

ANIMAL PRODUCTION SCIENCE

Comparison of dairy cow step activity under different milking schedules

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ABSTRACT

Context. Variations in the number of milkings each day and their timing are becoming increasingly common. How these changes affect cow behaviour is poorly understood. When cows are milked less frequently, their walking to and from the dairy is reduced and their amount of time spent at pasture increases; however, the impact on activity under different milking schedules has not been measured. Aims. The objective of this study was to identify any differences in cow walking activity (steps per hour) among three milking frequencies and three milking schedules of 3-in-2 (milking three times in 2 days), at two stages of lactation (34 and 136 days in milk), over a period of 6 weeks. Time spent eating was assessed to help explain differences in activity within a day. Methods. Data were collected from five groups of 40 cows (n = 200) milked, as follows: once a day (OAD); twice a day (TAD); 3-in-2 (three groups) at intervals of 12-18-18 h, 10-19-19 h, and 8-20-20 h. All cows were fitted with AfiAct pedometers, which recorded steps per hour. Eight cows in each treatment group were also fitted with CowManager SensOor™ ear tags, which recorded minutes per hour spent eating. Key results. Cow steps per hour increased with an increasing milking frequency in both trial periods. When data associated with walking to and from milking were removed, there were still differences in cow step activity. Cows milked OAD took 30% fewer steps than TAD cows. The diurnal pattern of eating time differed between these two trial groups. The effect of milking time among the 3-in-2 trials showed that the shorter the time between the milkings on the day the cows were milked twice, the greater the number of steps per hour. There were graphical eating differences between the 8-20-20 trial group and 12-18-18 trial group on the day that cows were milked twice. **Conclusions.** We conclude that both the number and timings of milkings affect a cow's step activity and grazing behaviour. Implications. Farmers should minimise the amount of time cows spend away from the paddock, especially in the afternoon, to minimise any changes to natural behaviour.

Keywords: activity, behaviour, cow, dairy, frequency, milking, step, timing.

Introduction

Consumers of animal products are increasingly taking animal welfare into consideration when making a purchase (European Commission 2007; Napolitano *et al.* 2008; Alonso *et al.* 2020). A widely accepted definition of animal welfare focuses on three aspects, namely, whether the animal is (1) functioning well, (2) feeling well, and (3) able to live a reasonably natural life (Fraser *et al.* 1997). Dairy farming has undergone intensification over time, which has contributed to an increase in average herd size (Clay *et al.* 2020). In pastoral systems, an increase in herd size has led to cows walking longer distances to the dairy, and an increase in time spent away from pasture (Beggs *et al.* 2019). Increasing time away from pasture, the amount of time spent standing on concrete, and the distance cows walk, may affect the expression of natural behaviours, and thus have an impact on animal welfare.

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The milking frequency and milking interval can influence the time cows spend away from pasture. The milking frequency is defined as the number of milkings per day, and the milking interval is the time between milkings. Dairy farmers can adapt both of these to suit their needs.

Twice-a-day (TAD) milking frequency is common in pastoral dairy farming systems, and it usually results in greater milk production than a once-a-day (OAD) milking frequency (Edwards 2018). Milking more than TAD is rare in pastoral systems as the revenue from the additional milk does not cover the cost of the additional milkings (Culotta and Schmidt 1988). Milking three times in 2 days (3-in-2) is an intermediate option, between TAD and OAD, that gives some of the benefits of OAD with a lesser impact on milk production (Edwards *et al.* 2022). Changes in cow behaviour have been identified between OAD and TAD milking; however, there has been little research investigating the effect of milking 3-in-2 on cow behaviour.

There has been little research around the effect of milking frequency on activity (Hall *et al.* 2021). Most research has focused on the effect of milking frequency on behaviours such as eating and lying time rather than activity, as they are seen as more important to the cow (Munksgaard *et al.* 2005). Most research on total walking distance has focused on farm productivity metrics, such as decreased milk production and profit (Thomson and Barnes 1993) or lameness incidence (Crossley *et al.* 2020), rather than aspects of cow behaviour.

