

ASSESSING COW COMFORT USING LYING BEHAVIOUR AND LAMENESS

by

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Abstract

Over the past decade, there has been growth in scientific research on welfare in modern dairy production systems. The issue of cow comfort and how it relates to the risk of lameness has received considerable interest. The objectives of this thesis were to establish reliable methods of using lying behaviour as a measure of cow comfort, to describe the variation in lying behaviour of individual cows within farm and between farms, and to evaluate the relationship between stall comfort, lying behaviour, and lameness. A cross-farm assessment was conducted on 43 commercial dairy farms in the Fraser Valley region of British Columbia. Electronic data loggers recorded lying behaviour of 2033 cows at 1-min intervals for 5 days. The first study established that monitoring at least 30 cows per farm for 3 days provides an accurate estimate of the lying behaviour of the lactating cows at that time. Cows averaged 11 h/d lying down, separated into 9 bouts/d with an average duration of 88 min/bout. Cows were scored for lameness using a 5-point Numerical Rating System (NRS) in which 1 = sound and 5 = severely lame. A subsample of 1319 cows from 28 farms using either deep-bedded stalls ($n = 11$) or mattress stalls ($n = 17$) were used for the second study. Overall, 21% of the cows were scored as NRS = 3 and 7% as NRS = 4; no cow was scored as NRS = 5. Mattress farms had higher prevalence of NRS = 4 compared to deep-bedded farms (9 vs. 4%, respectively). Cows with NRS = 4 housed on deep-bedded stalls spent 1.6 h/d more lying, and had longer bouts compared to cows with $\text{NRS} \leq 3$, but there were no behavioural differences among cows with different degrees of lameness housed on mattress stalls. Extreme lying behaviour, particularly the high lying times (≥ 14 h/d) and long lying bouts (≥ 99 min/bout) were associated with increased odds of lameness, regardless of stall surface. Stall comfort, lying behaviour, and lameness are interlinked, and should all be integrated as measures of cow comfort.

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List of Abbreviations

BCS = body condition score

CCI = cow comfort index

DB = deep-bedded

DHIA = Dairy Herd Improvement Association

DIM = days in milk

MAT = mattress

NAHMS = National Animal Health Monitoring System

NRS = numerical rating system

PMR = partial mixed ration

SD = standard deviation

SE = standard error of the mean

SUI = stall use index

TMR = total mixed ration

USDA = United States Department of Agriculture

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Kiyomi Ito, and Drs. Marina von Keyserlingk and Dan Weary designed the project collaboratively. Data collection, data analysis, interpretation, and manuscript preparation was performed by Kiyomi Ito under the supervision of Drs. Marina von Keyserlingk, Dan Weary (Chapters 2 and 3), and Stephen LeBlanc (Chapter 3).

CHAPTER 1: General Introduction

1.1. Introduction

Dairy production has become increasingly intensified. The number of dairy farms in Canada decreased from 24,603 farms in 1995 to 13,214 farms in 2009, while the number of cows remained relatively consistent (1.3 million to 1.0 million) and the production of milk increased from 7.2 billion to 7.6 billion litres per year (Canadian Dairy Information Centre, 2009). This increasing farm size and production per cow has been supported by technological changes in indoor housing systems where cows are kept inside the barn year around and fed a high energy ration to meet the nutritional requirements of high producing dairy cows. Two main dairy systems used in Canada are free-stall housing and tie-stall housing. In free-stall housing, more common in the western Canada, cows are housed in groups in indoor pens with access to a lying area and a feeding area. In tie-stall housing, more typical in the eastern Canada, cows are tied by a chain around the neck to the stall where they are fed and milked individually. Other types of intensive production systems include dry lot and bedded pack, in which cows are group housed, as in free-stall systems, but have an open area for lying down instead of stalls. Such intensive systems allow for better control over management and production compared to more extensive systems, but are also associated with a number of problems with regards to the health and welfare of the dairy cows.

Conceptions of animal welfare stem from three ethical questions over the quality of life of animals: 1) is the animal functioning well (biological functioning); 2) is the animal feeling well (affective state); and 3) is the animal able to live a natural life (natural living) (Fraser et al., 1997)? The criteria for judging the welfare of an animal depend on the priorities people place on each of these questions. Traditionally, producers and veterinarians have judged the welfare of

the animals based primarily on health conditions, as injuries and illnesses compromise production, which directly affects the viability of the farm. However, many people are also concerned about the emotional state of the animal, including negative emotions such as pain, fear, and distress, and the ability to experience positive emotions such as pleasure from opportunities to play and socialize. Furthermore, some people also question the ‘unnaturalness’ of the modern production systems, explaining some of the growth in the organic systems that are perceived to be more natural. The three questions are often overlapping. For instance, dairy cows that are kept under intensive production systems (unnatural living) may become lame (compromised biological functioning) which likely causes discomfort and pain (negative affective state) (von Keyserlingk et al., 2009).

Over the past decade, there has been growth in scientific research on welfare in modern dairy production systems, providing practical solutions that address all three aspects of animal welfare (von Keyserlingk et al., 2009). Among these, the issue of cow comfort and how it relates to the risk of lameness has received considerable interest.

1.2. Scientific Assessment of Cow Comfort

Cow comfort is an emotional state; in other words, it reflects how cows feel. Any factor related to how the barn is designed and managed, including but not limited to the lying area, influences how comfortable cows may be. For example, a cow can stand more comfortably on pasture than on concrete; a heat stressed cow may feel uncomfortably hot; and a lame cow is likely less comfortable than a healthy cow. Producers may spend millions of dollars building housing systems, with the aim of providing a comfortable environment for their animals - one that ensures protection from climatic extremes, ensures adequate rest, and minimizes the risk of injuries and diseases. Despite the effort, housing systems do not always function well from the

cows' perspective. Assessments of cow comfort typically involve one of three methods: 1) preference testing, 2) detailed analysis of behaviour, and 3) assessment of injuries and diseases.

1.2.1. Preference testing

Preference testing is a research method used to assess the subjective experiences of animals (i.e. how they feel about their environment). For instance, evaluating the amount of time cows spend using free-stalls with different features can help identify which features are important and which option cows prefer given the choice. When given free access to different stall surfaces, cows preferred deep-bedded stalls (sand or sawdust) over poorly bedded mattress stalls, spending over 90% of their time lying on their first choice (Tucker et al., 2003). Furthermore, cows also preferred ample bedding to bare mattresses (Tucker and Weary, 2004) and dry bedding to wet (Fregonesi et al., 2007b). In contrast, cows' preference for stall configuration was less clear; cows showed no clear preference for the position of the neckrail (Tucker et al., 2005) or stall size (Tucker et al., 2004) with individual cows preferring different options. These findings indicate that the lying surface of the free-stall is a particularly important feature of stall comfort from the cows' perspective.

1.2.2. Analysis of lying behaviour

An alternative approach is to analyze the cows' behaviour when they have only one option. The first step in using behaviour to assess comfort is to establish what behaviours change when cows are comfortable and uncomfortable. Haley et al. (2000) demonstrated that cows housed in individual large box pens with a mattress flooring bedded with straw lay down 4.2 h more per d and stood up and lay down more often than cows housed in conventional tie-stalls with concrete flooring also bedded with straw. Similarly, cows spent 12.2 h/d lying down

when provided with softer geotextile mattress compared to 10.4 h/d on hard concrete surface; they also changed position from lying to standing more frequently on mattress than on concrete (Haley et al., 2001). These studies identified the amount of time cows spend lying and the frequency of lying events as sensitive behavioural measures of stall comfort.

A series of experiments have established that stall features such as the lying surface and configuration affect stall usage and behaviour, including those indicative of stall comfort. For example, cows spent more time lying down and had higher frequency of lying events on their preferred lying surfaces: well bedded stalls over poorly bedded mattresses (Tucker et al., 2003; Tucker and Weary, 2004), suggesting that the cows are choosing the option that is more comfortable. Drissler et al. (2005) found that lying time decreased progressively when sand bedded stalls were poorly maintained; supporting the idea that ample bedding contributes to stall comfort. Moreover, when the stall was bedded with dry sawdust, cows spent 13.8 h/d lying down which reduced to 8.8 h/d when the bedding was wet (Fregonesi et al., 2007b). Cows also spent less time lying down and had shorter lying bouts in narrow stalls and in stalls with brisket board, suggesting that the contact with stall partitions while lying down causes discomfort and consequently reduce the duration of lying bouts (Tucker et al., 2004; 2006). Cows' lying behaviour also responds to changes in management. For instance, increasing stocking density from 100% (1 cow: 1 stall) to 150% (1.5 cows: 1 stall) resulted in a reduced lying time from 12.9 h/d to 11.2 h/d (Fregonesi et al., 2007a).

Free-stalls provide not only a place to lie down but also a place to stand, away from the concrete alley. The position of the neckrail did not affect lying time, but it restricted cows from standing with all four feet inside the stall, increasing the time cows spent standing with only two feet inside the stall (Bernardi et al., 2009; Fregonesi et al., 2009). Understanding cows'

behaviour in relation to facility design and management essentially provides a way to evaluate the housing environment from the cow comfort perspective.

1.2.3. Injuries and diseases

Features of the free-stalls are important risk factors for hock lesions and knee injuries. For example, Weary and Taszkun (2000) found that 73% of lactating dairy cows housed in free-stalls in British Columbia had at least one hock lesion, but these lesions were the most prevalent on farms using mattresses (91% of cows) and least prevalent on farms using sand bedding (24% of cows). Mattress stalls have been repeatedly associated with higher prevalence of hock lesions (Wechsler et al., 2000; Fulwider et al., 2007). In addition, a concrete stall surface can cause swollen knees due to impact when cows lie down (Rushen et al., 2007). These injuries directly reduce cow comfort, and may have long-term consequences including lameness.

Free-stall systems in general are associated with higher rates of lameness (Cook and Nordlund, 2009). For example, Cook (2003) found that lameness prevalence (during winter when the prevalence is highest) was 20% in tie-stalls compared to 28% in free-stalls. Haskell et al. (2006) reported a lameness prevalence of 17% in herds housed in free-stalls but allowed seasonal access to pasture compared to 39% in herds housed strictly in free-stalls. Straw yards (Somers et al., 2003) and bedded backs (Barberg et al., 2007) also tend to have lower rates of lameness. Within free-stall systems, lameness prevalence was 28% in herds using non-sand stalls compared to 17% in herds using sand stalls (Espejo et al., 2006). Furthermore, lameness was more prevalent when cows were exposed to concrete flooring (e.g. Somers et al., 2003; Vanegas et al., 2006). More specifically, lame cows recovered in a few weeks after allowed access to pasture (Hernandez-Mendo et al., 2007) and when the neckrail was removed from free-stalls (Bernardi et al., 2009). These studies showed that the design and management of housing

systems could affect the risks of injuries and lameness. Thus, evaluating the prevalence of injuries and diseases is also a promising method of assessing the housing environment with respect to cow comfort.

1.3. Lameness

1.3.1. The problem

Lameness is widely recognized as a serious production and animal welfare issue in the dairy industry. Recent studies have estimated that about 25% of lactating dairy cows in North America are clinically lame. For example, the prevalence of lameness was 28% during the winter and 23% during the summer among 15 free-stall herds in Wisconsin (Cook, 2003), and 25% among high production cows housed in free-stalls in Minnesota, ranging from 3 to 57% across farms (Espejo et al., 2006). Further, Cramer et al. (2008) found that 47% of cows in free-stall herds had lesions in at least one foot, and Bicalho et al. (2007a) found that 13% of cows in a free-stall herd had a painful lesion (i.e. reaction to digital pressure applied to the lesion). A recent study estimated lameness prevalence to be as high as 48%, ranging from 0 to 81%, among 33 free-stall herds in Germany (Dippel et al., 2009). Lameness is a significant economic problem as it results in reduced milk yield (Warnick et al., 2001; Green et al., 2002; Bicalho et al., 2008), reduced fertility, and increased risk of premature culling (Garbarino et al., 2004; Bicalho et al., 2007b). Lameness is often considered the biggest welfare concern for dairy cows due to pain associated with the injuries and its high prevalence (Whay et al., 2003).

1.3.2. Risk factors

Lameness a multi-dimensional problem triggered by a combination of factors: nutrition, hormonal changes around calving, external trauma, and infectious diseases of the hoof (Cook

and Nordlund, 2009). In the past 20 years, considerable work has gone into identifying risk factors for lameness associated with housing systems and management practices. Herd-level risk factors for lameness, within the free-stall system, include: stall features (Philipot et al., 1994; Espejo and Endres, 2007), lying surface (Cook, 2003; Espejo et al., 2006), overcrowding (Leonard et al., 1996), increased time spent away from the pen for milking (Espejo and Endres, 2007), and the use of automatic alley scrapers (Barker et al., 2007). Dippel et al. (2009) found that reduced lying comfort, measured by the frequency of abnormal lying down and rising behaviours (i.e. interrupted movements, lying down or standing up taking longer than 20 s, lying down with hindquarters first, or rising with forequarters first), was also a risk factor for lameness. It is generally understood that increased exposure to hard flooring surfaces between periods of rest, combined with reduced rest due to an uncomfortable lying environment, collectively contribute to lameness (Cook and Nordlund, 2009).

