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RESEARCH ARTICLE

Milk characteristics and milking efficiency in Italian Mediterranean buffalo

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ABSTRACT

The study aimed to verify the influence of different milking machine setting parameters on Italian Mediterranean buffalo milking performance. Data on milking machine settings (Milking Dry Tests [MDTs]) and milk Test-day (TD) were collected by the Italian Breeders Association (AIA). Data for each TD consisted of milk yield (MY), fat percentage (FP), protein percentage (PP), lactose percentage (LP) and linear score (LS). Working vacuum level (VL), pulsation ratio (PR), automatic cluster removal system (AC) and effective vacuum reserve (EVR) were recorded from each MDT. A total of 558 MDTs, 217,967 TD, collected from 43,593 buffaloes in 198 buffalo farms were utilised. MDT, TD and Animal data (AD) were analysed through a mixed linear model and a logistic regression model was used to evaluate the relationship between VL and EVR. Analysis of MY and quality revealed that an incorrect setting of the milking was responsible for a higher LS and lower MY and LP, along with higher FP and PP compared to farms with adequate EVR. Except for PP, the height of the milking system significantly influenced all milk parameters. Conversely, VL affected all milking traits. A higher PR (70:30) was responsible for significantly lower LS and higher FP compared to a 60:40 PR. Similarly, the presence of AC showed a significant effect on FP and MY and a slight reduction in LS. Finally, the diameter and length of the pipeline (DL) ratio lower than 2.5 m was associated with a decrease in LS. These results suggest that buffaloes require specific milking parameters.

HIGHLIGHTS

- Buffalo has a specific requirement for milking parameters.
- Incorrect milking setting increase somatic cells and reduce lactose content.
- Vacuum level (VL) affects milk yield (MY) and quality as well as somatic cells and lactose.

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Milking machine parameters; milking vacuum; milking efficiency; udder health; Mediterranean buffalo

Introduction

The mechanisation of milking is a valuable tool to reduce dairy farming costs and improve both milk quality and labour efficiency (Cogato et al. 2021). Nowadays, mechanised milking is widely used on dairy farms and still accounts for between 33% and 57% of total farm labour (Bach and Cabrera 2017). Monitoring milking routine and milking machine operating parameters is a crucial factor in ensuring optimal milk yield (MY), labour efficiency and udder health in dairy animals (Thomas et al. 2005; Besier et al. 2016; Odorčić et al. 2019). In addition, it is the basis for ensuring proper milk sampling and recording in animal husbandry. Indeed, the data collected during a normal Test-day (TD) can only be considered reliable if all the parameters are adequate and organised to best explain a specific production process. This interesting aspect is also particularly important from the point of view of genetic improvement.

For this reason, an appropriate milking parlour model must be adapted to each animal species or even breed, along with appropriate setting criteria and technical parameters for milking (Dzidic et al. 2019).

Differences in udder and teat morphology in dairy animals require the study of species-specific milking

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procedures and machine parameters (Thomas et al. 2004; Ambord et al. 2010). For example, the vacuum parameter is one of the milking machine settings that can seriously affect mammary gland health if not properly regulated, leading to changes in milk flow and teat damage (Besier et al. 2016; Nørstebø et al. 2019). In addition, vacuum fluctuations have been shown to increase the risk of bacterial transmission from one teat to another in dairy cows (Besier et al. 2016). The impact of mechanical milking to mastitis occurrence is often poor clear. In fact, it was demonstrated that machine-related effects usually are less important than the role of milking management, herd management and cow or teat characteristics in most herds (Mein 2012; Odorčić et al. 2019; Vermaak et al. 2023).

