



Lying behaviour of lactating dairy cows in a cow-calf contact system

Claire Sydney Wegner

Degree project • 30 credits
Swedish University of Agricultural Sciences, SLU
Department of Animal Nutrition and Management
Animal Science – Master's Programme
Uppsala 2021



Lying behaviour of lactating dairy cows in a cow-calf contact system

Claire Sydney Wegner

Supervisor: Emma Ternman, Swedish University of Agricultural Sciences, Department of Animal Nutrition and Management (HUV)
Examiner: Sigrid Agenäs, Swedish University of Agricultural Sciences, Department of Animal Nutrition and Management (HUV)

Credits: 30 credits
Level: Second cycle, A2E
Course title: Independent Project in Animal Science
Course code: EX0870
Programme/education: Animal Science – Master's Programme
Course coordinating dept: Department of Animal Nutrition and Management

Place of publication: Uppsala, Sweden
Year of publication: 2021

Keywords: Cow-calf contact system, dairy cattle, lying behaviour, welfare

Swedish University of Agricultural Sciences
Faculty of Veterinary Medicine and Animal Science
Department of Animal Nutrition and Management

Publishing and archiving

Approved students' theses at SLU are published electronically. As a student, you have the copyright to your own work and need to approve the electronic publishing. If you check the box for **YES**, the full text (pdf file) and metadata will be visible and searchable online. If you check the box for **NO**, only the metadata and the abstract will be visible and searchable online. Nevertheless, when the document is uploaded it will still be archived as a digital file.

If you are more than one author you all need to agree on a decision. Read about SLU's publishing agreement here: <https://www.slu.se/en/subweb/library/publish-and-analyse/register-and-publish/agreement-for-publishing/>.

YES, I/we hereby give permission to publish the present thesis in accordance with the SLU agreement regarding the transfer of the right to publish a work.

NO, I/we do not give permission to publish the present work. The work will still be archived and its metadata and abstract will be visible and searchable.

Abstract

Conventional dairy farming practices usually involve the early separation of calves from their dams. Cow-calf contact (CCC) systems may offer an alternative rearing solution that allows for the expression of natural behaviours, such as suckling and bonding. However, the effects of such systems on rest and lying behaviours of lactating cows are relatively unstudied. Cows are highly motivated to lie down, and a reduction in daily rest may have negative consequences for overall welfare. Thus, the aims of this study were: (1) to assess the effects of a CCC system on lying behaviours in lactating dairy cows, and (2) to determine if cubicle use of treatment dams and calves changes throughout the rearing period. Cow-calf pairs (n=37) were assigned 1 of 2 treatments after calving: dam-rearing (TREATMENT), where calves would be housed in the same facility as their dams, or separation shortly after parturition (CONTROL). Only TREATMENT cows had access to a modified lying area in which full CCC was available. Daily lying time – as well as the duration and frequency of lying bouts – was collected for cows automatically using leg-mounted tri-axial accelerometers (IceQube, IceRobotics, Edinburgh, UK). Video recordings were also collected and used to perform scan sampling at 10-minute intervals for a 24-h period each week. Behavioural data was collected during 14 weeks, starting when all cow-calf pairs had entered the experimental pen and continuing until weaning began. Access to full cow-calf contact did not affect lying behaviour. Overall, daily lying time and lying bout duration increased with consecutive time periods, while the frequency of lying bouts decreased. TREATMENT cows were observed to spend $76.6 \pm 29.2\%$ of their total lying time within the contact area across all weeks. These results combined indicate that lying behaviour in CCC systems is influenced by factors other than cow-calf contact, which cows are motivated to maintain throughout the rearing period.

Keywords: Cow-calf contact system, dairy cattle, lying behaviour, welfare

Table of contents

1. Introduction.....	10
2. Literature Review.....	12
2.1. Importance of rest for dairy cow welfare	12
2.1.1. Lying down as a behavioural need	12
2.1.2. Effects of reduced lying time.....	14
2.2. Factors that may affect lying behaviours.....	17
2.2.1. Cow-level factors	17
2.2.2. Management-based factors	21
3. Materials and Methods.....	25
3.1. Animals and treatments.....	25
3.2. Experimental facility and management	26
3.3. Collection of data.....	27
3.3.1. Behavioural observations	27
3.3.2. Lying behaviours.....	28
3.4. Statistical analysis	28
4. Results.....	30
4.1. Lying behaviour	30
4.1.1. Daily lying time.....	30
4.1.2. Frequency of lying bouts.....	30
4.1.3. Duration of lying bouts	32
4.2. Lying time within contact area	32
4.3. Stall use by calves.....	33
5. Discussion.....	34
6. Conclusions	39
References	40
Acknowledgements.....	48

List of tables

Table 1. Mean (SD) lactation weeks of cows during either of the 3 experimental periods. The week following and including a calving event was referred to as lactation week 0. Data is based on the final cleaned dataset.....	29
Table 2. Lying behaviours of cows during each of the 3 experimental periods. Data are presented as LSM means \pm S.E.M.....	31

List of figures

- Figure 1.** Mean proportion of total lying time spent in the cow-calf contact area during each of the experimental periods.32
- Figure 2.** Mean number of stalls occupied by calves (n=19) across all experimental weeks.33
- Figure 3.** Mean number of stalls occupied by calves (n=19) and cows (n=19) within the contact area each hour throughout a 24-h period. Cow data was summed in 10-min intervals and averaged per hour. All data is averaged across a period of 14 consecutive weeks.....33

Abbreviations

ACTH	Adrenocorticotropic Hormone
AMS	Automatic Milking System
CCC	Cow-Calf Contact
DIM	Days In Milk
HPA	Hypothalamic-Pituitary-Adrenal
NREM	Non-Rapid Eye Movement
REM	Rapid Eye Movement
SH	Swedish Holstein
SR	Swedish Red

1. Introduction

Under natural or semi-natural conditions, cows preparing to give birth will separate themselves from the herd and find a dry, sheltered place to use as a nesting site (Lidfors et al. 1994). In the first few days following birth, the calf will remain hidden in bushes or other tall vegetation while the mother grazes nearby (Vitale et al. 1986; Bouissou et al. 2001). During this time, a maternal-filial bond will begin to form through the engagement of licking and teat-seeking or suckling behaviours (Edwards & Broom 1982; Lidfors et al. 1994; von Keyserlingk & Weary 2007). The dam will rejoin the herd after a few days (Bouissou et al. 2001) and, quite quickly, the dynamic shifts from dam-initiated contact-seeking behaviours to the majority of social behaviours being initiated by the calf (Jensen 2011). The amount of time spent daily on suckling behaviours decreases as the calf grows older and ends altogether after weaning, a process that has been shown in semi-wild Zebu cattle to occur between 8-12 months of age (Reinhardt & Reinhardt 1981).

Conventional dairy farming practices the separation of cow and calf immediately after birth, thereafter raising the calf either in isolation or with conspecifics. Reasons cited for early separation are often related to economics and calf health, as milk intake and disease transmission can be more easily controlled in artificial rearing settings (Flower & Weary 2001). However, there are both production and welfare benefits to keeping dams and calves together. Delayed separation has been shown to result in higher calf growth rates (Grøndahl et al. 2007; Roth et al. 2009) and lower instances of abnormal behaviour (i.e., cross-suckling, tongue-rolling) (Margerison et al. 1999; Fröberg & Lidfors 2009; Roth et al. 2009). As a result, new housing systems have been developed that facilitate contact between dams and calves. These systems, known as dam-calf or cow-calf contact (CCC) systems, provide increased opportunities for cows and calves to express natural behaviours (as reviewed by de Oliveira et al. 2020). Within these housing systems, cow-calf pairs are housed together with either free or restricted contact, allowing for the development of a maternal-filial bond and natural suckling behaviours (Johnsen et al. 2016).

Previous research on alternative calf rearing systems has been largely focussed on calf behaviours and production measures, including pre- and post-weaning growth rates, separation stress response and cross-suckling or social behaviours (Le Neindre 1989; Johnsen et al. 2016; de Oliveira et al. 2020). To date, there has been

limited exploration on the potential effects of cow-calf contact systems on the cow, other than the amount of milk delivered to the milk tank. For example, very little is known about important rest-related behaviours in cows, such as the daily amount of time spent lying down. Lying time is a particularly important activity to monitor, as acute decreases in this behaviour may lead to impaired health and physiological functioning. Johnsen et al. (2021) recently evaluated the lying behaviours of both cows and calves in a CCC system, but the small sample size (8 cow-calf pairs in total) and lack of behavioural measures on control cows renders the study to be considered a pilot for further ethological research.

Lying behaviour is an important measure of cow health and comfort. As such, animal legislation and guidelines sometimes include outlines for 'healthy' resting times, with the implication that a failure to meet these recommendations may result in reduced welfare. For example, the Canadian Code of Practice advises that cows achieve daily lying times of at least 12 h/d (National Farm Animal Care Council, 2009). Many on-farm welfare assessments also utilize observations of lying behaviours to calculate estimates of stall use, such as the cow comfort index (Ito et al. 2009). This index represents the proportion of all cows within stalls that are fully lying down at a given point in time (Nelson 1996; Ito et al. 2009). However, this measure is based on a single estimation and – despite being widely used – has been shown to have little or no association with behavioural measures, including daily lying time as well as the frequency and duration of lying bouts performed (Ito et al. 2009).

Behavioural measures of rest may be used as indicators of underlying health issues; therefore, it is important to monitor individual variation (Weary et al. 2009). There is unfortunately a current lack information relating to lying patterns and behaviours in CCC systems. It is possible that cows may be forced to alter these behaviours when adapting to the added contact time with calves. Thus, the primary aim of this experiment was to determine how access to a cow-calf contact area affects the lying behaviours of lactating dairy cows. Additionally, the study aimed to explore how cubicle use and lying patterns of treatment dams and calves change throughout the rearing period.

2. Literature Review

2.1. Importance of rest for dairy cow welfare

In 1997, the concept of animal welfare as a trio of overlapping ethical concerns was first proposed, suggesting that quality of life can be determined by measuring the extent to which an animal is able to (1) live a natural life, (2) function well physiologically, and (3) maintain a positive affective state (Fraser et al. 1997; von Keyserlingk et al. 2009). It thus follows that certain natural behaviours are essential to an animal's welfare, which when prevented from being performed may result in poor biological functioning and behaviours suggestive of a negative affective state (i.e., stereotypies). The extent to which an ethological need may affect welfare is often dependent on the motivational strength to perform the behaviour(s) (Jensen & Pedersen 2008). It is often thought that animals are willing to work harder for access to resources that are essential for their well-being (Patterson-Kane et al. 2008). The following sections will explore the function of rest as a behavioural need, as well as the consequences of failing to adequately fulfill this need.

2.1.1. Lying down as a behavioural need

The relative importance of a particular resource to an animal can be quantified by its willingness to work for that resource (Munksgaard et al. 2005). To measure this motivation, animal ethologists often utilize techniques related to the economic concepts of elasticity of demand and income elasticity (Mason et al. 1998). The demand elasticity of a resource – in this case, the ability to perform lying behaviours – is usually measured using operant conditioning techniques, where access to a resource is offered at an increasing cost (Jensen & Pedersen 2008). This method allows for the creation of a behavioural demand function; the slope of the function represents the demand – or price – elasticity, and the area under the function can be used to calculate the maximum price paid to access a resource (Jensen & Pedersen 2008). This entire theory works on the idea that demand for a resource can be inelastic (even as the price of that resource rises, the demand stays constant) or elastic (Matthews & Ladewig 1994). Behavioural resources for which the demand

is inelastic are said to be necessities (Mason et al. 1998). Limiting the ability to access these resources can thus act negatively on overall welfare.

To date, a total of 3 studies have utilized the economic theory of demand elasticity in an attempt to quantify the motivation of dairy cows to rest (Jensen et al. 2004, 2005; Tucker et al. 2018). In all cases, cows were – to a certain extent – deprived of rest for a fixed period of time each day. Jensen et al. (2004, 2005) explored various aspects associated with the demand function for rest by training dairy heifers to press a panel in order to access the ability to lie down. In an initial set of experiments, heifers were tested under two different levels of rest deprivation, as well as controlled rest reward lengths of 10, 15 or 20 minutes (Jensen et al. 2004). Lying was prevented in all cases by means of a simple girth strap. Researchers found that the demand function with the longest reward period (20 minutes) had the lowest elasticity, meaning the cows were highly motivated to access these rest periods regardless of cost. Elasticity was also lower after longer lengths of rest deprivation. Further experiments tested heifers under a larger variation of reward durations, eventually leading to the conclusion that 50 minutes appears to be the most valid measure of the demand for lying (Jensen et al. 2005). While prescribing a fixed reward unit may pose limits on the external validity of the results (Mason et al. 1998) (i.e., applicability of results to situations beyond the scope of the study), longer reward periods come with a higher risk for interruption of the resting behaviour (Jensen et al. 2005). Ultimately, researchers determined that heifers have an inelastic demand of 12-13 hours of rest per day, and will work towards earning enough rest time (in the form of 50 min rewards) to maintain this (Jensen et al. 2005).

