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## Comparison between automatic and conventional milking systems for milk coagulation properties and fatty acid composition in commercial dairy herds

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### ABSTRACT

The aim of this study was to investigate the effect of milking dairy cows using conventional milking parlour (CMP) and automatic milking system (AMS) on milk coagulation properties and fatty acid (FA) composition. Milk coagulation traits were rennet coagulation time, curd-firming time and curd firmness. Data consisted of 10,476 individual milk samples collected from 918 Holstein–Friesian cows in 8 herds: four herds milked cows using a CMP and four using an AMS. A linear mixed model was used to investigate sources of variation for milk yield, traditional quality traits, coagulation properties and FA profile. On average, cows from AMS produced 1 kg/d more milk than cows from CMP. Rennet coagulation time was slightly longer (+1.2 min) and free FA content was greater (+0.16 mmol/100 g milk fat) in milk from cows milked in AMS than CMP. Overall, the milking system did not affect the FA concentration of milk. Results of the present study suggest that AMS can be adopted without detrimental effects on milk features.

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### KEYWORDS

Fatty acid; mid-infrared spectroscopy; milk acidity; milk technological trait; milking system

### Introduction



Automatic milking systems (AMS) have been available in Europe since the beginning of 1990s. The major advantages of AMS are the reduction of labour for milking (Dijkhuizen et al. 1997) and the enhanced production per cow due to higher milking frequency than conventional milking parlour (CMP) (Klei et al. 1997). Milk yield increases from 2% to 8% (Svennersten-Sjaunja & Pettersson 2008) and labour decreases by about 18%.

The effect of AMS on milk quality traits is negligible (Svennersten-Sjaunja & Pettersson 2008), whereas increasing milking frequency increases free fatty acids (FFA) content (Klei et al. 1997; Klungel et al. 2000; Wiking et al. 2006), which has a negative impact on milk flavour (Shipe et al. 1980) and cheese-making process (Sapru et al. 1997). The effect of AMS on somatic cell count (SCC) is contradictory; several authors reported an increase in SCC after the introduction of AMS (Kartinen et al. 1990; Rasmussen et al. 2002), whereas other studies reported no effect of AMS on SCC (Klungel et al. 2000; Berglund et al. 2002; Mollenhorst et al. 2011).

Italy has a long tradition in producing high-quality cheeses and thus milk coagulation properties (MCP),

protein composition, acidity and mineral composition are crucial to improve profitability of the dairy industry. Milk coagulation properties have been widely investigated (Pretto et al. 2013; Penasa et al. 2014) and proposed as innovative technological traits to enhance the efficiency of the dairy sector (De Marchi et al. 2013; Tiezzi et al. 2013). Moreover, mid-infrared spectroscopy (MIRS) prediction models have been implemented in several Italian milk quality laboratories to routinely predict MCP (De Marchi et al. 2014). The accuracies of these prediction models implemented by Foss (Foss Electric A/S, Hillerød, Denmark) were reported by Penasa et al. (2014) and Penasa et al. (2015) for MCP and fatty acid (FA) composition, respectively.

Although several researches have been focussed on the effect of AMS or milking frequency on milk chemical composition and SCC, very limited information is available on the effect of AMS on milk technological traits. Abeni et al. (2003) and Innocente and Biasutti (2013) did not find any significant differences for technological properties between milk derived from AMS and CMP. The aim of this study was to investigate the effect of CMP and AMS on MCP and FA composition, as predicted by MIRS, in Holstein–Friesian (HF) dairy cows.

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## Materials and methods

### Data and editing

Individual milk samples from HF cows were collected between September 2011 and July 2014, as part of the official monthly milk recording, by the Breeders Association of Veneto Region (Padova). Samples were from 8 dairy herds located across the Veneto Region (northeast Italy). Cows were milked in 4 herds using a CMP and in 4 other using an AMS. Herds selected for this study were commercial farms of comparable size and their management, feeding conditions and production area were similar. Briefly, cows were reared in indoor barn with access to exercise paddock and diet was based on corn silage (no grazing pasture) and concentrates. Farms with AMS had DeLaval (VMS, DeLaval, Tumba, Sweden) group milking system equipped with Milko-Scope MKII for milk sampling.