Several studies have analysed milking parameters in dairy cows (Stauffer et al. 2020), sheep (Romero et al. 2020) and goats (Bueso-Ródenas et al. 2016; Fernández et al. 2020), whereas little information is available for buffalo (Thomas et al. 2004; Caria et al. 2011). The buffalo species represents an important livestock resource in many countries, providing food of animal origin and draught power (D'Occhio et al. 2020). In Italy, the population of Italian Mediterranean buffalo reaches more than 430,000 heads and is specialised for milk production. In 2022, MY reached 2350 kg/lactation, with fat and protein percentages (PPs) of 7.72% and 4.65%, respectively. Italian buffalo farms are characterised by a high degree of innovation and animal husbandry is similar to that performed in dairy cows. Similarly, milking procedures and parameters applied to the buffalo species are also derived from those of dairy cows, although both the anatomy and physiology of the two species are different (Caria et al. 2011). The mammary gland of buffaloes is characterised by a smaller cistern and consequently a lower amount of available cisternal milk (5 *vs*. 30%) compared to cattle (Ambord et al. 2010). Furthermore, other morphological characteristics, such as longer teats and a stronger teat sphincter than in cattle udders should also be considered (Thomas et al. 2004).

Currently, only three commercial farms use automated milking systems (AMS), whereas conventional milking systems (CMS) are widely used on buffalo farms in Italy. This requires a large-scale study to verify the parameters that would be applied to buffalo species. Preliminary results have been published in abstract form by Matera et al. (2023). Based on the results obtained, the aims of the study were:

i. to provide an overview of the common milking conditions in a large population of Italian Mediterranean buffaloes;
ii. to investigate the influence of different milking conditions on milk quality traits;
iii. to define the most appropriate milking machine parameters for Italian Mediterranean buffaloes.

Material and methods

Milking system and setting parameters data collection

Data on milking machine and equipment settings were collected as part of the official activity of the Milking Control Service (MCS) of the Italian Breeders Association (AIA). The data were collected according to the ISO 5707:2007 and 6690:2007.

The data collection campaign was conducted in 213 buffalo farms in the Campania Region (Italy). A total of 558 Milking Dry Tests (MDT) were performed between December 2018 and February 2022 according to IDF (2005). The MDTs were performed with 6 different types of milking systems: Bucket (11), low milking jars (194), high milking jars (58), low milk pipeline (277), high milk pipeline (10) and AMS (3). For further statistical analysis, the height of the milking system (MSH) and the ratio between the diameter and length of the pipeline (DL) were considered. High and low milking systems were determined according to UNI ISO 3918:2007. A high MSH exists when the milk inlet is 1.25 m above the standing level of the animals. Low MSH occurs when the milk inlet is below the standing level of the animals.

During each MDT, the following milking parameters were recorded: Working vacuum level (VL), pulsation ratio (PR), automatic cluster removal system (AC) and effective vacuum reserve (EVR).

ISO 5707:2007 was used to determine milking machines with sufficient and insufficient EVR. Briefly, sufficient EVR was calculated considering the following factors: Number of units, presence of automatic shut-off valves and type of milking system.

Data collection and editing

In addition to the control of the milking system, individual production data from the AIA's official performance recording system were considered at each farm. In order to evaluate the effects of milking parameters, 3 consecutive records before and 3 after the date of milking system control were considered. A total of 267,305 TD samples from 46,605 buffaloes were used. Information on ID, parity (3.53 ± 2.46), and days in milk (DIM $-$ 129.71 ± 84.14) was available for each animal at each TD. Productive data for each TD consisted of

MY, milk fat percentage (FP), milk PP, lactose percentage (LP) and somatic cell count (SCC – cells/mL). MY was measured using milk metres according to ICAR Procedures (2022), while FP, PP and LP were determined by mid-infrared spectroscopy using a MilkoScan FT6000 (Foss Electric A/S, Hillerod, Denmark). SCC was assessed using the Fossomatic FC (Foss Electric A/S) and the value was log-transformed to calculate the linear score (LS) according to the following Equation (1) (Ali and Shook 1980):

$$LS = \log_2 * \left(\frac{SCC}{100.000}\right) + 3 \tag{1}$$

Statistical analysis

Data from each TD were analysed with the following mixed linear model (2):

$$y_{ijkmnopqrs} = \mu + MSH_i + parity_j + DIM_k + VL_m + PR_n + AC_o + DL_p + EVR_q + Farm_r + Animal_s + \varepsilon_{ijkmnopqrs} \tag{2}$$