In a slightly alternative approach to the demand elasticity theory, Tucker et al. (2018) used a pneumatic push gate to measure the motivation of lactating cows to lie down. This approach presents animals with an “entrance fee”, which the animal must “pay” by pushing on a weighted gate to gain access to a resource – in this case, a deep-bedded lying area. Experimental results determined that cows are highly motivated to access a deep-bedded pack, especially when deprived of rest for 4 hours. In fact, the true extent of their motivation may be unknown; 5 of the 16 cows tested reached the maximum pressure allowed by the gate, therefore potentially underestimating their motivation.

An economic measure that is closely related to the elasticity of demand is income elasticity. Rather than manipulating the price of the resource, as was just demonstrated, income elasticity is measured by instead controlling the income received (Mason et al. 1998). This technique usually involves the manipulation of an animal’s total time budget and measuring the relative changes in the activities performed (Munksgaard et al. 2005). The idea is that when presented with a constricted time budget, animals will prioritize certain activities or behaviours over others (Munksgaard et al. 2005).

In what was perhaps the first study to attempt to quantify the relative importance of lying to other activities (i.e., feeding), Metz (1985) prevented a group of 20 freestall-housed cows from accessing a lying area for 3 hours each day, restricting them instead to an alleyway containing feed, water and a brush. Compared to control weeks, cows increased their lying time significantly during the 3 hours immediately following deprivation. In a second experiment, Metz (1985) simultaneously deprived cows from both feeding and lying down, and found that cows prioritized lying over eating once access to both resources was reobtained. More recently, researchers attempted to measure the priority of rest relative to feeding and socializing behaviours, both of which are well-known to be of importance to dairy cows (Munksgaard et al. 2005). Lactating cows were given controlled access to a pen containing the opportunities to feed, lie down and socialize with conspecifics. Cows had access to the pen for 23, 15 or 12 hours per day, with the remainder of each day spent in tie-stalls and deprived of feed and social contact, as well as rest via girth straps attached to ceiling rafters. As access time to the resource pen decreased, cows – both in early and late lactation – spent an increasing proportion of time lying down, to the extent that some of the cows lost weight due to reduced feed intake. However, the study may not have presented the cows with a true “closed economy” scenario; cows, while deprived of social contact with other cows in a physical and visual sense, still had olfactory and auditory contact with conspecifics during the deprivation periods. Dwarf goats, another gregarious species, have been observed to display more behavioural indications of emotional distress and frustration when subjected to complete isolation rather than partial isolation, where both acoustic and olfactory contact were accessible (Siebert et al. 2011). It may be that the cows were able to gain some degree of social satisfaction during the deprivation period, possibly to the extent that their relative motivation to access social contact in the pen decreased (Munksgaard et al. 2005).

Overall, these studies all contribute to the idea that lying down is a behaviour that cows are highly motivated to perform, especially after a period of deprivation. In commercial settings, this may translate to the time spent waiting to be milked. Nonetheless, rest is a clear ethological need for dairy cows, one which producers are responsible for ensuring their animals have the ability to fulfil within their respective management systems.

2.1.2. Effects of reduced lying time

When dairy cows are unable to satisfy their motivation to lie down, their overall welfare may be negatively affected. A deprivation of sufficient lying time can lead to various adverse behavioural and physiological changes, as well as an increased risk for disease.

Effects on behaviour

Experimental rest deprivation has been shown to result in a variety of behavioural responses. Munksgaard and Simonson (1996) first scored the behavioural effects of depriving a group of mid-lactation cows from lying down for 14 h/d. They found throughout the experiment, which lasted a total of 8 weeks, cows deprived of lying performed acts of oral manipulation and leaning at a greater frequency and duration than control cows. These behaviours are stereotypic and may indicate that the animals are experiencing some degree of frustration or stress (Broom 1983). Similarly, young bulls experiencing the same level of deprivation, but for a slightly longer period (10 weeks), were also shown to perform higher rates of oral manipulation (Munksgaard et al. 1999). In both cases, animals were subjected to levels of deprivation likely not experienced in commercial settings. However, even short periods of deprivation (i.e., 2 or 4 hours) can result in an increase in leg stomping, repositioning, weight shifting and head swinging – all of which are behaviours thought to be indicative of discomfort, frustration and/or tiredness (Cooper et al. 2007, 2008).

Effects on limb and hoof health

The relationship between lying time and lameness is complicated; a reduction in the time spent lying down will, of course, result in increased time spent standing, which is a known risk factor for lameness. Modern dairy housing systems often use hard flooring substrates, such as concrete, due to their affordability and resistance to wear (Telezhenko & Bergsten 2005). However, the use of hard flooring surfaces can increase the risk for developing lameness-causing diseases, including white-line disease, digital dermatitis and sole ulcers (Bergsten 2001; Somers et al. 2003).

A cow that is lame will also alter her lying behaviours. High lying times are therefore not to be interpreted as a definitive indication of better welfare. Due to the difficulties in differentiating a change in lying time as being causal or consequential of lameness, many studies simply report potential associations between lying time and an increased risk of lameness (i.e., Ito et al. 2010).

A limited number of studies have attempted to test the direct effects of lying deprivation on hoof health, with varied results. Leonard et al. (1996) housed primiparous cows at a stocking density of 200 % for a 4-month period after calving. While claw health deteriorated for all cows, those with the shortest average daily lying times displayed higher claw haemorrhage scores than cows receiving more rest. The study also reported a significant negative correlation between foot lesion score at 4 months post-calving and lying behaviour in the month prior. In contrast, Ouweltjes et al. (2011) observed heifers for sole hemorrhages 1 and 6 weeks post-restriction and found no differences from control cows. However, the animals in this case were only deprived of lying for 6 hours each day and were able to

compensate by altering their time budgets. This, in combination with the relatively short restriction period (8 weeks), may explain the results.

Effects on physiological functioning

There is some evidence to suggest that insufficient rest – or potentially a combined deprivation of lying and sleep – may induce some physiological responses in cows and increase the risk for nutritionally-related disorders. The hypothalamic-pituitary-adrenal (**HPA**) axis is responsible for regulating the release of hormones – such as cortisol – in response to stress (Brown & Vosloo 2017). Exactly how the HPA axis responds to lying deprivation in cows is unclear, as the experimental results are quite varied. Young bulls forced to stand for 14 h/d displayed an increased cortisol response to injections of adrenocorticotrophic hormone (**ACTH**), the hormone responsible for cortisol regulation (Munksgaard et al. 1999). Meanwhile, lactating cows subject to 8 weeks of an identical forced standing treatment presented no notable changes in the HPA axis (Munksgaard & Simonsen 1996). It remains unclear as to what extent – if at all – lying deprivation influences the physiological stress response in cattle, despite clear behavioural indicators of stress.

Another potential consequence of rest deprivation in cattle may be a reduction in sleep. The effects of sleep deprivation are well documented in humans; a lack of sufficient sleep can have severe consequences on overall health and cognitive functioning (reviewed by Alvarez & Ayas 2004). In contrast, there exists very little research on sleep – and a lack of it – in cattle. Cows are known spend approximately 4 hours per 24 hours in either non-rapid eye movement (**NREM**) and rapid eye movement (**REM**) sleep states (Ruckebusch 1972), divided into short intervals of 3-5 minutes (Ternman et al. 2012). The remainder of each day is spent either fully awake or in a state of wakefulness known as drowsing (Ruckebusch 1972). This large difference in sleeping patterns – as well as the various documented effects of lying deprivation – leads to the belief that sleep in cows may not serve the same purpose as in humans. To date, a single study exists that has attempted to observe both the separate and combined effects of sleep and lying deprivation in dairy cows (Kull et al. 2019). Twelve mid-to-late lactation cows were subjected 1 of 2 treatments in a crossover design: a 24-hour lying deprivation period, or 24 hours of sleep deprivation. Since cows are thought to be unable to perform REM sleep while standing (Ruckebusch 1974), the forced standing treatment was essentially a combined deprivation of both lying and REM sleep. Cows deprived of lying down exhibited a slight decrease in milk production in the days immediately following deprivation (Kull et al. 2019). Additionally, these cows displayed higher levels of TNF and IL1B cytokines expressed by whole-blood leukocytes, a known physiological response indicating inflammation (Proudfoot et al. 2021). It is possible that these physiological changes may instead – or in part – be an effect of

forced standing for 24 hours. It should also be noted that these results may only be applicable to mid-to-late lactation cows. Further research is thus necessary in order to draw any conclusions regarding the effects of sleep deprivation in cows.

Cows that are unable to obtain sufficient rest may also be at a higher risk for nutritionally-related disorders, such as ruminal acidosis. Rumination occurs predominantly while cows are lying down (Albright 1993) and plays an important role in the remastication and reswallowing of food particles – processes through which saliva is produced (Beauchemin 1991). Saliva aids in buffering the volatile fatty acids in the rumen, thus preventing the pH from dropping too low (Beauchemin 1991). Cows that are prevented from lying down for an extended period of time (i.e., 2-4 hours) may decrease their total daily rumination time and instead spend more time standing without ruminating (Cooper et al. 2007, 2008). This may increase the risk for developing subacute ruminal acidosis, a condition with serious health and production consequences (Plaizier et al. 2008). Admittedly, examination of rumination activity in dry cows with regards to lying time has been unsuccessful in finding a correlation between the two measures (Schirmann et al. 2012). It is therefore currently unknown how short lying times may affect ruminal changes in other bovine demographics, including cows in different stages of lactation.

2.2. Factors that may affect lying behaviours

When offered the opportunity, cows have been shown to lie down 12-13 hours per day (Jensen et al. 2005). However, the levels of rest actually achieved in commercial settings is highly variable, both within herds and between farms (Ito et al. 2009; Gomez & Cook 2010; Charlton et al. 2014). Lying behaviours – including daily lying time, as well as the frequency and duration of individual lying bouts – have been associated with a vast variety of different animal- and management-based factors. Some of these factors can be controlled by producers (i.e., choice of stall bedding), while others, such as a cow's stage of lactation, can only be taken into consideration. The following sections will review some of the most common factors that have been experimentally linked to changes in lying behaviours.

2.2.1. Cow-level factors

Various aspects of a cow's current state can play a role in influencing their individual rest patterns. Dairy cows will alter their lying behaviours in response to sickness and changes in reproductive hormone levels. Furthermore, cows of different ages and stages of lactation may also differ in their behaviours.

Disease and injury

Animals are known to alter certain behaviours in response to sickness and pain (Weary et al. 2009). As such, behavioural changes are sometimes used as indicative measures to identify ill individuals (Weary et al. 2009). Extensive research has been conducted attempting to link changes in lying behaviours with an increased risk for lameness or mastitis. Therefore, these two maladies will be the focus of this review, specifically with regards to their interactions with lying time and lying bout characteristics in dairy cows.

As previously mentioned, the relationship between lying behaviour and lameness is complex; yet, it is generally agreed upon that lame cows will spend more time lying down than non-lame cows (Chapinal et al. 2009; Ito et al. 2010; Blackie et al. 2011; Westin et al. 2016). One epidemiological study collected data from 28 dairy farms through Western Canada, and reported that high lying times (>14.5 h/d) and long lying bouts (>90 min/bout) were associated with increased odds of a cow being severely lame (Ito et al. 2010). Similarly, Westin et al. (2016) assessed cows across 36 AMS farms and found associations of clinical lameness with longer daily lying times and fewer, but longer, lying bouts. Blackie et al. (2011) studied lying behaviours in chronically lame cows (lame for 3 consecutive months) and observed a 2.1 hour increase in daily lying time compared to nonlame cows.

However, a few studies have reported contradictory results. Galindo and Broom (2000) found a significant association between increased daily standing times and cows diagnosed with lameness or hoof lesions, suggesting that these cows were lying down less than healthy cows. Cook et al. (2004) selected focal cows from 12 different freestall herds and observed that moderately lame cows had lower daily lying times than nonlame cows. It should be noted that an extremely small sample of moderately lame cows (n=10) was used in this analysis, and thus the average lying time projected may not be representative of cows with that particular lameness score. Lying time is highly variable between individuals and previous estimates have suggested that a minimum of 30 cows are necessary to obtain accurate herd estimates (Ito et al. 2009). A third study observed equal numbers of lame and nonlame from 10 different freestall farms and discovered no difference in daily lying time or the daily frequency of lying bouts (Yunta et al. 2012). Lame cows did, however, perform significantly longer lying bouts than nonlame cows – a similar finding to those of Westin et al. (2016) and Ito et al. (2010). The inconsistent results depicted in these three studies are possibly due to interactions of one or more cow- or herd-level factors with lameness – a phenomenon that may thereafter have influenced the lying behaviours of lame cows. Furthermore, lameness was primarily diagnosed by locomotion scoring without identification of what factors may have caused the disease. Although not experimentally confirmed, it is possible that lame cows may behave differently depending on the causation for the condition. These

inconsistencies, in addition to the high degree of individual variation and lack of a threshold for defining lameness, provide arguments against utilizing lying behaviour as a sole indicator for detecting lameness (Ito et al. 2010).