Within the same housing environment, some individuals are more susceptible to lameness than others. Older cows are generally at higher risk (Espejo et al., 2006; Haskell et al., 2006; Bicalho et al., 2007b; Dippel et al., 2009), and incidence of lameness peaks at 3 to 4 months into lactation (Green et al., 2002). Thinner digital cushion (Bicalho et al., 2009), higher milk production at the beginning of lactation (Green et al., 2002; Bicalho et al., 2008), and low body condition (BCS < 3.0) before and at calving (Hoedemarker et al., 2009) have also been identified as cow-level risk factors for lameness. Low body condition (BCS < 2.5) has been repeatedly associated with increased risk of lameness, though it is unclear whether this is a risk factor or a consequence of lameness (Espejo et al., 2006; Dippel et al., 2009). In a study examining behavioural risks for lameness, Galindo et al. (2000) found that cows that became lame spent more time standing with only two front feet in the stall compared to cows that did not

become lame (6.2% vs. 5.6% of the day, respectively). Galindo and Broom (2000) also found that the incidence of lameness was related to standing behaviour such that the number of new cases of lameness was higher among cows that spent > 45% of the day standing, and those that spent > 10% of the day standing with two feet in the stall. The knowledge of risk factors helps in the design and management of facilities to reduce the risk of lameness, and also helps identify at-risk individuals before lameness becomes a problem.

1.3.3. Lameness detection

Early detection is the key to minimizing the negative impact of lameness. Behavioural assessment such as visual observations of cow gait is usually the first line of lameness detection before the causes of lameness (e.g. hoof lesions) are diagnosed through hoof trimming (Bicalho et al., 2007a; Chapinal et al., 2009). Several methods of gait scoring have been developed. Sprecher et al. (1997) developed a 5-point scale where 1 represents a sound cow with normal locomotion and 5 represents a severely lame cow with an inability or extreme reluctance to bear weight on one or more limbs. Further, Flower and Weary (2006) identified six specific behavioural criteria for assigning a gait score (Numerical Rating System; NRS) including 1) back arch, 2) head bob, 3) tracking of the front and back feet, 4) joint flexion, 5) asymmetric gait, and 6) reluctance to bear weight. Gait scoring identifies the subtle changes in cows' locomotion caused by pain associated with the underlying injuries; Flower et al. (2008) demonstrated that lame cows improved their gait score after receiving a dose of analgesic (ketoprofen) providing evidence that lameness is driven by pain. This system has been validated for reliability within and between observers, and for sensitivity to the presence of hoof injuries (Flower and Weary, 2006; Chapinal et al, 2009). Similar systems of gait scoring have been successfully used in epidemiological studies as methods of lameness detection (e.g. Cook, 2003;

Whay et al., 2003; Espejo et al., 2006). Gait scoring is a practical tool that can also be used by veterinarians and producers to identify lame cows before the problem becomes severe.

1.3.4. Lameness prevention

Early detection provides a tool for minimizing the harm due to lameness, but how can we prevent cows from becoming lame in the first place? Cow comfort is thought to play an integral part in lameness dynamics. Physical comfort of the housing environment and certain behaviours influence the risk of lameness, but lameness also affects the comfort of the lame cows leading them to modify their behaviour, which in turn may influence the severity and duration of the lameness event (Singh et al., 1993; Cook and Nordlund, 2009). In general, the less comfortable the stalls are, the longer the cows spend standing on slurry-covered concrete in the alley, which makes them more susceptible to hoof injuries and diseases especially if inadequate rest compromises their immune function (Cook and Nordlund, 2009).

1.4. On-farm Assessment

A useful starting point for addressing welfare issues is to benchmark the industry through on-farm assessments. There are several different parties that may be interested in animal welfare including producers, consumers, and the public; the welfare assessment can serve a different purpose for each group. For example, information gathered through welfare assessment can be used as an advisory tool to aid the producers in decision-making about their facilities and management practices. This allows the producers to identify specific areas of concern, and helps plan how to achieve the targeted level. Some consumers rely on certification programs (e.g. organics and welfare labeling) for assurance that the animals are raised according to animal welfare standards. Similarly, many citizens expect the authorities to ensure that farm animals are

raised humanely. Benchmarking can provide a backbone for developing effective and reasonable policies. Auditing for compliance to these standards and policies also requires a form of on-farm welfare assessment.

There are two broad approaches to welfare assessment: environment-based measures, and animal-based measures. For example, the “Animal Needs Index” (ANI) developed in Europe considers five husbandry conditions: 1) possibility of mobility, 2) social contact, 3) condition of flooring for lying, standing, and walking, 4) climate (including light, ventilation, and noise), and 5) human attitude (Bartussek, 1999). ANI value is generated as the total sum of points (-0.5 to +3.0) awarded for 24 criteria in each component, so that deficiency in one category can be compensated by success in another, as long as minimum standards in all of the components are met (Bartussek, 1999). Evaluation of environmental characteristics such as this can compare systems; for instance, free-stall systems by default would receive higher scores than tie-stall systems for possibility of mobility and social contact. However, considerable variation exists within system and also among individual cows within farm, and therefore welfare assessment should include measures that can be used within and across systems.

The University of Bristol in the United Kingdom has developed an animal-based welfare assurance program (Leeb et al., 2004). This system focuses primarily on animal-based measures, including physical conditions, behavioural observations, and lameness prevalence (Whay et al., 2003). These animal-based measures reflect the actual outcome of the environmental conditions regardless of which system is used; this method also allows for the assessment of individual animals within farm. A combination of environmental and animal-based measures may be most effective in developing a comprehensive on-farm assessment.

In recent years, there has been an increasing interest in the dairy industry about cow comfort and its link to lameness. A growing body of research has now confirmed that facility design and management has a major impact on lameness, which in turn affects cow welfare and longevity. Despite the knowledge in these areas, practical application of research findings requires additional work. For instance, the methods of assessing lying behaviour as described in earlier section may not be practical on commercial farms since it requires detailed observation of individual cows for extended periods of time. As well, lameness detection on commercial farms has been a challenge. For example, prevalence of lameness reported by herd managers was found to be three times lower than that estimated by researchers (8% compared to 25%) for the same groups of cows (Espejo et al., 2006). Gait scoring requires training and an additional time commitment, which may not always be possible on farms, especially as farm size continues to increase. This suggests that there is a need to establish practical method of assessing cow comfort and lameness that are scientifically valid and at the same time practical for producers on-farm.

1.5. Objectives

The objectives of this thesis were: to establish reliable methods of using lying behaviour as a measure of cow comfort, to describe the variation in lying behaviour of individual cows within farm and between farms (Chapter 2); and to evaluate the relationship between stall comfort, lying behaviour, and lameness (Chapter 3).

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CHAPTER 2: Lying Behaviour of Free-stall-housed Dairy Cows¹

2.1. Introduction

Dairy cows are highly motivated to lie down for approximately 12 h/d (Jensen et al., 2005) and lying is a higher priority behaviour than eating and social contact when opportunities to perform these behaviours 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 behaviour, 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 to a concrete stall base (Haley et al., 2001), and on deep-bedded surfaces compared to inadequately bedded mattresses (Tucker et al., 2003). Similarly, cows spend more time lying down and have longer bouts in wider stalls (132 cm 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/d 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 behaviour continuously over a few days, either 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 labour

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intensive. Alternatively, some studies have used 10-min, 15-min or even 1-h instantaneous scan sampling, recording the proportion of the group of animals that was 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 behaviour 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 behaviour of the entire group. Methods of sampling lying behaviour 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 Stall Standing Index (proportion of cows touching a stall that are standing or perching) (Cook et al., 2005). Cow comfort index was linked to lameness prevalence (Espejo and Endres, 2007), and was used to assess stall usage at a range of stocking densities (Krawczel et al., 2008). The National Animal Health Monitoring System (NAHMS) of the USDA estimated CCI and SUI on participating farms during the national 2007 cow comfort survey of the dairy industry. Despite the reliance upon these indices it is not clear if these indices provide reliable estimates of lying behaviour in dairy cows.

The diurnal pattern of lying behaviour 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) was observed (Overton et al., 2002). These authors recommended that

walk-through assessments be undertaken 1 h after the return from morning milking to capture maximum lying behaviour. 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 behaviour on farm.

Electronic data loggers are widely available and can be used to accurately measure lying behaviour including the total time spent lying down, the frequency of lying bouts, and the duration of each bout for individual cows (O'Driscoll et al., 2008). One aim was to determine how these loggers should be used to provide accurate estimates of lying behaviour for a herd, specifically investigating how the number of days and focal animals sampled affects the estimate of the herd mean. A 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 behaviour. The final aim was simply to describe within- and between-herd variation in measures of lying behaviour on commercial dairy farms using free-stall housing.

2.2. Materials and Methods

2.2.1. 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 PMR (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 (GEA Westfalia Surge GmbH, Bönen, Germany) software, but had no individual production records, and 5 farms had no production records available. The average herd size was 170 ± 80 (\pm SD, ranging from 71 to 511) milking cows producing $10,548 \pm 800$ (ranging from 8,991 to 12,080) kg annually (based on annual yield estimated by DHIA or equivalent value derived from VAMPP and DairyPlan). The majority of the farms ($n = 37$) milked twice daily, while the rest ($n = 6$) milked 3 times daily; and the majority ($n = 34$) fed once daily, while 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 there were a variety of other types including concrete, rubber mat, tires, wood, or a combination of multiple types ($n = 14$); stalls were bedded with sawdust ($n = 33$), sand ($n = 9$), or chopped straw ($n = 1$).

2.2.2. Data collection

Each farm was visited twice, with 5 d between visits. As heat stress is known to affect lying time (Cook et al., 2007), the data collection period was limited to days where the maximum temperature was < 25 °C. The maximum temperature across study days was 9.5 ± 5.3 °C (ranging from -1.7 to 24.5 °C). Upon arrival 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 vs. low producing cows, or multiparous vs. primiparous cows. The selected group size was 94 ± 31 (ranging from 32 to 187) cows, and the stall stocking rate was 104 ± 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 count of the number of cows lying down,

standing fully in the stall, standing with only two front feet in the stall, and feeding; these measures were used to calculate the CCI and SUI. Another 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 behaviour 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 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-axis at 1-min intervals for 5 d, pre-programmed to start at midnight following 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 g force readings into degrees of tilt. These data were exported into Microsoft Excel, 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 while readings $\geq 60^\circ$ indicated the cow lying down. A macro was used to calculate daily standing time (min/d) and the frequency of standing bouts (no./d) based on 1,440 observations from midnight until midnight the following day. Standing and lying bouts of ≤ 2 min were ignored, as these reading 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 (min/bout) 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 23:30 until 00:30 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.

Prior to 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 behaviour 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 position at 1-min intervals. 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 behavioural variables following the same methodology described above for the HOBO loggers. Measures of daily lying time (min/d) and bout frequency (no./d) 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 behaviour data; 76 cows were removed because they were either sold, became sick, moved to a different group during the assessment period, or 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 dataset. The final dataset consisted of 43 farms and 2,033 cows. On average, 47 ± 5 (ranging between 26 and 50) cows were sampled on each farm for 4.9 ± 0.4 (ranging

between 3 and 5) d. The focal cows averaged 2.6 ± 1.4 (ranging from 1 to 12) lactations and 150 ± 94 (ranging from 11 to 704) DIM at the time of assessment.

2.2.3. Data analyses

In order to determine how sample size affected the estimate of lying behaviour, 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) 5, 4, 3, 2, 1 d per cow, and b) 44, 40, 30, 20, 10, 5, and 1 cow(s) per farm, using the SURVEYSELECT procedure in SAS (SAS Institute Inc., 2004). Simple random sampling without replacement (method=SRS; seed=0) was used with strata specified as a) farm cow, or b) farm. Estimates of the mean lying time (h/d) and mean bout frequency (no./d) based on each subset were calculated using the SUMMARY procedure, and the relationship between the overall mean (based on 5 d and 44 cows per farm) and each estimate was tested using regression (REG; model df = 1). Appendix 1 provides examples of the SAS codes used for this analysis. The complete procedure was repeated 10 times for each subset, and the 10 R^2 values were used to estimate a mean R^2 and SD 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 (REG; model df = 1).

2.3. Results

2.3.1. Measuring behaviour

The estimate of the overall mean lying time (h/d) and bout frequency (no./d) based on the 5 d of observations declined progressively when fewer days were available (Figure 2.1). Measures of lying time (h/d) and bout frequency (no./d) based on 3 d of data provided excellent estimates of the overall means ($R^2 = 0.94$ 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 lying time (h/d) and bout frequency (no./d) declined when estimates were based on fewer cows per farm (Figure 2.2). Estimates of lying time (h/d) and bout frequency (no./d) based on 30 cows provided a reasonable estimate of the overall means ($R^2 = 0.88$ and 0.90 , respectively), but this relationship was less when estimates were based on 20 cows ($R^2 = 0.75$ and 0.82 , respectively) and declined further when the estimates were based on only 10, ($R^2 = 0.54$ and 0.60 , respectively), 5 ($R^2 = 0.39$ and 0.39 , respectively) and 1 cow ($R^2 = 0.08$ and 0.08 , respectively) per farm.