where $y_{ijkmnopqrs}$ = milk quality parameters (LS, fat, protein and LP, daily milk production), μ is the overall mean, MSH_i = fixed effect of the ith Milking System Height (Low, High – ISO 3918, 2007); $parity_j$ = fixed effect of the jth parity (first, second, third, ≥third); DIM_k = fixed effect of the kth class of days in milking (<90, 90–180, >180); VL_m = fixed effect of the mth class of VL (<41, 41–43, 44–45, >45); PR_n = fixed effect of the nth class of Pulse Ratio (60:40; 70:30); AC_o = fixed effect of the oth Automated cluster removers system (yes, no); DL_p = fixed effect of the pth Diameter Length ratio of pipeline (<2.5, ≥2.5); EVR_q = fixed effect of the qth Empty Useful Reserve status (yes, no); $Farm_r$ = random effect of the rth farm (157 levels); $Animal_s$ = random effect of the sth animal (52,689 levels); $\varepsilon_{ijkmnopqrs}$ = random residual. The effects were declared significant at $p < 0.05$. Multiple comparisons among means were performed by Tukey's test, with significance considered at $p < 0.05$.

A logistic regression model was used to evaluate the relationship between the levels of vacuum (<41, 41–43, 44–45, >45) and EVR.

The following parameters were involved in the canonical discriminant analysis (CDA) based on approach described by Conte et al. (2018): PR, VL, DL, EVR, Vacuum Reserve difference, LS, lactose, milk proteins percentage, milk FP and milk production. This technique was used to discriminate the different types of milking systems: carousel milking machine with low pipeline (CMLP), milking machine in parallel with low pipeline (PMLP), herringbone milking machine with high jars (HMH), herringbone milking machine with high jars (HML), herringbone milking machine with high pipeline (HMHP), herringbone milking machine with low pipeline (HMLP), tandem milking machine with high jars (TMH), tandem milking machine with low jars (TML), tandem milking machine with high pipeline (TMHP) and tandem milking machine with low pipeline (TMLP).

All analyses were performed by JMP Pro version 17 software (SAS, Cary, NC).

Results

After data editing, a total of 217,967 TD from 43,593 buffaloes in 198 farms were considered. An insufficient EVR was found in 160 MDT (28.6%). Analysis of MY and quality for these cases (63,917 TD – 29.4% of the total) revealed that inadequate EVR was responsible for a higher LS and lower MY along with higher fat and protein concentration compared to those with adequate EVR (Table 1). Therefore, these data were excluded from further analysis except for CDA and only MDT cases with sufficient EVR were further considered, yielding a total of 154,050 TD from 30,810 buffaloes.

Descriptive statistics for MY and milk traits are presented in Table 2, while the effects of parity and lactation stage on milk traits are showed in Supplementary Tables S1 and S2, respectively.

In particular, a higher LS was observed in buffaloes with more than 180 DIM and in those with more than 3 parities compared to their counterparts. LS was also affected by VL (Table 3): Indeed, a strong increase was observed at a VL higher than 45, while 2.04 LS was recorded at a VL lower than 41.

The various fixed effects included in the model are summarised in Table 4.

MSH significantly ($p < 0.001$) influenced LS and MY. MSH also significantly ($p < 0.001$) influenced LP with significant differences ($p < 0.001$) between high and low height. Conversely, different vacuum classes significantly ($p < 0.001$) affected all milk traits. A higher PR (70:30) was responsible for significantly ($p < 0.001$) lower LS and higher FP compared to a 60:40 PR. Similarly, the presence of AC showed a significant effect ($p < 0.001$) on FP and MY and a slight ($p < 0.05$) reduction in LS. Finally, DL lower than 2.5 m was associated with a LS reduction ($p < 0.001$).

The CDA extracted two canonicals functions (CAN) (describing the 87% of observed variance, 70% and

Table 1. Milk yield (kg) and characteristics in milking machines with sufficient and insufficient EVR.