A recent study reported that on days where cows walked further, grazing time increased and rumination time decreased (Neave *et al.* 2021). The authors hypothesised that this was due to the increased energy expenditure of walking further. However, the maximum distance walked in the study was 4 km/cow.day under a TAD milking regime. This is a conservative distance for farms of over 200 cows. Another study investigated the relationship between cow lying time and their steps and found that for every additional 1000 steps/day, lying time decreased by 0.49 h (Beggs *et al.* 2018). As lying time is an important resting behaviour, it is likely prioritised by cows (Munksgaard *et al.* 2005), and they change other behaviours to preserve it. These observations suggest that there may be an effect of milking frequency, through walking distance, on behaviour.

The choice of milking frequency and interval affects the timing of milkings within a day. Grazing behaviour has a significant diurnal pattern, with major grazing bouts occurring in the early morning and late afternoon into early evening (Gibb *et al.* 1998), with the later grazing event being more intense. However, dairy cows milked TAD are typically milked in the early morning, and late afternoon, potentially affecting these natural cycles. In a previous study, researchers observed major grazing bouts after milking (Brumby 1955) as cows tried to rectify the disruption to their natural routine. This is likely further influenced by whether fresh forage is offered later in

the morning, the morning feeding bout can be delayed (DeVries and von Keyserlingk 2005). With respect to milking frequency, despite no difference in grazing time being measured between OAD and TAD, TAD cows grazed later into the evening because of the disruption of a second milking in the afternoon (Tucker *et al.* 2007). Rumination follows a grazing bout, often with a period of idling in between (Gregorini 2012), and these activities are important for efficient digestion and supporting high intakes (Beauchemin 2018). Factors that influence the timing of grazing, including the timing of milkings, will also likely affect the timing of rumination within the day.

Previous studies have noted a stage of lactation effect on behaviour (Bewley et al. 2010; Munksgaard et al. 2020); however, this may be confounded by the other factors that change with the stage of lactation, particularly in block calving pasture-based systems. These factors could include cow milk production, age, and body condition score (BCS), as well as the environmental temperature. Milking frequency affects udder fill but the existing research does not indicate that udder fill significantly affects lying time (Tucker et al. 2007; O'Driscoll et al. 2010, 2011). Research has shown that cows past their first lactation increase their lying time in later lactation (Munksgaard et al. 2020). This suggests that age in combination with stage of lactation affects lying behaviour. Cows with a lower BCS decreased their lying time and increased their grazing time in late lactation, when compared with higher-BCS cows (Matthews et al. 2012). This could potentially be caused by a drive to increase the lower-BCS cows' heat production from rumination or because of hunger. Environmental temperatures that exceed a cow's upper thermal neutral zone of 25°C (Avendaño-Reyes 2012) reduced grazing time, because, to dissipate heat, cows prefer to stand, seek shade, and decrease digestion as that produces large amounts of heat (Kadzere et al. 2002; Herbut et al. 2021). Consequently, it is important to separate the stage of lactation and any other factors that may influence changes in behaviour.

The objective of this study was to identify any differences in cow step activity, measured as steps per hour, among three milking frequencies and three milking schedules of 3-in-2 (milking three times in 2 days) milking, at two stages of lactation. Time spent eating was assessed to help explain differences in step activity within a day. These were investigated using data from five herds of 40 cows (n = 200) milked OAD, TAD, and three 3-in-2 intervals of 12–18–18 h, 10–19–19 h, and 8–20–20 h. The information from this study will allow farmers to assess whether the milking frequency and milking interval they use on farm are altering their cows' behaviour.

Materials and methods

Experimental site and design

The study ran twice for 6 weeks, between 11 September 2020 and 22 October 2020 (early lactation) and 15 January 2021

and 25 February 2021 (mid-lactation) at Ashley Dene Research and Development Station (ADRDS; 43°38′48″S, 172°20′44″E; 35 m above sea level), Canterbury, New Zealand, under the authority of the Lincoln University Animal Ethics Committee (application 2020-12). The 75.5 ha experimental area was subdivided into 3.75 and 4.5 ha paddocks. Rainfall was supplemented with centre pivot irrigation for all paddocks during both experimental periods. Effluent was applied through the pivot weekly and following grazing. Urea was applied at a rate of 23 kg N/ha directly after each grazing event.