2.3.2. Reliability of cow comfort indices

The mean CCI was 75 ± 10 (ranging from 50 to 92) %, and the mean SUI was 58 ± 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 and the mean lying time (h/d) derived from continuous monitoring of the focal cows from the same group (Figure 2.3A; $R^2 = 0.00$, $P = 0.10$). The CCI was mildly associated with the mean bout frequency (no./d; Figure 2.3B; $R^2 = 0.16$, $P = 0.01$) and mean bout duration (min/bout; Figure 2.3C; $R^2 = 0.09$, $P = 0.05$). The SUI was not associated with lying time (h/d), bout frequency (no./d), or bout duration (min/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).

2.3.3. Lying behaviour – within- and between-herd variation

Across farms, cows spent, on average, 11.0 ± 2.1 h/d lying down separated into 9 ± 3 bouts/d. The farm means varied from 9.5 to 12.9 h/d and 7 to 10 bouts/d, while the individual means varied from 4.2 to 19.5 h/d and 1 to 28 bouts/d (Figures 2.4 and 2.5). The duration of lying bouts across farms averaged 88 ± 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 SD in lying time varied from 1.5 to 3.3 h/d. Similarly, the SD in bout frequency and bout duration varied from 2 to 4 bouts/d, and 17 to 49 min/bout.

2.4. Discussion

2.4.1. Measuring behaviour

Two aspects of sampling procedure must be considered when deciding on a method of measuring behaviour: 1) the sampling rule that specifies which subjects to observe, and 2) the recording rule that specifies how the behaviour 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 behaviour 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 influences the accuracy of measures of lying behaviour 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, Weschler 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 provides 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 accurately estimate lying behaviour. Even a sampling period of 2 d yielded

reasonable estimates with about 90% accuracy and a single d sampling yielded estimates with about 75% accuracy compared to the overall mean based on 5 d.

Variation among individual cows was considerable, such that the number of focal animals required to accurately estimate the farm mean was high. Focal sampling is generally the best approach for studying groups (Martin and Bateson, 1993), as 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 behaviour with about 90% accuracy; this was reduced to about 80% when the sample size decreased to 20, and less than 60% when the sample size decreased to 10 cows. From these results, we suggest that farm estimates of lying behaviour be obtained from at least 30 cows per group. Herd estimates appeared to plateau between 30 and 40 cows, suggesting that sampling additional cows would provide little extra information.

2.4.2. Reliability of cow comfort indices

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%, similar to values reported in WI and MN (Cook et al., 2005; Espejo et al., 2006). The 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.,

2005). Stall use index is generally higher when cows have access to more space and free-stalls (Krawczel et al., 2008), but the stocking densities in the farms in our study were comparable to those studied by Overton et al. (2002) and Cook et al. (2005).

Cow comfort index and SUI are frequently used as practical indicators 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 frequency 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 also found no association. In combination, these results indicate that CCI and SUI do not provide an accurate estimate of lying behaviour, and likely should not be used for on-farm assessments of this behaviour.

2.4.3. Lying behaviour – within- and between-herd variation

Mean lying times (11.0 h/d) in the current study were similar to values previously recorded on commercial farms using free-stall housing (Wechsler et al., 2000; Cook et al., 2005), although the frequency of lying bouts (9 bouts/d) was lower and the duration of individual bouts (88 min/bout) was longer compared to values reported by Wechsler et al. (2000; 12 to 15 bouts/d and 53 to 67 min/bouts). Lying behaviour 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 behaviour is influenced by her social ranking (Galindo and Broom, 2000) as well as production and health status (Fregonesi and Leaver, 2001; Walker et al., 2008). Variation in individual behaviour 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 daily lying time of 7.5 h/d for heifers housed at 2:1 stocking density (2 animals: 1 stall) with values ranging from 2.7 to 11.9 h/d for individual heifers. Overcrowding could contribute to a high variation in individual lying bouts due to increased opportunities for displacements from the stalls disrupting the normal lying behaviour (Fregonesi et al., 2007a). Another potential source of variation is estrous behaviour. In the present study there was no way of systematically accounting for estrus, but future work studying the effects of estrous activity on lying behaviour would be beneficial. Regardless of the cause, individual lying behaviours of cows housed together can be highly variable. The large cow-to-cow variation reduces the statistical power of tests relying on between-cow comparisons; within-cow comparisons are likely more sensitive in detecting management or design changes expected to affect lying behaviour.

2.5. Conclusions

Reliable estimates of lying behaviour 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 lying time, bout frequency, or bout duration; and thus, cannot be recommended as methods of assessing this behaviour. The range in lying behaviour among individual cows within farms was greater than differences across farms.

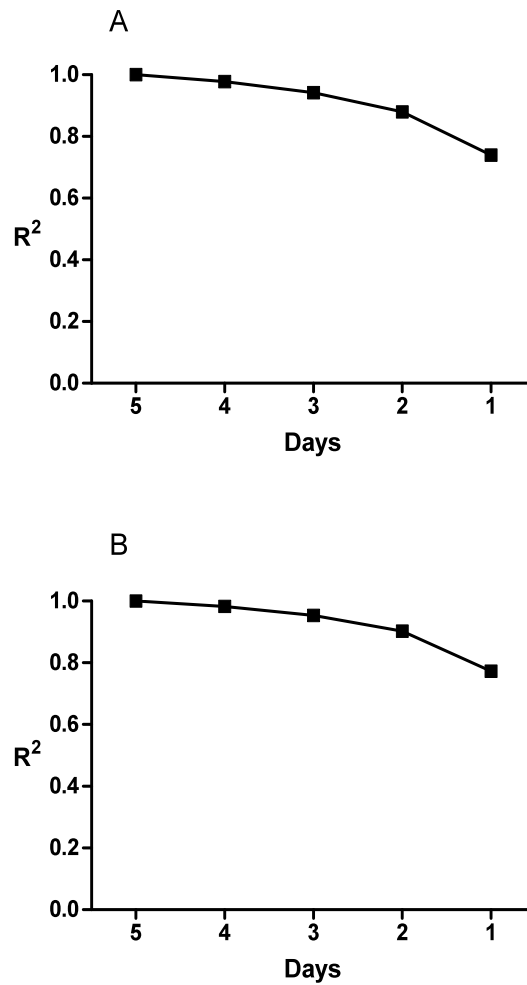


Figure 2.1. The relationship (R^2) between (A) mean lying time (h/d) and (B) mean bout frequency (no./d) based on 5 d per cow and estimates derived from 4, 3, 2, or 1 d randomly sampled for each cow ($n = 1,818$ cows; 38 farms). The SDs across the 10 random sampling events are too small to be visible.

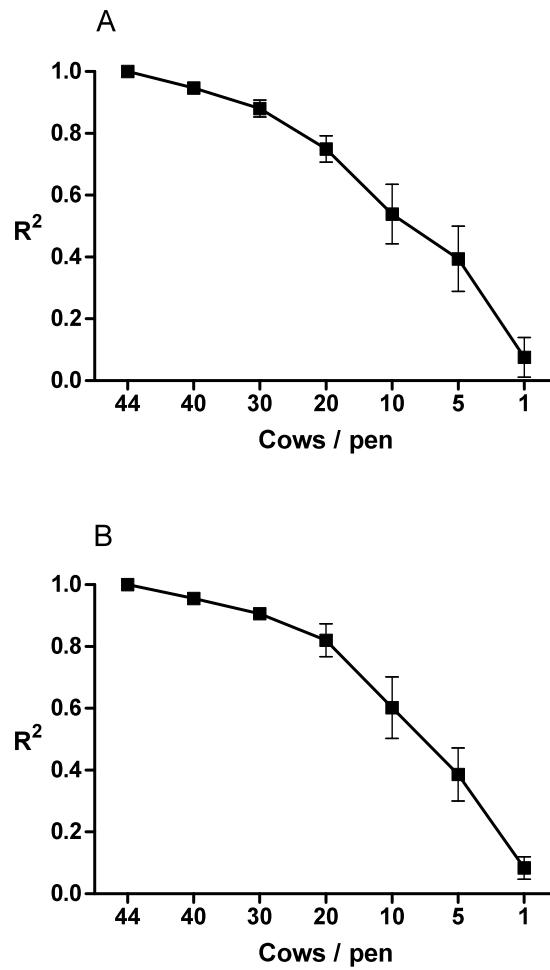


Figure 2.2. The relationship (R^2) between (A) mean lying time (h/d) and (B) mean bout frequency (no./d) based on 44 cows on each farm and estimates derived from 40, 30, 20, 10, 5 and 1 cow(s) randomly sampled on each farm ($n = 38$ farms; 1,818 cows). The error bars represent ± 1 SD of the mean generated from 10 random sampling events.

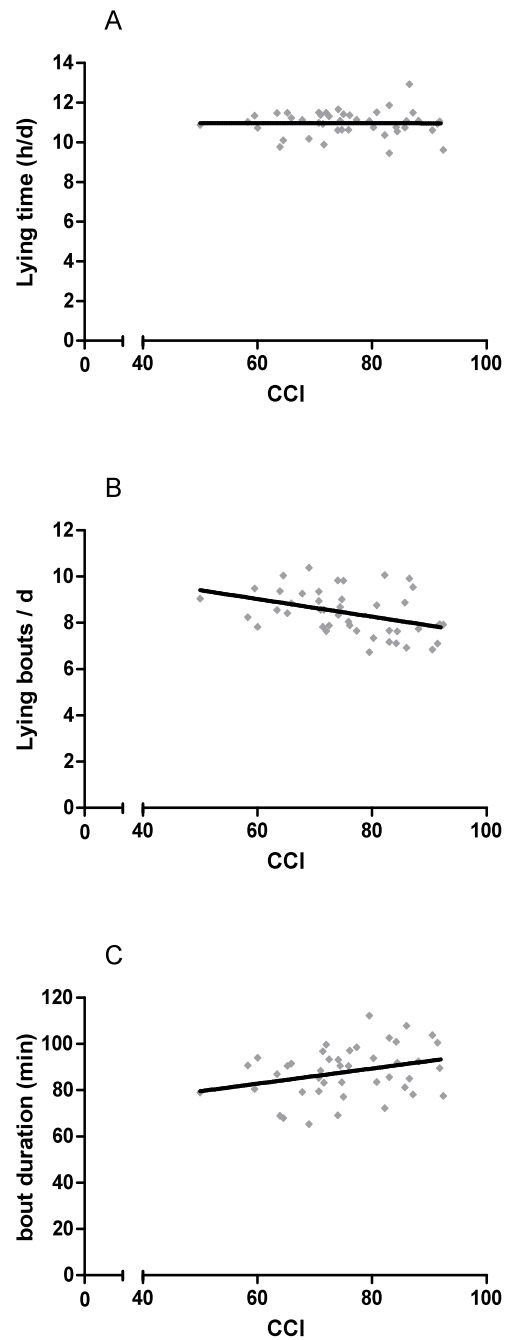


Figure 2.3. Association between Cow Comfort Index (CCI), derived from a point-count observation of the assessment group on each of 43 farms at 2 h before the afternoon milking, and (A) mean lying time (h/d; $R^2 = 0.00$, $P = 0.10$), (B) mean bout frequency (no./d; $R^2 = 0.16$, $P = 0.009$), and (C) 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.

