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Bimodal milk flow and overmilking in dairy cattle: risk factors and consequences

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ABSTRACT

To maximise the return on capital invested in the milking parlour, the largest number of cows should be milked gently and completely in the shortest possible time. Bimodal milk flow and overmilking negatively influence the efficiency of the milk removal process and teat health. This observational study had the objective of investigating the prevalence of bimodal milk flow and overmilking, determining which individual and farm-related variables are associated with these occurrences, and determining the association of overmilking and bimodal milk flow with milk yield and with short- and long-term teat changes. Twenty-one farms were visited once during the study period, wherein the milking routine was timed, the teat condition was assessed, and dynamic evaluation of the milking vacuum was performed. A total of 606 vacuum graphic records were obtained, with an average of 29 ± 3 records per farm, in order to indirectly evaluate the milk flow and thus determine the occurrence of bimodal milking and overmilking time. The average percentage of bimodality per farm was 41.7%. The median overmilking time was 59 seconds, and on average, 78.3% of the cows in a herd were overmilked longer than 30 seconds. An association was found at cow level between the occurrence of bimodal milk flow and days in milk, the total stimulation time, parity, and the preparation lag time. The increase in the mean total stimulation time and the number of passes during preparation were associated with a decrease in the proportion of bimodality in the herd. Parity, reattachment of the milking unit and milking in manual mode were associated with an increase in overmilking time of an individual cow. The presence of a clogged air bleed hole in the claw and the reduction of the cluster removal milk flow threshold were associated with an increase in the herd's median overmilking time. The average milk flow decreased with the increase in overmilking time and with the occurrence of bimodal milk flow. An association was also found between the occurrence of bimodal milk flow and decreased milk yield. A mean of 78.4% of cows per farm had short-term teat changes in at least one teat, and 33.6% of evaluated cows per farm displayed at least one teat with hyperkeratosis. These results emphasise the association of bimodality and overmilking on milking efficiency and reinforce the importance of the milkers' actions and the functioning of the milking parlour for its prevention.

Implications

Bimodal milk flow, characterised by a period of reduction or even transient absence of milk flow in the beginning of milking, and overmilking, characterised by milk flow below a certain threshold at the end of milking, negatively influence the efficiency of the milk removal process and teat health. A proper stimulation in the premilking routine and having a milking parlour with the correct settings and properly maintained were associated with lower observed bimodal milk flow and overmilking, potentially improving milking efficiency, teat health and animal welfare.

Introduction

Milking is the dairy farm activity with the highest labour expenditure (Hansen, 1999), with tasks related to milking reaching 50% of the total labour force available on farm (Chang et al., 1992). In 2018 in the USA, labour costs in dairy farms corresponded on average to 10.6% of operational income (USDA, 2020). Optimizing the

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milking process has therefore the potential to reduce dairy farm costs.

Milk secretion to the alveolar lumen is continual, but its removal occurs periodically, with milk being stored in the mammary gland between milkings (Nickerson and Akers, 2011). Milk is stored in the mammary gland in two compartments: part remains in the alveoli and small ducts – alveolar milk – and the remainder is stored in the lumen of larger ducts and in the gland and teat cisterns – cisternal milk (Knight et al., 1994). After milking, there is an initial period of alveolar filling before milk transfer to the cistern starts, due to secretion pressure in the following hours (Knight et al., 1994). Within 8–12 hours after milking, around 80% of the milk in the udder is stored as alveolar milk, whereas 20% is stored as cisternal milk (Pfeilsticker et al., 1996). Besides the time from last milking, milk distribution in the mammary gland is influenced by the rate of milk secretion and by the dimensions of the teat cistern. Milk in the alveolar compartment is unavailable for removal by machine milking, due to adhesive and capillary forces, so in order to be removed, it has to be transferred to the cisternal compartment by an oxytocin-dependent neuroendocrine reflex called milk ejection (Bruckmaier et al., 1993).

Tactile stimulation of the teats and udder, by either calf suction, milkers' hands or teat liner motion, leads to oxytocin release (Bruckmaier and Blum, 1996). Independently of the type and intensity of tactile stimulation, around 30 seconds after the beginning of such stimulation, there is a significant rise in the blood concentrations of oxytocin (Weiss and Bruckmaier, 2005). Transfer of alveolar milk to the cistern occurs around 40 seconds after the first contact with a teat in full udders, but it may take up to 150 seconds to happen in udders with low quantities of milk (Weiss and Bruckmaier, 2005).

The premilking routine includes a set of procedures, performed prior to attaching the milking units, aiming at effectively and efficiently improve milk quality, promote milk let-down and ensure the health of the udder. Premilking stimulation seeks to promote oxytocin release and milk let-down before unit attachment so that alveolar milk is transferred to the cisterns, before the removal of all the cisternal milk (Watters et al., 2012). When all the milk stored in the gland cistern is removed before ejection of alveolar milk, there is a period of reduction or even transient absence of milk flow, which is resumed after milk let-down, an occurrence termed bimodal milk flow (Bruckmaier and Hilger, 2001). In periods of reduced or absent milk flow, the vacuum levels at the tip of the teat increase, exacerbating stretching and compressive forces over the teats (Besier and Bruckmaier, 2016). An increase in the levels of vacuum at the mouthpiece chamber also occurs (Borkhus and Rønningen, 2003), potentially leading to the collapse of the teat wall, a deeper penetration of the teat into the milking unit, with the potential to compromise milk flow during milking (Bruckmaier and Blum, 1998).

Bimodal milk flow may be the result of inadequate stimulation before milking unit attachment or of a short latency period between stimulation and attachment (Weiss and Bruckmaier, 2005). Current recommendations suggest that for an adequate milk ejection to occur, teats should be stimulated for at least 15 seconds, with a preparation lag time of 90–120 seconds between the beginning of stimulation and unit attachment (NMC, 2013; Wieland et al., 2020a).

Besides bimodal milk flow, overmilking can also lead to the loss of milking efficiency by increasing the machine-on time at a low milk flow phase (Rasmussen, 1993). Overmilking starts when the milk flow to the teat cistern is less than the flow out of the milk canal (Rasmussen, 2004). During overmilking, there is a decrease in the intramammary pressure to around 90% of the milking vacuum (Rasmussen et al., 1994), with a rise in vacuum levels and in cyclic variations in the mouthpiece vacuum (Borkhus and Rønningen, 2003). Congestive changes in teat tissues occur throughout milking, but they are more severe at overmilking due to increased vacuum at the clawpiece and mouthpiece chamber (Penry et al., 2017).

It is possible to evaluate both the occurrence of bimodal milk flow and overmilking with different equipment during milking. Dodenhoff et al. (1999) used a continuous electronic milk flow meter (Lactocorder) to study the relationship between traits derived from the milk flow curve and somatic cell score. Using the same equipment, Sandrucci et al. (2007) characterised the association of individual cow-level variables (lactation number, days in milk (**DIM**), premilking delay time) with milking time, bimodal milk flow and milk production in Italian dairy herds. Using milking vacuum digital recorders (VaDia), a team from Michigan State University characterised which herd-level variables were associated with bimodal milk flow (Moore-Foster et al., 2019a) and overmilking time (Moore-Foster et al., 2019b) in Michigan dairy farms. Furthermore, in a recent study, Vierbauch et al. (2021) studied the relation of milking, overmilking and vacuum levels with the teat condition of front and rear quarters.

The present study aimed to characterise the prevalence of bimodal milk flow and overmilking in several dairy herds in Portugal, determine the association with herd and individual risk factors and characterise its consequences.

Material and methods

Data collection

The study included 606 cows from 21 dairy herds in Portugal, between February and June 2020. Enrolled farms were selected for their willingness to participate and for their ability to record milking data. Farms had cows housed all year round in either straw yards or cubicles, and feeding was performed according to the recommendations of the National Research Council (2001). Each farm was visited once, with milking data being gathered in the afternoon milking. During milking, information was obtained regarding the milking parlour, milking routine, teat health, milk production, with a dynamic vacuum evaluation and timing of routine protocols being performed for the animals being evaluated. After the evaluations performed during milking, information was sought from both the farm management software and milk recording, regarding parity and DIM for the animals previously undergoing vacuum dynamic evaluations. For parlours with automatic cluster removal, the cluster removal milk flow threshold was recorded.

Milking routine evaluation and timing

The milking protocol was observed on each farm, in order to identify the milking routine steps with a special focus on the stimulation procedures. Any procedure that involved physical contact with the teats was considered to have the potential to promote the necessary stimulation for oxytocin release, including forestripping, teat cleaning with either paper or cloth towel, or with a mechanical brush, and mechanical stimulation by the milking machine stimulator (Moore-Foster et al., 2019c).

