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COMPARATIVE STUDY OF BEHAVIOURAL AND MILKING TRAITS IN COWS MILKED WITH A CONVENTIONAL OR INDIVIDUAL QUARTER MILKING SYSTEM (MULTILACTOR®) AND WITH DIFFERENT MILKING PERSONS*

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Abstract

The aim of this study was to investigate the influence of a new type of milking system on the behaviour of cows during milking by comparing a conventional milking system (CON) with an individual quarter milking system (MUL), MultiLactor®. Sixty-eight dairy cows were observed during their milking times (32 cows in CON, 36 cows in MUL) using video recordings to analyse their behavioural traits. The udder preparation duration, milking duration and milk yield were also evaluated. No significant differences were found between the CON and the MUL regarding cows' head posture (P=0.38), body posture (P=0.85), number of steps (P=0.08) and number of kicks (P=0.56). However, the milk yield was lower (P=0.02), just as the udder preparation duration (P<0.01) and milking duration (P=0.01) were shorter in the CON compared to the MUL. In addition, in regard to the milking person, differences were displayed in the head posture of the milked cows, kick-off or loss of teat cup or milking cluster, and frequency of udder preparation. In conclusion, the investigated milking systems did not markedly influence the behaviour of dairy cows; however, udder preparation duration, milking duration and milk yield were significantly greater for the MUL than for the CON. However, the milking person appears to have a greater impact on the behaviour of the cows than the milking system.

Key words: behaviour, dairy cow, individual quarter milking, udder preparation

The relationship between humans and animals as well as familiar surroundings are important factors that influence the welfare and milk yield of dairy cows (Adam-

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czyk et al., 2015; Hemsworth et al., 1989; Waiblinger et al., 2003). Previous studies already showed that cows can discriminate between aversive and neutral/gentle persons (Hötzel et al., 2005; Munksgaard et al., 2001). In addition to the human-animal relationship, the reaction to environmental conditions, the individual age and milking temperament of the cows (Szentléleki et al., 2015), the kind of habituation to the milking routine (Kutzer et al., 2015) as well as the milking system itself influences the welfare and behaviour of the cows. The influence of the milking systems was examined in comparative studies between conventional and automatic milking systems (AMS). However, Gygax et al. (2008) found only minor differences in restlessness behaviour, heart rate and heart rate variability of dairy cows milked in an auto-tandem milking parlour or AMS. Their results indicate that milking in these two systems does not seriously impair the welfare of dairy cows, but the stress level in dairy cows slightly increases in AMSs as opposed to auto-tandem milking parlours (tandem milking parlours with automatic gates). Hopster et al. (2002), who compared as well cows milked in these two systems, also concluded that the differences between behavioural and physiological responses are relatively low. However, Wenzel et al. (2003) found significant differences in the behavioural (more step behaviour in the AMS) and physiological conditions (higher milk cortisol concentration and heart rate in the AMS) between cows milked in an AMS or in a milking parlour (tandem parlour).

An individual quarter milking system (MUL), MultiLactor® (Siliconform, Türkheim, Germany, market launch in 2009), was developed to improve the milk quality and to reduce negative impacts on the cows (e.g., reduction of turning, tilting and side forces at the teat (Rose-Meierhöfer et al., 2009) as well as a better adaption to the udder) during the milking process. The MUL uses no claw but a single milk tube per udder quarter, which is comparable to an AMS but designed for use in conventional milking parlours (tandem and herringbone parlours) with manual attachment of teat cups by the milking person. Recent studies (Müller et al., 2011; 2013) compared MUL and a conventional milking system (CON) with claw, each in a tandem parlour, in regard to milkability traits and udder health status of dairy cows. Differences ($P<0.05$) were found between both milking systems for all of the milk flow traits (a longer incline phase and shorter plateau phase in CON than in MUL), except for the decline phase. The milk yield differed not significantly between CON and MUL (Müller et al., 2011). Furthermore, the results of Müller et al. (2013) showed that the median somatic cell count of quarter foremilk samples did not exceed the threshold value of 100,000 cells/ml (as defined in Hamann, 2005) in either system. The median of somatic cell count was higher in MUL (40,000 cells/ml) than in CON (27,000 cells/ml), but the milking system had no significant effect ($P=0.06$) on the quarter health status. Other studies investigated the effects of the milk flow on the average liner vacuum in the MultiLactor® when compared to the CON (GEA Farm Technologies, Bönen, Germany) in a tandem parlour (Öz et al., 2010; Rose-Meierhöfer et al., 2010). The results showed that, at an average flow rate of approximately 5.0 l/udder/min, the average liner vacuum during the d-phase (when the liner is closed) was significantly higher for CON (28.5 kPa) when compared to MUL (16.3 kPa). This large difference is caused by the MultiLactor® using the BioMilker-system® (Siliconform GmbH, Türkheim, Germany), which allows the periodic ingress of air to the pulsation chamber. This result leads to the approach that there could also be an effect on milking duration and subsequently on the behaviour of animals suggesting to measure these criterions in our experiment. Furthermore, the MUL is represented by the manufacturer as an animal-friendly system, e.g., through an assisted relaxation of the udder tissue and a better adaption to different udder shapes.