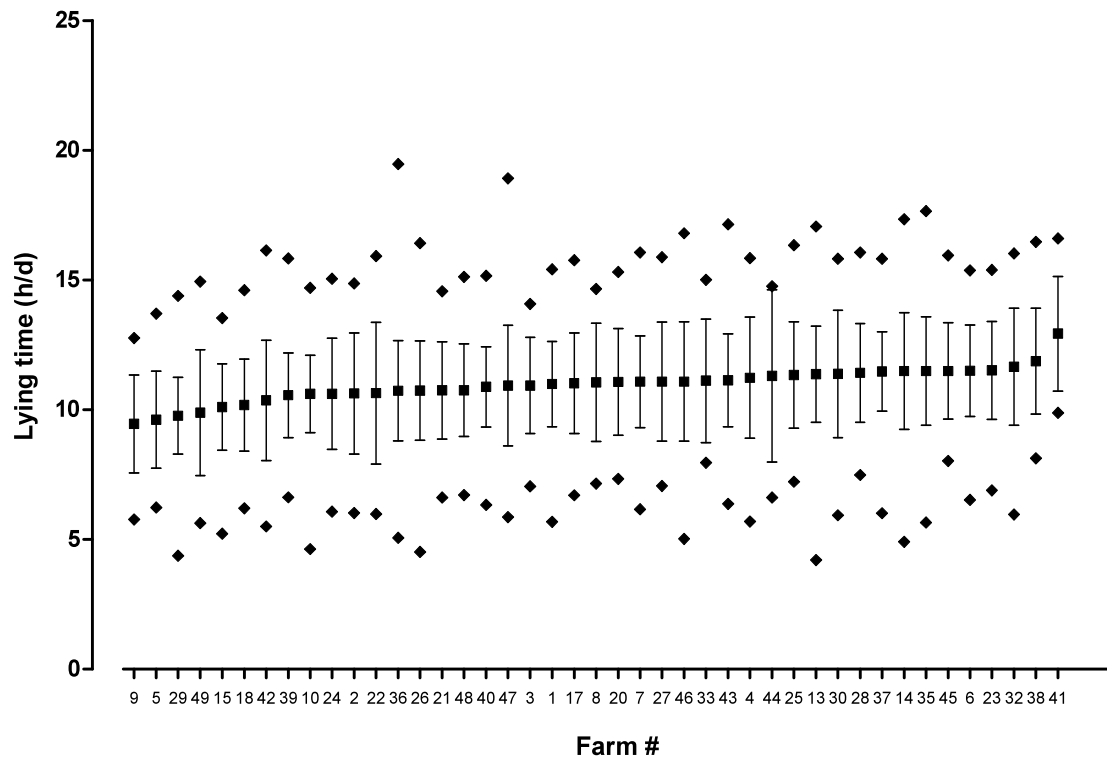


Figure 2.4. Mean (■), maximum and minimum (♦) lying time (h/d) on each of 43 farms. The error bars represent ± 1 SD of the means.

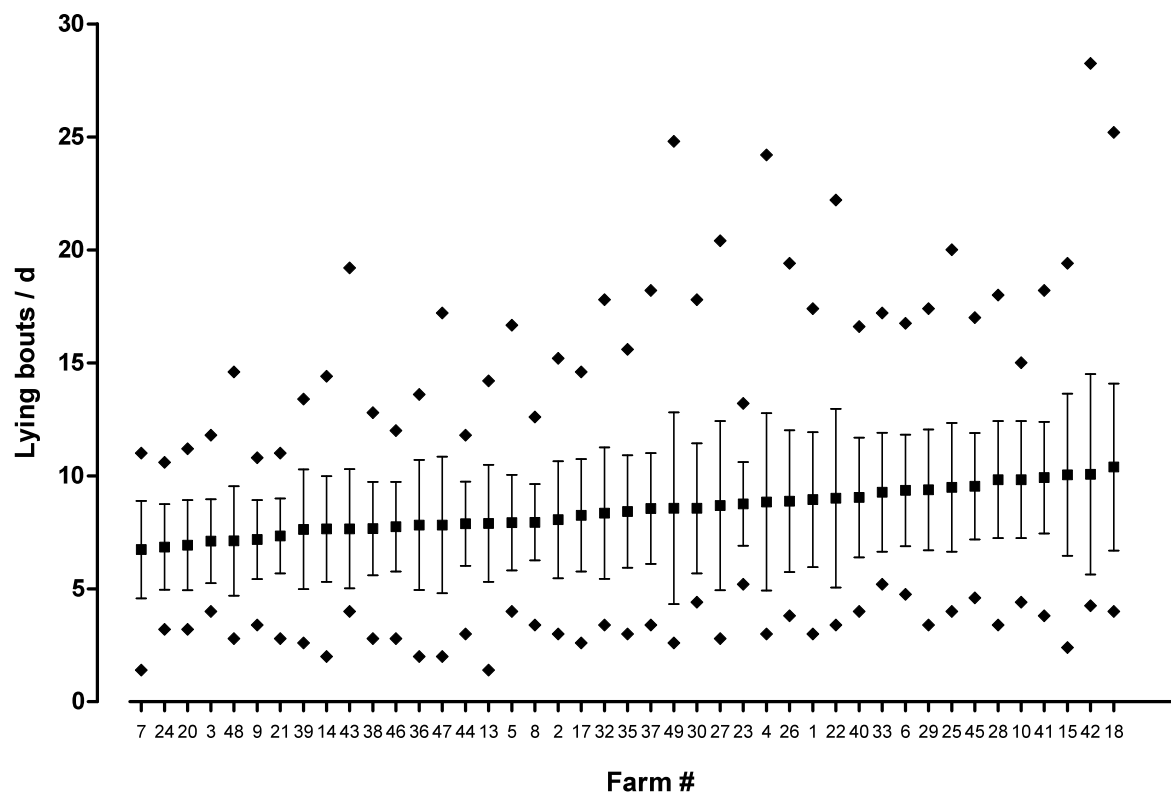


Figure 2.5. Mean (■), maximum and minimum (♦) bout frequency (no./d) on each of 43 farms. The error bars represent ± 1 SD of the means.

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CHAPTER 3: Lying Behaviour as an Indicator of Lameness²

3.1. Introduction

Lameness is widely recognized as a serious animal welfare and production issue in the dairy industry. Recent studies have estimated that 20 to 30% of lactating dairy cows in North America are clinically lame (Cook, 2003; Espejo et al., 2006), with the highest rates observed in herds housed in free-stalls (Cook, 2003; Haskell et al., 2006; Cook and Nordlund, 2009). For instance, Cramer et al. (2008) found that 47% of cows in free-stall herds had lesions on at least one foot, and Bicalho et al. (2007a) found that 13% of cows in a free-stall herd had a painful lesion (i.e. reaction to digital pressure applied to the lesion). Another study estimated lameness prevalence to be as high as 48%, ranging from 0 to 81%, among 33 free-stall herds in Germany (Dippel et al., 2009). Lameness compromises the welfare of the affected animals (Whay et al., 2003), and can result in reduced milk yield (Warnick et al., 2001; Green et al., 2002; Bicalho et al., 2008), reduced fertility and increased risk of premature culling (Garbarino et al., 2004; Bicalho et al., 2007b).

Herd-level risk factors for lameness, within the free-stall system, include: stall features (Philipot et al., 1994; Espejo and Endres, 2007), lying surface (Cook, 2003; Espejo et al., 2006), overcrowding (Leonard et al., 1996), increased time spent away from the pen for milking (Espejo and Endres, 2007), and the use of automatic alley scrapers (Barker et al., 2007). Dippel et al. (2009) found that reduced lying comfort, measured by the frequency of abnormal lying down and rising behaviours (i.e. interrupted movements, lying down or standing up taking longer than 20 s, lying down with hindquarters first, or rising with forequarters first), was also a risk

2. A version of this chapter has been submitted for publication. Ito, K., M. A. G. von Keyserlingk, S. J. LeBlanc, and D. M. Weary. Lying behavior as an indicator of lameness.

factor for lameness. Increased exposure to hard flooring surfaces between periods of rest, combined with reduced rest due to an uncomfortable lying environment, collectively contribute to lameness (Cook and Nordlund, 2009).

Within herds, some individuals are more susceptible to lameness than others. Older cows are generally at higher risk (Espejo et al., 2006; Haskell et al., 2006; Bicalho et al., 2007b; Dippel et al., 2009), and incidence of lameness peaks 3 to 4 months into lactation (Green et al., 2002). Thinner digital cushion (Bicalho et al., 2009), higher milk production at the beginning of lactation (Green et al., 2002; Bicalho et al., 2008), and low body condition (BCS < 3.0) before and at calving (Hoedemarker et al., 2009) have also been identified as cow-level risk factors for lameness. In a study examining behavioural risks for lameness, Galindo et al. (2000) found that cows that became lame spent more time standing with only two front feet in the stall compared to cows that did not become lame (6.2% vs. 5.6% of the day, respectively). Galindo and Broom (2000) also found that the incidence of lameness was related to standing behaviour such that the number of new cases of lameness was higher among cows that spent > 45% of the day standing.

The relationship between behaviour and lameness is complex, possibly due to the variation in the causes and severity of lameness. Lameness may be affected by behaviour, but it also modifies the behaviour of affected cows (Cook and Nordlund, 2009). In general, lame cows spend more time lying down (Singh et al., 1993; Walker et al., 2008; Chapinal et al., 2009) and less time feeding (Gonzalez et al., 2008), perform fewer aggressive interactions (Galindo and Broom, 2002), and are less active (O'Callaghan et al., 2003) compared to non-lame cows. Juarez et al. (2003) compared the lying behaviour of cows with varying degrees of lameness (locomotion scores 1 to 4), and found that the proportion of cows lying down within each category increased with the degree of lameness, with the greatest increase for the severely lame

cows. Cook et al. (2004, 2008) found that the type of lying surface affected the behavioural changes due to lameness; lame cows spent more time standing in stalls than non-lame cows, but this difference was greater on mattress stalls compared to sand stalls. These findings show that the resting environment plays an important role in the extent to which behaviour is modified by lameness.

Lameness, as diagnosed by producers, is often underestimated (Whay et al., 2003; Espejo et al., 2006). Behavioural assessment, such as visual observations of cow gait, is usually the first line of lameness detection before hoof lesions can be identified during trimming (Bicalho et al., 2007a; Chapinal et al., 2009). Gait scoring has been used to detect lameness by researchers (e.g. Cook, 2003; Espejo et al., 2006; Flower and Weary, 2006), but this method requires some training and some time to perform the assessments. Automated technology for measuring lying behaviour is readily available (Ito et al., 2009); inexpensive electronic data loggers can provide precise estimates of lying behaviour for individual cows. The objectives of the present study were to determine how lying behaviour of dairy cow is affected by lameness, and to evaluate the value of measures of lying behaviour in identifying lame cows.

3.2. Materials and Methods

3.2.1. 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 PMR (partially mixed ration with supplemental grain), and milking > 70 cows. Forty farms were recruited in this way; 3 other farms that met the same criteria were recruited

directly by the research team. The number of farms included in this study was determined predominantly by time constraints as the maximum number that could be recruited and assessed within the winter-spring period. Out of these farms, 11 farms using deep-bedded stalls (6 using sand and 5 using sawdust as bedding) and 17 farms using mattress stalls (geotextile mattress with minimal bedding) were selected for this study; 14 farms using other types of stall base (concrete, tires, rubber mat, wood, mixed types) were excluded, and 1 farm using deep-bedded stalls was removed due to the lack of production records. The average herd size was 177 ± 85 (\pm SD, ranging from 83 to 511) milking cows producing $10,434 \pm 799$ (ranging from 8,991 to 12,080) kg annually. These sample herds represent the larger dairies ranked in the top quartile of production in British Columbia. The farms milked twice ($n = 24$) or three times ($n = 4$) daily, and fed once ($n = 22$) or twice ($n = 6$) daily.

3.2.2. Data collection

Each farm was visited twice, with 5 d between visits. As heat stress can affect lying time (Cook et al., 2007), the data collection period was limited to days where the maximum temperature was $< 25^{\circ}\text{C}$. The average maximum temperature across study days was $9.4 \pm 5.3^{\circ}\text{C}$ (ranging from -1.0 to 24.5°C). Upon arrival at each farm, the producer was asked to identify 1 pen that housed the high producing cows; this pen was used for data collection. Six farms housed all lactating cows in a single group, and others separated high producing vs. low producing cows, or multiparous vs. primiparous cows. The selected group size averaged 99 ± 34 (mean \pm SD, ranging from 47 to 187) cows, and the average stall stocking rate for these groups was 104 ± 15 (ranging from 78 to 157) %.

During the afternoon milking on the first visit, up to 50 cows (47 ± 3 , ranging from 37 to 50) 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 selected. The sample size was decided based on the availability of the recording devices; however, retrospective analysis established that 30 cows per farm provided a reasonable sample for encompassing the variations in lying behaviour (Ito et al., 2009). The final sample included 1319 cows, averaging 2.6 ± 1.4 (ranging from 1 to 12) lactations and 150 ± 94 (ranging from 11 to 581) DIM. Lying behaviour of the focal cows was recorded using electronic data loggers (HOBO Pendant G Acceleration Data Loggers, Onset Computer Corporation, Pocasset, MA) at 1-min interval for 5 d (Ito et al., 2009). Durations of individual lying bouts were computed using Microsoft Excel macros for the 5-d period, from which daily lying time (h/d), frequency of lying bouts (no./d), average duration of lying bouts (min/bout), and the SD of lying bout duration (min/bout) were calculated for each cow.

Following the morning milking on the second visit, focal cows were gait scored as they exited the parlor, using a 5-point Numerical Rating System (NRS; Flower and Weary, 2006). For the purposes of the current study cows were categorized as $\text{NRS} \leq 2$, $\text{NRS} = 3$, or $\text{NRS} = 4$. No cow in this study was scored as $\text{NRS} = 5$. The NRS uses 6 behavioural criteria but was simplified for this study by focusing on 3 attributes: asymmetric gait, head bob, and reluctance to bear weight. These attributes are highly correlated with each other as well as with the overall NRS score based on the 6 attributes (Chapinal et al., 2009). A single trained observer performed all scoring. The observer was trained to gait score using recorded videos of cows walking in a straight line on a concrete alley (Chapinal et al., 2009); these same video passages were referenced every 2 months in an effort to minimize changes in the observer's subjective assessment of gait as the study progressed. Prior to the study, the observer scored 132 cows

simultaneously with another experienced observer, and achieved high inter-observer agreement (Kappa statistic = 0.94).