During milking, the time spent by the milkers in each step of the milking routine and the interval between them was recorded using a stopwatch for the animals subjected to the dynamic vacuum evaluation. Based on this timing, the total teat stimulation time (s) was determined by adding the total amount of time spent on each stimulation procedure. The preparation lag time was also determined by timing between the beginning of stimulation and attachment of the milking unit.

Dynamic vacuum evaluation and milk yield

Vacuum recordings were made with two VaDia (Biocontrol, Rakkestad, Norway), a portable digital vacuum logger. The VaDia logger presents four vacuum recording channels, allowing to record the vacuum dynamics in four distinct points of the milking unit. Using a 2.4 mm inner diameter silicone tube, the four channels were connected to the following points of the milking unit: (a) short pulsation tube of a rear teatcup, to evaluate pulsation; (b) rear quarter liner mouthpiece, to evaluate vacuum at the mouthpiece chamber; (c) front quarter liner mouthpiece, to evaluate mouthpiece chamber vacuum; (d) short milk tube of a teat liner for a rear quarter, to evaluate vacuum at the clawpiece. Vacuum recordings were performed continuously from unit attachment until the units were automatically or manually removed by the milkers in the milking units where the VaDia were assembled. All vacuum recordings were downloaded to a computer and analysed with the VaDia Suite software (Biocontrol, Rakkestad, Norway).

Based on the graphic analysis of the vacuum recordings made with the VaDia Suite software, it was possible to identify-four milk flow moments during milking: beginning of milking, beginning of peak milk flow, beginning of overmilking and end of milking. Milking time was determined by the difference between the beginning and end of milking in the vacuum recording. The beginning of milking, corresponding to the moment in which the teatcups were attached to the teats, was identified as the first time with a graphical recording of vacuum above 25 kPa (Biocontrol, 2011). Bimodal milk flow was identified when the milking vacuum recording showed a decrease in vacuum in the short milk tube and in both front and rear mouthpiece chambers, after the beginning of milking, followed by a marked increase in the three vacuum recordings (Erskine et al., 2019). To determine the beginning of peak flow, an automatic function of VaDia Suite was used, which evaluates the mean vacuum in the short milk tube for periods of 10 seconds after unit attachment. This automatic determination of the beginning of peak flow was confirmed and eventually corrected through visual assessment. When the difference in vacuum between two such periods is lower than 0.15 kPa, the middle point between these two periods is considered the beginning of peak flow (Biocontrol, 2011). This function uses the principle that claw vacuum is inversely proportional to the milk flow (Besier et al., 2016).

The beginning of overmilking was determined as the moment in which the mouthpiece chamber vacuum in both teats increased, to a plateau level above 15 kPa and vacuum fluctuations in the short milk tube were below 3 kPa, according to Moore-Foster et al. (2019b). The end of milking corresponded to the moment in which the vacuum in all four channels returned to zero. Overmilking time was recorded as the interval in seconds between beginning of overmilking and end of milking. The milk yield for each individual milking was retrieved from the milking parlour.

Teat condition evaluation

Presence of short- and long-term changes in teat condition was evaluated on all farms included in this study. To detect the presence of short-term teat changes, each teat was evaluated by a single evaluator in the first 30 to 60 seconds following milking unit removal for: (a) firmness (1-normal, 2-firm, 3-wedged); (b) colour in non-pigmented teats (1-pink, 2-reddened, 3-blue-coloured); (c) swelling near the teat base (1-no ring, 2-visible mouthpiece lip mark, 3-palpable thickened ring) as proposed by Teat Club International (Mein et al., 2001). It was considered that a cow had a short-term teat change whenever at least one of the teats presented a firm or wedged teat end, was reddened or blue-coloured or had a palpable thickened ring, according to Wieland et al. (2020b). To evaluate teat-end hyperkeratosis, the visual classification proposed by the Teat Club International was used, which presents four distinct levels of hyperkeratosis. It was considered that an animal presented long-term changes to teat condition whenever at least one teat presented a score of 3 or 4 in the scale (Mein et al., 2001). In farms with 400 or less lactating cows, all 4 teats from 80 cows were evaluated, whereas in farms with over 400 lactating animals, all 4 teats from 20% of the lactating cows were evaluated. Cows from all pens were evaluated, proportionately to the number of cows in each pen (Reinemann et al., 2001).

Statistical analysis

Data collected from farm visits and evaluated milkings were compiled in an Excel (Microsoft Corp., Redmond WA, USA) spreadsheet for data handling and descriptive statistical analysis. Statistical analysis was performed with SAS (SAS 9.4, SAS Institute Inc., Cary, NC).

Farm risk factors that could be associated with the proportion of cows exhibiting bimodal milk flow were analysed by fitting a generalised linear model with a logit link and a beta distribution using the procedure PROC GLIMMIX (SAS). Three groups of variables were considered as independent variables: farm characteristics (number of milkings per day (2 or 3) and average milk production recorded in the month of the visit (kg), premilking routine characteristics (total average stimulation time (s), average preparation lag time (s), number of cows prepared per pass and number of passes during the premilking routine) and milking parlour characteristics (number of milking stalls, number of milking stalls per milker and number of cows per milking stall). Average milk production, average stimulation time, average preparation lag time, number of cows prepared per pass, number of passes during the milking routine, number of milking stalls, number of milking stalls per milker and number of cows per milking stall were classified as continuous variables, whereas the number of milkings per day was classified as a categorical variable.

Farm risk factors that could be associated with the median overmilking time were analysed by fitting a linear model with the procedure PROC MIXED (SAS). Farms (n = 2) without automatic cluster removers were excluded from this analysis. Average milk production for the farm (kg), presence of obstructed air bleed hole (yes or no), number of milkings per day (2 or 3), number of milking stalls, cluster removal milk flow threshold (g/min) and size of the lactating herd were considered independent variables for inclusion in the model. Average milk production, number of milking stalls, size of the lactating herd and cluster removal milk flow threshold were classified as continuous variables, whereas the number of milkings per day and presence of obstructed air bleed holes were considered categorical variables.

To identify individual risk factors associated with bimodal milk flow, a generalised linear mixed model with a logit link and a binary distribution of the response variable was fitted with the PROC GLIMMIX (SAS) procedure. To control for the clustering of cows per farm, farm was included as a random effect on all individual models using cow as the observational unit. The independent continuous variables selected for this model were, stimulation time (s), and preparation lag time (s), whereas DIM (<100, 101–200, >201) and parity (primiparous or multiparous) were included as categorical variables.

To identify individual risk factors associated with the occurrence of overmilking, a linear mixed model was fitted with the PROC MIXED (SAS) procedure. Parity, DIM, use of manual cluster removal (yes or no) and unit reattachment (yes or no) were included in the model as categorical variables.

To investigate if there was an association between overmilking time and bimodal milking occurrence with average milk flow (kg/min), a linear mixed model was fitted with the PROC MIXED (SAS) procedure. Parity, DIM, bimodal milk flow (yes/no) and overmilking time (s) were included in the initial model, with overmilking time as a continuous variable and DIM, parity and bimodal milk flow as a categorical variable.

To investigate if there was an association between bimodal milk flow and individual milk yield (kg), a linear mixed model was fitted with the PROC MIXED (SAS) procedure. Occurrence of bimodal milk flow, DIM and parity were independent variables included in the model as categorical variables.

As suggested by Heinze and Dunkler (2017), manual backward elimination with a *P*-value criterion of 0.157 without a preceding univariable prefiltering was performed to reach the final models. The assumption of homoscedasticity was evaluated visually using the plots of studentised residuals against predicted values, and the assumption of normality of residuals was evaluated using the Q-Q plots. To satisfy these assumptions, overmilking time was naturally log-transformed. For models with farm included as a random effect, the covariance structure that resulted in the lowest Akaike information criteria was selected. In the final models, differences were considered significant when $P \leq 0.05$, whereas a tendency was defined as $0.05 < P \leq 0.10$. Mathematical equations and SAS statistical models used are provided in Supplementary Material S1.