Mastitis is another major disease common in dairy cows, known to negatively affect welfare and productivity. Research, while slightly more limited, is suggestive of cows responding behaviourally to mastitis in a manner that is opposite to what has typically been reported for lameness. Cows with induced acute (Siivonen et al. 2011) or naturally-occurring mastitis (Medrano-Galarza et al. 2012) spend less time lying down per day than control cows. While these findings are opposite to typical mammalian sickness behaviour – where rest is usually increased – the reduced lying times may be due to the cows having increasingly swollen udders (Siivonen et al. 2011). Mastitic cows may therefore find lying down to be painful as a result of the increased pressure. Regardless, associations of lying behaviour and mastitis, as well as other types of infections, have not been well studied. As such, lying behaviours should not currently be considered in the diagnosis or risk assessment of infectious diseases such as mastitis.

Estrus

Estrus – also known as standing estrus or standing heat – is characterized behaviourally in cows by increased levels of restlessness and activity, and can range from 7-18 hours in length (Diskin & Sreenan 2000). The intensity of displayed estrus behaviours, such as mounting, can be influenced by management factors, including flooring surface or stocking density (Diskin & Sreenan 2000). Therefore, the extent to which estrus influences rest may depend on these factors as well.

Numerous studies have illustrated clear associations between reproductive state and daily lying time. Cows in heat perform fewer (but not shorter) lying bouts, resulting in significant decreases in daily lying time compared to when not in estrus (Dolecheck et al. 2015; Silper et al. 2017). Heifers respond similarly to estrus; Silper et al. (2015) reported a 35% increase in standing time when heifers were in heat, translating to an almost 5 hour difference in daily lying time. More recently, a study compared the lying behaviours of cows either in behavioural or silent estrus, the latter of which is characterized by a lack of behavioural signs (Zebari et al. 2018). Cows in behavioural estrus decreased their daily lying time by approximately 3 hours compared to measures taken 3 days before or after the reproductive event. However, silent estrus events did not elicit any changes in lying time or the number of lying bouts performed.

It is clear that estrus events must be taken into consideration when studying activity patterns or time budgets in open cows.

Stage of lactation

Lying time has repeatedly been shown to be associated with the stage of lactation – also referred to as the number of days in milk (**DIM**) – a cow is in. Large-scale studies of freestall-housed dairy herds have generally reported that the average daily lying time increased with lactation stage (Bewley et al. 2010; Gomez & Cook 2010; Ito et al. 2014; Solano et al. 2016; Westin et al. 2016). Recent findings from Maselyne et al. (2017) suggest that this relationship between lying time and DIM is non-linear. Lying data collected from 200 cows across 4 Danish freestall farms showed that while lying time does increase from early to late lactation, it first decreases from the very start of lactation until approximately 28 DIM (Maselyne et al. 2017). However, lying times reported in the first 10 DIM were still significantly lower than those in a late stage of lactation (265 DIM), which could explain the seemingly linear findings of other large-scale association studies.

Lying behaviours within different stages of lactation have also been observed experimentally. Chaplin and Munksgaard (2001) found that in a 21.5-hour period, tethered cows in early lactation (< 100 DIM) spent significantly less time lying down than late lactation (> 200 DIM) or dry cows. Another study examined the lying behaviours of both tethered and freestall-housed cows in 3 different stages of lactation (Vasseur et al. 2012). Daily lying time was shown to increase with lactation stage in both housing systems. Vasseur et al. (2012) also noted an effect of DIM on the frequency and duration of lying bouts, although the extent of this influence was further dependent on parity.

Multiple explanations have been brought forth in attempts to explain the reduced lying time in early lactation. One suggestion is that cows in early lactation experience greater levels of discomfort while lying due to increased udder pressure, and that this distension decreases as cows progress through lactation (Vasseur et al. 2012). Alternatively, cows in early lactation may be spending more time eating to meet the demands of high milk production and, as a result, be spending less time on rest (Bewley et al. 2010). Regardless of why this difference occurs, lactation stage should be considered when designing or analyzing experiments related to lying behaviours.

Parity

Similar to findings on lactation stage, herd-level studies of freestall-housed cows have found increased daily lying times to be associated with an increase in parity (Solano et al. 2016; Westin et al. 2016). Short-term observational studies of lying behaviours in primi- and multiparous cows have reported slightly more varied results. Grazing cows observed in the first 3 weeks following calving displayed slight differences between the two parity classes, with multiparous cows lying down approximately 1 hour more per day than primiparous cows (Sepúlveda-Varas et al. 2014). In contrast, Neave et al. (2017) found no differences in lying time between

freestall-housed primi- and multiparous cows, also having examined a 3-week post-calving period.

While the effects of parity on daily lying time are not entirely clear, there is a substantial amount of evidence suggesting differences in the length and frequency of lying bouts performed by primiparous and multiparous cows. Particularly in early lactation, primiparous cows have been shown to lie down more frequently than older cows, but for shorter durations of time (Vasseur et al. 2012; Sepúlveda-Varas et al. 2014; Solano et al. 2016; Neave et al. 2017). A plausible explanation for this difference in lying behaviours is that primiparous cows may be experiencing higher levels of stress (Solano et al. 2016). This stress may be the result of various changes related to calving for the first time, such as the forced introduction into a novel social environment. The initial increased frequency of transitions from standing to lying seen in primiparous cows may be a reflection of restlessness (Solano et al. 2016). As lactation progresses, lying bouts become longer and less frequent, perhaps representing a gradual adaptation to the new environment (Vasseur et al. 2012). Lying behaviours in primiparous may also be influenced by more socially dominant individuals, as age has previously been correlated with social dominance in dairy cows (Beilharz & Zeeb 1982). Depending on the stocking density, there may have been high levels of competition to access lying space. However, as dominance has been shown to be resource-dependent (Val-Laillet et al. 2008), age alone cannot define submissiveness. Ultimately, there is a lack of knowledge regarding the social dynamics of stall use, and thus this explanation for the lying behaviours observed in primiparous cows can neither be confirmed nor rejected.

2.2.2. Management-based factors

Lying behaviour in herds of dairy cows can be influenced by any number of individual and interacting factors related to housing or management. Therefore, this section will focus on a selection of factors that have been extensively studied in relation to rest behaviours, including housing and milking system, stocking density and stall design and management. For the purpose of this review, housing system and stall design will be considered management-related factors rather than environmental factors, as the choices relating to designing either are ultimately a management decision.

Housing and milking system

Cows in conventional freestall housing systems spend between 10 and 12 hours per day lying down, as estimated in a previous literature review (Tucker et al. 2021). This is similar to average lying times reported for cows housed in tie-stalls (Vasseur et al. 2012) and in outwintering pad systems (O'Driscoll et al. 2009). However, this assessment was drawn from a number of large-scale studies that estimated herd

lying times across as many as 141 different farms. In reality, the between-farm variation exhibits a much greater range. For example, Ito et al. (2009) assessed lactating cow on 43 different freestall farms and reported a between-farm variation in lying time of 9.5 to 12.5 h/d. This means that some farms were achieving herd rest averages that were lower than recommended lying times. Furthermore, this indicates that housing system alone (i.e., freestall, tiestall) does not influence lying behaviours; instead, there are likely a large number of management and cow-level factors that influence the amount of rest cows are able to obtain.

Within the results of herd-level studies examining freestall housing systems, no obvious differences in lying time can be noted between farms that use milking parlours (Ito et al. 2009, 2014; Gomez & Cook 2010; Charlton et al. 2014; Solano et al. 2016) and those with automatic milking systems (**AMS**) (Deming et al. 2013; Helmreich et al. 2014; Westin et al. 2016; King et al. 2017). The possible effects of different milking systems have also been tested experimentally, with slightly varying results. Lexer et al. (2009) housed similarly-composed groups of cows in freestall barns equipped with either a herringbone parlour or a single automatic milking robot, and found no difference in the daily proportion of time spent lying down. Comparatively, in a separate study, Spolders et al. (2004) collected hourly lying estimates for an extended period of time (110 days) and observed that cows housed in parlour systems were lying down for a greater daily proportion compared to automatically milked cows. Furthermore, within AMS herds, whether cow traffic is forced, semi-forced or free does not appear to have an effect on the time cows spend resting each day (Hermans et al. 2003; Munksgaard et al. 2011).

Stocking density

Overstocking is often practiced as a means of increasing overall farm profitability (Krawczel & Lee 2019). As many as 60 % of North American freestall dairy producers maintain stocking densities greater than 100 %, meaning that they house more cows in a pen than there are available stalls (von Keyserlingk et al. 2012). While some herd-level studies have reported a lack of association between stocking density and lying times (Charlton et al. 2014; King et al. 2017), experimental manipulation of stocking density has garnered evidence that suggests an association does exist. Various studies have explored the effects of increasing the stocking density for a short period of time (i.e., 1-2 weeks) on daily lying time, particularly in systems utilizing milking parlours. Compared to a density of 100 % (1 cow per stall), cows housed at a 150 % stocking density show decreases in daily lying time of 1.2-1.7 hours (Fregonesi et al. 2007a; Winckler et al. 2015). Other studies have also reported significant decreases in lying time at densities of 131 % and 142 %, although the differences were not quite as large (0.5-0.9 h/day) (Hill et al. 2009; Krawczel et al. 2012).

More recently, a similar experimental approach was applied to freestall-housed cows with automatic milking (Witaifi et al. 2018). It was initially predicted that overstocking may have less of an effect on overstocked AMS-housed cattle, as their lying behaviours are likely less synchronous than that of parlour-milked cows (Witaifi et al. 2018). Yet, cows housed at a density of 150 % were still observed to spend less time lying down than those kept at densities of 120 % or 100 %. It is clear that stocking cows at densities greater than 1 cow per stall may negatively influence the daily amount of rest they are able to obtain, thus potentially outweighing the monetary profit gained from maintaining a higher overall milk production.

Stall design and management

Lying time is often considered to be a direct measure of cow comfort (Haley et al. 2000). The particular combination of material components utilized in freestalls – including the stall base and bedding type – will determine the level of comfort, thus directly influencing the daily lying time of freestall-housed cattle (McPherson & Vasseur 2020). Cows have been shown to have higher lying times in stalls that utilize soft rubber mats or mattresses as their base, as opposed to a concrete base (Haley et al. 2000; Rushen et al. 2007; Norring et al. 2010).

The bedding material used to cover the stall base can also play an influential role in cattle resting behaviour. However, out of the materials most often experimentally tested (straw, sand, sawdust and wood shavings), no one substrate has been determined to consistently result in increased lying times. Instead, cows have been shown to display a tendency for higher resting times when housed in soft, deep-bedded stalls, regardless of substrate (Tucker et al. 2003; Ito et al. 2014; Solano et al. 2016). A cross-sectional study of 40 different freestall farms across northeastern United States reported that herds housed in deep-bedded stalls had average daily lying times that were approximately 0.8 hours higher than in herds utilizing other types of bedding (Ito et al. 2014).

Researchers have also attempted to quantify the relative preferences of cows for various bedding substrates. Through a series of free-choice and restriction phases, Tucker et al. (2003) discovered that dry cows spent significantly less time lying down on mattresses covered with a thin layer of sawdust than on deep-bedded sawdust or sand stalls. In a separate study, Manninen et al. (2002) reported a clear preference for deep-bedded straw and lightly bedded rubber mats over deep-bedded sand stalls. In any case, cow preferences are likely to be influenced by prior experience (Duncan 1992). This has been shown to be true, particularly with sand bedding (Manninen et al. 2002; Tucker et al. 2003; Norring et al. 2010); cows that had not previously experienced sand bedding displayed visible reductions in their daily lying times when restricted to sand-bedded stalls.

Two additional factors that may influence daily lying time are the quantity and quality of the bedding used. Due to the high costs associated with bedding materials such as straw or wood shavings, the quantity of bedding utilized in freestalls is often kept to a minimum (McPherson & Vasseur 2020). Canadian freestall farms that bed their stalls at a depth of 2 cm or greater tend to have higher daily lying times than farms that use less bedding (Solano et al. 2016). Cows have also been experimentally tested on different depths of bedding material. Tucker et al. (2009) housed lactating cows with straw or shavings at a series of different depths and found that lying time increased as the amount of bedding increased. For every additional kilogram of straw or shavings, cows increased their daily lying time by 12 minutes and 3 minutes, respectively. Another study subjected cows to a variety of sand depths, noting that for every 1 cm decrease in depth, cows reduced their daily resting time by 11 minutes (Drissler et al. 2005).

Additionally, the cleanliness of a bedding substrate is crucial in upholding high levels of cow comfort and health. Poor stall management may lead to bedding becoming wet or soiled. Stalls bedded with wet (as opposed to dry) sawdust have been shown to result in rest decreases of up to 5 h/d (Fregonesi et al. 2007b). Instead, cows increased the amount of time spent standing half or fully in the stalls. Similarly, Chen et al. (2017) depicted a clear decrease in daily lying time as the muddiness of the designated lying area increased. In this case, dry cows decreased the frequency of lying bouts performed and instead increased the proportion of time they spent lying or standing on a concrete pad. The consequences of prolonged standing on hard flooring substrates, such as concrete, have previously been described as potentially contributing to an increased risk for lameness (see section 2.1.2).

Ultimately, it appears that dairy cows are able to achieve the highest lying times when offered stalls that are bedded with a soft, dry substrate. Moreover, caution should be maintained when making changes to bedding substrate, as novel materials may result in significant decreases in lying time.