3.2.3. Data analyses

Data were analyzed using cow as the observational unit. Descriptive summaries were obtained using the SUMMARY and FREQ procedures of SAS (SAS Institute Inc., 2004). Data from farms using deep-bedded stalls (DB; n = 11 farms and 526 cows) and farms using mattresses (MAT; n = 17 farms and 793 cows) were analyzed separately as there is evidence from previous work that the stall surface affects the behaviour of lame cows (Cook et al., 2004, 2008). Table 3.1 provides the descriptive summary of the cows used in analyses.

The differences in lameness prevalence and lying behaviours between the two stall surface categories, stratified by NRS, were tested using the TTEST procedure of SAS 9.1 (SAS Institute Inc., 2004) (Appendix 1). Normality of data was tested using the UNIVARIATE procedure, and heterogeneity of variance, where necessary, was accounted for by using the Satterthwaite method for computing the SE. All other statistical analyses were performed using the GLIMMIX procedure (SAS Institute Inc., 2008). Within each stall surface category, mixed linear regression models were created to test the differences in lying behaviour between cows with different degrees of lameness, controlling for the random effect of farm (Appendix 1). Compliance with the assumptions of normality and homogeneity of variances were checked visually through residual plots created by the UNIVARIATE and GPLOT procedures (SAS Institute Inc., 2004). Natural logarithm transformation was applied to bout frequency (no./d), bout duration (min/bout), and the SD of bout duration (min/bout) to normalize these distributions. Extreme outliers (3 times the interquartile range outside of the interquartile limits: n = 1 for bout frequency; n = 5 for bout duration; n = 2 for SD of bout duration) were removed

from analysis. Parity and DIM were forced into all models because they have been known to influence lying behaviour (Endres and Barberg, 2007) as well as risk of lameness (Warnick et al., 2001; Espejo et al., 2006; Bicalho et al., 2007b). Parity was classified as 4 categories: 1, 2, 3, and ≥ 4 , and DIM was treated as continuous. Interactions between the covariates were not included because the effects were never significant and did not confound the effect of lameness. Least square means of each lying variable for the 3 categories of lameness were contrasted, back-transformed where applicable, and reported with 95% confidence intervals.

Frequency distributions of the lying variables in each lameness category (including all data; $n = 28$ farms and 1319 cows) were generated using the CHART procedure (SAS Institute Inc., 2004), dividing the observed range for each variable into 15 bins of equal increment (Appendix 1). The distributions as the percentage of cows in each bin were used to create overlapping line graphs. For each lying variable, a cutoff point to define extreme behaviour was set as the value where the distributions of cows with $\text{NRS} \geq 3$ and cows with $\text{NRS} \leq 2$ intersected. Lameness outcome was dichotomized twice, as $\text{NRS} \geq 3$ and as $\text{NRS} = 4$. Odds ratios (OR) for $\text{NRS} \geq 3$ and $\text{NRS} = 4$ were estimated, for each stall type separately, using logistic regression (GLIMMIX; dist = binomial and link = logit) controlling for the random effect of farm (Appendix 1). Each extreme lying variable was tested individually for association with $\text{NRS} \geq 3$ and $\text{NRS} = 4$. Parity and DIM were forced into models as explanatory variables as in the linear models. Sensitivity and specificity of each extreme behavioural cutoff point to discriminate cows with $\text{NRS} \geq 3$ and $\text{NRS} = 4$ were calculated using the FREQ procedure.

3.3. Results

3.3.1. Effect of stall surface in relation to lameness and lying behaviour

Of the 1319 cows gait scored, 943 (71.5%) had $\text{NRS} \leq 2$, 280 (21.2%) had $\text{NRS} = 3$, and 96 (7.3%) had $\text{NRS} = 4$. At the farm level, the mean prevalence of $\text{NRS} = 4$ was $9.3 \pm 1.3\%$ (mean \pm SE) in MAT farms compared to $4.4 \pm 1.2\%$ in DB farms (Figure 3.1; $P = 0.02$). Cows with $\text{NRS} \leq 3$ in DB and MAT farms had similar mean lying times (h/d) but cows with $\text{NRS} = 4$ spent more time lying down on DB farms than on MAT farms (13.1 ± 0.8 vs. 10.9 ± 0.5 h/d, $P = 0.03$; Figure 3.2). The frequency of lying bouts (no./d), average bout duration (min/bout), and SD of bout duration (min/bout) were similar across lameness categories on both DB and MAT farms.

3.3.2. Lying behaviour in relation to lameness

Lying time (h/d), average bout duration (min/bout), and SD of bout duration (min/bout) differed between cows with $\text{NRS} \leq 2$ and cows with $\text{NRS} = 4$ on DB farms, but not on MAT farms (Table 3.2). On DB farms, cows with $\text{NRS} = 4$ lay down 1.6 h longer per d than cows with $\text{NRS} \leq 2$ ($P < 0.001$) and $\text{NRS} = 3$ ($P < 0.001$). Cows with $\text{NRS} = 4$ had longer lying bouts than cows with $\text{NRS} = 3$ ($P = 0.004$) and $\text{NRS} \leq 2$ ($P = 0.003$). The SD of bout duration was greater for cows with $\text{NRS} = 4$ than cows with $\text{NRS} \leq 2$ ($P = 0.02$).

3.3.3. Extreme lying behaviour as indicators of lameness

There were differences in the distribution of lying time (h/d), average bout duration (min/bout), and SD of bout duration (min/bout) between cows with $\text{NRS} \leq 2$, $\text{NRS} = 3$, and $\text{NRS} = 4$ (Figure 3.3; bout frequency is not reported because this variable was never significant). The intersections in these distributions were used to define extreme lying behaviours: low daily lying time ≤ 9 or high daily lying time ≥ 14 h/d, bout duration $\geq 4.6 \log(\text{min})/\text{bout}$ (back-transformed duration ≥ 99 min/bout), and SD of bout duration $\geq 4.1 \log(\text{min})/\text{bout}$ (back-transformed SD \geq

60 min/bout). High lying time and long bout duration were associated with increased odds of $\text{NRS} \geq 3$ and $\text{NRS} = 4$ on all farms; additionally, the SD of bout duration was associated with $\text{NRS} \geq 3$ and $\text{NRS} = 4$ on DB farms (Table 3.3 and 3.4). Within DB farms, low lying times were associated with increased odds of $\text{NRS} = 3$ but not $\text{NRS} = 4$ (Table 3.5). The behavioural cutoffs used in these analyses provided poor sensitivity, but high specificity in discriminating cows with $\text{NRS} \leq 2$ from cows with $\text{NRS} \geq 3$ or $\text{NRS} = 4$ (Tables 3.3 to 3.5).

3.4. Discussion

Most 5-point scoring systems define clinical lameness as ≥ 3 , but different authors use different terms to classify degrees of severity. For instance, Espejo et al. (2006) described cows scored as 3 as ‘lame’, 4 as ‘moderately to severely lame’, and 5 as ‘severely lame’. Bicalho et al. (2007a) described 3 as ‘moderately lame’, 4 as ‘severely lame’, and 5 as ‘extremely lame’. Cook et al. (2004, 2008), using a 4-point scale, described 3 as ‘moderately lame’ and 4 as ‘severely lame’. In this paper, we simply refer to the NRS score (i.e. ≤ 2 , 3, or 4) to avoid potential confusion with terminology.

Overall prevalence of $\text{NRS} \geq 3$ across the 28 farms in this study was 28.5% including 7.3% of cows classified as $\text{NRS} = 4$. These values are consistent with previously reported values of 24.6% among high-producing cows in 50 free-stall herds in Minnesota (Espejo et al., 2006), and 27.8% during winter and 22.8% during summer among 15 free-stall herds in Wisconsin (Cook, 2003). The prevalence was higher on MAT farms than DB farms (33.0% vs. 22.9%, respectively), again in agreement with previous findings (Cook, 2003; Cook et al., 2004; Espejo et al., 2006). In our study, the effect of stall surface was driven solely by the difference in the prevalence of $\text{NRS} = 4$, suggesting that MAT stalls are a risk particularly for more severe lameness.

Understanding the behavioural differences between lame and sound cows may contribute to developing practical tools for detecting lameness on-farm. On DB farms, cows with NRS = 4 spent more time lying down than did cows with $\text{NRS} \leq 3$, but there was no difference in lying times of cows of different lameness categories on the MAT farms. An effect of stall surface on the behaviour of lame cows has been reported previously. For instance, Cook et al. (2008) found that lame cows increased standing time in the stall by 3.3 h/d on mattress stalls compared to stalls bedded with 5 to 8 cm of sand. We found that the cows with NRS = 4 increased lying time by 1.6 h/d on DB stalls (comparable to sand stalls) compared to MAT stalls. Although the two studies measured different outcome variables (standing in the stall vs. lying in the stall), they both showed that cows' behaviour is affected by both lameness and stall surface.

Several studies have reported increased lying times by lame cows. Juarez et al. (2003) found that lame cows spent more time lying down; 25.2% of lame cows were lying down at the time of behavioural scans compared to 17.5% of sound cows. Similarly, Walker et al. (2008) found that lame cows spent 6.3 % more time lying down during estrus compared to sound cows. Chapinal et al. (2009) reported that cows with sole ulcers spent > 1 h more per d lying down compared with cows without any hoof lesions (13.8 vs. 12.6 h/d, respectively).

Changes in daily lying times associated with lameness were bi-directional; cows with $\text{NRS} \geq 3$ had both higher and lower lying times than cows with $\text{NRS} \leq 2$. This divergence may be explained by cows that are already lame spending more time lying down (perhaps due to difficulties in standing up) and cows with low lying times being at higher risk of becoming lame. Consistent with this idea is that high lying times were especially seen among cows with NRS = 4 and lower lying times were evident only among cows with NRS = 3. The particularly long lying times of cows with NRS = 4 on DB stalls suggests that these stalls provide a place where lame

cows can lie down more comfortably. Increasing lying times may be more difficult on mattress stalls that potentially restrict the lying down and standing up movement, leading to increased standing times as demonstrated by Cook et al. (2004, 2008). In general, when stalls are restrictive (again limiting transitions from lying to standing and vice versa), cows spend more time standing in the stall instead of lying down (Tucker et al., 2004, 2005). These results highlight the importance of providing a comfortable lying place, especially for lame cows.

Within DB farms, cows with NRS = 4 also had longer lying bouts compared to cows with $\text{NRS} \leq 3$. This finding agrees with Chapinal et al. (2009) who reported that cows with sole ulcers had longer bouts compared with cows with hemorrhages, dermatitis, or no lesions (93.3 vs. 82.7, 81.1, or 71.0 min, respectively). Although our data showed that lame cows had longer bouts, the frequency of bouts was unchanged. This result is at least partly explained by increased variability (i.e. SD) in bout durations. Cook et al. (2004) found that the standing bout structure of lame cows was modified so that the proportion of short bouts was reduced and the proportion of long bouts was increased, especially for cows on mattress stalls. In the current study, the behavioural differences were evident only on DB stalls. Previous work has shown that mattresses reduce lying time, especially if managed with little or no bedding (Tucker et al., 2003; Tucker and Weary, 2004) as was the case for the MAT farms included in the current study. Reduced stall comfort, particularly for the lame cows, may explain the higher prevalence of more severe lameness (i.e. NRS = 4) on MAT farms.

Stall standing behaviour has been identified as a key behaviour that changes in response to lameness (Cook et al., 2004, 2008). Furthermore, increased time spent perching, with only two front feet inside the stall, is a known risk factor for lameness (Galindo et al., 2000). Unfortunately, the data loggers used in this study could not discriminate between standing

locations (inside or outside the stall) or positions (two feet or four feet inside the stall).

Development of technology that identifies standing position is desirable.

Extreme lying behaviour was not a sensitive diagnostic tool for lameness; clear cutoff points to accurately identify cows with $\text{NRS} \geq 3$ or $\text{NRS} = 4$ were not established. This is because most cows, regardless of NRS, spent between 9 and 14 h/d lying down. However, the cutoff values did provide high specificity for correctly distinguishing cows with $\text{NRS} \leq 2$, suggesting that extreme lying behaviour could be a useful element in a more sophisticated system for identifying lame cows. Our results indicate that cows with high lying times or long lying bouts are more likely to be lame and should be flagged for closer examination. It is likely that technology for automated measurement of lying behaviour integrated with existing cow identification and activity monitoring systems will become commercially available in the near future. The present data may be applied with such systems as a component of lameness detection.

3.5. Conclusions

The prevalence of $\text{NRS} = 4$ was higher in farms using MAT stalls compared to those using DB stalls. Cows with $\text{NRS} = 4$ housed on DB stalls spent more time lying down, and had longer lying bouts compared to cows with $\text{NRS} \leq 3$, but there were no behavioural differences among cows with different degrees of lameness housed on MAT stalls. Extreme lying behaviour, especially high lying times (≥ 14 h/d) and long lying bouts (≥ 99 min/bout), was associated with increased odds of both $\text{NRS} \geq 3$ and $\text{NRS} = 4$ for both types of stall surface. These results indicate that automated measurement of lying behaviour may contribute to detection of lameness.