Results

Farm, milking parlour and milking routine

Among the 21 farms included in the study, average herd size was 406 lactating cows, ranging from 160 to 864 cows and mean milk yield per cow/day was 34.2 (Table 1). Twelve farms (57%) milked two times per day, whereas nine (43%) milked three times per day. Fourteen milking parlours (66.6%) were parallel, three were herringbone (14.3%), two were rotary (9.5%), onewas tandem (4.8%) and one was herringbone with swing-over arm (4.8%). The average number of milking stalls in each parlour was 30, and 50% of the parlours with automatic cluster removers had 400 g/min or lower as cluster removal milk flow threshold (Table 1).

Thirteen (61.9%) of the premilking routines evaluated had a territorial routine, six (28.6%) had a sequential routine and two (9.5%) had a grouping routine. As for the number of passes per milking stall, in two farms (9.5%) workers performed a single pass, in five farms workers performed two passes (23.8%), in 10 farms (47.6%) workers performed three passes and in four farms (19.1%) workers performed four passes. In nine farms (43%), there was forestripping and in six farms (29%), there was mechanical stimulation (Stimuplus, GEA) as additional teat stimulation. The individual herd and parlour characteristics of the 21 herds enrolled in the study are shown in Supplementary Table S1.

Table 1
Herd descriptive data of 21 dairy cattle farms enrolled in the study.

Herd descriptive data	Mean[1]	Minimum	Maximum
Number of lactating cows per farm	406 ± 44	160	864
Individual milk yield (kg/day)	34.2 ± 1.1	24.1	44.1
Number of parlour milking stalls	30 ± 3	10	70
Cluster removal milk flow threshold (g/m)	445 ± 43	200	900
Mean stimulation time (s)	16.0 ± 3.6	2.9	65.5
Mean preparation lag time (s)	94.5 ± 10.9	14.7	261.0
Proportion of cows with bimodal milk flow (%)	41.7 ± 4.9	7.1	86.4
Median overmilking time (s)	83.2 ± 17.5	24.0	393.5
Proportion of cows with >30 s of overmilking time (%)	78.3 ± 4.4	29.3	100
Proportion of cows with short-term teat changes (%)	78.4 ± 2.7	55.8	98.8
Proportion of cows with hyperkeratosis (%)	33.6 ± 2.2	20.2	59.0

[1] Mean ± SEM.

Total stimulation and preparation lag time

Median stimulation time of the premilking routines that were monitored (n = 606) was 8.6 seconds, with a minimum and maximum total stimulation time of 0.7 and 97.8 seconds, respectively. Median preparation lag time was 83.4 seconds, with a minimum and maximum of 9.2 and 342.2, respectively.

Bimodal milk flow

Among the 606 digital graphic recordings of individual milkings evaluated, 260 (42.9%) were identified as bimodal milkings and mean proportion of cows with bimodal milk flow per farm was 41.7% (Table 1).

Overmilking

For the same amount of digital graphic recordings of individual milkings, there was a median overmilking time of 59 seconds, with three animals with no overmilking time, while the longest overmilking time was 932 seconds. Moreover, 467 (77.1%) of the observed animals were overmilked more than 30 seconds.

Average milk flow-associated risk factors

Average milk flow was associated with overmilking time ($P < 0.001$), bimodal milk flow ($P < 0.001$), parity ($P = 0.002$) and the interaction between parity and DIM ($P = 0.004$), whereas it tended to be related to DIM ($P = 0.063$) (Table 2). Each additional second of overmilking was associated with a 0.004 kg/min decrease in average milk flow. Cows exhibiting bimodal milk flow had a 0.28 kg/min lower milk flow. Cows between 101 and 200 DIM tended to exhibit the highest milk flow (2.64 kg/min), whereas cows less than 101 DIM and more than 201 DIM exhibited 2.57 kg/min and 2.47 kg/min, respectively. Multiparous cows displayed an average milk flow 0.52 kg/min higher than primiparous cows, but the more DIM the lesser this difference.

Teat condition evaluation

Regarding short-term teat changes, in each farm, 78.4 ± 2.7% of evaluated cows presented at least one teat with a change in colour, firmness or with a swelling at the base of the teat. Moreover, there was a moderate positive correlation (coefficient: 0.65; $P = 0.0015$) between the farms' median overmilking time and the proportion of cows exhibiting short-term teat changes. In 33.6 ± 2.2% of evaluated cows, there was at least one teat with a hyperkeratosis score of 3 or 4, and no correlations were detected between the proportion of cows exhibiting hyperkeratosis and farms' median overmilking time or proportion of cows with bimodal milk flow.

Bimodal milk flow herd-level risk factors

Regarding herd-related variables, herd mean stimulation time ($P = 0.020$) and number of passes per milking unit ($P = 0.029$) were associated with the proportion of cows with bimodal milk flow in the herd (Table 3). Maintaining all other variables constant, each additional second of mean stimulation in the milking routine was linked to a 0.97 odd of a cow from that herd exhibiting bimodal milk flow. Likewise, each additional pass per milking stall was linked to a 0.65 odd of a cow from that herd exhibiting bimodal

Table 2
Multivariable analysis model of average milk flow (kg/min) for 606 cows enrolled in the study.

Parameter	Estimate	SE	*P*-value	Overall *P*-value	LSM	LSM 95% CI
Intersect	2.79	0.12	<0.001	-	–	–
Overmilking (seconds)	−0.004	0.0005	<0.001	<0.001	–	–
Bimodal milk flow	–	–	–	<0.001	–	–
No (referent)	–	–	–	–	2.70	2.58–2.82
Yes	−0.28	0.07	<0.001	-	2.42	2.29–2.55
DIM	–	–	–	0.063	–	–
1–100 (referent)	–	–	–	–	2.57	2.42–2.72
101–200	0.28	0.14	0.04	–	2.64	2.51–2.78
>201	0.17	0.13	0.19	–	2.47	2.34–2.59
Parity	–	–	–	0.002	–	–
Primiparous (referent)	–	–	–	–	2.45	2.32–2.59
Multiparous	0.52	0.13	<0.001	-	2.66	2.55–2.78
Parity × DIM	–	–	–	0.004	–	–
Primiparous × 1–100	–	–	–	–	–	–
Primiparous × 101–200	–	–	–	–	–	–
Primiparous × > 200	–	–	–	–	–	–
Multiparous × 1–100	–	–	–	–	–	–
Multiparous × 101–200	−0.41	0.17	0.017	–	–	–
Multiparous × > 200	−0.54	0.16	0.001	–	–	–

Abbreviations: DIM = Days in milk; LSM = Least square means; CI = Confidence interval.

Table 3
Multivariable analysis model of bimodal milk flow proportion (%) for 21 dairy cattle farms enrolled in the study.

Parameter	Estimate	SE	*P*-value	LSM	Odds[1]	Probability[2]
Intersect	1.06	0.48	0.042	–	–	–
Mean stimulation time (seconds)	−0.029	0.011	0.020	–	0.97	0.49
Number of passages per milking unit	−0.43	0.18	0.029	–	0.65	0.39
Number of milkings	–	–	0.098	–	–	–
Two (referent)	–	–	–	−0.605	0.546	0.35
Three	0.58	0.33	0.098	−0.029	0.971	0.49

Abbreviations: LSM = Least square means.
[1] Estimates back transformed for interpretation in text as in beta regression estimates are interpreted as changes in log-odds.
[2] Probability was calculated as: Probability = odds/(odds + 1).

milk flow. Additionally, farms milking three times daily tended (P = 0.098) to have a higher proportion of cows with bimodal milk flow, as represented by the 49% probability of cows displaying bimodal milk flow, compared to the 0.35% of farms milking twice a day.

Overmilking herd-level risk factors

Herd median overmilking time was related to cluster removal milk flow threshold (P = 0.005) and presence of obstructed air bleed holes (P = 0.034) (Table 4). Each g/min increment in cluster removal milk flow threshold was associated with 0.09 seconds decrease in median overmilking time, and presence of an obstructed air bleed hole in the clawpiece was linked to an increase in median overmilking time of 37.40 seconds.

Bimodal milk flow individual risk factors

Preparation lag time (P = 0.001) and DIM (P < 0.001) were related with the occurrence of bimodal milk flow, whereas parity (P = 0.090) and stimulation time (P = 0.054) tended to be related (Table 5). Compared to cows with less than 101 DIM, the odds of cows between 101 and 200 DIM and the odds of cows with more than 201 DIM exhibiting bimodal milk flow were 1.78 and 3.07 higher, respectively. Each additional second of preparation lag time represented an OR 0.99 lower of the cow exhibiting bimodal milk flow. Compared to primiparous cows, multiparous cows tended to have an OR of 0.68 for displaying bimodal milk flow. Maintaining all other variables constant, for each additional second of stimulation time, the OR of exhibiting bimodal milk flow was 0.98.