3. Materials and Methods

This study was conducted at the Swedish University of Agricultural Sciences' Swedish Livestock Research Centre (Lövsta lantbruksforskning) in Uppsala, Sweden from Sept 2020 to Jan 2021 as part of a larger research project. All procedures outlined were approved by the regional ethics committee (ID-No: 5.8.18-18138/2019).

3.1. Animals and treatments

A total of 40 multi- (n = 19) and primiparous (n = 21) cows were used in this experiment. All cows in the herd that calved within the time frame of Sep 1 and Oct 15, 2020 were eligible for inclusion. Upon calving, cow-calf pairs were assigned 1 of 2 treatments: dam-rearing (TREATMENT), where calves would be housed in the same facility as their dams, or separation shortly after parturition (CONTROL). Treatment groups were balanced for calf age and gender, such as that every other heifer calf or bull calf born was separated following parturition. Dam breed (Swedish Holstein (**SH**), n = 15; Swedish Red (**SR**), n = 25) and parity were also considered during treatment allocation as 2nd and 3rd tier criteria. On average, cows were a parity of 2 (± 1) and produced 2726 (± 1057) kg of milk in the first 100 days after first entering the experimental pen. CONTROL calf housing and management will not be described in further detail, as they are not relevant to the aims of this study.

The experimental period began once the last cow-calf pair entered the pen and lasted for a total of 14 weeks, after which calves were weaned and consequentially removed from the experimental pen. Calves were an average 24 (± 12) days old when observations began. During the study, 1 calf was euthanized due to severe lameness, 1 cow died following treatment for *E. coli* mastitis and another cow was removed after being diagnosed with mastitis. The final number of cow-calf pairs available for statistical analysis was 37 (19 TREATMENT and 18 CONTROL).

3.2. Experimental facility and management

Cows were housed in an insulated building with freestalls and automatic milking. The experimental pen was divided into 4 distinct areas: the feed alley, the automatic milking system and two lying areas that differed in the degree of cow-calf contact. Cow traffic through the pen was partly managed with the use of an automatic selection gate (DeLaval smart selection gate SSG, DeLaval International AB, Tumba, Sweden), operating a Feed FirstTM system (DeLaval International AB, Tumba, Sweden). Upon exiting the feeding alley, cows entered the selection gate and were sorted in one of three directions, depending on treatment and milking permission status. Cows that were due to be milked were sent into a holding pen, which led to an automatic milking robot (DeLaval VMSTM Classic, DeLaval International AB, Tumba, Sweden). Milking permission was granted 6 hours following the previous milking session. TREATMENT cows were otherwise directed forwards to the CCC area, while CONTROL cows were sent to the general lying area. The maximum number of cows housed within the pen at any given point was 58, although this number was often lower.

The CCC area was accessible by both calves and TREATMENT cows and contained a total of 24 freestalls. In this area, dams had full, unrestricted access to their calves, as well as controlled access to 2 concentrate feeding stations (DeLaval feed station FSC400, DeLaval International AB, Tumba, Sweden). Calves also had access to a separate calf creep: a 73.2 m² deep-bedded wood-shavings area that ran the entire length of the pen. Roughage and water were available ad libitum to calves within the creep. Access to the creep was limited to calves by way of horizontally placed wooden boards. Cows were able to exit the contact area through a pair of spring-loaded one-way gates (FeedSelect, GEA Farm Technologies GmbH, Bönen, Germany), which led directly to the general lying area.

The general lying area consisted of 38 stalls, distributed unevenly across 4 rows. Two of these rows lined a center alley that ran adjacent to the feed alley, while the remaining rows of stalls were situated in an alley alongside the calf creep. As such, cows could access visual, auditory and olfactory contact with the calves from this area; physical contact was, however, largely restricted due to the placement of wooden beams at multiple heights. Cows in the general lying area also had access to concentrate via 2 concentrate feeding stations. From the lying area, cows could access the feed alley through 1 of 2 different one-way gates.

In the feed alley, cows had free access to 7 water cups, 20 individual forage bins (CRFI, BioControl AS, Rakkestad, Norway) and 2 swinging brushes (DeLaval SCB, DeLaval International AB, Tumba, Sweden). Fresh feed – a specially formulated TMR – was delivered to the bins 5 times per day via a rail-suspended distribution wagon (DeLaval FS1600, DeLaval International AB, Tumba, Sweden). The only way for cows to exit the feed alley was to enter the automatic selection gate, thus maintaining circularity of cow traffic within the housing system.

Flooring throughout the alleyways encompassing the lying areas was grooved concrete, while the feed alley was equipped with rubber mats. The holding pen leading to the AMS was the only area containing rubber slatted flooring. Manure was removed using automatic scrapers, which ran through the feed alley once every 30 minutes, and through all alleys encompassing the general lying area once per hour. In the contact area, the scraper was initially run manually during day hours to minimize the risk of injury to young calves. On Nov 11, the scraper was set to start running automatically during the day while still being turned off at night. From Nov 23 onwards, manure scraping in the contact area was conducted automatically during both day and night hours.

Stalls were bedded with rubber mats and a layer of sawdust, which was topped up 4 times per day by use of a rail-suspended shavings dispenser (JH miniStrø COW, MAFA i Ängelholm AB, Ängelholm, Sweden). Stalls in the contact area received 3 additional distributions of sawdust throughout the day.

3.3. Collection of data

3.3.1. Behavioural observations

A total of 8 fisheye cameras (Samsung SNF-8010VM, Samsung Techwin Co., Ltd., Seoul, South Korea) were used to continuously monitor the experimental areas throughout the duration of the study. All cameras were mounted to provide an overhead view, with 4 placed over both the calf creep and cow-calf contact area. Additionally, 2 cameras were positioned over the general lying areas, one overlooking the VMS holding pen and selection gate passageway, while the final camera was placed above the feed alley. Video recordings were later viewed using the program BackupViewer (v2 1.4.6_M190708).

Each TREATMENT cow was marked with a unique symbol on her sides and back using blue (KRUUSE Marking Spray, Jørgen Kruuse A/S, Langeskov, Denmark) or yellow (RAIDEX Animal Marking Spray, RAIDEX GmbH, Dettingen an der Erms, Germany) animal-safe paint to facilitate identification of individuals. Markings were refreshed every week (± 1 day). Scan sampling was performed at 10-minute intervals for a 24-h period each week using the video recordings, starting at 0000 h and ending at 2350 h. During each scan, the IDs of all cows lying down in stalls that were located within the contact area were recorded. Cows in the process of lying down or getting up (i.e., not in a fully horizontal position) were not included. The number of stalls occupied by resting calves was also logged at each scan.

3.3.2. Lying behaviours

All experimental cows were equipped with leg-mounted tri-axial accelerometers (IceQube, IceRobotics, Edinburgh, UK) which automatically recorded the time spent lying (min), as well as the frequency and length of individual lying bouts. Once every week, data was manually downloaded from each cow using an IceReader (IceRobotics, Edinburgh, UK) and laptop containing the IceManager program (IceRobotics, Edinburgh, UK).

3.4. Statistical analysis

All data analysis was carried out using SAS OnDemand for Academics (v. 3.1.0, SAS Institute Inc., Cary, NC). Prior to analysis, IceQube data was manually transformed into .csv files using the IceManager program. Daily summaries were obtained for lying time (min/d), lying bout duration (min/bout) and lying bout frequency (bouts/d) using the SUMMARY procedure in SAS. For lying bout duration data, outliers were removed to correct for the overlap in bouts between days. Each lying behaviour was then averaged weekly across a 3-day period, based on previous research that suggests this is the minimum observation time necessary to attain an accurate estimate (Ito et al. 2009). Means composed of fewer than 3 complete days of data were not included in the final dataset. Lactation weeks were calculated for cows using their calving dates (Table 1); observations with a lactation week less than 1 were removed from the dataset. Furthermore, all observations during which a cow was in estrus were removed. Experimental weeks were then grouped into 1 of 3 time periods: early (wk 1-5), peak (wk 6-9) or post-peak (wk 10-14). This was done so as to be able to analyze and compare the lying behaviours during distinct periods throughout the trial, each of which corresponds to a stage of lactation. Parity data was used to classify cows either as being primiparous (parity = 1) or multiparous (parity > 1). Finally, one cow was temporarily moved to a sick pen while undergoing treatment for mastitis; observations occurring during this time period were subsequently removed. The final dataset consisted of 376 observations and was checked for normality using the UNIVARIATE procedure of SAS.

Each of the three lying behaviours were analyzed as continuous dependent variables using the MIXED procedure, with individual cow identity included as a repeated measure. Breed (SH and SR), parity (primi- and multiparous), treatment (TREATMENT and CONTROL) and experimental time period (early, peak and post-peak) were included as fixed effects in each model. Lactation week was ultimately not included due to the confounding effect on time period. As it was assumed that the measures of each individual cow across time were correlated, a first-order autoregressive covariance structure was specified in the model. To test

for differences within treatment or time period, additional models were run where either variable was removed from the fixed effects and instead used to create a data subset through input of a WHERE statement. Differences in LSMeans were adjusted using the Tukey-Kramer option within the MIXED procedure.

Observational data collected via the video recordings was summarized to obtain the daily frequency of lying events for each treatment cow. This data was then combined with the 1-day mean lying data corresponding to the observation dates, and thereafter used to calculate the average weekly proportion of time spent lying down within the cow-calf contact area. A MIXED procedure was used to test the fixed effects of breed, parity and time period on the daily frequency of lying events. Cow identity was specified as a repeated measure and the covariance structure stated to be autoregressive.

Calf stall use data, also collected via video recordings, was averaged per hour and fitted into a simple MIXED procedure to test the fixed effects of hour and experimental week on the number of stalls occupied by calves within the contact area. Observational cow data was used to calculate corresponding cow stall use, which was thereafter averaged per hour and plotted against hourly stall use by calves for visual comparison. Estrus events were not removed for this comparison of stall occupancy.

Within this study, significant differences were declared when $P < 0.05$, and results presents as LSMeans \pm SEM or mean(SD) if not else stated.

Table 1. Mean (SD) lactation weeks of cows during either of the 3 experimental periods. The week following and including a calving event was referred to as lactation week 0. Data is based on the final cleaned dataset.

Time Period	TREAMTENT	CONTROL
Early	5.0 (2.3)	5.4 (2.3)
Peak	9.8 (2.2)	10.1 (2.2)
Post-peak	13.9 (1.9)	14.2 (2.1)

4. Results

4.1. Lying behaviour

There was no overall effect of treatment on daily lying time ($F_{1,337} = 0.97$, $P = 0.33$), the frequency of lying bouts ($F_{1,337} = 0.17$, $P = 0.68$) or lying bout duration ($F_{1,337} = 0.00$, $P = 0.97$).

4.1.1. Daily lying time

Cows spent on average 11.6(1.9) h/d lying down throughout the experiment, regardless of treatment. No further treatment differences were found within any of the experimental time periods (Table 2). Moreover, neither parity ($F_{1,337} = 0.79$, $P = 0.37$) nor breed ($F_{1,337} = 2.17$, $P = 0.14$) were significant in the model. However, lying time increased significantly with time period for all cows ($F_{2,337} = 43.14$, $P < 0.01$). The post-hoc analysis showed that TREATMENT cows increased their lying time by approximately 1.3 h/d from the early to post-peak period in the experiment (SE = 0.21, $P < 0.01$), while CONTROL cows saw a similar increase of approximately 1.4 h/d (SE = 0.21, $P < 0.01$).

4.1.2. Frequency of lying bouts

No overall effects were found of either parity ($F_{1,337} = 2.82$, $P = 0.09$) or breed ($F_{1,337} = 2.48$, $P = 0.12$) on the frequency of lying bouts. In contrast, the number of daily lying bouts performed decreased over time ($F_{2,337} = 8.17$, $P < 0.01$). However, post-hoc analysis revealed no significant differences in bout frequency between time periods for TREATMENT cows. For CONTROL cows, lying bout frequency in the early period was significantly higher when compared to the peak ($P = 0.01$) or post-peak ($P < 0.01$) periods, but no difference was evident between peak and post-peak ($P = 0.74$).

Additionally, no treatment differences were evident within time periods. Within the peak period of the study, there was a tendency for multiparous cows to perform slightly more lying bouts each day than younger cows, but this difference did not reach significance ($P = 0.07$).

Table 2. Lying behaviours of cows during each of the 3 experimental periods. Data are presented as LSMeans \pm S.E.M.

	TREATMENT				CONTROL			
	Early	Peak	Post-peak	<i>P</i> -value	Early	Peak	Post-peak	<i>P</i> -value
Lying time (h/d)	10.9 \pm 0.2 ^a	11.5 \pm 0.3 ^b	12.2 \pm 0.2 ^c	< 0.001	11.3 \pm 1.0 ^a	12.0 \pm 1.0 ^b	12.7 \pm 1.0 ^c	< 0.001
Length of lying bouts (min/bout)	49.5 \pm 2.4 ^a	53.3 \pm 2.4 ^a	58.9 \pm 2.3 ^b	< 0.001	46.7 \pm 4.5 ^a	54.2 \pm 4.6 ^b	59.9 \pm 4.5 ^c	< 0.001
Frequency of lying bouts (no./d)	14.9 \pm 0.8	14.9 \pm 0.8	13.7 \pm 0.8	0.058	16.5 \pm 2.7 ^a	15.2 \pm 2.7 ^b	14.9 \pm 2.7 ^b	0.001

^{a,b,c}Means within rows and treatments with different letters differ ($P < 0.05$).