	Sufficient EVR milking machine ($n = 154,050$)	Insufficient EVR milking machine ($n = 63,917$)	SEM	p Value
MY (kg)	8.82	8.47	0.03	0.003
PP (%)	4.70	4.73	0.01	0.01
FP (%)	8.15	8.24	0.03	<0.001
LP (%)	4.67	4.63	0.01	<0.001
LS	3.29^{B}	3.34^{A}	0.06	<0.001

EVR: effective vacuum reserve; SEM: standard error of the mean; MY: milk yield; PP: protein percentage; FP: fat percentage; LP: lactose percentage; LS: linear score
Values within a row with different superscripts differ significantly at $p < 0.001$.

Table 2. Descriptive statistics (mean, standard deviation, coefficient of variation, minimum and maximum values) for milk yield and characteristics ($n°$ of test days = 154,050).

	Mean	SD	CV	Min	Max
LS	3.41	1.65	48.54	0.06	11.13
LP (%)	4.67	0.28	5.95	3.00	5.59
PP (%)	4.69	0.40	8.44	1.64	6.00
FP (%)	8.08	1.38	17.04	5.00	11.00
MY (kg/d)	9.24	3.62	39.24	1.00	27.70

SD: standard deviation; CV: coefficient of variation; Min: minimum value; Max: maximum value; LS: linear score; LP: lactose percentage; PP: protein percentage; FP: fat percentage; MY: milk yield

Table 3. Descriptive statistics (mean, standard deviation, coefficient of variation, minimum and maximum values) of linear score per vacuum level ($n°$ of test days= 154,050).

	Vacuum level			
	<41	41–43	44–45	>45
Mean	2.04	3.10	3.43	4.13
SD	0.98	1.62	1.66	1.51
CV	48.04	52.26	48.40	36.56
Min	0.06	0.05	0.06	0.05
Max	5.36	11.10	10.98	10.88

SD: standard deviation; CV: coefficient of variation; Min: minimum value; Max: maximum value

17% for CAN_1 and CAN_2, respectively) which discriminated the 11 groups as demonstrated in Figure 1 (p value Hotelling's t-test < 0.0001). The first CAN separated milking systems with pipeline at a lower level (CMLP, PMLP and HMLP) with a positive score, from the other ones (Figure 1).

Otherwise, the second CAN discriminated CMLP and TMH (positive scores) from the other milking systems. Based on this discrimination, the variables principally associated with the first CAN were EVR and Vacuum Reserve difference with a positive and negative score, respectively (Supplementary Table S3). The second CAN associated diameter of vacuum tube and Vacuum Reserve difference with a positive score and diameter of pipeline with negative score (Supplementary Table S3).

Discussion

The aim of this study was to verify the influence of different milking parameters on MY and quality in buffalo species, in which the practices routinely utilised in dairy cows have usually been transposed. The evaluation of milk let-down is probably the most important aspect to be considered in a dairy farm. A proper setting of the milking machine allows to obtain both high MY, preserving animal health, and quality, reducing SCC and maintaining the secreting capability of the mammary gland. A milking machine should have enough airflow capacity to supply the normal operating requirements of the milking machine plus unintended air admission that may occur during unit attachment or detachment (Reinemann et al. 2021). If the volume of admitted air is greater than the vacuum effective reserve a drop in the system vacuum will be observed (Brazil and Britten 2001).

As mentioned above, there are few and limited studies on the proper setting of milking parameters in buffalo species. Some authors showed that milking buffaloes can be laborious because milk output is delayed due to a slow reflex and thicker sphincter around the teat canal (Caria et al. 2011).

The first important finding of the study was the evidence that about 30% of buffalo milking machines carry out a milking with an inadequate EVR. This was responsible for both an increase in LS and a decrease in MY. Indeed, low effective reserve is one of the causes of vacuum fluctuations, leading to easier spread of infectious agents and increased risk of intramammary infection in different ruminant species (Fox et al. 2009; Bava et al. 2017; Romero et al. 2020).