However, the previously conducted studies have focused on the milkability traits (Müller et al., 2011; 2013) and physical workload of the milking persons (Jakob and Liebers, 2011); the behavioural traits of the cows milked during MUL milkings have not been considered to date. Therefore, the objective of this study was to compare the CON and the MUL systems based on the behaviour of the cows during milking. Furthermore, the udder preparation duration, the duration of the milking process and the amount of milk were also evaluated.

Material and methods

Animals and housing

Four weeks prior to the onset of the trial, two groups of 37 animals each were randomly selected from a German herd of 660 lactating Holstein-Friesians cows and uniformly divided. After grouping, the cows had four weeks for habituation before the trial began. The study exclusively included cows without clinical indications of udder inflammation (inconspicuous udder palpation and appearance as well as a not pathological microbiological status of quarter foremilk samples) and cows between the 150th and 270th day of lactation at the beginning of the trial. The study was conducted in autumn within a period of thirteen weeks. All cows were between 1.4 and 8.9 years of age and in their first to seventh lactation. The characteristics of the cows are shown in Table 1.

Table 1. Characteristics of the cows (mean ± standard deviation) and count of cows per lactation number

	Conventional milking system (CON)	Individual quarter milking system (MUL)	All animals
Days in milk	199.6±45.6	206.2±34.8	202.9±40.4
Age (in years)	3.9±1.4	3.7±0.9	3.8±1.1
Time of cows' familiarity with milking system during the present lactation (in days)	199.6±45.6	90.0±0.0	144.8±63.8
Lactation number	2.1±1.3	1.8±0.8	1.9±1.0
cows in 1st lactation	14	15	29
cows in 2nd lactation	10	15	25
cows in 3rd or higher lactation	8	6	14

The two groups were stabled in the same building in two separated, adjacent, and nearly identical barn areas with comparable microclimatic conditions (moderate climate). The cows were fed the same mixed ration, supplemented with concentrates that were fed animal-specific, according to the milk production level. During the habituation period and the whole trial, the two groups were milked separately, and the daily handling and milking management processes (herding animals to the milking parlours and pre-milking preparations) were as similar as possible. The cows were generally housed in groups of 30 to 50 animals on this farm and therefore were accustomed to this type of housing.

Milking

The cows were milked in the morning and in the afternoon in two separate tandem milking parlours without concentrate feeders. The parlour of one group was equipped with a conventional milking system (CON) (GEA Farm Technologies, Bönen, Germany) with 2x6 milking boxes. The parlour of the other group was equipped with an individual quarter milking system (MUL), MultiLactor® (Siliconform, Türkheim, Germany), with 2x4 milking boxes. The main differences in the milking equipment of the CON and the MUL are shown in Table 2. In addition, the MUL was equipped with a pneumatic arm that moves the four milk tubes regularly and should help to relax the udder tissue. Silicone liners were used in both milking systems.