A factorial design was used, consisting of five milking times and two stages of lactation. The five milking times were as follows:

- one OAD group with a 24 h milking interval milked at 07:00 hours;
- one TAD group with a 10–14 h interval milked at 06:30 hours and 16:30 hours; and
- three 3-in-2 groups with milking intervals of
 - 12-18-18 (05:00 hours, 17:00 hours and 11:00 hours),
 - 10–19–19 (05:30 hours, 15:30 hours and 10:30 hours) and
 - 8–20–20 (06:00 hours, 14:00 hours and 10:00 hours).

In the covariate period, 10 days prior to the experimental period, cows (n = 200) were allocated into their treatments to allow social groups to form, and milked TAD. Groups were blocked at early and mid-lactation on days in milk (DIM) (34 days \pm 6.7; 136 days \pm 17.5), combined fat and protein yield (1.67 kg/cow ± 0.29; 1.93 kg/cow ± 0.26), somatic cell count (113 000 cells/mL \pm 221 000; 66 500 cells/mL \pm 112 000), parity (20% primiparous; fourth lactation average), breed (Friesian-Jersey cross), genetic merit $(125 \pm 50 \text{ breeding worth}; 157 \pm 107 \text{ production worth})$ and BCS $(4.4 \pm 0.28; 4.4 \pm 0.19 \text{ on a 10-point scale})$ and randomly allocated to a treatment group. There was a smaller pool of animals to select from for the mid-lactation trial period and so 90 cows from the early lactation trial period were reused. The previous treatment was used as an additional blocking factor for these animals. Nine cows were replaced in the early lactation trial period due to mastitis; six cows were replaced in the mid-lactation trial period, including four due to lameness, one for mastitis, and one for other health reasons. Replacement cows were not sampled. Between trial periods cows re-joined the main herd and were milked TAD.

The average temperature during the early lactation trial period was 11.5°C, with the average maximum temperature per day being 17.7°C and minimum 6.3°C. The average temperature was 16.8°C during the mid-lactation trial period, with the daily average maximum being 22.8°C and minimum 11.5°C. A total of 20 mm rain fell over the early lactation trial period and 39 mm during the mid-lactation trial period.

Experimental area management

The groups were rotationally grazed around the experimental area. Each week the pasture mass of all the paddocks was calculated using a calibrated rising plate metre (RPM), and the paddocks were ranked from highest to lowest mass to determine their grazing order. The target dry matter (DM) allocation was 17 kg DM/cow.day over the early lactation trial period and 16 kg DM/cow.day over the mid-lactation trial period.

Occasionally, small adjustments to the grazing area were made to ensure consistency of pasture DM allocation among groups. If the pasture target height post-grazing was exceeded, cows not participating in the trial grazed the paddock for up to 2 days after the experimental cows had finished, to get as close to the target as possible. This was to maintain the pasture quality of any paddocks that would be grazed a second time during the experimental period. When there was insufficient pasture to meet the target allocation, pasture silage was used to appropriately supplement the respective treatment groups.

Temporary fencing was used to subdivide each experimental paddock into five subpaddocks and each treatment group was randomly allocated to one of the five subpaddocks. These subpaddocks were split again to provide grazing for 2 days. All the treatment groups grazed the same paddock on the same days for ease of management and consistency of diet.

The allocation of groups to subpaddocks was randomised among paddocks to ensure that herd-to-herd interaction did not bias results. A random number generator was used to assign a value to each herd for each paddock; the value of the random numbers, from highest to lowest, dictated the break the herd was assigned, with Break 1 being the closest to the lane and the dairy, and Break 5 the furthest away.

In the early lactation period, cows received a fresh pasture allocation after each milking and the size of the allocation was proportional to the milking interval for each treatment group. In the mid-lactation period, each subpaddock was divided into two allocations (63:37 split). Treatment groups spent 30 h in the first allocation and the remaining 18 h in both allocations. All the treatment groups were given their second allocation after the mid-morning milking of the groups milked 3-in-2. The pasture allocation was changed for the mid-lactation period to allow greater access to water troughs during the warmer summer weather. No back fencing was used during either trial period. The average distances between the paddock and the dairy were 1402 m and 1480 m one direction for the early lactation and mid-lactation trial periods respectively.