Overmilking individual risk factors

Milking with automatic cluster removal (P < 0.001), parity (P < 0.002), and unit reattachment after the automatic cluster removal being activated (P = 0.006) were all associated with overmilking time (Table 6). Cows milked with automatic cluster removal had an average of 64.42 seconds in overmilking whereas cows with manual cluster removal had an average of 222.96 seconds in overmilking. Compared to multiparous cows (132.67 seconds), primiparous cows were subjected to less overmilking time (108.24 seconds). In cows which units were reattached after automatic cluster removal activation (162.47 seconds), overmilking time increased compared to non-reattached cows (88.40 seconds).

Milk yield-associated factors

Milk yield in the recorded milking (kg) was associated with DIM (P < 0.001), parity (P < 0.001), and the interaction between DIM and parity (P = 0.017) (Table 7). Cows with > 201 DIM (12.85 kg) produced less milk than cows between 101 and 200 DIM (15.18 kg), and both produced less milk than cows with less than 101 DIM (16.14 kg). Multiparous cows produced more milk (15.79 kg) than primiparous cows (13.65 kg), but that increment was 0.86 kg lower in cows between 101 and 200 DIM, and 2.14 kg lower after 200 DIM. Bimodal milk flow was associated with a decrease in milk

Table 4
Multivariable analysis model of median overmilking time (s) for 21 dairy cattle farms enrolled in the study.

Parameter	Estimate	SE	*P*-value	LSM	LSM 95% CI
Intersect	96.69	13.66	<0.001	–	–
Cluster removal milk flow threshold (g/min)	−0.090	0.028	0.005	–	–
Obstructed air bleed holes	–	–	0.034	–	–
No (referent)	–	–	–	55.50	44.32–66.69
Yes	37.73	16.27	0.034	93.24	60.61–125.87

Abbreviations: LSM = Least square means; CI = Confidence interval.

Table 5
Multivariable analysis model of bimodal milk flow for 606 cows enrolled in the study.

Parameter	Estimate	SE	*P*-value	Overall *P*-value	OR	OR (95% CI)
DIM	–	–	–	<0.001	–	–
1–100 (referent)	–	–	–	–	–	–
101–200	0.58	0.31	0.060	-	1.78	0.97–3.25
>201	1.12	0.30	<0.001	-	3.07	1.70–5.50
Parity	–	–	–	0.090	–	–
Primiparous (referent)	–	–	–	–	–	–
Multiparous	−0.39	0.23	0.090	-	0.68	0.43–1.06
Preparation lag time (s)	−0.009	0.003	0.001	0.001	0.99	0.98–1.00
Stimulation time (s)	−0.025	0.013	0.050	0.054	0.98	0.95–1.00

Abbreviations: DIM = Days in milk; OR = Odds ratio; CI = Confidence interval.

Table 6
Multivariable analysis model of individual overmilking time (s) for 606 cows enrolled in the study.

Parameter	Estimate	SE	*P*-value	Overall *P*-value	LSM[1]	LSM 95% CI[1]
Intersect	3.76	0.11	<0.001	–	–	–
Automatic cluster removal	–	–	–	<0.001	–	–
Yes (referent)	–	–	–	–	64.42	48.17–86.14
No	1.24	0.15	<0.001	-	222.96	150.60–330.07
Parity	–	–	–	0.002	–	–
Primiparous (referent)	–	–	–	–	108.24	78.66–148.96
Multiparous	0.20	0.06	0.002	–	132.67	96.59–182.23
Unit reattached	–	–	–	0.006	–	–
No (referent)	–	–	–	–	88.40	69.65–112.19
Yes	0.61	0.22	0.006	–	162.47	100.13–263.62

Abbreviations: LSM = Least square means; CI = Confidence interval.
[1] Least square means and 95% CI back transformed from the original estimates due to the natural log-transformation of overmilking time data.

Table 7
Multivariable analysis model of milk yield (kg) in the recorded milking for 606 cows enrolled in the study.

Parameter	Estimate	SE	*P*-value	Overall *P*-value	LSM	LSM 95% CI
Intersect	15.51	0.70	<0.001	–	–	–
Bimodal milk flow	–	–	–	<0.001	–	–
No (referent)	–	–	–	–	15.66	14.55–16.77
Yes	−1.88	0.34	<0.001	–	12.64	12.64–14.92
DIM	–	–	–	<0.001	–	–
1–100 (referent)	–	–	–	–	16.14	14.95–17.32
101–200	−0.53	0.66	0.420	–	15.18	14.03–16.33
>201	−2.22	0.61	<0.001	–	12.85	11.72–13.97
Parity	–	–	–	<0.001	–	–
Primiparous (referent)	–	–	–	–	13.65	12.51–14.79
Multiparous	3.14	0.62	<0.001	-	15.79	14.69–16.89
Parity × DIM	–	–	–	0.017	–	–
Primiparous × 1–100	–	–	–	–	–	–
Primiparous × 101–200	–	–	–	–	–	–
Primiparous × >200	–	–	–	–	–	–
Multiparous × 1–100	–	–	–	–	–	–
Multiparous × 101–200	−0.86	0.82	0.290	–	–	–
Multiparous × >200	−2.14	0.77	0.006	–	–	–

Abbreviations: DIM = Days in milk; LSM = Least square means; CI = Confidence interval.

yield ($P < 0.001$) as cows with bimodal milk flow produced on average 12.64 kg compared to the 15.66 kg produced by cows with non-bimodal milk flow.

Discussion

The percentage of cows with bimodal milk flow in the present study was 41.7%, a higher value than reported in other studies (Dodenhoff et al., 1999; Sandrucci et al., 2007; Samoré et al., 2011; Moore-Foster et al., 2019a), despite some variation in case definition. Moore-Foster et al. (2019a), using the VaDia logger, considered bimodal milking when cows had alveolar milk ejection over 30 seconds after milking unit attachment, a situation that was followed 97% of the times by a graphic record of bimodal milk flow. Applying our case definition, the Michigan study would have a proportion of bimodal milk flow of 24.3%. Other researchers, using a Lactocorder (WMB, Balgach, Switzerland), another recording equipment, reported 35.1% (Sandrucci et al., 2007), 33.8% (Samoré et al., 2011) and 22% (Dodenhoff et al., 1999) of bimodal milk flow.

The observed higher OR of bimodal milk flow at cow level occurring with increasing DIM ($P < 0.001$) is in agreement with what was found both in observational studies (Sandrucci et al., 2007; Samoré et al., 2011) and experimental trials (Bruckmaier and Hilger, 2001; Weiss and Bruckmaier, 2005). The degree of udder filling decreases after peak production (Bruckmaier and Hilger, 2001), due to a lower milk secretion rate with increasing DIM (Knight et al., 1994; Pfeilsticker et al., 1996), which probably results in a need for a stronger and more complete myoepithelial contraction to transfer the milk of the alveoli with an increase of the time needed for milk ejection (Weiss and Bruckmaier 2005). Also, there is a lower milk transfer from the alveolar compartment to the gland cistern between milkings, resulting in a lower quantity of milk being present in the cistern and large ducts in the moments before milking (Pfeilsticker et al., 1996; Ayadi et al., 2003).

In the present study, there was a decreased probability of bimodal milk flow occurring when the preparation lag time between the beginning of stimulation and unit attachment increased ($P = 0.001$). Blood concentration of oxytocin significantly increases around 30 seconds after initial stimulation, with the increase of intramammary cistern pressure occurring 50–120 seconds after initial stimulation, depending on the amount of milk present in the alveolar compartment (Bruckmaier and Hilger, 2001; Weiss and Bruckmaier, 2005). Therefore, a time interval after initial stimulation is necessary to allow for the transfer of alveolar milk to occur before milking unit attachment.

Field trials, in which several combinations of stimulation and preparation lag times were used, report a decrease in bimodality with increasing preparation lag time from 0 to 60 seconds (Rasmussen et al., 1992; Kaskous and Bruckmaier, 2011; Watters et al., 2012), with that decrease being more evident in later stages of lactation (Kaskous and Bruckmaier, 2011; Watters et al., 2012). Watters et al. (2012) also report that cows in later stages of lactation would benefit from a preparation lag time increased to 90–120 seconds. An even longer increase in preparation lag time to 240 seconds resulted in a significant increase in bimodality across any lactational stage. In the present study, 457 (75.4%) out of 606 cows had a preparation lag time between 0 and 120 seconds and only 3.5% had a preparation lag time of 240 seconds or higher, which may have undervalued the effect of a longer preparation lag time in our model. Also, 102 (68.5%) out of 149 cows with a preparation lag time longer than 120 seconds were evaluated in farms where the stimulation procedures were split in the milking routine.