Table 2. Milking equipment

Conventional milking system (CON)	Individual quarter milking system (MUL)
Conventional cluster (capacity: 300 ccm)	Single tube guidance
Weight of milking cluster: 2.4 kg + one long milk tube	Weight of 4 teat cups: 1.2 kg + 4 long milk tubes
Machine vacuum: 40 kPa	Machine vacuum: 37 kPa
Automatic teat cup cleaning and disinfection (backflush)	Automatic teat cup cleaning and disinfection
Alternating pulsation	Sequential pulsation (each quarter consecutively, counterclockwise)
Pulsation rate: 60:40	Pulsation rate: 65:35
Without periodic air inlet	With periodic air inlet (BioMilker®)

When the trial began, the MUL was already in use on that farm for three months. Therefore, no matter what the day of lactation, the cows were familiar with both milking systems (Table 1). They were also familiar with all of the milking persons as they had been working there for at least ten months prior to the trial. Altogether, eleven milking persons were working on the farm, and all of them operated both the CON and the MUL. During one milking time, two milking persons always worked together, and they first performed the milking in the MUL followed by the CON. During the trial period, 21 milking times were analysed. Every milking person was present at 4 (median) milking times (minimum: 2, maximum: 7). For the entire trial, every milking person performed 13 (median) evaluable milkings in the CON (minimum: 3, maximum: 24) and 24 milkings in the MUL (minimum: 1, maximum: 35).

Video recordings and analysis

The video recording was performed using two analogue colour cameras (Exwave HAD, SSC-DC 58 AP, Sony Electronics Inc., San Diego, CA, USA, lens: 4–10 mm/F1.8) in each parlour, placed above the milker pit to film the cows from the side. One milking box was recorded with one camera, and the camera was placed in such a position to enable the best possible view of the cow. However, the number of analysed cows was limited to the cows that happened to get into those two boxes per milking system and milking time. As a result, the number of available cows in each milking system was lower than the total number of cows per group, with 32 cows analysed in the CON and 36 cows in the MUL. A multiplexer (DPLEX-16-ECO, Ganz, CBC Corp., Commack, NY, USA) and a video recorder (HS-1024, Mitsubishi Electric Corp., Tokyo, Japan) were used to save the videos. The data for one morning and one afternoon milking per week were saved. However, for the behaviour analysis, only the milking times without disturbances (e.g., unknown persons in the parlour or poor visibility of the cow in the video) were used. Altogether, videos of 104 milkings in the CON and 177 milkings in the MUL were analysed using continuous sampling. The identification of the cows (which cow in which milking box) and the milk yield was achieved using herd management software (Dairy-Plan, GEA Farm Technologies, Bönen, Germany). The identity of the milking person was established by the use of videos for each milking event of interest.

The following traits were analysed qualitatively or quantitatively through the use of the videos. All traits, except the *time point of restless body posture* and the *udder preparation duration,* were observed in the time span from attaching the first teat cup until the automatic removal of the milking cluster (CON) or the 4 teat cups (MUL):

– *head posture* (calm or restless presence, whereby the head posture was defined as calm when the cow held her head above in one position almost all the time and as restless if she often changed the position of her head), *body posture* (calm or restless presence, whereby calm was defined as a cow with less movement and restless as a cow with an overall nervous appearance, often signalled by beating of the tail) and *time point of restless body posture* (before the start of milking, during the main milking phase or a constant restless body posture if the cow showed the restlessness during the whole stay in the milking box);

– *elimination* (number of defecation and urination events during the time in the milking box, counted separately);

– *rumination* (whether any rumination behaviour or no rumination behaviour was displayed by a cow during the stay in the milking box);

– *kick-off of teat cup or milking cluster* (a kick of the cow caused the equipment to fall off) or *loss of teat cup or cluster* (the cup or cluster fell off before the regular end of milking without an obvious cause, whereby this was signalled when even one cup fell off and/or if the milking person came to attach the cluster again);

– *number of steps* (defined as shifting weight from one hind leg to the other or the lifting of one leg forward, backward or sideways, with a low lifting of the leg) and *number of kicks* (defined as a higher, powerful lifting of the leg, often directed at the udder, milking cluster or milking person);

– *udder preparation duration* (time from the first touch of the udder by the milking person until the attachment of the first teat cup);

– *milking duration* (time from attaching the first teat cup until the automatic removal of the milking cluster, whereby in the CON and MUL, all teat cups stayed on the cow until milking was complete for all quarters).

Unlike udder preparation duration, milking duration is expected to be influenced by milk yield; hence, both traits were analysed separately instead of analysing total milking duration.