4.1.3. Duration of lying bouts

There was a significant effect of time on the overall duration of lying bouts ($F_{2,337} = 41.40$, $P < 0.01$). For both TREATMENT and CONTROL cows, lying bout length increased significantly with progressing time periods.

Breed was not found to have a significant effect on the length of lying bouts ($F_{1,337} = 2.42$, $P = 0.12$). Conversely, lying bout duration was found to have an overall weak interaction with parity ($F_{1,337} = 4.69$, $P = 0.03$); multiparous cows tended to have longer lying bouts than primiparous cows. This effect was also evident within treatment groups, but only for TREATMENT cows ($P < 0.01$), as well as within the earliest time period ($P = 0.01$).

4.2. Lying time within contact area

TREATMENT cows spent on average 76.6(29.2) % of their total lying time within the cow-calf contact area (Figure 1). The proportion of lying time in this area did not differ throughout the experiment ($F_{2,209} = 0.50$, $P = 0.61$). Parity did not have an overall effect on the proportional lying time ($F_{1,209} = 0.35$, $P = 0.56$), while breed was found to have a significant influence ($F_{1,209} = 5.08$, $P = 0.03$). Overall, SH cows spent proportionately more time lying down in the CCC area than SR cows. Post-hoc analysis of means revealed higher lying times within the contact area for SH dams only in the peak ($P = 0.02$) and post-peak ($P < 0.01$) experimental periods.

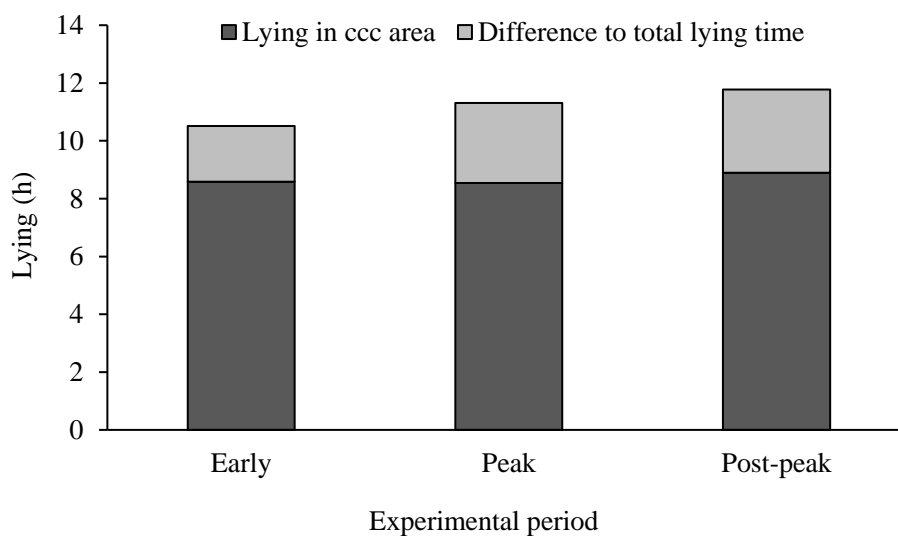


Figure 1. Mean proportion of total lying time spent in the cow-calf contact area during each of the experimental periods.

4.3. Stall use by calves

Across all weeks, calves occupied approximately 6.1(2.2) stalls at any given hour within the cow-calf contact area. The occupancy rate was found to be dependent both on week ($F_{13,299} = 11.42, P < 0.01$) and hour ($F_{23,299} = 3.78, P < 0.01$). No obvious pattern of stall use was evident across experimental weeks, despite significant differences between various weeks (Figure 2). In a 24-h period, calves tended to occupy the fewest stalls between 0400 h and 0600h, while consistently reaching a maximum stall occupancy rate between approximately 1300h and 1500h. Average stall use by calves was lower at hours where a cows occupied a higher percentage of stalls (Figure 3).

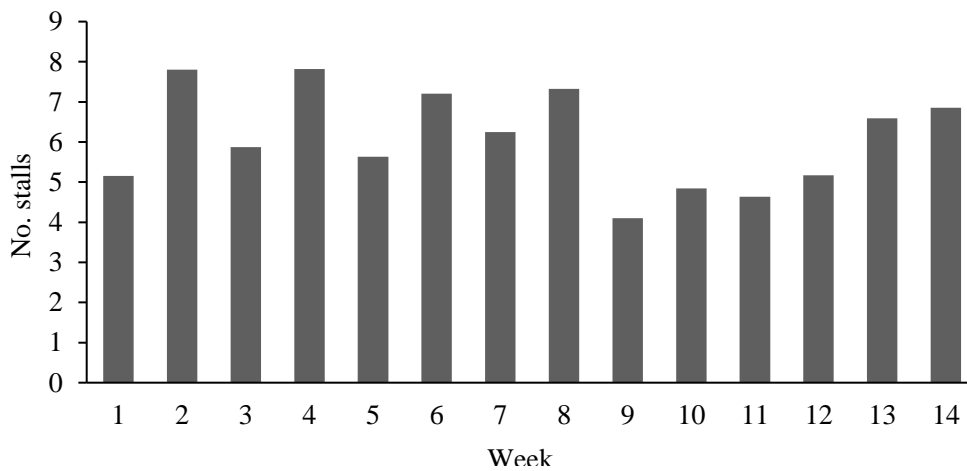


Figure 3. Mean number of stalls occupied by calves ($n=19$) across all experimental weeks.

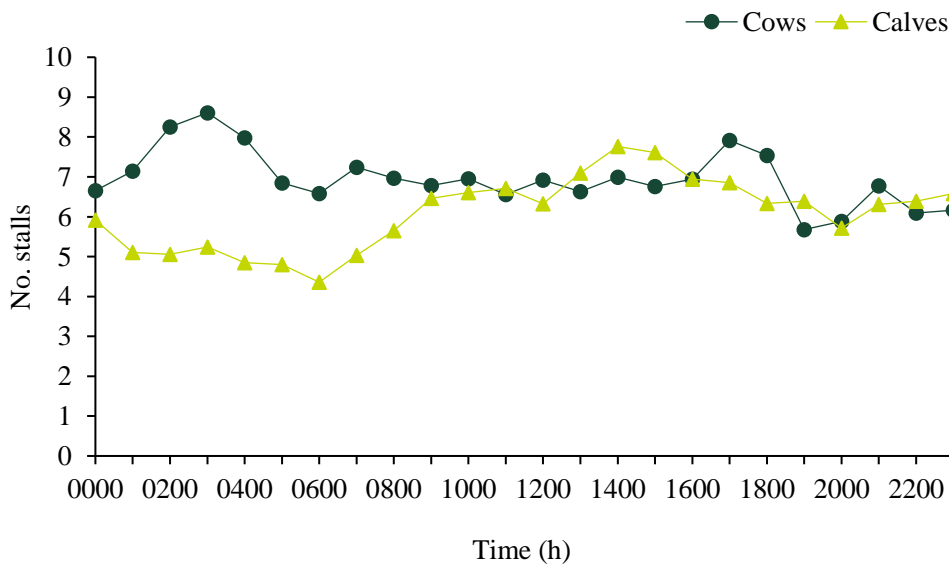


Figure 2. Mean number of stalls occupied by calves ($n=19$) and cows ($n=19$) within the contact area each hour throughout a 24-h period. Cow data was summed in 10-min intervals and averaged per hour. All data is averaged across a period of 14 consecutive weeks.

5. Discussion

This study is believed to be the first to directly compare the lying behaviours of cows housed in a CCC system, with and without their calves. Daily lying time was not affected by treatment; cows with access to full contact with calves maintained similar levels of rest as cows without. The average lying times in this study were comparable to those previously reported for freestall-housed herds with automatic milking (Deming et al. 2013; Westin et al. 2016; King et al. 2017). Similar lying times were reported by Johnsen et al. (2021) for dams housed in a CCC system with either free or partially-controlled access to calves. However, the cow-calf contact area in their system did not contain any stalls. Moreover, the small sample size (4 cow-calf pairs per access treatment) and lack of true controls renders it difficult to further compare the results to our own.

Similar to the lack of treatment difference for resting times seen in our study, Margerison et al. (1999) reported no difference in lying time within a 24-h period for cows with and without restricted calf contact. The contact in this case was in the form of 15-minute suckling periods prior to each milking session; thus, cows were likely able to compensate this reduction in time by altering their time budgets. A similar explanation is perhaps possible for the cows within our own study. Cattle are known to prioritize lying over other activities (i.e., feeding, socializing) when time constraints are implemented, and will increase the relative proportion of time dedicated to lying in order to maintain adequate levels of rest (Metz 1985; Munksgaard et al. 2005). In our study, TREATMENT cows were likely experiencing time budget reductions due to their participation in cow-calf interactions, such as suckling. At 10 weeks of age, calves with full, unrestricted access to their dams have been shown to spend approximately 13 minutes per 4 hours suckling (Roth et al. 2009). It is possible the cows in our study may have adjusted the time spent on other activities in order to attain sufficient rest. However, without close analysis of the daily time budgets for cows within a CCC system, this explanatory theory cannot be confirmed.

Cows increased their daily lying time by as much as 1.4 h/d from the start to the end of the 14-week period. As this effect was visible regardless of treatment, it is assumed that part or most of this can be explained by exploring the relationship between lying time and stage of lactation. It is known from previous experimental (Vasseur et al. 2012) and association studies (Ito et al. 2014; Solano et al. 2016;

Westin et al. 2016) that beyond the first few weeks of lactation, daily lying time generally increases with DIM.

Across all weeks, cows performed approximately 14-17 lying bouts each day, which each bout lasting an average 47-60 minutes. Comparatively, observations of AMS herds have reported cows to perform longer, less frequent bouts, with durations ranging from 71 to 78 minutes and frequencies of about 9-10 bouts/d (Deming et al. 2013; Westin et al. 2016; King et al. 2017). One explanation for the differences in behaviour is our decision to exclude events of estrus from our dataset. When in heat, cows are known to be more active and lie down less frequently relative to days not in estrus (Dolecheck et al. 2015; Silper et al. 2017). This decrease in lying bout frequency (but not duration) results in an overall decrease in daily lying time during estrus events, which can last anywhere from 7-18 hours (Diskin & Shreenan 2000). The removal of these cows likely resulted in the higher mean bout frequencies observed. However, without confirmation of how many cows were in heat during each of the herd-level studies, this theory cannot be tested for certainty.

As the weeks progressed, cows – both with and without calves present – changed their resting patterns by lying down fewer times per day, but for longer durations of time. However, the observed decrease in lying bout frequency was only significant for CONTROL cows, and only between the early and two later time periods. Vasseur et al. (2012) found a similar pattern of increased bout duration and decreased bout frequency between early (10-40 DIM) and middle (100-140 DIM) stages of lactation. Further investigation found this pattern to only be significant for primiparous cows. It has been suggested that primiparous cows may be experiencing higher levels of stress in early lactation due to changes related to calving for the first time, which may translate to increased restlessness and a greater frequency of transitions from lying to standing (Solano et al. 2016). Within our study, the early time period encompassed cows that were an average 35-38 DIM, while the peak and post-peak periods contained average DIM values of 70 and 98, respectively. As approximately half of the cows in each treatment were primiparous, it is possible that the pattern we observed was in part due to the effect of lactation, much like that observed by Vasseur et al. (2012). The relationship between lying bout measures and lactation stage has also been explored in various herd-level association studies. Westin et al. (2016) reported an associated increase in lying bout duration with increasing lactation month across 36 North American dairy herds. In contrast, Gomez and Cook (2010) and Ito et al. (2014) sampled cows from 16 farms and 79 farms, respectively, and found no association of DIM with either lying parameter. Ultimately, it is likely the singular treatment difference observed for lying bout frequency during the earlier stages of our experiment was due to the relatively small number of observation available for testing after stringent data editing.

Cows with access to the CCC area spent a large majority of their total daily lying time resting within this area – a behaviour that did not change throughout the entirety of our study. This is interesting, as it suggests that cows may be choosing to remain in close proximity to their calves for a large portion of each day, even as calves near 3-4 months of age. In semi-wild, free-ranging herds of Maremma cattle, Vitale et al. (1986) observed calves to spend an increasing proportion of time away from their dams as they grew older. Moreover, relatively soon after calving, the majority of contact behaviours have been shown to be initiated by calves rather than dams (Jensen 2011). While these previous findings of maternal-filial contact behaviours contradict the behaviour observed in our study, it is important to point out that the selection gate directed dams directly towards the contact area. Thus, it is also possible that cows simply chose to lie down in the first available stall, remaining in the contact area to obtain rest rather than contact with calves. Recently, Johnsen et al. (2021) observed that within the first month after calving, dams visited a CCC area approximately 4.6 times per day for 28 min/visit when access was granted upon successful milking, and nearly 8 times per day (20 min/visit) when access was free. These frequent visits occurred despite the fact that the CCC area did not contain any stalls. This finding suggests that the motivation for cows in our study to remain within the contact area may not have been entirely based on the availability of stalls. Future ethological research on CCC systems should focus on the latency for cows to lie down upon entering the contact area, as well as whether cows immediately seek out a stall or instead engage in contact with calves.