An excessively long preparation lag time may determine the reflux of milk from the cistern back to the alveolar compartment (Caja et al., 2004). Those authors recorded this observation 15 minutes after oxytocin injection, whereas in the present work, the longest preparation lag time recorded was 342 seconds (slightly under 6 minutes).

The increase in total stimulation time in the present study tended to be associated with a decrease in the probability of bimodal milking occurring at individual cow level ($P = 0.054$) and at farm level ($P = 0.020$), similar to other reported observations (Moore-Foster et al., 2019a). Wieland et al. (2020b) while comparing two milking routines with different stimulation procedures, one wiping the teats with a towel cloth and another with a combination of forestripping and wiping also found a lower incidence of bimodality in cows with a longer stimulation time, in their case for the routine that also included forestripping. Watters et al. (2012) on the other hand, while also comparing milking routines with and without forestripping, only found a significant reduction in bimodality in cows between 174 and 428 DIM. Tactile stimulation of the teats leads to oxytocin release from the posterior pituitary lobe (Bruckmaier and Blum, 1996). The association of increased stimulation time with bimodal milk flow may be the effect of a higher concentration of oxytocin, which leads to a stronger contraction or contraction of a higher number of myoepithelial cells, as hypothesised by Wieland et al. (2020b). However, Kaskous and Bruckmaier (2011) did not find significant differences in oxytocin concentration in animals subject to 15, 30 or 45-second stimulation during milking routine. In the present study, 424 (70%) of cows were stimulated for less than 15 seconds, which suggests that the increase in stimulation time between 0 and 15 seconds may be more important than in the intervals considered by Kaskous and Bruckmaier (2011).

We also found a decrease in the probability of a farm having a high proportion of bimodal cows with an increase in the number of passes during udder preparation. All farms that performed four passes and three (30%) of the farms that performed three passes during the premilking routine had a complete preparation, including forestripping. Alternatively, none of the farms that performed one or two passes during the milking routine performed a complete udder preparation, which might explain the observed difference. There was a correlation between the number of passes and mean preparation lag time (Spearman correlation coefficient = 0.6, $P = 0.004$), which indicates that farms that performed milking procedures in different passes had a higher preparation lag time. The effect of a longer preparation lag time may explain the recorded decreased bimodality. Moore-Foster et al. (2019a) reported an increase in the proportion of animals with a delayed milk letdown with an increase in the number of passes, an association that was deemed paradoxical by the authors that speculated that farms in the study with fewer passages could however have a higher stimulation during the first pass.

Among the 606 cows with a vacuum graphic recording, median time in overmilking was 59 seconds, with 77.1% of the animals undergoing at least 30 seconds in overmilking. These numbers are higher than those reported by Moore-Foster et al. (2019a), who reported a median overmilking time of 47 seconds and 55% of evaluated cows being overmilked at least 30 seconds.

There was an association between overmilking time and parity ($P < 0.001$) with primiparous animals showing significantly less overmilking. There are reports that the decline phase of milk flow in primiparous animals is significantly shorter than in multiparous animals (Tančin et al., 2006; Sandrucci et al., 2007). This means that during the first lactation, milk flow reaches a value of 300 g/min faster, which might be due to the maximum flow and milk production being lower (Sandrucci et al., 2007). Therefore, the reduction in overmilking found in the present study may have occurred because these animals reached the milk flow threshold (445 g/min average) that activated the automatic cluster remover sooner.

The association found between reattachment of the milking unit and increased overmilking time (P = 0.006) has not been reported in previous publications. Moore-Foster et al. (2019b) reports reattachment as a potential risk factor for overmilking but without having found any association between the proportion of reattachments on a farm and median time in overmilking. This might have been due to the low number of reattachments on the farms, and to the fact that animals to which milking units were reattached being outliers, so they were not reflected in the median overmilking time. When the milking unit is reattached, the milking system does not activate the automatic cluster remover for a predetermined time, independently of milk flow. Among the milked animals in automatic mode not subject to reattachment, 355 (73%) out of 490 were at least 30 seconds in overmilking and only three (0.6%) were not overmilked for a single second. This indicates that on these farms, automatic cluster remover is activated when the animals are already being overmilked. The forced vacuum time after a reattachment will therefore lead to further overmilking. Milking unit reattachment is an action taken by the milker, implying a decision that milking has not occurred in a complete fashion. The completeness of milking is generally ascertained by observation, palpation and manual milking of the udder. The present study shows those criteria to be subjective and inadequate, leading to overmilking.

Use of manual mode for cluster removal was associated with marked overmilking (P < 0.001). This finding agrees with what was found in an observational study in which researchers found a relation between use of manual mode and increase in total unit-on time as well as time with milk flow under 1 kg/min (Wieland et al., 2017). In manual mode, the end of milking is determined by the milker either because the milking parlour does not have automatic cluster removers or because the milker decides to use this mode. With the manual mode, milkers evaluate milk flow in the claw periodically and decide when to remove the milking unit. The increase in overmilking probably occurs due to some degree of subjectivity and imprecision of the evaluation made by the milker, sometimes aided by the belief that a complete milking reduces the risk of mastitis occurring (Wieland et al., 2020b). Several studies have demonstrated, however, that there is no association between milk present in the udder at the end of milking and incidence of clinical or subclinical mastitis (Rasmussen, 1993; Clarke et al., 2008; Wieland et al., 2020b). Additionally, in milking parlours without automatic cluster removers, the extra task of removing the units, in addition to the multiple jobs performed by the milkers (moving cows in and out of the parlour, premilking routine, etc), may further compound the occurrence of overmilking, as they only remove the units after concluding an ongoing task.

At farm level, there was a significant increase in median overmilking time whenever air bleed holes were obstructed (P = 0.034), an association not previously reported. The main task of air bleed holes in the clawpiece is to speed up the milk outflow of milk from the claw to the tubes of the milking system, and to reduce the density of the milk and air mixture stream which flows through the milk line (Wiercioch et al., 2016). Standard 5707:2007 (ISO, 2007) determines that to guarantee vacuum stability and an adequate milk flow, the claw should allow for 4–12 litres of air to enter per minute. Clogging of the air entrance at the cluster leads to an increase in cyclic vacuum fluctuations of around 7 kPa and an increase of mean claw vacuum level of around 1.2 kPa (Thiel et al., 1964; Rasmussen et al., 2006). Vacuum instability due to excessive increase in vacuum cyclic variations determines a decrease in the average milk flow, consequently reducing milking efficiency (Schmidt et al., 1964). Therefore, the increase in overmilking resulting from the presence of obstructed air bleed holes may have been the result of a deficient milk transport through the milking system and of vacuum instability. The slowing down of the milk flow may prolong the time necessary to reach the threshold for automatic cluster removal, and consequently increase the overmilking time.

Finally, there was an association between the increase in the farm cluster removal milk flow threshold and a decrease in median time in overmilking (P = 0.005). Automatic cluster removers are not activated during a set minimum period, allowing them not to be activated while the milk flow is in the increase phase, whereas the detaching flow and postmilking time are selected so that the cluster remover is activated in the decline phase (Stewart et al., 2002; Tančin et al., 2006; Krawczel et al., 2017). The postmilking vacuum time corresponds to the time interval for which milk flow should be below the detaching threshold, for the unit to be removed (Stewart et al., 2002). Different controlled randomised trials comparing two or three cluster removal milk flow thresholds report findings that support the conclusions of the present study: all the trials found a significant reduction in individual milking time with an increasing cluster removal milk flow threshold (Rasmussen, 1993; Stewart et al., 2002; Magliaro and Kensinger, 2005; Jago et al., 2010; Edwards et al., 2013a and 2013b; Besier and Bruckmaier, 2016; Krawczel et al., 2017; Wieland et al., 2020b). Apart from two such studies that reported a significant reduction in milk yield (Magliaro and Kensinger, 2005; Jago et al., 2010) and from one study that reported an increase in milk yield in two out of five farms included in the trial (Stewart et al., 2002), all the others observed no significant differences in milk production in cows subjected to higher cluster removal milk flow thresholds (Rasmussen, 1993; Edwards et al., 2013a and 2013b; Wieland et al., 2020b). Decreasing individual milking time, without decreasing milk yield as observed by most authors, suggests a reduction in overmilking. It is also clear from the aforementioned publications, there is a tendency for the dairy industry to adopt progressively higher cluster removal milk flow thresholds, reinforced by several recent publications using cluster removal milk flow thresholds of 1100 g/min (Erskine et al., 2019) or even 1300 g/min (Wieland et al., 2017; Melvin et al., 2019). In the 19 farms with automatic cluster removers included in the present study, mean and median removal flows were 445 and 400 g/min, respectively, well below what is mentioned, which could help explain the high level of overmilking observed, presenting one of the main opportunities for improvement. There is no standard from the International Standardisation Organisation that defines how to adjust the forced vacuum time and the cluster removal milk flow thresholds. Some publications suggest gradual adjustments, around 100 g/min or 2 seconds for forced vacuum time every-two weeks, allowing for animals to adapt (Stewart et al., 2002; Rasmussen, 2004).