All of the behavioural traits were observed by using video techniques, so the animals were not disturbed during the observation. Only one observer performed the video analysis to ensure reliable results. This person was already familiar with the analysis of videos.

The milking of the cows in both of the milking parlours was a part of the normal daily routine. Therefore, the study complies with the current laws relating to animal ethics.

Statistical analyses

SAS 9.3 software (SAS Institute Inc. Cary, NC, USA) was used for the statistical analyses. The influence of the cows, the milking system and the milking person were tested on the observed traits by using generalised linear mixed models with the GLIMMIX procedure. Depending on the trait, additional factors and covariables were included and were specified with the relevant traits (see below).

Binary (or Bernoulli) distribution with a Logit link function was used for analysis of binary traits such as head and body posture, rumination, cluster or teat cup kick-off and cluster or teat cup loss as well as udder preparation (0 usually referring to "calm posture" or "no event" and 1 to a "restless posture" or an "occurring event", such as teat cup loss or kick-off, except udder preparation where 0 refers to the event "no preparation" and 1 refers to "udder preparation happened"). The number of steps, number of kicks, udder preparation duration and milking duration were discrete count data and therefore were analysed using a Poisson distribution with a Log link function. For milk yield, a normal distribution with an identity link was used. For all traits, the basic model equation for the linear predictor η was as follows:

$$\eta_{ijk} = \mu + MS_1 + (MS \times COW)_{ij} + (MS \times MP \times COW)_{ijk} + \varepsilon_{ijk}$$

where:

μ as the general mean,

MS_i as the fixed effect of the i-th milking system (i=1, 2),

$(MS \times COW)_{ij}$ as the random effect of cow j nested in milking system i (j=1, …, 68),

$(MS \times MP \times COW)_{ijk}$ as the random effect of the interaction between cow j, and milking person k nested in milking system i (k=1, …, 11),

ε_{ijk} as the random residual.

The milking duration (in seconds) was added to the model for the number of steps and number of kicks traits as a covariable to test the hypothesis that there were more steps or kicks when there was more time available. For the trait milking duration, the milk yield (in kg) was added to the model as a covariable. The model for milk yield was extended with fixed effects for lactation with m=3 levels for 1st, 2nd and 3rd or higher lactation, test day milkings with n=21 levels and days in milk as a covariable. Because all cows were at least 150 days in milk during the trials, a simple linear approach was considered for the decrease of milk yield per milking during that stage of lactation.

For all cows in the CON, between 1 and 8 milkings were available (with a median of 3, with 8 of the 32 cows in the CON where only 1 milking was available), while for all cows in the MUL, there were between 2 and 9 milkings per cow available (with a median of 5 milkings for the 36 cows in the MUL). Those repeated measurements were included with appropriate variance-covariance matrices in the models for the traits udder preparation and step count (with a standard variance components structure in which the milkings were considered independently for those traits) as well as milking duration (with a spatial power structure based on the number of milkings between milkings with camera observation) to account for variability within a cow. The models that considered the repeated measurements did not converge for the other traits. In those cases, the model without repeated measurements was used while considering the residual effect was inflated by the within cow variability.

Unlike the binary traits, where the model was predetermined, model assumptions were checked by the visual inspection of the conditional residuals (i.e., including random effects) for the models with count data, durations and milk yield.

A Spearman rank correlation coefficient (r_s) between the traits was calculated using the CORR procedure.

The null hypotheses for all of the tests were that there is no significant factor influence and the significance level for the hypothesis tests was established at 5%.

Results

The comparison of the CON and the MUL with regard to behavioural and milking traits is shown in Table 3.

When comparing the *head* and *body posture*, the cows behaved similarly in both systems with no significant difference between the CON and the MUL (*head posture*: P=0.38, *body posture*: P=0.85). Most of the cows (89%) showed a calm head posture during the CON milking process, whereas 11% showed a restless head posture. The proportions of calm and restless head postures during the MUL milking process were 94% and 6%, respectively. As for the body posture, the proportion of restless cows was always higher (CON: 65%, MUL: 60%) than the proportion of calm cows. The *time point of the restless body posture* was also analysed, with nearly all of the cows (98%) in a calm state before the start of milking. The time point of restlessness was mostly (CON: 42%, MUL: 35%) during the main milking phase. The cows with a constant restless body posture during milking were observed more in the CON (9%) than in the MUL (4%).