In addition, vacuum reserve is necessary for vacuum stability in all tracts of the milking machine (International Dairy Federation 2005). As demonstrated by the discriminant analysis (Figure 1) the different types of milking systems differ in the reserve of voids. The herringbone milking machine with high pipeline milking system was found to be the most effective for buffalo milking.

The importance of maintaining vacuum stability during milking has been widely recognised in dairy cows for several years (Rønningen 2002). Thus, fluctuations in the milking system, such as those occurring when vacuum reserve is inadequate, seriously affect

Table 4. Least square means ± standard error of milk characteristics for each milking machine parameters.

MSH						
		Low ($n = 141,421$)	High ($n = 11,813$)		SEM	p Value
LS		3.40	3.33		0.01	<0.001
LP (%)		4.68	4.65		0.02	<0.001
PP (%)		4.72	4.70		0.01	0.240
FP (%)		8.04	8.06		0.01	0.310
MY (kg/d)		9.07	8.87		0.02	<0.001
VL						
	<41 ($n = 12,064$)	41–43 ($n = 60,616$)	44–45 ($n = 53,246$)	>45 ($n = 27,298$)	SEM	p Value
LS	3.19^D	3.23^C	3.29^B	3.37^A	0.01	<0.001
LP (%)	4.61^B	4.65^A	4.63^B	4.66^A	0.01	<0.001
PP (%)	4.71^A	4.73^A	4.72^A	4.69^B	0.01	<0.001
FP (%)	8.09^B	8.23^A	8.22^A	8.18^B	0.01	<0.001
MY (kg/d)	8.52^C	8.52^C	8.59^B	8.90^A	0.03	<0.001
PR						
		60:40 ($n = 81,925$)		70:30 ($n = 71,309$)	SEM	p Value
LS		3.55		3.44	0.01	<0.001
LP (%)		4.65		4.63	0.01	0.330
PP (%)		4.71		4.71	0.01	0.300
FP (%)		8.17		8.22	0.01	<0.001
MY (kg/d)		8.68		8.66	0.02	0.440
AC						
		No ($n = 77,622$)		Yes ($n = 75,612$)	SEM	p Value
LS		3.50		3.48	0.01	0.030
LP (%)		4.64		4.63	0.01	0.150
PP (%)		4.73		4.69	0.02	0.489
FP (%)		8.11		8.28	0.01	<0.001
MY (kg/d)		8.41		8.87	0.10	<0.001
DL						
		<2.5		>2.5	SEM	p Value
LS		3.58		3.38	0.02	<0.001
LP (%)		4.63		4.63	0.01	0.58
PP (%)		4.68		4.69	0.01	0.62
FP (%)		8.29		8.26	0.01	0.26
MY (kg/d)		8.72		8.66	0.02	0.45

SEM: standard error of mean; LS: linear Score; LP: lactose percentage; PP: protein percentage; FP: fat percentage; MY: milk yield; MSH: milking system height; VL: vacuum level; PR: pulse ratio; AC: automated cluster removers system; DL: diameter length ratio
Values within a row with different superscripts differ significantly at $p < 0.001$.

the health of the mammary gland, leading to both keratinisation of teat ends and an increase in SCC (Odorčić et al. 2019).

It is worth noting that our study also found a decrease in lactose concentration when the vacuum reserve was not adequate. Although the role of lactose as mastitis indicator has been studied since 30 years ago, controversial results have been reported (for review see Pyörälä 2003). Recently, some studies carried out in both dairy cattle (Costa, Bovenhuis, et al. 2020) and dairy buffaloes (Costa, De Marchi, et al. 2020) seem to support the hypothesis that a decrease in lactose concentration may be indicative of an unhealthy status of the mammary gland. These interesting findings confirm the importance of vacuum reserve and supporting our decision to exclude data with inadequate EVR from further analysis.