The current study observed a decrease in milk yield associated with bimodal milk flow (P < 0.001) an observation also recently reported by Erskine et al. (2019). The milk loss associated with bimodal milk flow may be the result of vacuum increase at the mouthpiece chamber at the beginning of milking of these animals (Erskine et al., 2019). The absence or reduction of milk flow after the removal of cisternal milk promotes the collapse of the teat wall inside the teat liner (Mein et al., 1973) with consequent increase of the mouthpiece chamber vacuum (Borkhus and Rønningen, 2003). Reduction of the friction forces between the wall of the teat and the body of the teat liner, as well as the increase in the mouthpiece chamber vacuum, lead to the penetration of the tissue at the base of the teat into the mouthpiece, with consequent constriction and possible partial occlusion of the communication between the gland cistern and the teat cistern (Mein et al., 1973; Borkhus and

Rønningen, 2003). In a complementary way, the rise in mouthpiece chamber vacuum leads to teat congestion and reduction of the teat canal cross-sectional area (Penry et al., 2017), which in turn results in a decrease in milk flow (Williams et al., 1981). The combined effects of the repositioning of the milking unit and the teat congestion, induced by the absence of milk flow, may justify the decrease in milk yield in bimodal milking (Erskine et al., 2019). In the current study, just like in the study by Erskine et al. (2019), bimodality was only assessed at a single milking, which does not allow clarification if daily milk yield is affected. As expected, our model showed a decrease in milk yield in primiparous cows and along lactation, with a significant interaction between lactation and parity reflecting the higher persistency of the lactation curve in first lactations.

The average milk flow translates all four milk flow phases, allowing to evaluate the suitability of premilking protocols and milking system settings (Stewart et al., 2001).

The increase in overmilking was associated with a decrease in the average milk flow ($P < 0.001$). The overmilking period occurs after the peak flow period, at the end of milking, corresponding to a period of low milk flow (Tančin et al. 2006). Therefore, prolonging overmilking time increases total unit-on time (Moore-Foster et al., 2019b) without proportionally increasing milk yield, and consequently reducing average milk flow. Wieland et al. (2020b), Edwards et al. (2013a and 2013b) and Stewart et al. (2002) all reported an increase in average milk flow when cluster removal milk flow thresholds were increased. The three research groups attributed this observation to an earlier termination of milking, in the low flow phase, shortening the less efficient phase of milk flow, and increasing the relative duration of the peak milk flow phase in the total milking time; these conclusions agreed with our findings.

An association between the occurrence of bimodal milk flow and reduction of the average milk flow was also found ($P < 0.001$), which is consistent with results from multiple trials (Weiss and Bruckmaier, 2005; Kaskous and Bruckmaier, 2011; Watters et al., 2012), although the way bimodality negatively influences the average milk flow differs between studies. Results of this study point to an increase in the average milk flow through the reduction of the time necessary to milk the same amount of milk, because there is a greater amount of milk harvested at the beginning of milking and there is no period with low or absent milk flow between removal of cisternal milk and let-down of alveolar milk. Erskine et al. (2019), however, did not find any association between bimodality and unit-on time, but did find that bimodality was associated with a decrease in milk yield. These results point to a reduction in the average milk flow through the decrease in milk yield for the same milking time. The answer is probably a combination between reduction in milk yield and total milking time increase.

We also report a reduction of the average milk flow as lactation proceeds in multiparous cows, which is due to the decrease in milk production and shortening of the peak flow phase. Contrarily, primiparous cows showed an increase in the average milk flow along lactation, these findings reflecting the adaptation of primiparous cows to a new environment as the first lactation begins, and the persistency of the lactation curve of these cows. Parity also significantly influenced the average milk flow as multiparous cows showed a higher average milk flow, this finding being justified by the higher milk production and maximum milk flow of these cows, which are the main determinants of the average milk flow (Tančin et al. 2006).

Regarding short-term teat changes, a mean of 78.4% of the cows per farm had such changes in at least one teat. The farm with the lowest prevalence of such changes showed 56.2% of teats with at least one change detectable immediately after milking. Teat Club International proposes a threshold of 20% of evaluated cows (Mein et al., 2001) with changes in firmness, colour or presence of swelling near the base of the teat, to determine the need for intervention in a herd, rendering every participating farm in the present study in need of corrections. According to Mein et al. (2001), an excessive number of teats displaying this type of changes results from faults in milking machines or milking management, resulting in overmilking or long periods with a milk flow under 1 kg/min. The vacuum level in the clawpiece and pulsation characteristics were evaluated on every farm of the present study, with all the farms successfully fulfilling the requirements of Standard 5707:2007 (ISO, 2007). Likewise, the vacuum level in the clawpiece at peak flow, recorded in the 21 farms, fulfilled the requirements of both ISO (2007) and NMC (2012). Therefore, the main faults in milking machines were excluded as potential causes of short-term changes in teat condition (Hamann et al., 1993; Hamann and Mein, 1996). Thus, it is fair to assume that long periods of overmilking or milk flow under 1 kg/min in the farms evaluated were playing an important role in the high prevalence of congestive changes to the teats, as evidenced by a moderate positive correlation between farms' median overmilking time and proportion of cows exhibiting short-term teat changes. Overmilking and bimodal milking have been associated with prolonging total milking time (Moore-Foster et al., 2019b; Wieland et al., 2020a), time with milk flow under 1 kg/min (Wieland et al., 2020a; 2020b), and therefore prolonging and intensifying the mechanical forces produced by vacuum (Bade et al., 2009; Besier and Bruckmaier, 2016). Wieland et al. (2020b) observed a decrease in the probability of short-term changes appearing in animals stimulated for 16 seconds during premilking routine, as opposed to animals stimulated for 7 seconds. These differences could have been the result of the significant reduction in the proportion of bimodality in animals stimulated for 16 seconds. A total of 10 (48%) of the 21 farms enrolled in the current study presented a proportion of bimodality above 40%. Considering that in the study by Wieland et al. (2020b), 41.5% of the cows stimulated for 6–7 seconds showed bimodal milk flow, it can be speculated that the observed bimodality in the present study is playing an important role in triggering the short-term teat changes.

Regarding teat-end hyperkeratosis, a mean of 33.6% of evaluated cows per farm presented at least one teat with a score of 3 or 4. Every farm had a prevalence of hyperkeratosis above 20%, the threshold beyond which Teat Club International (Mein et al., 2001) considers hyperkeratosis as unacceptable. Total milking time (Neijenhuis et al., 2000) and overmilking time (Edwards et al., 2013c) have been associated with the presence of high levels of hyperkeratosis. Rasmussen (1993) observed a reduction in the levels of hyperkeratosis with an increase in cluster removal milk flow thresholds from 200 to 400 g/min. Mein et al. (2001) mention that whenever there is a high prevalence of short-term teat changes, presence of hyperkeratosis is generally the result of long periods of milk flow below 1 kg/min. The high levels of bimodality and overmilking observed in the current study are therefore likely to be playing a major role in deteriorating teat health. Congestive changes to the teats have been linked to increased odds of microbial colonisation of the teat canal (Zecconi et al., 1992) and intramammary infection (Zecconi et al., 1996). Teat congestion leads to pain and uneasiness, which compromise animal welfare during milking (Hillerton et al., 2002). Although somatic cell counts were not evaluated in the present study, the presence of high levels of hyperkeratosis has also been linked to deteriorating udder health (Neijenhuis et al., 2001) and discomfort during milking (Cerqueira et al., 2018). Therefore, it is important to implement corrective measures on farms aiming to minimise such changes, to guarantee good milk quality and animal welfare during milking, while striving to achieve milking efficiency.