Elimination behaviour during the stay in the milking box was rarely observed. Neither defecation nor urination was observed during the time in the CON milking box. Defecation occurred during four milking times using the MUL (2% of all MUL milkings), but there was no urination by the cows.

Table 3. Results for Type III tests of fixed effects by trait

Trait	Factor	Numerator DF	Denominator DF	F Value	Pr > F
Head posture	MS	1	66	0.78	0.3814
Body posture	MS	1	66	0.04	0.8477
Kick-off of teat cup/cluster	MS	1	66	2.81	0.0985
Loss of teat cup/cluster	MS	1	66	0.01	0.9089
Number of steps	MS	1	66	0.42	0.0778
	MD	1	57	3.22	0.5631
Number of kicks	MS	1	66	0.34	0.5631
	MD	1	57	3.1	0.0835
Udder preparation	MS	1	66	0.02	0.8834
Udder preparation duration	MS	1	64	15.93	0.0002
Milking duration	MS	1	65	6.95	0.0105
	MY	1	57	26.45	<0.0001
Milk yield	MS	1	62	6.24	0.0152
	LN	2	37	3.11	0.0566
	Mdate	20	37	1.67	0.0878
	DIM	1	37	12.15	0.0013

MS=milking system, MD=milking duration, MY=milk yield, LN=lactation, Mdate=milking date, DIM=days in milk.

The proportion of cows exhibiting *rumination* during milking was higher in the MUL (60%) than in the CON (45%), but the difference between the two milking systems was not significant (P=0.09).

The *kick-off* and *loss of teat cup or milking cluster* also showed no significant differences between the two systems (P=0.10 and P=0.91, respectively) and was observed only rarely. The kicking off of a teat cup or milking cluster occurred in 2.5% of all CON milkings and in 6.4% of all MUL milkings, with the percentage of losses higher in the CON (1.7%) than in the MUL (1.1%).

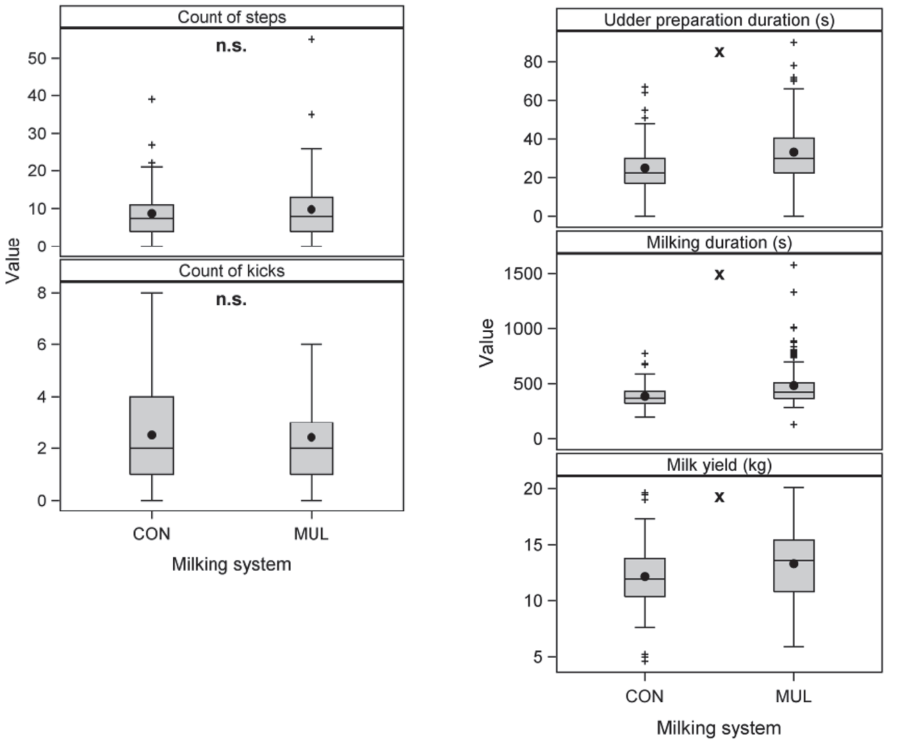
There was no significant difference between the CON and the MUL regarding the *number of steps*, the *number of kicks* and the *udder preparation* (P=0.08, P=0.56 and P=0.88, respectively). However, there was a significant difference in the *udder preparation duration* (P<0.01), the *milking duration* (P=0.01) and the *milk yield* (P=0.02) between the CON and the MUL (Table 3), whereby the respective values were always higher in the MUL compared with the CON. The distribution of values is shown in Figure 1.