It should be noted that the CCC system in our study was understocked at all times (cow:stall < 1:1). While the act of understocking itself does not appear to influence daily lying time (Winckler et al. 2015), overstocking has repeatedly been demonstrated to result in a decrease of lying time (Fregonesi et al. 2007a; Hill et al. 2009; Krawczel et al. 2012). It is currently unknown how housing cows in such a system at a stocking density of 100% or greater would affect lying behaviour. This is an important question to answer if CCC systems are ever to be implemented throughout North America, where overstocking is a common practice (von Keyserlingk et al. 2012). Despite having access to a dedicated lying space with nearly 3.85 m² per individual, calves consistently chose to lie down in stalls intended for cow use. Yet, within the contact area alone there were 5 more stalls than treatment cows, adding to the likelihood that competition for stall use was relatively low. Previous research has demonstrated that as stocking density increases, so does the direct competition for stalls (Fregonesi et al. 2007a). It would be interesting to see how stall use within the contact area would change if cows and calves were housed at higher densities. Additionally, it could be relevant to investigate the interactions of cows and calves in stalls, as it is currently unknown if cows actively displace calves like they do with adult conspecifics.

Within a 24-h period, numerically calves occupied the fewest number of stalls at times when stall occupation by dams was the highest. However, this observation was not analyzed further for significance.

Parity was generally found not to influence the lying behaviours of cows within our study, with one exception: in the first 5 weeks, multiparous TREATMENT cows performed longer lying bouts than primiparous cows. This is in line with the findings of Vasseur et al. (2012), who experimentally determined that parity was positively associated with bout duration during early and mid-lactation stages. Further studies exploring lying behaviour within the first 3 weeks of lactation have confirmed this relationship (Sepúlveda-Varas et al. 2014; Neave et al. 2017). However, they also reported a negative association between parity and bout frequency, which does not explain the tendency for multiparous cows within our study having performed slightly more lying bouts per day than primiparous cows. This contrast in our findings, as well as the overall lack of parity effect, may be the result of one or a combination of factors. Cows were introduced to the AMS – including the contact area and selection gate – prior to calving. This may have reduced some of the stress usually experienced by first-parity heifers when entering a new housing after calving for the first time. Furthermore, the low stocking density – combined with the fact that milking sessions did not occur simultaneously for all cows like in parlour systems – likely reduced the direct competition for lying space. Age is thought to influence social dominance in cows (Beilharz & Zeeb 1982), but in this case, younger cows may not have had to compete with older cows for lying space.

Within our study, whether a cow was Swedish Holstein or Swedish Red did not affect the daily lying time, nor the duration and frequency of individual lying bouts. However, Swedish Holstein cows spent a greater proportion of time lying within the contact area during the peak and post-peak experimental periods compared to Swedish Reds. While the lying behaviours of these two particular breeds have not previously been experimentally observed, O’Driscoll et al. (2009) compared two similar breeds – Norwegian Red and Holstein-Friesian – and found no differences in behaviour. Conversely, Le Neindre (1989) reported differences in maternal behaviour between Friesian and Salers (a traditional French breed) cow-calf pairs – specifically for suckling bout length and licking during the first 2 months post-calving. In the case of our study, the difference in behaviour by the two breeds was possibly due to the fact that nearly half as many cows were Swedish Holstein; therefore, any individual variation would have been much more pronounced.

The results of this study are suggestive of the fact that a combined cow-calf housing system does not appear to alter the various lying behaviours of lactating cows. However, there were a few constraints that may limit the applicability of these findings to CCC systems in a broader setting. Firstly, only calves born within a 6-week period were housed together in the experimental pen, resulting in all

calves being relatively close in age. In commercial settings, calves are born on a continuous basis. The setup of the CCC system would likely need to be adapted to better fit commercial dairy practices, as well as to accommodate differences in management (i.e., parlour-based systems opposed to AMS). Furthermore, due to time constraints, the lying behaviours of calves were not explored to any extent beyond daily stall usage. Although the importance of rest relative to calves is not as well-known as it is for adult cows, lying behaviours – including those explored in our study – have not previously been explored within a CCC system. Finally, after stringent editing of the dataset, some experimental weeks contained observations for as a few as 9 TREATMENT dams and 10 CONTROL cows. Further research with additional cows is thus needed to confirm the results of our study.

6. Conclusions

Overall, access to a CCC area did not affect the lying behaviours of lactating cows. Cows altered their lying behaviours as time progressed, generally increasing their daily lying time by performing fewer, but longer, lying bouts. Cows with access to CCC were found to spend a large proportion of their daily resting times within the contact area – an observation that remained constant throughout the rearing period. These results suggest that cows were able to adapt to the added contact with calves without making changes to lying behaviour beyond the scope of what is already observed in freestall-housed, AMS herds. CCC systems, while still a relatively novel concept, may offer an alternative to early cow-calf separation and allow cows and calves to be housed together without compromising important lying behaviours in dams. Further research is needed on these systems in order to make sound recommendations for their use on a larger scale.

References

- Albright, J.L. (1993). Feeding behavior of dairy cattle. *Journal of Dairy Science*. 76 (2), 485–498. [https://doi.org/10.3168/jds.S0022-0302\(93\)77369-5](https://doi.org/10.3168/jds.S0022-0302(93)77369-5)
- Alvarez, G.G. & Ayas, N.T. (2004). The impact of daily sleep duration on health: a review of the literature. *Progress in Cardiovascular Nursing*. 19 (2), 56–59. <https://doi.org/10.1111/j.0889-7204.2004.02422.x>
- Beauchemin, K.A. (1991). Ingestion and mastication of feed by dairy cattle. *Veterinary Clinics of North America: Food Animal Practice*. 7 (2), 439–463. [https://doi.org/10.1016/S0749-0720\(15\)30794-5](https://doi.org/10.1016/S0749-0720(15)30794-5)
- Beilharz, R.G. & Zeeb, K. (1982). Social dominance in dairy cattle. *Applied Animal Ethology*. 8 (1–2), 79–97. [https://doi.org/10.1016/0304-3762\(82\)90134-1](https://doi.org/10.1016/0304-3762(82)90134-1)
- Bergsten, Christe. r. (2001). Effects of conformation and management system on hoof and leg diseases and lameness in dairy cows. *Veterinary Clinics of North America: Food Animal Practice*. 17 (1), 1–23. [https://doi.org/10.1016/S0749-0720\(15\)30051-7](https://doi.org/10.1016/S0749-0720(15)30051-7)
- Bewley, J.M., Boyce, R.E., Hockin, J., Munksgaard, L., Eicher, S.D., Einstein, M.E. & Schutz, M.M. (2010). Influence of milk yield, stage of lactation, and body condition on dairy cattle lying behaviour measured using an automated activity monitoring sensor. *Journal of Dairy Research*. 77 (1), 1–6. <https://doi.org/10.1017/S0022029909990227>
- Blackie, N., Amory, J., Bleach, E. & Scaife, J. (2011). The effect of lameness on lying behaviour of zero grazed Holstein dairy cattle. *Applied Animal Behaviour Science*. 134 (3–4), 85–91. <https://doi.org/10.1016/j.applanim.2011.08.004>
- Bouissou, M.F., Boissy, A., Neindre, P. le & Veissier, I. (2001). The social behaviour of cattle. In: Keeling, L.J. & Gonyou, H.W. (eds.) *Social Behaviour in Farm Animals*. Wallingford: CABI Publishing. 113–145. <https://doi.org/10.1079/9780851993973.0113>
- Broom, D.M. (1983). Stereotypies as animal welfare indicators. In: Smidt, D. (ed.) *Indicators Relevant to Farm Animal Welfare*. Dordrecht: Springer. 81–87. https://doi.org/10.1007/978-94-009-6738-0_11
- Brown, E.J. & Vosloo, A. (2017). The involvement of the hypothalamo- pituitary- adrenocortical axis in stress physiology and its significance in the assessment of animal welfare in cattle. *Onderstepoort Journal of Veterinary Research*. 84 (1), a1398. <https://doi.org/10.4102/ojvr.v84i1.1398>
- Chapinal, N., de Passillé, A.M., Weary, D.M., von Keyserlingk, M.A.G. & Rushen, J. (2009). Using gait score, walking speed, and lying behavior to detect hoof lesions in dairy cows. *Journal of Dairy Science*. 92 (9), 4365–4374. <https://doi.org/10.3168/jds.2009-2115>
- Chaplin, S. & Munksgaard, L. (2001). Evaluation of a simple method for assessment of rising behaviour in tethered dairy cows. *Animal Science*. 72 (1), 191–197. <https://doi.org/10.1017/S1357729800055685>
- Charlton, G.L., Haley, D.B., Rushen, J. & de Passillé, A.M. (2014). Stocking density, milking duration, and lying times of lactating cows on Canadian

- freestall dairy farms. *Journal of Dairy Science*. 97 (5), 2694–2700. <https://doi.org/10.3168/jds.2013-6923>
- Chen, J.M., Stull, C.L., Ledgerwood, D.N. & Tucker, C.B. (2017). Muddy conditions reduce hygiene and lying time in dairy cattle and increase time spent on concrete. *Journal of Dairy Science*. 100 (3), 2090–2103. <https://doi.org/10.3168/jds.2016-11972>
- Cook, N.B., Bennett, T.B. & Nordlund, K.V. (2004). Effect of free stall surface on daily activity patterns in dairy cows with relevance to lameness prevalence. *Journal of Dairy Science*. 87 (9), 2912–2922. [https://doi.org/10.3168/jds.S0022-0302\(04\)73422-0](https://doi.org/10.3168/jds.S0022-0302(04)73422-0)
- Cooper, M.D., Arney, D.R. & Phillips, C.J.C. (2007). Two- or four-hour lying deprivation on the behavior of lactating dairy cows. *Journal of Dairy Science*. 90 (3), 1149–1158. [https://doi.org/10.3168/jds.S0022-0302\(07\)71601-6](https://doi.org/10.3168/jds.S0022-0302(07)71601-6)
- Cooper, M.D., Arney, D.R. & Phillips, C.J.C. (2008). The effect of temporary deprivation of lying and feeding on the behaviour and production of lactating dairy cows. *Animal*. 2 (2), 275–283. <https://doi.org/10.1017/S1751731107001164>
- Deming, J.A., Bergeron, R., Leslie, K.E. & DeVries, T.J. (2013). Associations of housing, management, milking activity, and standing and lying behavior of dairy cows milked in automatic systems. *Journal of Dairy Science*. 96 (1), 344–351. <https://doi.org/10.3168/jds.2012-5985>
- de Oliveira, D., Barth, K., Haskell, M.J., Hillmann, E., Jensen, M.B., Johnsen, J.F., Mejdell, C., Waiblinger, S. & Ferneborg, S. (2020). Methodology for experimental and observational animal studies in cow-calf contact systems. *Journal of Dairy Research*. 87 (S1), 115–121. <https://doi.org/10.1017/S0022029920000552>
- Diskin, M.G. & Sreenan, J.M. (2000). Expression and detection of oestrus in cattle. *Reproduction Nutrition Development*. 40 (5), 481–491. <https://doi.org/10.1051/rnd:2000112>
- Dolecheck, K.A., Silvia, W.J., Heersche, G., Chang, Y.M., Ray, D.L., Stone, A.E., Wadsworth, B.A. & Bewley, J.M. (2015). Behavioral and physiological changes around estrus events identified using multiple automated monitoring technologies. *Journal of Dairy Science*. 98 (12), 8723–8731. <https://doi.org/10.3168/jds.2015-9645>
- Drissler, M., Gaworski, M., Tucker, C.B. & Weary, D.M. (2005). Freestall maintenance: effects on lying behavior of dairy cattle. *Journal of Dairy Science*. 88 (7), 2381–2387. [https://doi.org/10.3168/jds.S0022-0302\(05\)72916-7](https://doi.org/10.3168/jds.S0022-0302(05)72916-7)
- Duncan, I.J.H. (1992). Measuring preferences and the strength of preferences. *Poultry Science*. 71 (4), 658–663. <https://doi.org/10.3382/ps.0710658>
- Edwards, S.A. & Broom, D.M. (1982). Behavioural interactions of dairy cows with their newborn calves and the effects of parity. *Animal Behaviour*. 30 (2), 525–535. [https://doi.org/10.1016/S0003-3472\(82\)80065-1](https://doi.org/10.1016/S0003-3472(82)80065-1)
- Flower, F.C. & Weary, D.M. (2001). Effects of early separation on the dairy cow and calf: 2. Separation at 1 day and 2 weeks after birth. *Applied Animal Behaviour Science*. 70 (4), 275–284. [https://doi.org/10.1016/S0168-1591\(00\)00164-7](https://doi.org/10.1016/S0168-1591(00)00164-7)
- Fraser, D., Weary, D.M., Pajor, E.A. & Milligan, B.N. (1997). A scientific conception of animal welfare that reflects ethical concerns. *Animal Welfare*. 6, 187–205
- Fregonesi, J.A., Tucker, C.B. & Weary, D.M. (2007a). Overstocking reduces lying time in dairy cows. *Journal of Dairy Science*. 90 (7), 3349–3354. <https://doi.org/10.3168/jds.2006-794>

