services Ltd. (Caesarea, Israel) who visited the farm regularly twice a week and added visits on request. Medication was given when appropriate.

Data Collection

Locomotion Score. Cows' individual locomotion was visually assessed once a week, for 14 consecutive weeks, using a 5-point scoring system (1 = non lame to 5 = Severe lameness; Thomsen et al., 2008, Table 1). Locomotion was assessed at the exit of the milking parlor following the noon milking while the cows walked a 20m long concrete pathway covered by dried manure. Two-thirds of this pathway was fenced with a crossbeam, yet still enabled a good view of the walking cow, while the middle third was not fenced and provided a full view of the walking cow. Cows' locomotion was scored by a well-trained experimenter (trained by “Hachaklait” hoof health expert for three weeks). The experimenter stood approximately 11m from the cow’s pathway in order to allow recognition of individual cows identification (by its unique 3- or 4-digit number that had been applied as a brand at a younger age), and to allow proper view of cow’s walking gait. Prior to the beginning of the study, intra-observer reliability was calculated using an Intra-class Correlation (ICC) test. This test was based on four independent ratings of video recordings of 123 cows walking down the aforementioned path ($ICC_{(2,1)}$=0.823, CI 95% 0.775-0.864). The locomotion scoring data was recorded directly on a tablet computer (Nexus 9, HTC, New Taipei City, Taiwan) using an android based software developed for this study. The software recorded the date and time of each rating that was entered. Scoring sessions were recorded using a video camera (Panasonic HC-V160 Full HD Camcorder) installed 7 m from the pathway, in order to verify the correct recognition of cows in cases where the number branded on the cow was not completely clear during the locomotion assessment. IDs of cow from 26 ratings (from a total of 1436 ratings) were traced back and validated using this method. Cows were habituated to the presence of the observer (while standing at the observation point) for 2 hrs/d, during 5 consecutive days in the week prior the beginning of the observations. Lame cows which were detected by the farmers were treated by the farm's trained hoof trimmer and received veterinary medical care if needed. A total of 1436 locomotion ratings were collected throughout the observation period: 672 ratings of score one ('normal' - non-lame, 154 cows), 620 ratings of score two ('uneven gait', 159 cows), 128 ratings of score three ('mild lameness', 60 cows), 14 ratings of score four ('lameness', 10 cows) and 2 ratings of score five ('severe lameness', 2 cows). Cow scoring could stay constant or vary (improve/ worsen) between weeks throughout the observation period. 66 cows received the same score, 115 cows received 2 different scores, 25 cows received 3 different scores' and 3 cows received 4 different score.

Brushing Activity. Ten months prior to the experiment, six rotating brushes (swinging cow brush SCB, DeLaval International AB, Tumba, Sweden) were installed in the dairy farm, two in each cowshed. As shown in figure 1, one brush was installed close to the feed-bunk (3 m from the feed-bunk, "brush near the feed bunk") and the other on the opposite side of the shed (16 m from the feed-bunk, "brush away from the feed bunk"). This brush layout (one brush near and the other away from the feed source) makes it possible to assess the effect of the brush location on its utilization. As shown in previous studies (Mandel et al., 2017, 2013), increasing the cost (i.e. walking distance) involved in using the brush improves the ability to detect stress and morbidity. The brushes were equipped with a revolving head and a pivoting arm that allows them to move freely in different directions. The brush started revolving at a speed of 26 rpm when a mechanical pressure was applied to it and continued to rotate for 10s after the cow departed. Cows daily brush usage was collected automatically using a monitoring system validated during a previous study (Mandel et al., 2017). In order to minimize false registration of brush usage, i.e. when a cow was crossing under the brush but not using it, data was retained for analysis only if the following criteria was met: a cow was considered to be using the brush if present in a radius of 1m from the brush (the range of the infra-red beam) for at least 10 s, while the brush was rotating at least 1s during this time period (Mandel et al., 2017). Daily brush usage was collected from 209 lactating cows.