Kicking behaviour was rarely observed, and there were no kicks observed during the stay in the milking box in 63% of the CON milkings and in 75% of the MUL milkings.

Table 4. Covariance parameters (Estimate ± Standard Error) for the different statistical models by trait in the conventional milking system (CON) and the individual quarter milking system (MUL)

Milking system	CON				MUL				
Covariance parameter	COW	COW×MP	RM COW	ρ	COW	COW×MP	RM COW	ρ	Residual
Trait									
Head posture	5.94±3.59	9.88±2.89			0	11.88±2.05			0.06±0.01
Body posture	2.29±1.20	1.37±0.89			0.66±0.47	1.56±0.70			0.62±0.10
Kick-off of teat cup/cluster	0.02±10.33	11.96±10.92			1.38±2.12	11.11±2.55			0.04±0.004
Loss of teat cup/cluster	0	9.71±3.61			0.61±6.87	15.68±7.45			0.01±0.001
Number of steps	0.31±0.12	0.07±0.06	1.28±0.39		0.17±0.07	0.23±0.06	1.14±0.27		0.42
Number of kicks	2.19±0.95	1.13±0.42			0.57±0.48	2.47±0.54			0.33±0.04
Udder preparation	0.27±1.88	7.84±2.49			0	8.63±1.47			0.19±0.02
Udder preparation duration	0	0	2.68±2.06		0.01±0.02	0.02±0.03	2.68		2.34±0.96
Milking duration	0.02±0.01	0	0	1	0.07±0.02	0	14.81±2.89	–0.24±0.32	7.30±1.21
Milk yield	4.66±1.51	1.14±0.71			4.48±1.28	1.25±0.59			1.57±0.40

MP=milking person, RM=repeated measurements.



n.s.: not significant (P>0.05)
x: significant (P≤0.05)

Figure 1. Distribution of values of selected traits for the conventional milking system (CON) and the individual quarter milking system (MUL)

The results for the covariance parameter estimation from the statistical models for the different traits can be found in Table 4. These measures were carried out to determine how important the influences of the cows and milking persons on various traits were.

The head posture variability in both milking systems was highest for the interaction between the cows and milking persons. Regarding the body posture, the variability between cows was higher than the variability between the cows and milking persons in the CON; while for the MUL, the opposite was true. The source of variability for the kick-off or loss of the teat cup or cluster as well as the occurrence of udder preparation comprised mostly the interactions between the cows and milking persons in both milking systems. The influence of the milking person on the variability in the number of steps, udder preparation duration, milking duration or milk yield was lower when compared to residual variability. The estimates for the within-cow variability were available for those traits by including repeated measurements into the statistical model, and in these four traits, they accounted for the major part of variance. For the number of kicks, the variability between cows was higher than for the interaction between the cows and milking persons in the CON, while the opposite was displayed in the MUL. The major source of variance in the milk yield was the variability between cows in both milking systems.

All of the milking persons, except one, spent more time preparing the udders for MUL than CON. There were also milking operations without udder preparation; however, the milk yield was not significantly different (P=0.36) between the milkings without (n=44) and with (n=263) udder preparation.

An analysis of the relationship between the *udder preparation duration* and the *milking duration* showed no significant correlation (P>0.05) for the MUL, whereas a significant negative correlation existed between both traits for the CON (r_s=–0.27; P<0.01).

Conversely, significant positive correlations were found between the *milking duration* and *step count* (r_s=0.32; P<0.01) and between the *milking duration* and *milk yield* (r_s=0.29; P<0.01). However, an analysis of the milking duration and corresponding milk yield (according to the rule of Hamann, 2001) showed that the milking duration was excessive in both systems. The mean difference between the analysed and the ideal milking duration was 1.5 min for the CON and 2.7 min for the MUL according to the corresponding milk yield.