- Fregonesi, J.A., Veira, D.M., von Keyserlingk, M.A.G. & Weary, D.M. (2007b). Effects of bedding quality on lying behavior of dairy cows. *Journal of Dairy Science*. 90 (12), 5468–5472. <https://doi.org/10.3168/jds.2007-0494>
- Fröberg, S. & Lidfors, L. (2009). Behaviour of dairy calves suckling the dam in a barn with automatic milking or being fed milk substitute from an automatic feeder in a group pen. *Applied Animal Behaviour Science*. 117 (3–4), 150–158. <https://doi.org/10.1016/j.applanim.2008.12.015>
- Galindo, F. & Broom, D.M. (2000). The relationships between social behaviour of dairy cows and the occurrence of lameness in three herds. *Research in Veterinary Science*. 69 (1), 75–79. <https://doi.org/10.1053/rvsc.2000.0391>
- Gomez, A. & Cook, N.B. (2010). Time budgets of lactating dairy cattle in commercial freestall herds. *Journal of Dairy Science*. 93 (12), 5772–5781. <https://doi.org/10.3168/jds.2010-3436>
- Grøndahl, A.M., Skancke, E.M., Mejdell, C.M. & Jansen, J.H. (2007). Growth rate, health and welfare in a dairy herd with natural suckling until 6–8 weeks of age: a case report. *Acta Veterinaria Scandinavica*. 49, 16. <https://doi.org/10.1186/1751-0147-49-16>
- Haley, D.B., Rushen, J. & Passillé, A.M. de (2000). Behavioural indicators of cow comfort: activity and resting behaviour of dairy cows in two types of housing. *Canadian Journal of Animal Science*. 80 (2), 257–263. <https://doi.org/10.4141/A99-084>
- Helmreich, S., Hauser, R., Jungbluth, T., Wechsler, B. & Gygax, L. (2014). Time-budget constraints for cows with high milking frequency on farms with automatic milking systems. *Livestock Science*. 167, 315–322. <https://doi.org/10.1016/j.livsci.2014.06.014>
- Hermans, G.G.N., Ipema, A.H., Stefanowska, J. & Metz, J.H.M. (2003). The effect of two traffic situations on the behavior and performance of cows in an automatic milking system. *Journal of Dairy Science*. 86 (6), 1997–2004. [https://doi.org/10.3168/jds.S0022-0302\(03\)73788-6](https://doi.org/10.3168/jds.S0022-0302(03)73788-6)
- Hill, C.T., Krawczel, P.D., Dann, H.M., Ballard, C.S., Hovey, R.C., Falls, W.A. & Grant, R.J. (2009). Effect of stocking density on the short-term behavioural responses of dairy cows. *Applied Animal Behaviour Science*. 117 (3–4), 144–149. <https://doi.org/10.1016/j.applanim.2008.12.012>
- Ito, K., Chapinal, N., Weary, D.M. & von Keyserlingk, M.A.G. (2014). Associations between herd-level factors and lying behavior of freestall-housed dairy cows. *Journal of Dairy Science*. 97 (4), 2081–2089. <https://doi.org/10.3168/jds.2013-6861>
- Ito, K., von Keyserlingk, M.A.G., LeBlanc, S.J. & Weary, D.M. (2010). Lying behavior as an indicator of lameness in dairy cows. *Journal of Dairy Science*. 93 (8), 3553–3560. <https://doi.org/10.3168/jds.2009-2951>
- Ito, K., Weary, D.M. & von Keyserlingk, M.A.G. (2009). Lying behavior: assessing within- and between-herd variation in free-stall-housed dairy cows. *Journal of Dairy Science*. 92 (9), 4412–4420. <https://doi.org/10.3168/jds.2009-2235>
- Jensen, M.B. (2011). The early behaviour of cow and calf in an individual calving pen. *Applied Animal Behaviour Science*. 134 (3–4), 92–99. <https://doi.org/10.1016/j.applanim.2011.06.017>
- Jensen, M.B., Munksgaard, L., Pedersen, L.J., Ladewig, J. & Matthews, L. (2004). Prior deprivation and reward duration affect the demand function for rest in dairy heifers. *Applied Animal Behaviour Science*. 88 (1–2), 1–11. <https://doi.org/10.1016/j.applanim.2004.02.019>
- Jensen, M.B. & Pedersen, L.J. (2008). Using motivation tests to assess ethological needs and preferences. *Applied Animal Behaviour Science*. 113 (4), 340–356. <https://doi.org/10.1016/j.applanim.2008.02.001>
- Jensen, M.B., Pedersen, L.J. & Munksgaard, L. (2005). The effect of reward duration on demand functions for rest in dairy heifers and lying

- requirements as measured by demand functions. *Applied Animal Behaviour Science*. 90 (3–4), 207–217. <https://doi.org/10.1016/j.applanim.2004.08.006>
- Johnsen, J.F., Johanssen, J.R.E., Aaby, A.V., Kischel, S.G., Ruud, L.E., Soki-Makilutila, A., Kristiansen, T.B., Wibe, A.G., Bøe, K.E. & Ferneborg, S. (2021). Investigating cow-calf contact in cow-driven systems: behaviour of the dairy cow and calf. *Journal of Dairy Research*. 88 (1), 52–55. <https://doi.org/10.1017/S0022029921000194>
- Johnsen, J.F., Zipp, K.A., Kälber, T., Passillé, A.M. de, Knierim, U., Barth, K. & Mejdell, C.M. (2016). Is rearing calves with the dam a feasible option for dairy farms?—current and future research. *Applied Animal Behaviour Science*. 181, 1–11. <https://doi.org/10.1016/j.applanim.2015.11.011>
- King, M.T.M., LeBlanc, S.J., Pajor, E.A. & DeVries, T.J. (2017). Cow-level associations of lameness, behavior, and milk yield of cows milked in automated systems. *Journal of Dairy Science*. 100 (6), 4818–4828. <https://doi.org/10.3168/jds.2016-12281>
- Krawczel, P.D., Klaiber, L.B., Butzler, R.E., Klaiber, L.M., Dann, H.M., Mooney, C.S. & Grant, R.J. (2012). Short-term increases in stocking density affect the lying and social behavior, but not the productivity, of lactating Holstein dairy cows. *Journal of Dairy Science*. 95 (8), 4298–4308. <https://doi.org/10.3168/jds.2011-4687>
- Krawczel, P.D. & Lee, A.R. (2019). Lying time and its importance to the dairy cow: impact of stocking density and time budget stresses. *Veterinary Clinics of North America: Food Animal Practice*. 35 (1), 47–60. <https://doi.org/10.1016/j.cvfa.2018.11.002>
- Kull, J.A., Proudfoot, K.L., Pighetti, G.M., Bewley, J.M., O’Hara, B.F., Donohue, K.D. & Krawczel, P.D. (2019). Effects of acute lying and sleep deprivation on the behavior of lactating dairy cows. *PLoS ONE*. 14 (8), e0212823. <https://doi.org/10.1371/journal.pone.0212823>
- Le Neindre, P. (1989). Influence of cattle rearing conditions and breed on social relationships of mother and young. *Applied Animal Behaviour Science*. 23 (1), 117–127. [https://doi.org/10.1016/0168-1591\(89\)90012-9](https://doi.org/10.1016/0168-1591(89)90012-9)
- Leonard, F.C., O’Connell, J.M. & O’Farrell, K.J. (1996). Effect of overcrowding on claw health in first-calved Friesian heifers. *British Veterinary Journal*. 152 (4), 459–472. [https://doi.org/10.1016/S0007-1935\(96\)80040-6](https://doi.org/10.1016/S0007-1935(96)80040-6)
- Lexer, D., Hagen, K., Palme, R., Troxler, J. & Waiblinger, S. (2009). Time budgets and adrenocortical activity of cows milked in a robot or a milking parlour: interrelationships and influence of social rank. *Animal Welfare*. 18 (1), 73–80.
- Lidfors, L.M., Moran, D., Jung, J., Jensen, P. & Castren, H. (1994). Behaviour at calving and choice of calving place in cattle kept in different environments. *Applied Animal Behaviour Science*. 42 (1), 11–28. [https://doi.org/10.1016/0168-1591\(94\)90003-5](https://doi.org/10.1016/0168-1591(94)90003-5)
- Manninen, E., de Passillé, A.M., Rushen, J., Norrington, M. & Saloniemi, H. (2002). Preferences of dairy cows kept in unheated buildings for different kind of cubicle flooring. *Applied Animal Behaviour Science*. 75 (4), 281–292. [https://doi.org/10.1016/S0168-1591\(01\)00206-4](https://doi.org/10.1016/S0168-1591(01)00206-4)
- Margerison, J.K., Phillips, C.J.C. & Preston, T.R. (1999). The effect of cow-calf separation in dairy cattle on animal behaviour. *BSAP Occasional Publication*. 23, 113–115. <https://doi.org/10.1017/S0263967X00033346>
- Maselyne, J., Pastell, M., Thomsen, P.T., Thorup, V.M., Hänninen, L., Vangeyte, J., Van Nuffel, A. & Munksgaard, L. (2017). Daily lying time, motion index and step frequency in dairy cows change throughout lactation. *Research in Veterinary Science*. 110, 1–3. <https://doi.org/10.1016/j.rvsc.2016.10.003>

- Mason, G., McFarland, D. & Garner, J. (1998). A demanding task: using economic techniques to assess animal priorities. *Animal Behaviour*. 55, 1071–1075
- Matthews, L.R. & Ladewig, J. (1994). Environmental requirements of pigs measured by behavioural demand functions. *Animal Behaviour*. 47 (3), 713–719. <https://doi.org/10.1006/anbe.1994.1096>
- McPherson, S.E. & Vasseur, E. (2020). Graduate student literature review: the effects of bedding, stall length, and manger wall height on common outcome measures of dairy cow welfare in stall-based housing systems. *Journal of Dairy Science*. 103 (11), 10940–10950. <https://doi.org/10.3168/jds.2020-18332>
- Medrano-Galarza, C., Gibbons, J., Wagner, S., de Passillé, A.M. & Rushen, J. (2012). Behavioral changes in dairy cows with mastitis. *Journal of Dairy Science*. 95 (12), 6994–7002. <https://doi.org/10.3168/jds.2011-5247>
- Metz, J.H.M. (1985). The reaction of cows to a short-term deprivation of lying. *Applied Animal Behaviour Science*. 13 (4), 301–307. [https://doi.org/10.1016/0168-1591\(85\)90010-3](https://doi.org/10.1016/0168-1591(85)90010-3)
- Munksgaard, L., Ingvarsten, K.L., Pedersen, L.J. & Nielsen, V.K.M. (1999). Deprivation of lying down affects behaviour and pituitary-adrenal axis responses in young bulls. *Acta Agriculturae Scandinavica, Section A - Animal Science*. 49 (3), 172–178. <https://doi.org/10.1080/090647099424088>
- Munksgaard, L., Jensen, M.B., Pedersen, L.J., Hansen, S.W. & Matthews, L. (2005). Quantifying behavioural priorities—effects of time constraints on behaviour of dairy cows, *Bos taurus*. *Applied Animal Behaviour Science*. 92 (1–2), 3–14. <https://doi.org/10.1016/j.applanim.2004.11.005>
- Munksgaard, L., Rushen, J., de Passillé, A.M. & Krohn, C.C. (2011). Forced versus free traffic in an automated milking system. *Livestock Science*. 138 (1–3), 244–250. <https://doi.org/10.1016/j.livsci.2010.12.023>
- Munksgaard, L. & Simonsen, H.B. (1996). Behavioral and pituitary adrenal-axis responses of dairy cows to social isolation and deprivation of lying down. *Journal of Animal Science*. 74 (4), 769–778. <https://doi.org/10.2527/1996.744769x>
- National Farm Animal Care Council. (2009). *Code of practice for the care and handling of dairy cattle*. Available at: http://www.nfacc.ca/pdfs/codes/dairy_code_of_practice.pdf [2021-04-20]
- Neave, H.W., Lomb, J., von Keyserlingk, M.A.G., Behnam-Shabahang, A. & Weary, D.M. (2017). Parity differences in the behavior of transition dairy cows. *Journal of Dairy Science*. 100 (1), 548–561. <https://doi.org/10.3168/jds.2016-10987>
- Nelson, A.J. (1996). On-farm nutrition diagnostics: nutrition management involvement opportunities for dairy practitioners. *American Association of Bovine Practitioners Proceedings of the Annual Conference*. 29, 76–85. <https://doi.org/10.21423/aabppro19965940>
- Norring, M., Manninen, E., de Passillé, A.M., Rushen, J. & Saloniemi, H. (2010). Preferences of dairy cows for three stall surface materials with small amounts of bedding. *Journal of Dairy Science*. 93 (1), 70–74. <https://doi.org/10.3168/jds.2009-2164>
- O’Driscoll, K.K., Hanlon, A., French, P. & Boyle, L.A. (2009). The effects of two out-wintering pad systems compared with free-stalls on dairy cow hoof and limb health. *Journal of Dairy Research*. 76 (1), 59–65. <https://doi.org/10.1017/S0022029908003695>
- Ouweltjes, W., van der Werf, J.T.N., Frankena, K. & van Leeuwen, J.L. (2011). Effects of flooring and restricted freestall access on behavior and claw health of dairy heifers. *Journal of Dairy Science*. 94 (2), 705–715. <https://doi.org/10.3168/jds.2010-3208>