Milk yield, activity and rumination. Daily milk yield was recorded by the parlor milking system. Daily activity and rumination was collected continuously by HR-Tags (SCR Engineers Ltd., Netanya, Israel) collared to the cows neck.

Statistical Analysis

Statistical analysis was performed using R (version 3.0.2, R. Core Team, 2016). Linear and generalized linear mixed-effects models (lmer and glmer functions, lme4 library; Bates et al., 2015) were used to evaluate the outcome variables. Due to relatively small sample size, locomotion scores from the fourth ('lameness'; 14 ratings), and fifth category ('severe lameness'; 2 ratings) were merged into one category.

Association between locomotion scores and brush use, on the day of locomotion assessment, were analyzed in two ways, in order to identify the most sensitive method measurement. Brush use, as the outcome variable was analyzed as daily duration of brush usage (sec/d), and as daily occurrence (binary; 0: no use, 1: use at least once a day for each cow). While lameness score (4-level factor), brush location (near/away from feed bunk), and DIM [fitted as 1/log (DIM) based on Mandel and Nicol, 2017], and all possible interactions between these 3 factors, were the explanatory factors in each model. Lactation was not included in the model due to its overlap with cows' group. Cow identity nested within cows' group was used as a random effect, while date of observation was used as cross random effect.

The associations between locomotion scores and milk yield, rumination, and activity level collected on the day of locomotion assessment, as an outcome variable, were analyzed separately using 3 different models. Locomotion score and DIM (fitted as a quadratic term) was used as explanatory factors, while the random and crossed-random effect was as in the brush use model.

In each model, assessment of the explanatory factors and the interactions between them was made by comparing the model with and without the relevant explanatory factor, using likelihood-ratio tests (LRT). Non-significant terms were removed using a standard model simplification procedure (i.e. stepwise backwards elimination). The level indicating statistical significance was set at $\alpha = 0.05$. The residuals were checked graphically for normal distribution and homoscedasticity. To satisfy assumptions, a log transformation was used for daily duration of brush usage. Bonferroni correction was conducted for post-hoc pair-wise comparisons between non lame cows and cows with higher locomotion scores. The results are presented as model estimates and 95% confidence intervals (CI).

Table 1. Description of the 5-point ordinal lameness scoring system for dairy cows used in the study (adopted from Thomsen et al., 2008).

Score/level	Description of level
1- Normal	The cow walks normally. In most cases, the back is flat, both when the cow is standing and when walking. No signs of lameness or uneven gait. No signs of uneven weight bearing between legs. No signs of head bob when the cow is walking.
2- Uneven gait	The cow walks (almost) normally. In most cases, the back is flat when the cow is standing, but arched when walking. No signs of head bob when walking. The gait might be slightly uneven and the cow may walk with short strides, but there are no evident signs of lameness.
3- Mild lameness	Abnormal gait with short strides on 1 or more legs. In most cases, the back is arched both when the cow is standing and walking. In most cases, there are no signs of head bob when walking. In most cases, an observer will not be able to tell which leg is affected.
4- Lameness	The cow is obviously lame on 1 or more legs. An observer will, in most cases, be able to tell which leg is affected. In most cases, the back is arched both when the cow is standing and walking. In most cases, head bob will be evident when walking.
5- Severe lameness	The cow is obviously lame on 1 or more legs. The cow is unable, unwilling, or very reluctant to bear weight on the affected leg. In most cases, the back is arched both when the cow is standing and walking. In most cases, head bob will be evident when walking.