Discussion

The results from this study showed only minor differences between the two milking systems, the CON and the MUL, and the analysed behavioural traits showed no significant differences between the two systems. The present study was the first investigation of the influence of an individual quarter milking system in a conventional milking parlour on the behaviour of the cows. However, the results are comparable to investigations of individual quarter milking systems in automatic milking systems (AMS) in regard to the influence of the milking system on behavioural and milking traits. Hagen et al. (2004) could not find indications of more stress in an AMS than in a conventional milking system (herringbone milking parlour). However, we agree with Gygax et al. (2008) and Hopster et al. (2002) who concluded that only minor differences can be found between a conventional and an individual quarter milking system (AMS in those studies) and that the cows showed relatively low behavioural responses. In contrast, Wenzel et al. (2003) found higher levels of restlessness behaviour, such as stepping and kicking, in an AMS than in an auto-tandem milking parlour but could not identify the reasons for these observations. Thereby, the experimental design should be taken into account: Hagen et al. (2004) observed cows in first or second lactation who had experience of one system only and were at least 8 days in lactation; Gygax et al. (2008) used multiparous cows and milking systems which were in use for at least 6 months; Hopster et al. (2002) used only primiparous cows with 4 weeks of adaptation and no experience with either milking system; whereas Wenzel et al. (2003) used cows between first and fifth lactation who were familiar with the milking parlour, but at least 2 months adapted to the AMS. The relevance of the parity was already studied by Szentléleki et al. (2015), who observed significant differences between primiparous and multiparous cows in behaviour (leg movements scored by direct human observation on a 5-point-scale) during udder preparation, but no significant differences in behaviour during milking (herringbone milking parlour). Furthermore, the cow traffic of the AMS groups was different between and within the mentioned studies. Cows either had to pass through the AMS to get from the resting area to the feeding area (guided cow traffic: Gygax et al., 2008; Hagen et al., 2004; Hopster et al., 2002) or could freely alternate between both areas (free cow traffic: Gygax et al., 2008; Wenzel et al., 2003). Studies about the influence of cow traffic on the behaviour of dairy cows in an automatic milking system are rare and described more the behaviour outside the AMS, but significant differences were already shown. Cows spend more time in the feeding area, spend more time with standing and made less journeys from the lying to the feeding area when cow traffic was guided. Furthermore, the frequency of visits to the AMS may be increased through guided cow traffic (Hermans et al., 2003; Ketelaar-de Lauwere et al., 1998). However, in the present study it was also a challenge to create a reliable experimental design. The cows were in their first to seventh lactation. Cows milked with the CON were familiar with this milking system and cows milked with the MUL were 3 months adapted to it. All cows were familiar with all of the milking persons, but differences in their daily working routine were not avoidable. Therefore, differences in experimental designs could be one possible explanation for differences between study results.

The present study showed that the milking persons had a major influence regarding the head posture of the cows, the kick-off and loss of teat cups and milking clusters as well as on the occurrence of udder preparation. During the milking times in the MUL, there was also a significant influence of the milking person on the body posture of the cows and the number of kicks. Although the behaviour of the milking persons towards the cows was not analysed, it seemed that the cows behaved differently depending on the type of treatment by the milking person. This finding is also described by Hötzel et al. (2005) as well as by Munksgaard et al. (2001), who observed that cows can discriminate between aversive and neutral/gentle individuals. Rushen et al. (2001) also showed that human contact reduces some behavioural signs of agitation. However, the studies found no significant effect between the presence of an aversive handler at milking (Munksgaard et al., 2001) or the milking person itself (Hötzel et al., 2005; Rushen et al., 2001) on the milk yield. This is in accordance with our results, where the influence of the cows was higher than the influence of the milking persons on the milk yield. In addition, different behavioural reactions of the cows during milking can also be explained as their individual direct response to environmental stimulus during milking as described by Szentléleki et al. (2015) as the difference in cows' milking temperament. This individual milking temperament could also be influenced by the type of habituation to the milking routine itself, as shown by Kutzer et al. (2015). In their study, heifers were habituated prepartum to the milking parlour, and this habituation results in less restlessness behaviour, such as stepping and kicking during the udder preparation phase.