VL is probably one of the most important parameters to be considered in milking management. It is known that high vacuum may be responsible for serious damages of the teats, while low vacuum can cause low pression at teat level, resulting in altered massage phase and reduced milk let-down (Odorčić et al. 2019). Thomas et al. (2004) studied the milking parameters applied to buffaloes in different countries and reported a range of vacuum values from 45 to 68 kPa. In Italy, the most applied vacuum values are 44–46 kPa, although Caria et al. (2011) reported a range of 40–53 kPa. Several studies demonstrated that buffaloes are often exposed to vacuum for long periods of time without milk ejection, especially if an adequate mammary gland stimulation is not provided prior to cluster application (Boselli et al. 2020). Ambord et al. (2010) found that vacuum up to 45 kPa is generally ineffective in the absence of alveolar milk ejection. In contrast, other authors (Caria et al. 2011) have clearly demonstrated that even a vacuum of 36 kPa can be more than sufficient to ensure teat canal opening and milk ejection in buffaloes, although a lower flow rate is recorded (0.79 kg/min *vs*. 0.69 kg/min at a vacuum of 42 and 36 kPa, respectively).

Both peak milk flow and milking speed increase with increasing milking vacuum (Fernandez et al. 2020; Stauffer et al. 2020). However, high vacuum settings are likely responsible for increased hyperkeratosis, increased post-milking teat end plugging, and