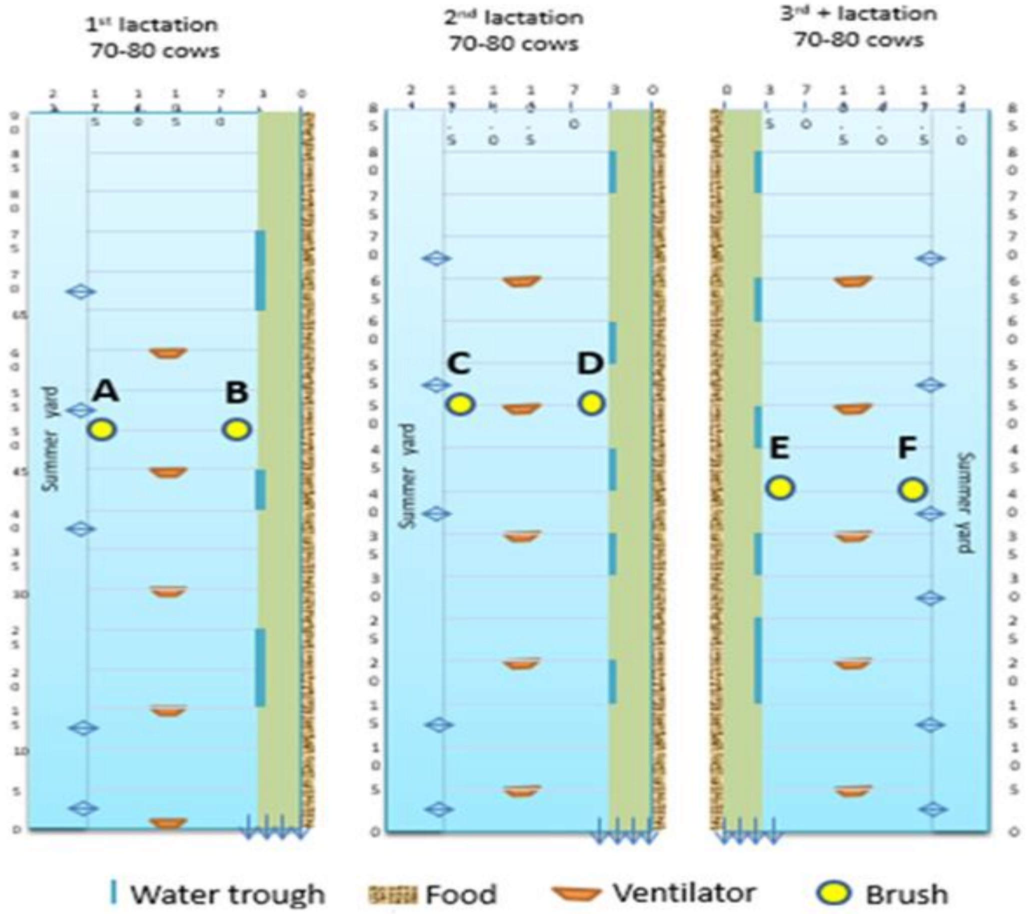


Figure 1. The layout of the experimental cow's sheds, displaying the location of the water troughs (), foodbank (), ventilators () and brushes ().

RESULTS

We found an interaction between locomotion score and brush location (near/away from feed-bunk) on both parameters of brush use, proportion of cows using the brush (χ^2_3 = 9.41, p = 0.025) and daily duration of brush usage (χ^2_3 = 11.19, p = 0.011). In order to understand more clearly the relationship and influence of these explanatory factors, and based on previous findings by Mandel et al 2013, 2017, we then split the data by brush location. For brushes installed away from the feed-bunk, we found significant association between locomotion scores and brush use in both parameters, daily proportion of cows using the brush (χ^2_3 = 24.15, p < 0.0001), and daily duration of brush usage (χ^2_3 = 9.92, p = 0.019).

Specifically, these two parameters of brush usage were lower among the lame and severely lame cows compared to non-lame cow (see Table 2 for model estimates). For brushes installed near the feed-bunk, locomotion scores was not statistically associated with neither daily proportion of cows using the brush, (χ^2_3 = 3.81, p = 0.28) nor daily duration of brush usage, (χ^2_3 = 6.70, p = 0.08).