Animal measurements

All the cows wore AfiAct monitors on the right rear leg from the beginning of the covariate period. The monitors measured the number of steps made over 15 min and data were download at every milking (Fan *et al.* 2022).

CowManager SensOor[™] ear tags (Cowmanager BV, Harmelen, Netherlands) were fitted in the right ear, over the electronic identification tags, of a cohort of eight cows per treatment group, a total of 40 cows per trial period. The cohort was selected from the blocking groups (described in the experimental site and design section above). The CowManager tags were attached in Week 2 of the early lactation trial period and in the covariate portion of the mid-lactation trial period. Data were downloaded when cows passed a solar-powered transmission unit on the way to milking. The tags measured the number of minutes per hour spent in one of the following five behaviour categories: active, not active, high active, eating, and ruminating (Pereira et al. 2018). Active was when the cow was stood on all four feet and walked or moved her body, high active referred to oestrus behaviour. Calibrated in-line milk metres (AfiMilk, Kibbutz Afikim, Israel) recorded each cow's milk weight at each milking.

Data analysis

The cow was the observational unit and the treatment group was the experimental unit, with repeated measures through time (n = 6 weeks). Step data from the AfiAct monitors were summarised as steps per hour per cow, on average, over a week. To achieve comparable means per treatment group per week, groups milked 3-in-2 (where the number of milkings were uneven each week) had odd numbered weeks defined as 6 days and even numbered weeks defined as 8 days. Data were analysed using a repeated measures model (R Core Team version 4.2.1). This was followed by a pairwise comparison using Tukey adjustment to account for multiple comparisons. The fixed effects included in the model were covariate period step activity, days in milk, breed, age, treatment, week, and treatment \times week. The two trial periods were analysed separately. To pinpoint paddock step-activity data, data peaks that coincided with a milking were removed on a treatment basis, to leave the step-activity data associated with when the cows were in a paddock. This included the hour prior to and the hour of milking, as this period was associated with walking to and from milking.

CowManager data were summarised at a cow level, as minutes per hour means by behaviour category, as classified by the CowManager tags, and treatment group, over each trial period. Raw hourly means were then multiplied by 24 to give values as minutes per day. Eating time was summarised as a mean per treatment per hour value and plotted over a period of 24 h. The 3-in-2 group data was subdivided into Day 1 data, when cows were milked twice, and Day 2 data, when cows were milked once. Cow-level data were removed at a daily level when the total eating time over 24 h was equal to zero (a logger error), or when values were biologically impossible, i.e. total eating time over 24 h/cow was less than 20 min or more than 780 min. Milk weight data were analysed using a model with the same fixed effects as for the step-activity analysis (SAS Institute Inc., Cary, NC, USA). Milk weight data were averaged over morning, afternoon, and 'mid'-morning milkings for the 3-in-2 groups to achieve a per day average milk yield.

Results

Step activity

Weekly mean steps per hour increased with an increasing milking frequency during both the early and mid-lactation trial periods (Table 1). The number of steps was greater during mid- than early lactation. Cows milked OAD averaged 95 and 59 fewer steps per hour than TAD and 3-in-2 herds (respectively) during the early lactation trial period, and 115 and 78 fewer steps per hour during the mid-lactation trial period. There was a significant difference between the 3-in-2 groups in cow-step activity on Day 1 due to the effect of time of milking. The shorter the time between the morning and afternoon milking on Day 1, the greater the number of steps per hour. No difference in steps per hour was detected during both the early and mid-lactation trial periods on Day 2 where the 3-in-2 groups were all milked once.

Cows milked OAD took 30% fewer steps per hour than those milked TAD, regardless of whether the steps related to milking were included or not. Cows milked 3-in-2 took 10% fewer steps than cows milked TAD when averaged over the whole day. However, when the steps associated with walking to and from milking were removed, 3-in-2 cows took 8% more steps than TAD cows during the early lactation trial period. Over mid-lactation, the amount of steps TAD cows took fell between the 8–20–20 and 10–19– 19 3-in-2 groups. Milk weight increased with an increased milking frequency (Table 1).