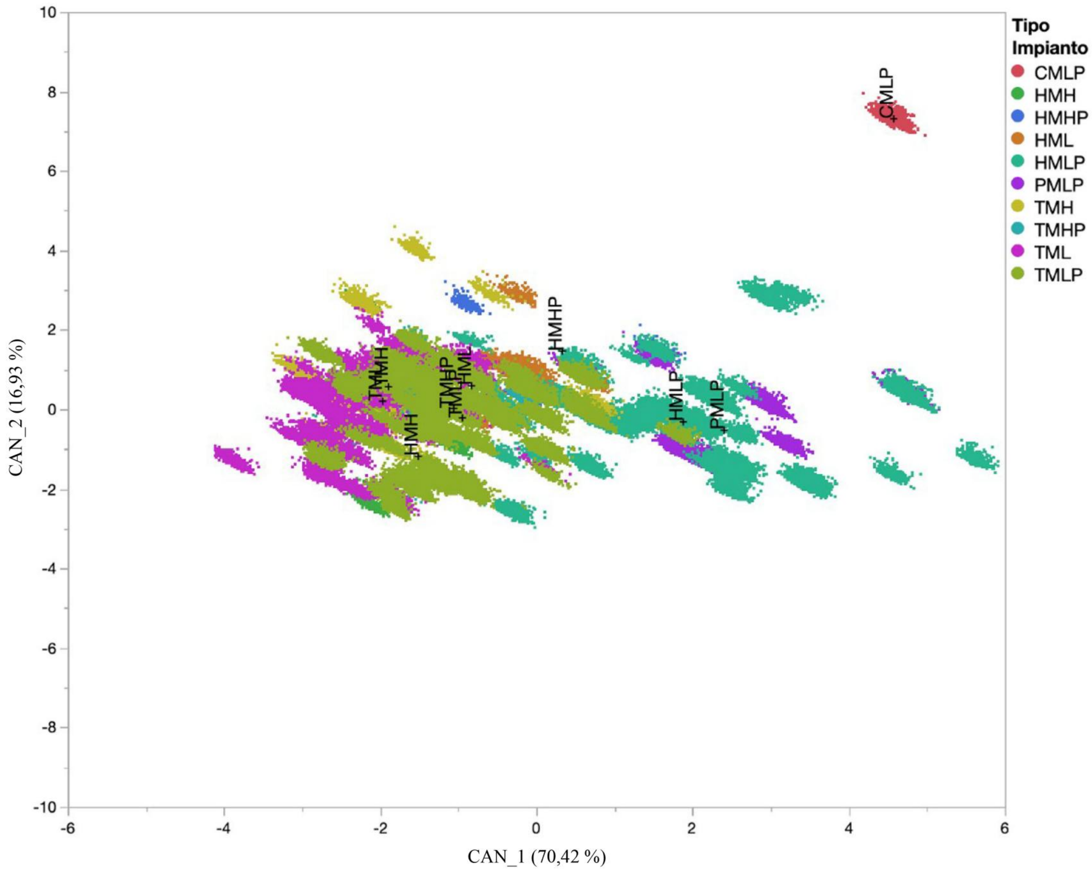

Figure 1. Graph of the canonical functions (CAN) for milking systems discriminations. CMLP: carousel milking machine with low pipeline; PMLP milking machine in parallel with low pipeline; HMH: herringbone milking machine with high jars; HML: herringbone milking machine with high jars; HMHP: herringbone milking machine with high pipeline; HMLP: herringbone milking machine with low pipeline; TMH: tandem milking machine with high jars; TML: tandem milking machine with low jars; TMHP tandem milking machine with high pipeline and TMLP: tandem milking machine with low pipeline.

possibly prolonged post-milking teat end opening (Fernandez et al. 2020). Particularly, the latter, is again a risk factor for clinical mastitis (Romero et al. 2020).

When the dataset was edited, the importance of the VL during milking became even more apparent in the buffalo species. Indeed, a significantly lower LS was recorded when a vacuum below 41 kPa was applied, while an increase in vacuum was associated with an increase in milk somatic cell score. In addition, several studies (Kunc et al. 2000) found that 45 kPa VL evoke higher teat temperatures than 40 kPa, resulting in both an increased stress on the mammary gland and higher susceptibility to mastitis. The effect of VL recorded in this study indicates that higher VLs could also increase the prevalence of mammary inflammation in Italian Mediterranean buffaloes, as previously observed in dairy cows (Mahle et al. 1982), sheep (Romero et al. 2020) and goats (Fernandez et al. 2020).

Different VLs also affected MY, although this aspect has not been recorded in other studies carried out on goats (Bueso-Ródenas et al. 2016) or buffaloes (Caria et al. 2011). In particular, low VLs were associated with low milk production. Significant differences in milk quality were also observed between the different VL, with a higher percentage recorded mainly when medium VLs (41–43 kPa and 44–45 kPa) were considered. Fat showed the lowest percentage at the lowest and highest VLs tested. A possible explanation for this phenomenon is that low vacuum prolongs milking time and favours the extraction of alveolar milk, which is the richest in fat. Indeed, during mechanical milking of dairy cows, the fat concentration of the milk increases over time because the milk obtained at the end of a single milking corresponds to alveolar milk, which is 2.5–5 times richer in fat than cysternal milk (Lollivier et al. 2002). At the same time, however, too low VL cannot guarantee proper extraction.

As mentioned above, in this study, we examined 10 different types of milking systems. As expected, multivariate analysis showed that the different types of systems affected milk quality and quantity characteristics differently. In particular, it was interesting to note that LS was lower when a herringbone system was used compared to a tandem system, despite no data are available in literature. A hypothesis may be that herringbone are applied in larger and newer herds where also milker may have a better training and tandem parlour are used in older and smaller herd where milkers' capability and management have poorer, thus leading to lower milk quality and quantity. However, in our studies, farms with herringbone and tandem parlour were similar as dimension of herd, so further investigations will be required. We can speculate that this result could be explained by the different management of the milking routine, that is commonly carried out in these two systems. Indeed, in a herringbone system, the animals enter and leave the milking parlour at the same time: this leads to both higher utilisation and more efficient milking routine (disinfection, cleaning and teats drying) compared to the tandem system. This condition ensures better udder stimulation before cluster attachment, as the udders of all the animals of one side are cleaned and disinfected before attachment. In tandem milking systems, on the contrary, animals enter and exit the milking stalls individually, without waiting for all animals to finish milking. Although this system ensures low utilisation of the milking parlour, improper preparation and stimulation of the udder by the milkers may occur.

Most (*n* 141,421) of the milking systems analysed in this study have a low milking line. Farmers often choose a low milking line because installation costs are lower (25–35%, depending on the manufacturer) and milking efficiency is higher with the same number of operators (Diaz et al. 2004). In addition, the low system allows milking at a lower vacuum, avoiding the effect of a higher VL mentioned above.