Conclusion

Out of 606 cows evaluated in 21 farms, bimodality was observed in 41.7% of milkings per farm and 78.3% of the cows on average in each herd were overmilked for over 30 seconds. Bimodal milking seemed mostly to be linked to insufficient stimulation and preparation lag times. Occurrence of overmilking was mostly linked to low cluster removal milk flow thresholds, milking in manual mode, unit reattachment, lower parity and presence of clogged air bleed holes. There are opportunities to improve milking efficiency, teat health and animal welfare during milking through measures aiming at reducing the occurrence of bimodal milking and overmilking.

Supplementary material

Supplementary material to this article can be found online at https://doi.org/10.1016/j.animal.2023.100716.

Ethics approval

The study was not submitted to the Faculty's Ethics and Welfare Committee because it derives from milk quality consultancy practice and all the procedures were within the current practice for trouble shooting milking routine and parlour problems. Informed consent was obtained from the owners of the farms studied.

Data and model availability statement

None of the data were deposited in an official repository. The data that support the study findings are available from the authors upon reasonable request.

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Author contributions

SF and **RB** contributed to the conception and design of the study. **SF** contributed to data collection. **SF**, **GP** and **RB** performed statistical analysis. **SF**, **GP** and **RB** drafted the manuscript. All authors read and approved the final manuscript.

Declaration of interest

The authors have not stated any conflicts of interest.

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References

Ayadi, M., Caja, G., Such, X., Knight, C.H., 2003. Use of ultrasonography to estimate cistern size and milk storage at different milking intervals in the udder of dairy cows. Journal of Dairy Research 70, 1–7.
Bade, R.D., Reinemann, D.J., Zucali, M., Ruegg, P.L., Thompson, P.D., 2009. Interactions of vacuum, b-phase duration, and liner compression on milk flow rates in dairy cows. Journal of Dairy Science 92, 913–921.
Besier, J., Bruckmaier, R.M., 2016. Vacuum levels and milk-flow-dependent vacuum drops affect machine milking performance and teat condition in dairy cows. Journal of Dairy Science 99, 3096–3102.
Besier, J., Lind, O., Bruckmaier, R.M., 2016. Dynamics of teat-end vacuum during machine milking : types, causes and impacts on teat condition and udder health – a literature review. Journal of Applied Animal Research 44, 263–272.
Biocontrol, 2011. VaDia Suite User Manual v3.7. Rakkestad, Norway. Retrieved on 20 Oct 2020. http://www.biocontrol.no/down/VaDiaUserManual.pdf.
Borkhus, M., Rønningen, O., 2003. Factors affecting mouthpiece chamber vacuum in machine milking. Journal of Dairy Research 70, 283–288.
Bruckmaier, R.M., Schams, D., Blum, J.W., 1993. Milk removal in familiar and unfamiliar surroundings: concentrations of oxytocin, prolactin, cortisol and B-endorphin. Journal of Dairy Research 60, 449–456.
Bruckmaier, R.M., Blum, J.W., 1996. Simultaneous recording of oxytocin release, milk ejection and milk flow during milking of dairy cows with and without prestimulation. Journal of Dairy Research 63, 201–208.
Bruckmaier, R.M., Blum, J.W., 1998. Oxytocin Release and Milk Removal in Ruminants. Journal of Dairy Science 81, 939–949.
Bruckmaier, R.M., Hilger, M., 2001. Milk ejection in dairy cows at different degrees of udder filling. Journal of Dairy Research 68, 369–376.
Caja, G., Ayadi, M., Knight, C.H., 2004. Changes in Cisternal Compartment Based on Stage of Lactation and Time Since Milk Ejection in the Udder of Dairy Cows. Journal of Dairy Science 87, 2409–2415.
Cerqueira, J.L., Araújo, J.P., Cantalapiedra, J., 2018. How is the association of teat-end severe hyperkeratosis on udder health and dairy cow behavior? Revue de Médecine Vétérinaire 169, 30–37.
Chang, W., Barry, M.C., Jones, L.R., Merrill, W.G., 1992. Compares: A Computerized Milking Parlor Evaluation System. Journal of Dairy Science 75, 2578–2586.
Clarke, T., Cuthbertson, E.M., Greenall, R.K., Hannah, M.C., Shoesmith, D., 2008. Incomplete milking has no detectable effect on somatic cell count but increased cell count appears to increase strip yield. Australian Journal of Experimental Agriculture 48, 1161–1167.
Dodenhoff, J., Sprengel, D., Duda, J., Dempfle, L., 1999. Studies on genetic evaluation of udder health using the Lacto Corder. Zuchtungskunde 71, 459–472.
Edwards, J.P., Jago, J.G., Lopez-Villalobos, N., 2013a. Milking efficiency for grazing dairy cows can be improved by increasing automatic cluster remover thresholds without applying premilking stimulation. Journal of Dairy Science 96, 3766–3773.
Edwards, J.P., Jago, J.G., Lopez-Villalobos, N., 2013b. Short-term application of prestimulation and increased automatic cluster remover threshold affect milking characteristics of grazing dairy cows in late lactation. Journal of Dairy Science 96, 1886–1893.
Edwards, J.P., Brien, B.O., Lopez-Villalobos, N., Jago, J.G., 2013c. Overmilking causes deterioration in teat-end condition of dairy cows in late lactation. Journal of Dairy Research 80, 344–348.
Erskine, R.J., Norby, B., Neuder, L.M., Thomson, R.S., 2019. Decreased milk yield is associated with delayed milk ejection. Journal of Dairy Science 102, 6477–6484.
Hamann, J., Mein, G.A., 1996. Teat thickness changes may provide biological test for effective pulsation. Journal of Dairy Research 63, 179–189.
Hamann, J., Mein, G.A., Wetzel, S., 1993. Teat Tissue Reactions to Milking : Effects of Vacuum Level. Journal of Dairy Science 76, 1040–1046.
Hansen, M.N., 1999. Optimal Number of Clusters per Milker. Journal of Agricultural Engineering Research 72, 341–346.
Heinze, G., Dunkler, D., 2017. Five myths about variable selection. Transplant International 30, 6–10.
Hillerton, J.E., Pankey, J.W., Pankey, P., 2002. Effect of over-milking on teat condition. Journal of Dairy Research 69, 81–84.
ISO (International Organization for Standardization), 2007. Milking machine installations: Construction and performance. Standard 5707:2007(E). 3rd ed. ISO, Geneva, Switzerland.
Jago, J.G., Burke, J.L., Williamson, J.H., 2010. Effect of automatic cluster remover settings on production, udder health, and milking duration. Journal of Dairy Science 93, 2541–2549.
Kaskous, S., Bruckmaier, R.M., 2011. Best combination of pre-stimulation and latency period duration before cluster attachment for efficient oxytocin release and milk ejection in cows with low to high udder-filling levels. Journal of Dairy Research 78, 97–104.
Knight, C.H., Hirst, D., Dewhurstf, R.J., 1994. Milk accumulation and distribution in the bovine udder during the interval between milkings. Journal of Dairy Research 61, 167–177.
Krawczel, P., Ferneborg, S., Wiking, L., Dalsgaard, T.K., Gregersen, S., Black, R., Larsen, T., Agenäs, S., Svennersten-Sjaunja, K., Ternman, E., 2017. Milking time and risk of over-milking can be decreased with early teat cup removal based on udder quarter milk flow without loss in milk yield. Journal of Dairy Science 100, 6640–6647.