Table 3.1. Parity and stage of lactation (DIM) (mean \pm SD) of cows with NRS ≤ 2 , NRS = 3, and NRS = 4 on 11 farms using deep-bedded stalls (DB) and 17 farms using mattress stalls (MAT).

Stall surface	Variable	NRS ≤ 2	NRS = 3	NRS = 4
DB	Number of cows	397	106	23
	Parity	2.5 \pm 1.4	3.8 \pm 1.8	3.6 \pm 1.6
	DIM ¹	146 \pm 95	158 \pm 109	140 \pm 98
MAT	Number of cows	546	174	73
	Parity	2.3 \pm 1.2	3.0 \pm 1.4	3.6 \pm 1.7
	DIM ¹	145 \pm 91	159 \pm 87	157 \pm 105

¹ DIM = days in milk

Table 3.2. Least square means (95% confidence intervals) of lying time (h/d), bout frequency (no./d), bout duration (min/bout), and SD of bout duration (min/bout) for cows with NRS ≤ 2 , NRS = 3, and NRS = 4 on 11 farms using deep-bedded stalls (DB) and 17 farms using mattress stalls (MAT). Values are from linear regression models accounting for parity, DIM, and a random effect of farm. Different superscripts indicate significant difference at $P < 0.05$ contrasted across row.

Stall surface	Variable	NRS ≤ 2	NRS = 3	NRS = 4
DB	Number of cows	397	106	23
	Lying time (h/d)	11.1 ^a (10.6 to 11.7)	11.1 ^a (10.5 to 11.8)	12.7 ^b (11.8 to 13.5)
	Bout frequency (no./d) ¹	8.1 (7.4 to 8.8)	8.0 (7.2 to 8.9)	7.7 (6.7 to 8.9)
	Bout duration (min/bout) ¹	79.9 ^a (74.5 to 85.8)	79.8 ^a (73.4 to 86.7)	93.6 ^b (83.0 to 105.4)
	SD bout duration (min/bout) ¹	44.1 ^a (40.8 to 47.6)	46.1 ^{ab} (42.0 to 51.6)	50.7 ^b (44.2 to 58.3)
MAT	Number of cows	546	174	73
	Lying time (h/d)	10.8 (10.4 to 11.2)	11.1 (10.7 to 11.6)	11.3 (10.7 to 11.8)
	Bout frequency (no./d) ¹	8.5 (8.0 to 8.9)	8.5 (8.0 to 9.2)	8.7 (7.9 to 9.5)
	Bout duration (min/bout) ¹	74.0 (69.6 to 78.8)	75.6 (70.2 to 81.3)	78.6 (71.7 to 86.2)
	SD bout duration (min/bout) ¹	44.2 (41.9 to 46.7)	44.5 (41.6 to 47.6)	47.3 (43.4 to 51.6)

¹ Back-transformed least square means (95% confidence intervals) where natural log transformation was applied to the variable.

Table 3.3. Numbers of cows with $\text{NRS} \geq 3$ and $\text{NRS} \leq 2$ in each of the cut-off categories, for 11 farms using deep-bedded stalls (DB) and 17 farms using mattress stalls (MAT), sensitivity and specificity of each cut-off point, and odds ratios (OR) for lameness estimated by logistic regression including parity and DIM as covariates in all models.

Stall surface	Variable (cut-off)	# cows		Lameness				
		$\text{NRS} \geq 3$	$\text{NRS} \leq 2$	Se ¹	Sp ²	OR	95% CI	P
		N = 129	N = 397					
DB	Lying time (≥ 14 h/d)	20	16	16	96	4.0	1.8 to 9.3	0.001
	Lying time (≤ 9 h/d)	23	41	18	90	0.2	0.8 to 3.1	0.2
	Bout duration (≥ 99 min/bout)	38	53	29	87	1.9	1.1 to 3.4	0.03
	SD bout duration (≥ 60 min/bout)	39	44	30	89	2.7	1.5 to 4.8	0.001
		N = 247	N = 546					
MAT	Lying time (≥ 14 h/d)	36	28	15	95	3.7	2.0 to 6.8	< 0.0001
	Lying time (≤ 9 h/d)	55	98	22	82	1.1	0.7 to 1.7	0.8
	Bout duration (≥ 99 min/bout)	56	80	23	85	2.0	1.3 to 3.1	0.003
	SD bout duration (≥ 60 min/bout)	52	83	21	85	1.4	0.9 to 2.2	0.1

¹ Sensitivity = the proportion of lame cows identified by the cut-off of lying behaviour

² Specificity = the proportion of non-lame cows correctly classified by the cut-off of lying behaviour

Table 3.4. Numbers of cows with NRS = 4 and NRS ≤ 3 in each of the cut-off categories, for 11 farms using deep-bedded stalls (DB) and 17 farms using mattress stalls (MAT), sensitivity and specificity of each cut-off point, and odds ratios (OR) for lameness estimated by logistic regression including parity and DIM as covariates in all models.

Stall surface	Variable (cut-off)	# cows		Lameness				
		NRS = 4	NRS ≤ 3	Se ¹	Sp ²	OR	95% CI	<i>P</i>
		N = 23	N = 503					
DB	Lying time (≥ 14 h/d)	9	27	39	95	13.4	4.4 to 40.4	< 0.0001
	Lying time (≤ 9 h/d)	1	63	4	87	0.3	0.0 to 2.2	0.2
	Bout duration (≥ 99 min/bout)	10	81	43	84	3.2	1.3 to 8.0	0.01
	SD bout duration (≥ 60 min/bout)	12	71	52	86	5.8	2.4 to 14.2	0.0001
		N = 73	N = 720					
MAT	Lying time (≥ 14 h/d)	16	48	22	93	3.9	1.9 to 7.8	0.0001
	Lying time (≤ 9 h/d)	18	135	25	81	1.3	0.7 to 2.3	0.4
	Bout duration (≥ 99 min/bout)	20	116	27	84	1.9	1.1 to 3.5	0.03
	SD bout duration (≥ 60 min/bout)	19	116	26	84	1.6	0.9 to 2.8	0.1

¹ Sensitivity = the proportion of lame cows identified by the cut-off of lying behaviour

² Specificity = the proportion of non-lame cows correctly classified by the cut-off of lying behaviour

Table 3.5. Numbers of cows with $\text{NRS} = 3$ and $\text{NRS} \leq 2$ in each of the cut-off categories for 11 farms using deep-bedded stalls (DB), sensitivity and specificity of each cut-off point, and odds ratios (OR) for lameness estimated by logistic regression including parity and DIM as covariates in all models.

Stall surface	Variable (cut-off)	# cows		Lameness				
		NRS = 3	NRS \leq 2	Se ¹	Sp ²	OR	95% CI	<i>P</i>
		N = 106	N = 397					
	Lying time (≥ 14 h/d)	11	16	10	96	1.7	0.7 to 4.4	0.3
DB	Lying time (≤ 9 h/d)	22	41	21	90	2.0	1.0 to 4.1	0.05
	Bout duration (≥ 99 min/bout)	28	53	26	87	1.1	0.6 to 2.0	0.5
	SD bout duration (≥ 60 min/bout)	27	44	25	89	1.9	1.0 to 3.6	0.07

¹ Sensitivity = the proportion of lame cows identified by the cut-off of lying behaviour

² Specificity = the proportion of non-lame cows correctly classified by the cut-off of lying behaviour

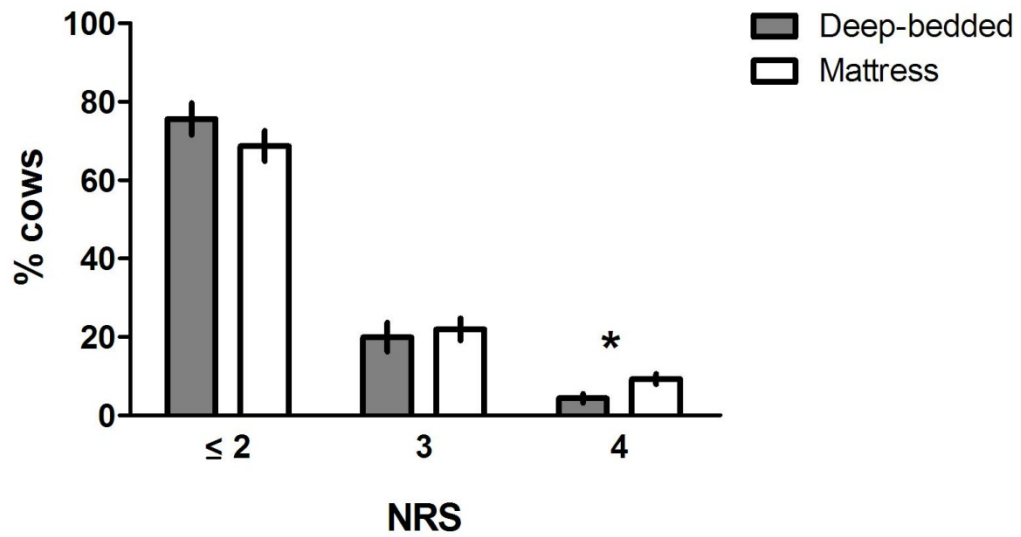


Figure 3.1. Percentages of cows with NRS ≤ 2 , NRS = 3, and NRS = 4 on 11 farms using deep-bedded stalls and 17 farms using mattress stalls. * indicates significant difference at $P < 0.05$.

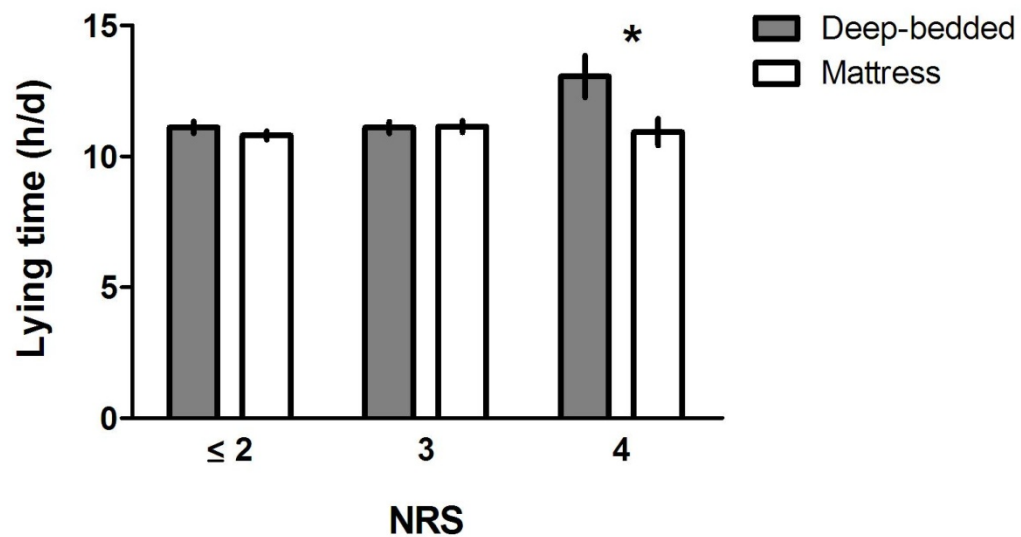


Figure 3.2. Mean lying time (h/d) of cows with NRS ≤ 2 , NRS = 3, and NRS = 4 on 11 farms using deep-bedded stalls and 17 farms using mattress stalls. * indicates significant difference at $P < 0.05$.