CowManager behaviour data

Due to the small number of tags available per treatment group, raw means for each behaviour category are presented by treatment group and experimental period (Table 2).

The eating time data were analysed to attempt to explain differences in the step activity of different trial groups (Table 1). In the early lactation trial period, cows' eating time increased after milking for all the treatment groups (Fig. 1). After 10 am, when compared with the TAD treatment group, the OAD treatment group had an extended period of eating throughout the afternoon into the evening, with a less defined afternoon peak than for the TAD treatment group. The 3-in-2 8–20–20 and 12–18–18 intervals were also compared, as they had the largest difference in milking time and they had a significant difference in step activity

Table 1. A comparison of early lactation and mid-lactation least square means by milking frequency and interval: once a day (OAD); twice a day (TAD); and three intervals of milking three times in 2 days (3-in-2); 8–20–20; 10–19–19 and 12–18–18 (<i>n</i> = 40 cows/treatment) for step per hour per cow values as a whole day average and paddock time averages, divided into Day 1
(two milkings) and Day 2 (one milking) values for the 3-in-2 herds over the experimental period (n = 6 weeks). As well as least square mean values for milk weight as an average per cow per day by treatment groups described below.

Parameter				Early lactation	ation							Mid-lactation	tation			
			Treatment			s.e.d.	s.e.d. P-value	P-value			Treatment			s.e.d.	P-value	P-value
	OAD	8-20-20	OAD 8-20-20 10-19-19 12-18-18		TAD		(treatment) (week)	(week)	OAD	8-20-20	OAD 8-20-20 10-19-19 12-18-18 TAD	12-18-18	TAD		(treatment)	(week)
Steps (per cow/h)	237a	302c	299bc	287b	332d 4.82	4.82	<0.001	<0.001	284A	381C	360B	344B	399D	6.37	<0.001	<0.001
Day I		329b	337b	309a		7.60	0.029	<0.001		422B	391A	373A		8.2 I	<0.001	<0.001
Day 2		277	274	275		7.13	0.549	<0.001		339	335	332		8.10	0.444	<0.001
Paddock steps (per cow/h)	184a	276d	272cd	262c	249b	4.67	0.001	<0.001	208A	37ID	327B	325B	343C	5.20	<0.001	<0.001
Paddock Day I		291b	292b	265a		5.81	<0.001	<0.001		426B	348A	337A		5.91	<0.001	<0.001
Paddock Day 2		255	245	257		5.62	0.080	<0.001		322	313	314		5.81	0.182	<0.001
Milk yield (kg/cow.day)	18.12c	20.75b	20.86b	21.46b	22.91a 0.364	0.364	<0.001	<0.001	16.75C	18.58B	I 8.40B	18.89AB	19.16A	0.208	<0.001	<0.001
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Means within a row followed by different letters are signficantly different (at P = 0.05; Tukey test) s.e.d., maximum standard error of the difference.

(Table 1). The influence of milking times was evident. On Day 1, the 12–18–18 treatment group was milked at 17:00 hours and had a peak in eating time later in the afternoon, while the 8–20–20 treatment group was milked at 14:00 hours and had an extended period of afternoon eating. On Day 2, both treatment groups were milked in the mid-morning and their eating patterns were similar.

Discussion

The primary objective of this study was to identify the effect of milking frequency, including 3-in-2 milking intervals, on cows' step activity, as indicated by the differences in mean steps per hour per cow. A secondary objective was to investigate diurnal differences in eating time to assist with the explanation of within-paddock differences in step activity.

An increase in milking frequency resulted in an increase in mean steps per hour in both the early and mid-lactation trial periods. As milking frequency dictated the number of times the cow walked between paddock and the dairy, with an average distance of between 1.4 and 1.5 km one way, the relationship between the number of milkings per day and average number of steps is not unexpected. However, when the data from the period that included travel to and from milking was removed, leaving step per hour values for the time spent in the paddock, a relationship remained between milking frequency and steps per hour.