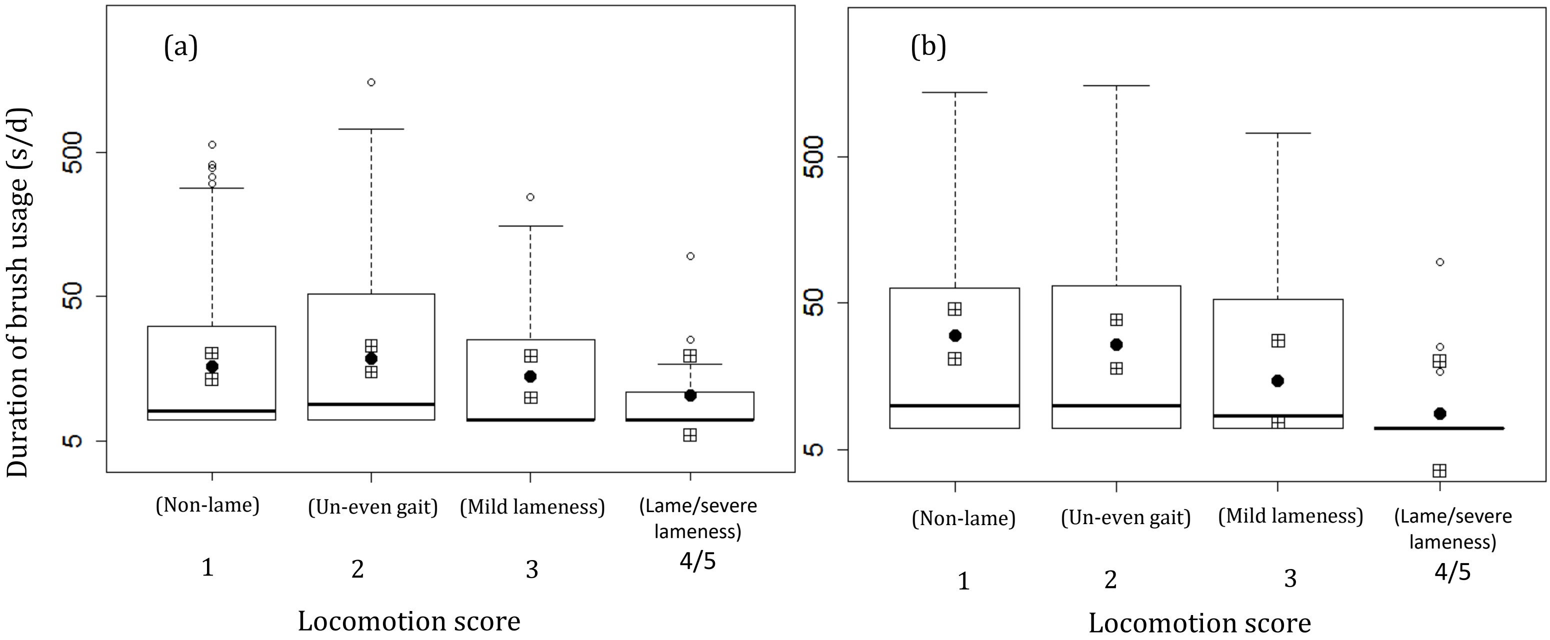
Locomotion scores were found to be associated with all three of the core activities, daily milk yield (χ^2_3 = 14.30, p = 0.026), daily rumination (χ^2_3 = 7.96, p = 0.047), and daily activity (χ^2_3 = 10.48, p = 0.015, (see Table 2 for means and pair-wise comparisons between lameness scores). Daily milk yield was found to be lower in cows with uneven gait and in lame and severely lame cow compared to non-lame cow (Table 2). While for daily rumination and daily activity, post-hoc comparisons revealed that there was no statistical difference between non lame cow and cows with uneven gait, mild lameness and lame and severely lame cows (Table 2).

Table 2. Association between locomotion scores and brush usage (near and away), milk yield, rumination and activity (model estimates with 95% CI in parentheses).