Magliaro, A.L., Kensinger, R.S., 2005. Automatic cluster remover setting affects milk yield and machine-on time in dairy cows. Journal of Dairy Science 88, 148–153.
Mein, G.A., Thiel, C.C., Westgarth, D.R., Fulford, R.J., 1973. Friction between the teat and teatcup liner during milking. Journal of Dairy Research 40, 191–206.
Mein, G.A., Neijenhuis, F., Morgan, W.F., Reinemann, D.J., Hillerton, J.E., Baines, J.R., Ohnstad, I., Rasmussen, M.D., Timms, L.L., Britt, J.S., Farnsworth, R., Cook, N., Hemling, T., 2001. Evaluation of bovine teat condition in commercial dairy herds: 1. non-infectious factors. Proceedings of the 2nd International Symposium of Mastitis and Milk Quality, 13–15 September 2001, Vancouver, BC, Canada, pp. 347–351.
Melvin, J.M., Heuwieser, W., Virkler, P.D., Nydam, D.V., Wieland, M., 2019. Machine milking–induced changes in teat canal dimensions as assessed by ultrasonography. Journal of Dairy Science 102, 2657–2669.
Moore-Foster, R., Norby, B., Schewe, R.L., Thomson, R., Bartlett, P.C., Erskine, R.J., 2019a. Herd-level variables associated with delayed milk ejection in Michigan dairy herds. Journal of Dairy Science 102, 696–705.
Moore-Foster, R., Norby, B., Schewe, R.L., Thomson, R., Bartlett, P.C., Erskine, R.J., 2019b. Short communication: Herd-level variables associated with overmilking in Michigan dairy herds. Journal of Dairy Science 102, 8400–8404.
Moore-Foster, R., Norby, B., Schewe, R.L., Thomson, R., Bartlett, P.C., Erskine, R.J., 2019c. Herd-level variables associated with premilking stimulation time in Michigan dairy herds. Journal of Dairy Science 102, 2544–2550.
Neijenhuis, F., Barkema, H.W., Hogeveen, H., Noordhuizen, J.P.T.M., 2000. Classification and longitudinal examination of callused teat ends in dairy cows. Journal of Dairy Science 83, 2795–2804.
Neijenhuis, F., Barkema, H.W., Hogeveen, H., Noordhuizen, J.P.T.M., 2001. Relationship between teat-end callosity and occurrence of clinical mastitis. Journal of Dairy Science 84, 2664–2672.
Nickerson, S.C., Akers, R.M., 2011. Mammary Gland: Anatomy. In: Fuquay, J.W., Fox, P.F., McSweeney, P.L.H. (Eds.), Encyclopedia of Dairy Sciences. 2nd ed. Academic Press Inc., London, UK, pp. 328–337.
NMC (National Mastitis Council), 2012. Procedures for Evaluating Vacuum Levels and Air Flow in Milking Systems. National Mastitis Council, Verona, WI, USA.
NMC, 2013. Recommended milking procedures. Retrieved on 20 Oct 2020. http://www.nmconline.org/wp-content/uploads/2016/09/Recommended-Milking-Procedures.pdf.
NRC, 2001. Nutrient Requirements of Dairy Cattle. National Academies Press, Washington, DC, USA.
Penry, J.F., Upton, J., Mein, G.A., Rasmussen, M.D., Ohnstad, I., Thompson, P.D., Reinemann, D.J., 2017. Estimating teat canal cross-sectional area to determine the effects of teat-end and mouthpiece chamber vacuum on teat congestion. Journal of Dairy Science 100, 821–827.
Pfeilsticker, H.U., Bruckmaier, R.M., Blum, J.W., 1996. Cisternal milk in the dairy cow during lactation and after preceding teat stimulation. Journal of Dairy Research 63, 509–515.
Rasmussen, M.D., Frimer, E.S., Decker, E.L., 1994. Reverse Pressure Gradients Across the Teat Canal Related to Machine Milking. Journal of Dairy Science 77, 984–993.
Rasmussen, M.D., 1993. Influence of Switch Level of Automatic Cluster Removers on Milking Performance and Udder Health. Journal of Dairy Research 60, 287–297.
Rasmussen, M.D., 2004. Overmilking and teat condition. Proceedings of the 43rd Annual Meeting of the National Mastitis Council, 1–4 February 2004, Charlotte, NC, USA, pp. 169–175.
Rasmussen, M.D., Frimer, E.S., Galton, D.M., Petersson, L.G., 1992. The Influence of Premilking Teat Preparation and Attachment Delay on Milk Yield and Milking Performance. Journal of Dairy Science 75, 2131–2141.
Rasmussen, M.D., Wiking, L., Bjerring, M., Larsen, H.C., 2006. Influence of air intake on the concentration of free fatty acids and vacuum fluctuations during automatic milking. Journal of Dairy Science 89, 4596–4605.
Reinemann, D.J., Rasmussen, M.D., LeMire, S., Neijenhuis, F., Mein, G.A., Hillerton, J.E., Morgan, W.F., Timms, L., Cook, N., Farnsworth, R., Baines, J.R., Hemling, T., 2001. Evaluation of bovine teat condition in commercial dairy herds: 3. getting the numbers right. Proceedings of the 2nd International Symposium of Mastitis and Milk Quality, 13–15 September 2001, Vancouver, BC, Canada, pp. 357–361.
Samoré, A.B., Román-Ponce, S.I., Vacirca, F., Frigo, E., Canavesi, F., Bagnato, A., Maltecca, C., 2011. Bimodality and the genetics of milk flow traits in the Italian Holstein-Friesian breed. Journal of Dairy Science 94, 4081–4089.
Sandrucci, A., Tamburini, A., Bava, L., Zucali, M., 2007. Factors Affecting Milk Flow Traits in Dairy Cows: Results of a Field Study. Journal of Dairy Science 90, 1159–1167.
Schmidt, G.H., Switzer, K.O., Guest, R.W., Guthrie, R.S., 1964. Effect of Teat-End Vacuum Fluctuations on Milking Rate and Mastitis. Journal of Dairy Science 47, 761–765.
Stewart, S., Eicker, S., Rapnicki, P., 2001. Automated collection of parlor performance data: information needed and proposed standardized definitions. Proceedings of the 2nd International Symposium of Mastitis and Milk Quality, 13–15 September 2001, Vancouver, BC, Canada, pp. 377–381.
Stewart, S., Godden, S., Rapnicki, P., Reid, D., Johnson, A., Eicker, S., 2002. Effects of automatic cluster remover settings on average milking duration, milk flow, and milk yield. Journal of Dairy Science 85, 818–823.
Tančin, V., Ipema, B., Hogewerf, P., Mačuhová, J., 2006. Sources of variation in milk flow characteristics at udder and quarter levels. Journal of Dairy Science 89, 978–988.
Thiel, C.C., Clough, P.A., Akam, D.N., 1964. Mechanics of machine milking: I. Pressures in the teatcup assembly and liner wall movement. Journal of Dairy Research 31, 303–313.
USDA, 2020. Farm Labor. Labor Cost Share Total Gross Revenues. Retrieved on 20 Oct 2020. https://www.ers.usda.gov/topics/farm-economy/farm-labor/#employment.
Vierbauch, T., Peinhopf-Petz, W., Wittek, T., 2021. Effects of milking, over-milking and vacuum levels on front and rear quarter teats in dairy cows. Journal of Dairy Research 88, 396–400.
Watters, R.D., Schuring, N., Erb, H.N., Schukken, Y.H., Galton, D.M., 2012. The effect of premilking udder preparation on Holstein cows milked 3 times daily. Journal of Dairy Science 95, 1170–1176.
Weiss, D., Bruckmaier, R.M., 2005. Optimization of individual prestimulation in dairy cows. Journal of Dairy Science 88, 137–147.
Wieland, M., Nydam, D.V., Virkler, P.D., 2017. A longitudinal field study investigating the association between teat-end shape and two minute milk yield, milking unit-on time, and time in low flow rate. Livestock Science 205, 88–97.
Wieland, M., Nydam, D.V., Heuwieser, W., Morrill, K.M., Ferlito, L., Watters, R.D., Virkler, P.D., 2020b. A randomized trial to study the effect of automatic cluster remover settings on milking performance, teat condition, and udder health. Journal of Dairy Science 103, 3668–3682.
Wieland, M., Virkler, P.D., Weld, A., Melvin, J.M., Wettstein, M.R., Oswald, M.F., Geary, C.M., Watters, R.D., Lynch, R., Nydam, D.V., 2020a. The effect of 2 different premilking stimulation regimens, with and without manual forestripping, on teat tissue condition and milking performance in Holstein dairy cows milked 3 times daily. Journal of Dairy Science 103, 9548–9560.
Wiercioch, M., Luberański, A., Krzyś, A., Skalska, D., Szlachta, J., 2016. Impact of the System of Air Supply to a Milking Unit on Selected Parameters of Milking Machine Operation. Agricultural Engineering 20, 195–205.
Williams, D.M., Mein, G.A., Brown, M.R., 1981. Biological responses of the bovine teat to milking: Information from measurements of milk flow-rate within single pulsation cycles. Journal of Dairy Research 48, 7–21.
Zecconi, A., Bronzo, V., Ruffo, G., Hamann, J., 1992. Machine-induced teat tissue reactions and infection risk in a dairy herd free from contagious mastitis pathogens. Journal of Dairy Research 59, 265–271.
Zecconi, A., Bronzo, V., Piccinini, R., Moroni, P., Ruffo, G., 1996. Field study on the relationship between teat thickness changes and intramammary infections. Journal of Dairy Research 63, 361–368.