Cows in the OAD treatment group had the lowest number of steps per hour during both the early and mid-lactation trial periods whilst in the paddock, despite having the greatest amount of time there due to their milking schedule. Differences in feed searching behaviour may be an explanation for this observation. Fig. 1 illustrates that the eating time of the OAD treatment group was more prolonged, with a slow rate of increase across the afternoon, when compared with the TAD treatment group. Perhaps the cows milked OAD felt a lesser need for rapid pasture intake as they were not removed for afternoon milking and were used to this routine, and other behaviours, such as grooming, ruminating or resting, were prioritised over the afternoon. It is also possible that due to less milk production from the OAD cows, their appetite was reduced and therefore grazing time was reduced, resulting in fewer steps. Previous research investigating the relationship between eating time and milking frequency (Tucker et al. 2007) or prolonged milking (Beggs et al. 2018) has not shown any differences in daily eating time, but diurnal variation and grazing intensity were not explored.

An alternative hypothesis could be that OAD cows were less active due to discomfort, as their udders were distended due to higher milk volumes caused by lower milking frequency. One study reported decreased lying time due to udder discomfort when a milking was omitted, showing that udder discomfort can change behaviour (O'Driscoll *et al.* 2011).

Table 2. A comparison of early lactation and mid-lactation raw means by milking frequency and interval: once a day (OAD); twice a day (TAD); and
three intervals of milking three times in 2 days (3-in-2); $8-20-20$; $10-19-19$ and $12-18-18$ ($n = 8$ cows/treatment) for total minutes per day in the
following behaviour categories: eating, ruminating, active, high active and not active, as defined by the CowManager tags, over the experimental
period $(n = 6 \text{ weeks})$.

Parameter			Early lactation			Mid lactation				
	OAD	8–20–20	10-19-19	12-18-18	TAD	OAD	8–20–20	10-19-19	12-18-18	TAD
Eating	305	389	456	439	336	444	382	338	403	446
Ruminating	454	403	427	420	492	463	427	454	458	437
Active	164	168	131	114	196	134	243	211	213	160
High active	218	199	191	200	171	153	145	194	149	147
Not active	305	295	250	286	262	248	249	269	222	254

3-in-2 12-18-18

3-in-2 8-20-20

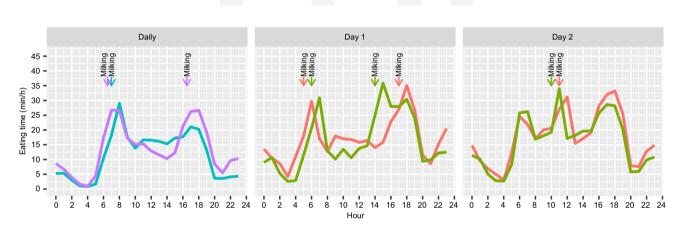


Fig. 1. A graphical representation of early lactation eating time in minutes per hour per cow (n = 8) over a period of 24 h by miking frequency; once a day (OAD) vs twice a day (TAD) and milking three times in 2 days (3-in-2) intervals; 8–20–20 and 12–18–18, compared with milking times and divided into Day I (when cows are milked twice) and Day 2 (when cows are milked once) data for 3-in-2 treatments.

In this trial, step activity was higher during the mid-lactation trial period, when cows produced less milk, than during the the early lactation trial period, and cows in the 3-in-2 treatment groups walked fewer steps on the day they were milked once. Only one study has investigated the correlation between activity and milking frequency. The study reported a relationship between decreased milking frequency from TAD to 3-in-2 and increased activity (Hall *et al.* 2021). More research is needed to understand which behaviours cows substitute for movement on days with only one milking, and the reasons for those differences.