		Locomotion score[1]			
		1 Non-lame	2 Uneven gait	3 Mild lame	4+5 Lame and severely lame
Number of cows		154	159	60	10
Number of scores		672	620	128	16
Brush away from feed bunk	Proportion (cows/d)	0.24 (0.1-0.4)	0.21 (0.1-0.4)	0.17 (0.0-0.4)	0.0 (0.0-0.0)***
	Duration (sec/d)	30.3 (20.1-45.5)	26.0 (17.7-39.0)	22.5 (13.5-38.4)	8.8 (3.6-20.6)*
Brush near the feed bunk[2]	Proportion (cows/d)	0.24 (0.1-0.5)	0.23 (0.1-0.5)	0.20 (0.0-0.5)	0.08 (0.0-0.4)
	Duration (sec/d)	16.4 (13.4-20.3)	18.6 (15.0-22.8)	14.0 (9.9-19.4)	10.4 (5.5-19.4)
Milk	L/d	37.1 (32.9-40.7)	36.0 (31.9-39.8)*	35.5 (31.3-39.3)	32.9 (27.9-37.9)*
Rumination	Min/d	521.3 (497.8-546.4)	511.6 (487.2-535.3)	500.7 (472.3-527.8)	494.5 (453.9-535.7)
Activity	Per day	664.7 (621.9-708.7)	678.9 (635.2-722.1)	645.3 (595.1-695.0)	630.2 (561.6-701.0)

[1]Locomotion was assessed up to 14 times per cow (repeated-measures design).
[2]Post hoc comparisons were not carried out because none of the measures of brush usage (daily duration and daily proportion of cows using the brush) were significantly associated with locomotion scores in brushes located near the feed bunk.
Statistical significance of pair-wise comparisons with non-lame cows (locomotion score 1) after applying Bonferroni correction for post hoc multiple comparisons, *P < 0.05, ***P < 0.001.

Figure 2. Daily duration of usage (in sec) as a function of locomotion score for (a) brushes installed near the feed bunk, (b) brushes installed away from the feed bunk. Box plots represent raw data (minimum, lower quartile, median, upper quartile, maximum). Filled black circles represent model estimates and square plus represent upper and lower 95% confidence intervals. Lame and severely lame cows did not use the distant brushes during the days when locomotion scores were assessed.



DISCUSSION

In this study we assessed the role of monitoring brush use as an indicator of lameness in dairy cows.

Our results show a statistically significant decline in brush usage only among lame and severely lame cows compared to non-lame cows, and only in brushes that were installed away from the feed bunk but not in those installed near the feed bunk. Moreover, in contrast to our prediction, brush usage of mildly lame cows and cows with uneven gait (locomotion score 3 and 2, respectively), did not statistically differ from that of non-lame cows. With regard to the association between locomotion scores and core behaviors, we found lower daily milk yield among lame and severely lame cows (locomotion score 4+5), as well as in cows with uneven gait (locomotion score 2), compared to non-lame cows. Daily rumination and daily activity of cows with uneven gait, mild lameness, lame and severely lame (locomotion score 2-5) did not differ from that of non-lame cows.

Lame and severely lame cows did not use brushes that were located away from the feed-bunk at all, but continued to use brushes that were installed near the feed-bunk (see figure 2). Approaching the farther brush would require more effort than lame and severely lame cows may be willing to invest. These findings are compatible with previous studies, which showed that brush use is more indicative of stress and disease when brush is located away from the food source. For example Mandel et al., 2013 showed reduced brush usage on days of heat load (i.e stress) when food was located away from brush compared with days that food was located near the brush. The same pattern of reduced brush usage in brushes located away from the feed bunk has been shown in cows diagnosed with metritis (i.e disease; Mandel et al., 2017).