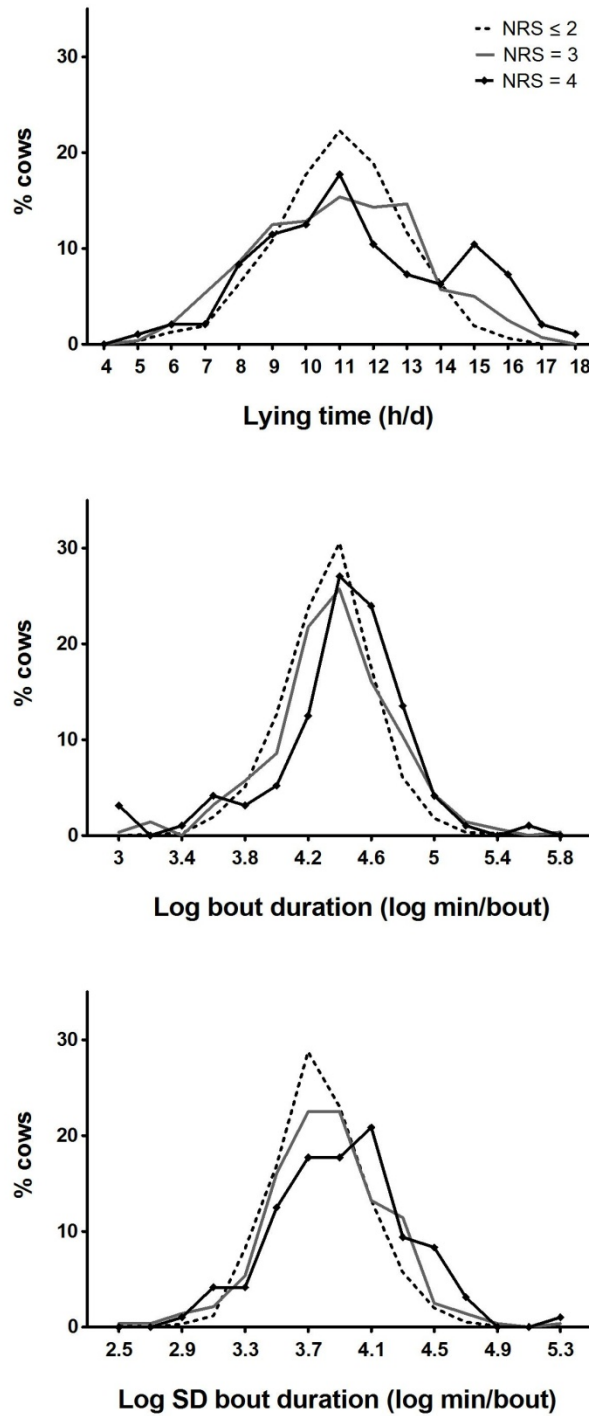


Figure 3.3. Percentage distributions of cows with $\text{NRS} \leq 2$, $\text{NRS} = 3$, and $\text{NRS} = 4$ as % of cows for (A) lying time (h/d), (B) bout duration (log min/bout), and (C) SD of bout duration (log min/bout). Cut-off points for extreme behaviour were defined as the value where distributions for $\text{NRS} \geq 3$ and $\text{NRS} \leq 2$ intersected.

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CHAPTER 4: General Discussion

4.1. Assessing Cow Comfort

The issue of cow comfort has received considerable interest in the dairy industry in recent years. The term ‘cow comfort’ may be most commonly perceived as the ability of cows to lie down; however, it should encompass any factor that affect how comfortable cows may feel. In this sense, the traditional approach to assessing cow comfort focusing on how much cows lie down is limiting. Lying behaviour clearly provides valuable information about stall comfort, but injuries and diseases, such as lameness, should also be integral parts of assessing cow comfort. Moreover, the interactions between behaviour, environmental factors (e.g. stall surface), and the health status of the cows (e.g. lameness), make the assessment of cow comfort a multi-dimensional task.

My research was the first of its kind to provide a detailed description of the lying behaviour of dairy cows housed in free-stall systems, and to establish a practical method of measuring this behaviour on commercial farms (Chapter 2). The common approach to assessing cow comfort has been to estimate the stall use at the group-level based on single observation (e.g. Cow Comfort Index). My data showed that this method does not reflect the cows’ actual lying behaviour and is not a reliable tool. Simple technology (e.g. electronic data loggers) is available to continuously monitor the lying behaviour of individual animals housed in groups. There was considerable variation in lying times and the frequency of lying bouts among cows within the same farm, suggesting that evaluation of cow comfort should focus on the analysis of behaviour of individual animals rather than farm averages. These results led to the development of a methodological paper (Chapter 2) that will be of value to future research on lying behaviour.

Daily lying times averaged 11.0 h/d, but varied from 4.2 to 19.5 h/d among individual cows. Some authors have suggested that dairy cows housed in free-stall systems should be allowed to lie down for at least 12 h/d (DeVries et al., 2005; Jensen et al., 2005; Munksgaard et al., 2005). By this standard, the majority of the cows in B.C. are not getting adequate lying time. Increased lying time is generally an indicator of stall comfort (e.g. Haley et al., 2000; Tucker et al., 2003; Fregonesi et al., 2007). However, my data demonstrated that extremely high lying times are associated with lameness, if the stalls allow for this behaviour (Chapter 3); therefore, lying time alone does not tell the whole story.

The frequency of lying bouts has also been considered a sensitive measure of stall comfort (e.g. Haley et al., 2000; Tucker et al., 2003). In my study, this variable did not differ between cows housed on different stall surfaces (i.e. deep-bedded vs. mattress) and was not associated with lameness (Chapter 3). The duration of individual bouts, on the other hand, provided promising results. Long lying bouts, as well as high variation in the duration of lying bouts, were associated with lameness, but only among cows on deep-bedded stalls. A combination of lying time and the duration of lying bouts likely provide a more comprehensive approach. Observations of cows lying down and standing up can add another dimension to assessing the functionality of free-stalls (Dippel et al., 2009). In any case, the type of stall surface must be taken into consideration as it affects both behaviour and lameness.

There is no gold standard for how many hours per day cows need to lie down, or how to design a perfect facility that allows for the optimal lying behaviour for all cows. Instead, behavioural research provides an understanding of stall features that cows like and how they modify behaviour in relation to these factors (e.g. Tucker et al., 2003; Fregonesi et al., 2007), as well as how much space cows require to perform a natural lying down movement (Ceballos et

al., 2005). All this contributes to guidelines for designing and evaluating facilities (e.g. Nordlund and Cook, 2003). Animal-based measures such as body condition and prevalence of injuries can also provide outcome-based measures for assessing how well the facilities are working for the cows.

My research provided the first data on lameness prevalence on dairy farms in British Columbia. In free-stall farms in B.C., 21% of cows were scored as NRS = 3 and a further 7% as NRS = 4. These results agree with previous reports of lameness prevalence in similar systems in North America and Europe (Cook, 2003; Whay et al., 2003; Espejo et al., 2006; Dippel et al., 2009). The recently published Code of Practice for the Care and Handling of Dairy Cattle (National Farm Animal Care Council, 2009) recommends < 10% obvious or severe lameness (i.e. NRS \geq 4) at the farm-level as a reasonable target, which the majority of farms in my study achieved. However, the average prevalence of NRS = 4 in farms using mattress stalls was over 9%, in comparison to 4% for farms using deep-bedded stalls. Higher lameness prevalence in mattress herds has been reported previously (e.g. Cook et al., 2004).

4.2. Linking Stall Comfort, Behaviour, and Lameness

Lameness can be painful, which likely affects how comfortable cows may feel. Moreover, housing comfort has been suggested to influence the risk of lameness as well as the severity and duration of lameness events (Cook and Nordlund, 2009). Early work on lameness suggested that insufficient rest due to uncomfortable stalls might contribute to increased incidences of lameness (Leonard et al., 1994). My study allowed for an evaluation of lying behaviour in relation to lameness on farms using different types of stalls. Low lying times in farms using deep-bedded stalls was driven by cows with NRS = 3, but not by those with NRS = 4. I suggest that these cows with low lying times are at risk for becoming more lame, and that

after cows become more severely lame, lying time increases. However, a causal relationship cannot be established from a cross-sectional study.

Lame cows may spend more time lying down because it is difficult for them to transition between lying and standing positions. It is also plausible that lying down allows the injured leg or foot to rest, and helps the cow recover from lameness. Cows with NRS = 4 housed on deep-bedded stalls lay down for 1.6 h more per d compared cows with $\text{NRS} \leq 2$. Interestingly, there was no difference in lying times among lameness categories for farms using mattress stalls. Long lying bouts and high variation in the duration of lying bouts were associated with lameness, but again only on farms using deep-bedded stalls. Previous studies have also found different behavioural responses by lame cows depending on the stall surface (Cook et al., 2004, 2008). Cook et al. (2004) found that lame cows on mattress stalls spent more time standing inside the stall, at the expense of lying down, compared to healthy cows; and the prevalence of lameness was higher among cows housed on mattress compared to cows housed on sand (24% vs. 11%, respectively). My data suggests that deep-bedded stalls (sand or sawdust) may allow lame cows to spend more time lying down as needed, whereas mattress stalls do not provide this opportunity, prolonging lameness events and thus resulting in a higher prevalence of lameness. These results highlight the importance of providing a comfortable place to lie down, especially for the lame cows.

Alternatively, many studies have suggested that increased standing behaviour increases the risk for lameness, perhaps more so than reduced lying behaviour (e.g. Leonard et al., 1994; Galindo and Broom, 2000; Cook et al., 2004). Exposure to concrete flooring that is wet and slippery, and contains infectious agents, exacerbates claw lesions and lameness (Cook and Nordlund, 2009). Cows on pasture spent less time lying down compared to cows in free-stalls,

yet lame cows recovered in a few weeks of pasture access (Hernandez-Mendo et al., 2007). This suggests that the uncomfortable standing surface inside the barn may drive cows to lie down in addition to the need for rest alone; or the reduced rest on pasture is compensated by a comfortable standing surface (Cook and Nordlund, 2009). The free-stall surface could provide a place inside the barn where cows can get away from the wet concrete; however, the neckrail often prevents cows from standing fully inside the stall to keep the bedding clean. Bernardi et al. (2009) demonstrated that an aggressive neckrail placement contributes to the occurrence of lameness while removing the neckrail helps lame cows recover. Lying behaviour was not affected by the position of the neckrail, but the neckrail forced cows to perch, with only the two front feet inside the stall, instead of standing fully inside the stall (Bernardi et al., 2009). Rubber mats in the alley may provide a softer surface for standing, but does not alleviate exposure to wet manure. A truly comfortable stall is one that not only optimizes lying but one that also provides a dry, comfortable place to stand. Collectively, these findings indicate that improved stall comfort contributes to reduced lameness.

4.3. Future Research

My study provided an insight into the possible link between stall comfort, lying behaviour, and lameness; however, further research is required to fully understand this complex relationship. Longitudinal research is necessary to tease apart the behavioural causes and consequences of lameness, and to document the development of lameness; for example, does NRS = 3 advance to NRS = 4, and how does the behaviour of these cows change in the process? The considerable variation among individual cows suggests that changes in behaviour within cow provide the most accurate data.

One limitation of my study was that I was not able to record the location or position of the cows while standing. This behaviour is much more challenging to measure on-farm compared to simple lying or standing that can be recorded by automated devices. There is potential, however, for developing automated methods of recording stall standing behaviour. Data loggers used in this study can be modified, or simply attached to the back of the cow to measure the degree of incline that would distinguish standing and perching in the stall. Future researchers are encouraged to include standing behaviour as indicators of lameness.

Finally, the effect of social behaviour on lameness deserves future study. Galindo and Broom (2002) hypothesized that lame cows are less able to cope with competitive environment; these cows performed fewer aggressive interactions and spent more time lying down outside the stall rather than competing to get access to a stall. Social behaviour is known to play a role in the development of other transition diseases (Huzzey et al., 2007), and may also be important for lameness.

4.4. Practical Application

Prior to this study, there was very limited knowledge of the facilities and management practices used on B.C. dairy farms, and where the industry stood for cow comfort. This thesis was part of a cross-farm study that we conducted in collaboration with the local dairy industry. One objective of the farm survey was to benchmark the industry on measures of cow comfort (i.e. lying time, prevalence of lameness and hock injuries). These results were communicated to each participating producer through individual reports, allowing them to compare their farm to the average across other farms in the study (Appendix). There was a large variation across farms in the prevalence of lameness and hock injuries; while some farms were experiencing challenges, many farms showed considerable success in these areas. This benchmarking information

provides a reference point and paves the way for new research. The first step in making a change is to identify the problem; baseline assessments on other farms can be compared to the industry benchmarks collected in my study to identify specific areas in need of improvement. The next step is intervention: implementing changes in facilities or management practices that are predicted to address the problem. The third step is a follow-up assessment to evaluate progress. This three-step approach provides a backbone for practical application of our research that is aimed to improve comfort and welfare of dairy cows.