When the step data associated with walking to and from milking were removed, the TAD trial group completed fewer average steps per hour than did the 3-in-2 trial groups, including on the day where 3-in-2 trial groups were milked twice. Greater activity in cows switched from TAD to 3-in-2 milking has been observed previously (Hall *et al.* 2021). This could have been due to an increase in feed searching behaviour, particularly in the evening, as a result of the larger area given per cow at each feed allocation. This hypothesis could be supported by the data from the early lactation trial period in our experiment; however, the pasture management changed for the mid-lactation trial period, where all treatments received the same-sized allocation each day, and the 8–20–20 herd remained more active. As the afternoon milking for the 8–20–20 herd occurred earlier than for the TAD herd (at 14:00 hours versus 16:30 hours), it is possible that the 8–20–20 herd had two grazing bouts, one occurring after milking and another in the early evening, resulting in more steps. Consequently, we speculate that the amount of walking TAD cows were required to do, due to their milking schedule, resulted in less step activity in the paddock.

TAD 10-14

OAD

A difference in steps per hour was also measured between 3-in-2 milking intervals, driven by differences on Day 1, when cows were milked twice. This suggests that the timing of milking influences step activity. The 8–20–20 trial group was milked at 06:00 hours and 14:00 hours on Day 1, and at 10:00 hours on Day 2, while the 12–18–18 trial group was milked at 05:00 hours and 17:00 hours on Day 1, and at 11:00 hours on Day 2. Cows have preferential grazing

bouts early in the morning, and late afternoon into early evening (Gibb et al. 1998), with the afternoon bout being more intense. Milking times of the 12-18-18 trial group on Day 1 appear to have coincided with the cows' preferred grazing time (Fig. 1). The 8-20-20 trial group, which was less disrupted by milking, clearly shows two separate peaks of eating over an extended period in the afternoon after milking, while the 12-18-18 herd had only one. The pattern of eating followed a similar trend when both trial groups were milked once on Day 2. Consequently, we hypothesise that the step-activity differences seen in the different 3-in-2 trial groups were driven by changes in grazing behaviour. Although there is little research on this topic, it is supported by one study that showed TAD cows grazing further into the evening than OAD cows (Tucker et al. 2007). The expression of natural behaviour is an important component of animal welfare, and thus the impact of milking timing on grazing behaviour is a consideration.

A key reason why farmers wish to reduce milking frequencies and extend milking intervals is to reduce work hours and early morning work starts, for their own welfare and so that they can attract and retain staff. However, it is important to also consider whether altering milking frequencies and intervals has a behavioural impact on cows. When looking at the impacts of milking frequency on a cow, most research has focused on farm metrics, such as milk production (Edwards 2018), or traditional welfare metrics, such as BCS (Roche et al. 2009). From our research, if we assume that the average length of the cow's step is 1.5 m (Alsaaod et al. 2017), and the energy expenditure per kilometre walking over flat ground is 2 MJ (Nicol and Brookes 2007), then a difference between groups of 27 steps/h would provide a difference of energy expenditure of 2 MJ/cow.day. The largest significant difference we saw between the OAD and TAD herds was 115 steps/h at mid-lactation. If this is extrapolated to increased pasture demand with a large herd, this could result in a large energy deficit if step-activity differences are not considered when changing milking frequencies. Welfare also covers many facets, including normal behaviour expression. When farmers consider adjusting their milking times and frequency to suit their goals, they should ensure they account for potential changes in normal grazing behaviour and grazing allocation. Putting systems in place, such as limiting herd sizes to ensure that cows are not away from the paddock any longer than necessary and that efficient milking practices are used (Edwards 2013), will also help ensure that disruption to natural behaviour is minimised.

Conclusions

Milking frequency affected cow step activity, both when walking to the dairy was included and excluded from the data. Changes in diurnal eating patterns were also evident when comparing OAD and TAD trial groups. We believe that these differences were likely to be due to the amount of walking between the paddock and dairy, and changes in grazing behaviour or another explanation such as udder discomfort. However, because milking frequency also affects milk production, more research is needed to understand the relationship among milk production, milking frequency and behaviour. The timings of milkings also affected cow step activity, with significant differences seen amongst the 3-in-2 trial groups, driven by differences on the day cows were milked twice. There were differences in the eating pattern amongst the 3-in-2 trial groups, suggesting the timing of milkings, in addition to the frequency, affects grazing behaviour and, thus, step activity. More research is needed to determine the impact of these differences on animal welfare. Farmers should offset these changes in grazing behaviour due to milking frequency and timing by minimising the time cows spend out of the paddock, especially in the afternoon.

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