The fact that we observed a decline in brush use only in lame and severely lame cows, but not in cows with an uneven gait and mild lameness may be explained by the following reason. It has been suggested that stiffness in gait may be observed in certain conditions which may not be associated with pain, for example after recovery from joint injuries (Weary et al., 2006). If so it can be speculated that at least for some cows less severe locomotion score, which characterized in loss of normal gait functioning with no obvious lameness, might be observed regardless of pain. While higher locomotion scores tended to be associated with more chronic lesions which cause more pain that is not as easy to ignore (O'Callaghan et al., 2003). Therefore it can be assumed that the cost of experiencing pain, when walking a greater distance, for lame and severely lame cows, overcomes the motivation to engage in brush usage. While the motivation of mildly lame cows and cows with uneven gait to engage in brush usage, still exceeds the cost when walking a greater distance. Unlike our result, Weigele et al., 2018 study showed a reduced number of daily visit to the brush in moderately lame cows compared with non-lame cows. Their findings demonstrate the potential of monitoring brush usage for detecting lameness at an early stage. Unfortunately, they did not provide details on the location of the brush in relation to the food, which as mentioned before, has a considerable effect on brush use, and may reveal the difference compared to our result. Indeed one way which may improve the sensitivity of brush use as an indicator for less severe lameness, is by increasing the cost involved in such activity. That is by installing the brush even farther from the feed bunk. It may be the case in Weigele et al., 2018 study which may reveal the different result compared to our study. However, this would also make the brush less accessible for cows as an enrichment tool (Mandel et al., 2016). The study's results reveal a decrease in milk yield in lame and severely lame cows, as well as cows with uneven gait compared with non-lame cow. Many studies have previously assessed the effect of lameness on milk production. Some studies show significant negative association between lameness in cattle and milk yield (Kocak and Ekiz, 2006; Van Hertem et al., 2013; Warnick et al., 2001), while others showed no significant association (Archer et al., 2011; Thorup et al., 2016). Thus, conflicting findings suggest that milk yield is an inconsistent indicator for lameness in cows.

In this study, daily rumination did not statistically differ between cows with different locomotion scores. Given limited energy reserves in cases of lameness, it might be more beneficial for cows to invest in core activities such as eating than brushing activity. Same as milk yield, evidence in the literature regarding the association between lameness and rumination are inconclusive. Van Hertem et al., 2013 report a negative association between lameness and rumination activity in cows, while Thorup et al., 2016 and

Weigele et al., 2018 report that lameness has no significant effect on rumination activity. Moreover Thorup et al., 2016 found that the lame cows reduce daily feeding time and feeding frequency, but it did not affect the cows' daily consumption of dry matter. It seems that lame cows tend to compensate by having a faster rate of eating. As expected for core behavior, the lame cow rumination activity tends not to change, because the cow alters her eating behavior as compensation (Walker et al., 2008).

Our results show no significant association between locomotion scores and cows' daily activity. The reasons for that can be the expression of lameness, severely lame cows tend to take smaller steps, accompanied with an expressive head bob, hence they make more steps to cover the same distance (Van Hertem et al., 2013). In addition, the lame cows would try to restrict their movements by lying down as close to the pen entrance as possible upon their return from the milking parlor (Juarez et al., 2003). Therefore, when taken thus two finding in consideration, overall activity level of lame and non-lame cows may not differ. Unlike the results of our study, most studies show decrease in the activity of lame cows (Reader et al., 2011; Van Hertem et al., 2013; Weigele et al., 2018). Nevertheless, those studies eventually came to conclude that activity level is not sensitive enough as an indicator to detect lameness.

In conclusion, our results show that brush use, by itself, is not sensitive enough tool to detect the very mild change in cow's behavior that occurs in early stages of lameness. Monitoring brush use, when installed away from the food bunk, can be useful as an indicator for lameness in cases of lame and severely lame cows, but its ability to detect mild lameness and uneven gait is less promising in this stage. Such a tool could potentially be useful as a lameness indicator in situations when other monitoring measures such as milk yield, are not available or possible (heifer or dry cows). Moreover, it appears from our results, that milk yield is also not a reliable (due to its inconsistent behavior in the different locomotion scores) measurement by itself for detection of lameness. However a combination of core behavior together with luxury activity, such as brush use, may improve our ability to detect lameness even in early stage.

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