4.5. References

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Appendix 1: SAS codes

The SURVEYSELECT Procedure (Chapter 2 - Figure 2.1)

```
proc surveyselect data=start out=sample5 n=5 method=srs seed=0;
  strata farm cow;
  id _all_;
proc summary;
  by farm cow;
  var bouts time;
  output out=samp5 mean=b5 t5;

proc surveyselect data=start out=sample4 n=4 method=srs seed=0;
  strata farm cow;
  id _all_;
proc summary;
  by farm cow;
  var bouts time;
  output out=samp4 mean=b4 t4;

proc surveyselect data=start out=sample3 n=3 method=srs seed=0;
  strata farm cow;
  id _all_;
proc summary;
  by farm cow;
  var bouts time;
  output out=samp3 mean=b3 t3;

proc surveyselect data=start out=sample2 n=2 method=srs seed=0;
  strata farm cow;
  id _all_;
proc summary;
  by farm cow;
  var bouts time;
  output out=samp2 mean=b2 t2;

proc surveyselect data=start out=sample1 n=1 method=srs seed=0;
  strata farm cow;
  id _all_;
proc summary;
  by farm cow;
  var bouts time;
  output out=samp1 mean=b1 t1;

data merged;
  merge samp5 samp4 samp3 samp2 samp1;
  drop _type_ _freq_;

proc reg;
  model t5=t4; model t5=t3; model t5=t2; model t5=t1;
  model b5=b4; model b5=b3; model b5=b2; model b5=b1;
run;
```

The TTEST Procedure (Chapter 3 – Figure 3.1 and 3.2)

Title "effect of stall surface on lameness and lying behaviour";

```
proc ttest data=cows;
  by nrs;
  class stall;
  var prev time bouts dur sddur;
run;
```

The GLIMMIX Procedure – Mixed Models (Chapter 3 – Table 3.2)

Title "effect of NRS on lying time";

```
DATA NRStime;
  set all;
  if stall=1 *Mattress farms;
  run;

proc GLIMMIX;
  class nrs farm parity;
  model time = parity dim nrs/ htype=1 solution;
  random farm;
  lsmeans nrs / adjust=smm adjdfe=row cl *For contrasted means with
  95% CI;
  output out=residual1 pred=p1 resid=r1;
  run;

DATA NRStime;
  set all;
  if stall=3 *Deep-bedded farms;
  run;

proc GLIMMIX;
  class nrs farm parity;
  model time = parity dim nrs/ htype=1 solution;
  random farm;
  lsmeans nrs / adjust=smm adjdfe=row cl;
  output out=residual2 pred=p2 resid=r2;
  run;

*To check for normal residual distribution;
proc UNIVARIATE data=residual1 normal plot;
  var r1;
  run;

*To check for homogeneity of residual variance;
proc GPLOT data=residual1;
  plot r1*p1;
  run;
```

The FREQ Procedure (Chapter 3 – Figure 3.3)

```
Title "frequency distribution for NRS=2";

DATA freq2;
    set all;
    if nrs=2;
    run;

proc chart;
    hbar time / midpoints= 4 to 18 by 1 *lying time;
run;

proc chart;
    hbar ldur / midpoints= 3.0 to 5.8 by 0.2 *log bout duration;
run;

proc chart;
    hbar lsddur / midpoints=2.5 to 5.3 by 0.2 *log SD of bout duration;
run;
```

NOTE: Same codes were repeated for NRS = 3 and NRS = 4.

The GLIMMIX Procedure – Logistic Regression (Chapter 3 – Table 3.3 to 3.5)

```
Title "extreme lying time to distinguish NRS=3+4 vs. NRS=2";

DATA lame;
    set all;
    if stall=1 *Mattress farms;
    lame=nrs ge 3;
    xhtime=time ge 14 *Cut-off point based on figure 3.3;
    xltime=time le 9;
    xdur=ldur ge 4.6;
    xsddur=lsddur ge 4.1;
    run;

proc GLIMMIX data=lame;
    class farm parity;
    model lame (event='1') = parity dim xhtime / dist=binomial
    link=logit oddsratio (diff=first) htype=1;
    random intercept / subject=farm;
run;
```

NOTE: Models for low lying time (xltime), bout duration (xdur) and SD of bout duration (xsddur) were omitted here due to space, but were identical except for the main variable. Same models were repeated for stall = 3. For Table 3.4, lameness was defined as NRS = 4; for Table 3.5, NRS =4 was deleted and lameness was defined as NRS = 3.

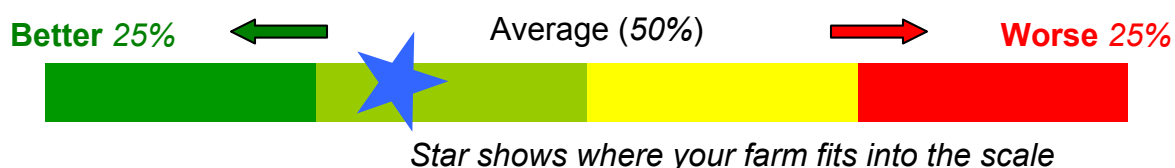


Appendix 2: Example of Producer Report

For:
FARM NAME

Cow Measures

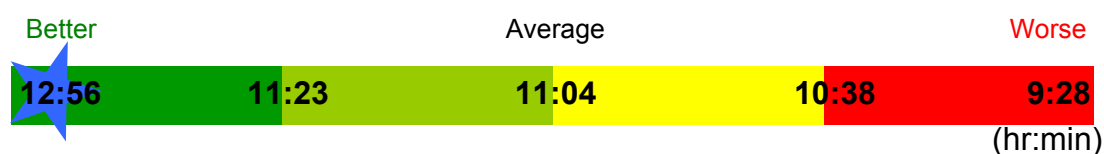
The colour scale shows the overall distribution of scores from the 43 farms assessed, ranging from best / better (green) to worse / worst (red). The blue star indicates where your farm fits into the range. The overall averages and your farm's scores are also included in the table below the scale.



Lying time

The amount of time cows spent lying down was recorded continuously for 5 days using electronic 'data loggers' attached to the hind leg of 50 cows randomly selected from the assessment pen. This data gives a quantitative measure of cow comfort on your farm.

Lying time (Hr:Min per 24 hours)



Your farm	Average	Comments
12 hr 56 min Min: 9 hr 54 min Max: 16 hr 36 min	11 hr 04 min	Cows should rest about 12 hours per day. Increased standing time in the alley puts the cow at risk for lameness. Keep up the good work!

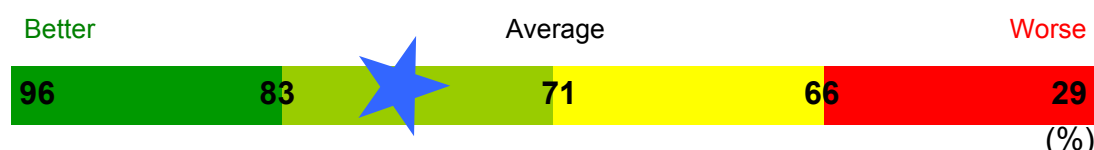


Lameness

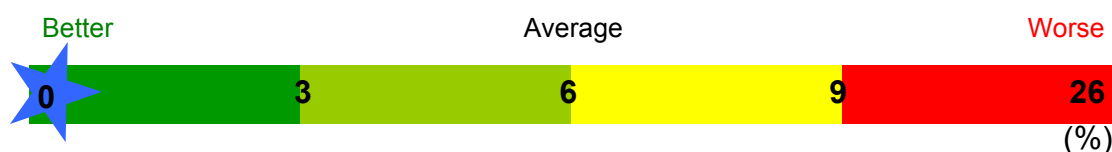
All cows in the assessment pen were gait scored, on a scale of 1 (normal locomotion) to 5 (severely lame), upon exit from the parlour after morning milking. Those cows scored as 1 or 2 were considered 'sound', while those scored as 4 or 5 were considered 'severely lame'. The cows scored as 3 were considered 'mildly lame' but are not included in this report.

Description	Behavioural signs	Action
Sound (Gait score 1 & 2)	<ul style="list-style-type: none"> Walks with a flat back Steady stride No limp – equal weight bearing on all legs 	<ul style="list-style-type: none"> Does not require treatment Routine monitoring and trimming
Mildly lame (Gait score 3)	<ul style="list-style-type: none"> Walks with a slightly arched back Short stride Slight limp on a leg that is not immediately identifiable 	<ul style="list-style-type: none"> Likely to benefit from treatment Foot should be lifted to diagnose the problem as soon as practically possible
Severely lame (Gait score 4 & 5)	<ul style="list-style-type: none"> Arched back Walks with a head bob (jerky movements of the head up or down) Obvious limp on a leg that is immediately identifiable 	<ul style="list-style-type: none"> Requires urgent attention! Should not be made to walk far Should be housed on pack or pasture until recovery

Percentage of cows scored SOUND



Percentage of cows scored SEVERELY LAME



Your farm	Average	Comments
Sound: 78 % Severely Lame: 0 %	Sound: 71 % Severely Lame: 6 %	There were no severely lame cows in the assessment group (pen 11-15), but 22% were mildly lame. Regular monitoring and prompt treatment/trimming will help reduce lameness even further.



Hock Scores

The hock condition of the 50 cows randomly selected for lying time assessment were also scored on the scale of 1 (normal) to 3 (swollen).



Hock Assessment Chart for Cattle



Printed with permission from
Cornell University,
Cooperative Extension

Score = 1
No Swelling. No hair is missing.

Score = 2
No swelling. Bald area on the hock.

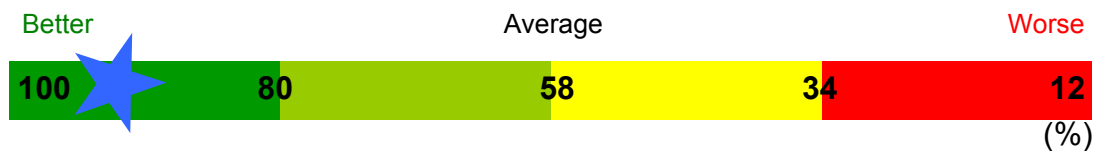
Score = 3
Swelling is evident or there is a lesion through the hide.

The normal, healthy hock is free from skin lesions and swelling. Ideally, the hair coat in that area is smooth and continuous with the rest of the leg.

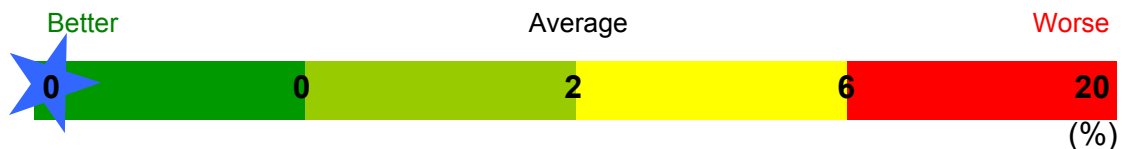
Hock health is an important indicator of the abrasiveness of stall bedding and cow comfort. Injury is usually the result of prolonged exposure to an abrasive stall surface. Skin breakage provides an opportunity for infection to occur, which can lead to swelling, discomfort, and possibly lameness.

A consistent method of scoring hocks for swelling and hair loss allows you to assess the need to modify your stall management and can help you evaluate the effect of management changes.

Percentage of cows with HEALTHY hock (% of cows in the pen assessed)



Percentage of cows with SWOLLEN hock (% of cows in the pen assessed)



Your farm	Average	Comments
Normal: 92 % Swollen: 0 %	Normal: 58 % Swollen: 2 %	Hocks of your cows were in good shape! Deep-bedded stalls and good bedding maintenance likely contribute to these good hock conditions. Keep up the good work!



FARM NAME

Assessment Date

Facility Design and Management Measures

Lying Area	Your Measurement	Average	Comments
Bedding frequency	14 days	10 days	Increased frequency and quantity of bedding may lead to increased lying time.
Bedding quantity – on a scale of 1 (base covered) to 3 (more than 50% of base exposed)	1.0	1.6	GOOD! Stall use improves with the addition of more bedding. The stall base or type of bedding is as important as bedding quantity or quality.
Bedding cleanliness – on a scale of 1 (clean) to 3 (dirty)	1.3	1.6	GOOD. Stall design, frequency of new bedding and cleaning all contribute to stall cleanliness.
Bedding dry matter	97 %	74 %	GOOD! Cows prefer to lie down on dry surface. Increased frequency of new bedding and stall cleaning helps maintain dry bedding.
Number of Cows / Stall *100 (Stocking rate)	100 %	104 % (Target: 100 %)	GOOD! Stocking rates of more than 100 % (not enough stalls for every cow) can reduce lying times and milk yield.
Percent of cows touching a stall that are perching (both front feet in stall, both rear feet in alleyway)	8 %	19 %	Perching in the stall indicates that cows find the stalls uncomfortable. Adding bedding, making the neck rail (if present) less restrictive (moving it towards the front of the stall) and widening stalls will reduce perching and risk of lameness.



FARM NAME

Assessment Date

Stall Dimensions	Your Measurement	Target	Comments
Curb height	7 in	8 in or less	GOOD. High curbs are associated with increased risk of lameness.
Stall width	44 in	50 in	Cows spend more time lying and are less likely to perch in wider stalls. Larger cows require larger stalls.
Stall length	101 in (single row) 96 in (double row)	120 in (single row) 102 in (double row)	Stalls should have adequate lunge space in front to allow cows to easily stand up and lie down.
Lunge barrier	Wall in front of single row stalls	none	
Neck rail height from bedding	46 in	50 in or greater	Neck rails increase the time cows spend standing in the alley or perching. Moving the neck rail up and further from the curb reduces these behaviours and the risk of lameness.
Neck rail distance from curb	61 in	70 in or greater	
Brisket board height	6 in	4 in or less	Brisket board reduces the amount of time cows spend lying in the stalls. High brisket boards prevent the forward thrust of the front leg when the cow rises.
Brisket board distance from curb	73 in	70 in or greater	



FARM NAME

Assessment Date

Feeding Area	Your Measurement	Target	Comments
Number of Cows / Headlock *100 (stocking rate)	n/a	100 %	Overstocking at the feed bunk increases competition. In turn, this reduces the time cows spend eating and increases the time cows spend standing in the alley.
Bunkspace per cow	23 in	24 in (mid lactation) 30 in (close up and fresh)	