The analysis of the videos in the present study showed significant differences in the performing of udder preparation among the milking persons. The milking operations without udder preparation had no significant effect on the milk yield using the MUL and the CON or on the milking duration of the MUL, whereas a significant negative correlation existed between the udder preparation duration and the milking duration for the CON (r_s=–0.27; P<0.01). Therefore, for the CON, we are in agreement with Bruckmaier and Blum (1996), who compared dairy cow milkings with and without prestimulation in a conventional milking system. They concluded that the milk yield is not significantly lower in the milkings without stimulation but that milking without stimulation leads to a prolonged machine-on time, confirming our results for the CON. Furthermore, Bruckmaier and Blum (1996) showed that the milk flow curves were generally bimodal without stimulation. A negative influence of a lack of teat preparation on milking characteristics (average milk flow rate and milking duration) is also described by Kanswohl et al. (2007), who concluded that a teat preparation of 60 s was sufficient, whereas times markedly under or over 60 s had a negative influence on the milking characteristics. Similar results were also described by Tančin et al. (2007), who showed a positive influence of pre-stimulation on the milk flow pattern and a reduction in the milking time.

The udder preparation duration was longer (P<0.01) in the MUL than in the CON. Although there was no obvious reason for this, it appeared to be attributable to the engagement of the milking persons or the MUL function (positioning of the milking cluster and teat cups under the udder) and construction of the MUL-milking parlour (e.g., less milking stations per side, the position of the teat-cleansing tissues and water hoses).

Furthermore, there was a significant difference in the milking duration (P=0.01) between the CON (mean ± SD: 386 ± 93 s) and the MUL (mean ± SD: 483±195 s) by almost 100 seconds. This tendency was also described by Müller et al. (2011), whose study was conducted in the same milking parlours with a similar number of cows, though the mean milking duration in their investigation was, overall, longer (mean ± SD of CON: 439±108 s, mean ± SD of MUL: 514±155 s) than in the present study. An explanation for this difference between the studies could be that Müller et al. (2011) used data from more milking operations (CON: n=497, MUL: n=556) and used the milking duration according to herd management software, whereas the present study determined the milking durations by video observations. Differences in milking duration in the present study as well as in the study of Müller et al. (2011) could also be explained through the differences in the milking equipment (Table 2). In comparison to the machine vacuum of 40 kPA in the CON, the lower vacuum under the teat due to periodic air inlet (BioMilker®) in the MUL (less than the 37 kPa of machine vacuum) can reduce milk flow. Additionally, through the sequential pulsation (MUL), milk flow is more regular than with alternating pulsation (CON) and could be an explanation for the longer milking duration observed in the MUL. However, according to the corresponding milk yield, the milking durations of both CON and MUL were longer than the ideal milking duration, but the milking duration for the MUL differs more from the ideal milking duration than for the CON in comparison to the general rule of Hamann (2001) with regard to the milking-induced mastitis risk.

Furthermore, other reasons for differences in milking duration must be taken into account like, for example, animal individual differences in milk yield and milk flow. Although the cows were randomly distributed to the two groups and milk yield was added as a covariable in the model, the experimental design could have been improved through measurements of the individual milk flow. The use of the same cows in both milking systems (cross-over design) would have been another improvement and should be applied in further studies, but was not possible under the practical conditions of the present study.

The difference in the milk yield (P=0.02) between the CON (mean ± SD: 12.2±2.9 l) and the MUL (mean ± SD: 13.3±2.9 l) was also significant as well as the difference in the milk yield between different days in milk (P<0.01), with the latter in accordance with the study of Müller et al. (2011). Another result of the present study showed that a longer milking duration is positively correlated with a higher number of steps and with a higher milk yield, as was expected.

In conclusion, the behavioural traits showed no significant differences between the two milking systems, the CON and the MUL, and no signs of strain were observed during the milking operations in either system. Therefore, the milking of cows using the CON and the MUL did not notably influence the behaviour of dairy cows, although the udder preparation and milking for the MUL were of significantly longer durations than for the CON. However, the milking person appeared to have a greater impact on the cows than the milking system, especially concerning their head posture, the kick-off of teat cups and milking clusters, the loss of teat cups and milking clusters, and furthermore, in the MUL, also on their body posture and the number of kicks. Performing of udder preparation was also significantly different among the milking persons. Although the milk yield was not significantly different among the milking persons, the results showed an impact of the milking person on the behaviour of the dairy cows.

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