- Patterson-Kane, E., Pittman, M. & Pajor, E. (2008). Operant animal welfare: productive approaches and persistent difficulties. *Animal Welfare*. 17 (2), 139–148
- Plaizier, J.C., Krause, D.O., Gozho, G.N. & McBride, B.W. (2008). Subacute ruminal acidosis in dairy cows: the physiological causes, incidence and consequences. *The Veterinary Journal*. 176 (1), 21–31. <https://doi.org/10.1016/j.tvjl.2007.12.016>
- Proudfoot, K.L., Kull, J.A., Krawczel, P.D., Bewley, J.M., O’Hara, B.F., Donohue, K.D. & Pighetti, G.M. (2021). Effects of acute lying and sleep deprivation on metabolic and inflammatory responses of lactating dairy cows. *Journal of Dairy Science*. 104 (4), 4764–4774. <https://doi.org/10.3168/jds.2020-19332>
- Reinhardt, C., Reinhardt, A. & Reinhardt, V. (1986). Social behaviour and reproductive performance in semi-wild Scottish Highland cattle. *Applied Animal Behaviour Science*. 15 (2), 125–136. [https://doi.org/10.1016/0168-1591\(86\)90058-4](https://doi.org/10.1016/0168-1591(86)90058-4)
- Reinhardt, V. & Reinhardt, A. (1981). Natural sucking performance and age of weaning in zebu cattle (*Bos indicus*). *The Journal of Agricultural Science*. 96 (2), 309–312. <https://doi.org/10.1017/S0021859600066089>
- Roth, B.A., Barth, K., Gygax, L. & Hillmann, E. (2009). Influence of artificial vs. mother-bonded rearing on sucking behaviour, health and weight gain in calves. *Applied Animal Behaviour Science*. 119 (3–4), 143–150. <https://doi.org/10.1016/j.applanim.2009.03.004>
- Ruckebusch, Y. (1972). The relevance of drowsiness in the circadian cycle of farm animals. *Animal Behaviour*. 20 (4), 637–643. [https://doi.org/10.1016/S0003-3472\(72\)80136-2](https://doi.org/10.1016/S0003-3472(72)80136-2)
- Ruckebusch, Y. (1974). Sleep deprivation in cattle. *Brain Research*. 78 (3), 495–499. [https://doi.org/10.1016/0006-8993\(74\)90932-9](https://doi.org/10.1016/0006-8993(74)90932-9)
- Rushen, J., Haley, D. & de Passillé, A.M. (2007). Effect of softer flooring in tie stalls on resting behavior and leg injuries of lactating cows. *Journal of Dairy Science*. 90 (8), 3647–3651. <https://doi.org/10.3168/jds.2006-463>
- Schirmann, K., Chapinal, N., Weary, D.M., Heuwieser, W. & von Keyserlingk, M.A.G. (2012). Rumination and its relationship to feeding and lying behavior in Holstein dairy cows. *Journal of Dairy Science*. 95 (6), 3212–3217. <https://doi.org/10.3168/jds.2011-4741>
- Sepúlveda-Varas, P., Weary, D.M. & von Keyserlingk, M.A.G. (2014). Lying behavior and postpartum health status in grazing dairy cows. *Journal of Dairy Science*. 97 (10), 6334–6343. <https://doi.org/10.3168/jds.2014-8357>
- Siebert, K., Langbein, J., Schön, P.-C., Tuchscherer, A. & Puppe, B. (2011). Degree of social isolation affects behavioural and vocal response patterns in dwarf goats (*Capra hircus*). *Applied Animal Behaviour Science*. 131 (1–2), 53–62. <https://doi.org/10.1016/j.applanim.2011.01.003>
- Siivonen, J., Taponen, S., Hovinen, M., Pastell, M., Lensink, B.J., Pyörälä, S. & Hänninen, L. (2011). Impact of acute clinical mastitis on cow behaviour. *Applied Animal Behaviour Science*. 132 (3–4), 101–106. <https://doi.org/10.1016/j.applanim.2011.04.005>
- Silper, B.F., Madureira, A.M.L., Polsky, L.B., Soriano, S., Sica, A.F., Vasconcelos, J.L.M. & Cerri, R.L.A. (2017). Daily lying behavior of lactating Holstein cows during an estrus synchronization protocol and its associations with fertility. *Journal of Dairy Science*. 100 (10), 8484–8495. <https://doi.org/10.3168/jds.2016-12160>
- Silper, B.F., Polsky, L., Luu, J., Burnett, T.A., Rushen, J., de Passillé, A.M. & Cerri, R.L.A. (2015). Automated and visual measurements of estrous behavior and their sources of variation in Holstein heifers. II: Standing and lying patterns.

- Theriogenology*. 84 (3), 333–341.
<https://doi.org/10.1016/j.theriogenology.2014.12.030>
- Solano, L., Barkema, H.W., Pajor, E.A., Mason, S., LeBlanc, S.J., Nash, C.G.R., Haley, D.B., Pellerin, D., Rushen, J., de Passillé, A.M., Vasseur, E. & Orsel, K. (2016). Associations between lying behavior and lameness in Canadian Holstein-Friesian cows housed in freestall barns. *Journal of Dairy Science*. 99 (3), 2086–2101. <https://doi.org/10.3168/jds.2015-10336>
- Somers, J.G.C.J., Frankena, K., Noordhuizen-Stassen, E.N. & Metz, J.H.M. (2003). Prevalence of claw disorders in Dutch dairy cows exposed to several floor systems. *Journal of Dairy Science*. 86 (6), 2082–2093. [https://doi.org/10.3168/jds.S0022-0302\(03\)73797-7](https://doi.org/10.3168/jds.S0022-0302(03)73797-7)
- Spolders, M., Meyer, U., Flachowsky, G. & Coenen, M. (2004). Comparison of resting and feeding behaviour for cows milked by an automatic milking system versus by a milking parlour. *Journal of Applied Animal Research*. 25 (2), 69–80. <https://doi.org/10.1080/09712119.2004.9706480>
- Telezhenko, E. & Bergsten, C. (2005). Influence of floor type on the locomotion of dairy cows. *Applied Animal Behaviour Science*. 93 (3), 183–197. <https://doi.org/10.1016/j.applanim.2004.11.021>
- Ternman, E., Hänninen, L., Pastell, M., Agenäs, S. & Nielsen, P.P. (2012). Sleep in dairy cows recorded with a non-invasive EEG technique. *Applied Animal Behaviour Science*. 140 (1–2), 25–32. <https://doi.org/10.1016/j.applanim.2012.05.005>
- Tucker, C.B., Jensen, M.B., de Passillé, A.M., Hänninen, L. & Rushen, J. (2021). Invited review: lying time and the welfare of dairy cows. *Journal of Dairy Science*. 104 (1), 20–46. <https://doi.org/10.3168/jds.2019-18074>
- Tucker, C.B., Munksgaard, L., Mintline, E.M. & Jensen, M.B. (2018). Use of a pneumatic push gate to measure dairy cattle motivation to lie down in a deep-bedded area. *Applied Animal Behaviour Science*. 201, 15–24. <https://doi.org/10.1016/j.applanim.2017.12.018>
- Tucker, C.B., Weary, D.M. & Fraser, D. (2003). Effects of three types of free-stall surfaces on preferences and stall usage by dairy cows. *Journal of Dairy Science*. 86 (2), 521–529. [https://doi.org/10.3168/jds.S0022-0302\(03\)73630-3](https://doi.org/10.3168/jds.S0022-0302(03)73630-3)
- Tucker, C.B., Weary, D.M., von Keyserlingk, M.A.G. & Beauchemin, K.A. (2009). Cow comfort in tie-stalls: increased depth of shavings or straw bedding increases lying time. *Journal of Dairy Science*. 92 (6), 2684–2690. <https://doi.org/10.3168/jds.2008-1926>
- Val-Laillet, D., Veira, D.M. & von Keyserlingk, M.A.G. (2008). Short communication: dominance in free-stall-housed dairy cattle is dependent upon resource. *Journal of Dairy Science*. 91 (10), 3922–3926. <https://doi.org/10.3168/jds.2008-1332>
- Vasseur, E., Rushen, J., Haley, D.B. & de Passillé, A.M. (2012). Sampling cows to assess lying time for on-farm animal welfare assessment. *Journal of Dairy Science*. 95 (9), 4968–4977. <https://doi.org/10.3168/jds.2011-5176>
- Vitale, A.F., Tenucci, M., Papini, M. & Lovari, S. (1986). Social behaviour of the calves of semi-wild Maremma cattle, *Bos primigenius taurus*. *Applied Animal Behaviour Science*. 16 (3), 217–231. [https://doi.org/10.1016/0168-1591\(86\)90115-2](https://doi.org/10.1016/0168-1591(86)90115-2)
- von Keyserlingk, M.A.G., Barrientos, A., Ito, K., Galo, E. & Weary, D.M. (2012). Benchmarking cow comfort on North American freestall dairies: lameness, leg injuries, lying time, facility design, and management for high-producing Holstein dairy cows. *Journal of Dairy Science*. 95 (12), 7399–7408. <https://doi.org/10.3168/jds.2012-5807>
- von Keyserlingk, M.A.G., Rushen, J., de Passillé, A.M. & Weary, D.M. (2009). Invited review: the welfare of dairy cattle—key concepts and the role of

- science. *Journal of Dairy Science*. 92 (9), 4101–4111.
<https://doi.org/10.3168/jds.2009-2326>
- von Keyserlingk, M.A.G. & Weary, D.M. (2007). Maternal behavior in cattle. *Hormones and Behavior*. 52 (1), 106–113.
<https://doi.org/10.1016/j.yhbeh.2007.03.015>
- Weary, D.M., Huzzey, J.M. & von Keyserlingk, M.A.G. (2009). BOARD-INVITED REVIEW: Using behavior to predict and identify ill health in animals. *Journal of Animal Science*. 87 (2), 770–777.
<https://doi.org/10.2527/jas.2008-1297>
- Westin, R., Vaughan, A., de Passillé, A.M., DeVries, T.J., Pajor, E.A., Pellerin, D., Siegford, J.M., Vasseur, E. & Rushen, J. (2016). Lying times of lactating cows on dairy farms with automatic milking systems and the relation to lameness, leg lesions, and body condition score. *Journal of Dairy Science*. 99 (1), 551–561. <https://doi.org/10.3168/jds.2015-9737>
- Winckler, C., Tucker, C.B. & Weary, D.M. (2015). Effects of under- and overstocking freestalls on dairy cattle behaviour. *Applied Animal Behaviour Science*. 170, 14–19. <https://doi.org/10.1016/j.applanim.2015.06.003>
- Witaifi, A.A., Ali, A.B.A. & Siegford, J.M. (2018). Stall and feed bunk stocking rates impact cows' diurnal behavior and activity in automatic milking system farms. *Journal of Veterinary Behavior*. 24, 48–55.
<https://doi.org/10.1016/j.jveb.2018.01.004>
- Yunta, C., Guasch, I. & Bach, A. (2012). Short communication: lying behavior of lactating dairy cows is influenced by lameness especially around feeding time. *Journal of Dairy Science*. 95 (11), 6546–6549.
<https://doi.org/10.3168/jds.2012-5670>
- Zebari, H.M., Rutter, S.M. & Bleach, E.C.L. (2018). Characterizing changes in activity and feeding behaviour of lactating dairy cows during behavioural and silent oestrus. *Applied Animal Behaviour Science*. 206, 12–17.
<https://doi.org/10.1016/j.applanim.2018.06.002>

Acknowledgements

First and foremost, to Emma Ternman, without whose guidance and ethological (as well as statistical) expertise this project would not have been possible. Thank you for your seemingly unlimited patience as I fumbled my way through data analysis, as well as for replying to what felt like my hundreds of questions regarding everything from experimental design to poster aesthetics. I truly hope you are proud of what this project has blossomed into.

Thank you to my cow-marking partner-in-crime and fellow animal welfare enthusiast, Teresa Johansson. Making the trek out to Lövsta as often as we did would have been a lot less fun if you hadn't been by my side. Thank you to Gunilla, Hanna and Malin, for providing warm rides to and from Lövsta during the snowy winter months, as well as for your help in gathering the data needed to bring this thesis into fruition.

To my friends, both in Uppsala and back home in Canada – thank you for your patience with me when I postponed group dinners and facetime calls. Thank you to my wonderful partner, Adrien, for lending an ear to my daily frustrations and triumphs regarding this project. I am eternally grateful for your unwavering emotional support through yet another (and hopefully not last) research endeavor. Thank you to my parents, for encouraging me to take the leap and move to Sweden without knowing if or when I would return.

Finally, to the 37 lovely ladies of VMS 1, without whom this entire project would not have been possible. May your efforts have been for the betterment of welfare for cows worldwide.