Although a previous study in ewes (Diaz et al. 2004) and in goats (Manzur et al. 2012) showed that milking system had no effect on SCC, we found a significant influence of MSH on inflammatory target parameters, such as LS and lactose. Similarly, Romero et al. (2020) observed that milkline height had no relationship with SCC when the other settings were adequate, confirming that there is not yet a best milking setting in buffaloes. Nevertheless, the lack of difference between high and low MSH for LS suggests that there are good milking prospects for both milking line systems when EVR is adequate. Differences from other studies available in the literature were also found with respect to milk production and milk quality. For example, Diaz et al. (2004) showed that low MSH had no significant effect on milk production. In contrast, in this study, it was observed that both high and low MSH had a significant effect on MY in dairy buffaloes. Moreover, no relationship was observed between FP and MSH. These results contradict the findings of other authors (Meffe 1994), who stated a decreased lipolysis rate when low milking system were used in dairy cows.

The use of automatic cluster removal (AC) on buffalo farms is not widespread: about 50% of the milking parlours involved in this study have AC installed. However, even when it is present, AC is rarely used by milkers, possibly due to the large variation in both MY and emission among lactating buffaloes (Boselli et al. 2020). It is also known that water buffalo is very sensitive to environmental stimuli before and during milking, which also explains why interrupted milk ejections are common in this species due to unstable blood oxytocin levels (Thomas et al. 2005), and which also makes it difficult to determine a correct milking routine. In this study, AC did not significantly affect parameters related to mammary gland status, although previous research in dairy cows have shown improved teat condition and udder health (Rasmussen 1993). Contrasting results have been reported in the literature regarding the effect of AC on MY. In agreement with some authors (Tangorra et al. 2010) we found a significant effect of AC on MY. On the contrary, Rasmussen (1993) showed that AC had no effect on MY. Theoretically, decreased udder emptying with increased AC decrease should result in lower fat content of milk and higher or equal protein and lactose content because residual milk has high fat content and lower protein and lactose content than available milk (Ontsouka et al. 2003). The fat content of buffalo milk appears to be related to AC function compared to lactose and protein content. Further studies are needed to evaluate the effects of AC equipment for these species.

Many authors have studied the effects of the PR. Some experiments (O'Callaghan 1998) showed that a wide PR resulted in faster milking than a narrow PR. Widening the PR up to 75:25 (Ferneborg and Svennersten-Sjaunja 2015) an increase in peak and average milk flow and a shorter machine duty cycle were recorded. They also observed that SCC was not affected by increasing PR. These results were confirmed by other authors (Gleeson et al. 2004), who demonstrated that an increased PR (from 60:40 to 67:33) had no negative effect on teat tissue but had a positive effect on milking time. Also in our work, despite the influence of PR on LS and FP, no differences were found between the two PRs studied.

Conclusions

These results suggested that buffalo have a specific requirement for milking parameters and that, as already recognised in dairy cattle, an incorrect setting of the milking was responsible for both an increase of LS and a reduction of lactose. In milking dairy buffaloes, VL also affects MY and quality per milking as well as somatic cells and lactose, which are indicative of udder health. Since higher MYs have been found with higher VLs, it would be advisable to use lower VLs to avoid udder damage and mastitis outbreaks. Similarly, since no differences in performance were identified, an extension of the PR from 60:40 to 70:30 and the use of a low-line milking equipment could be envisaged as a good opportunity to decrease milking time and cost without affecting the LS. Since, there are not data literature, these results are innovative for buffalo specie and confirm what just demonstrated in bovine. In any case, further studies are needed to investigate the effect of the various combinations of milking parameters on the quality and quantity characteristics of milk, as well as to verify their effect on milk flow and milking times.

Ethics approval

The approval of the Animal welfare and use Committee was not required for this study. All data were collected from pre-existing databases during routine animal recording procedures and standard controls of milking.

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Disclosure statement

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Data availability statement

None of the data were deposited in an official repository. The data that support the findings of this study are available upon reasonable request to the corresponding author.

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