Samples were processed according to International Committee for Animal Recording (ICAR 2014) procedures, combined with preservative immediately after collection (Bronopol; Knoll Pharmaceuticals, Nottingham, UK) and analysed in the laboratory of the Breeders Association of Veneto Region (Padova, Italy). Traditional milk quality traits (protein, fat, casein and lactose percentages), and pH, MCP and FA composition were assessed using Milko-Scan FT6000 (Foss Electric A/S, Hillerød, Denmark). Prediction models were implemented for routine prediction of MCP, FFA and FA composition, as reported by Tiezzi et al. (2013) and Penasa et al. (2015).

A production record consisted of cow identity code and parity, dates of calving and sampling, herd code and the following traits: milk yield (MY, kg/d), fat percentage (FP, %), protein percentage (PP, %), casein percentage (CP, %), lactose percentage (LP, %), SCC (n/mL), pH, rennet coagulation time (RCT, min), curd-firming time ( $k_{20}$ , min), curd firmness ( $a_{30}$ , mm), saturated FA (SFA, g/100 g milk), unsaturated FA (UFA, g/100 g milk), monounsaturated FA (MUFA, g/100 g milk), polyunsaturated FA (PUFA, g/100 g milk), FFA (mmol/100 g milk fat) and four single FA, namely C14:0, C16:0, C18:0 and the sum of C18:1 (g/100 g milk). Somatic cell count was log-transformed to somatic cell score (SCS) using the following formula:  $SCS = 3 + \log_2(SCC/100,000)$ .

Cows were retained if they were from parity 1 to 9 and from 5 to 480 days in milk. Moreover, lactations with less than 3 tests and herd-test-date (HTD) with less than 3 cows were removed from the dataset. After editing procedure, 10,476 records from 918 cows were available for statistical analysis. The average herd size during the period of the study was 115

(range: 81–179), and the average number of records per cow and HTD was 11 (range: 3–27) and 53 (range: 21–102), respectively.

### Statistical analysis

A preliminary analysis indicated that all studied variables were normally distributed and thus a linear mixed model (SAS Institute Inc., Cary, NC) was used to investigate sources of variation for MY, quality traits, MCP and FA composition:

$$y_{ijklm} = \mu + MS_i + PAR_j + DIM_k + (MS \times PAR)_{ij} + (MS \times DIM)_{ik} + (PAR \times DIM)_{jk} + HTD_l(MS_i) + cow_m + e_{ijklm},$$

where  $y_{ijklm}$  is the dependent variable,  $\mu$  is the overall intercept of the model,  $MS_i$  is the fixed effect of the  $i$ th milking system ( $i = \text{CMP, AMS}$ ),  $PAR_j$  is the fixed effect of the  $j$ th parity of the cow ( $j = \text{first, second, third, fourth and fifth and later parities}$ ),  $DIM_k$  is the fixed effect of the  $k$ th class of stage of lactation of the cow ( $k = 1-12$ , the first being a class from 5 to 35 d, followed by 9 classes of 30 d each, and the last two being classes of 45 and 130 d, respectively),  $(MS \times PAR)_{ij}$  is the fixed interaction effect between milking system and parity,  $(MS \times DIM)_{ik}$  is the fixed interaction effect between milking system and DIM,  $(PAR \times DIM)_{jk}$  is the fixed interaction effect between parity and DIM,  $HTD_l(MS_i)$  is the random effect of the  $l$ th herd-test-date ( $l = 1-197$ ) nested within the  $i$ th milking system  $\sim N(0, \sigma_{HTD(MS_i)}^2)$ ,  $cow_m$  is the random effect of the  $m$ th cow ( $m = 1-918$ )  $\sim N(0, \sigma_{cow}^2)$ , and  $e_{ijklm}$  is the random residual  $\sim N(0, \sigma_e^2)$ . Significance of milking system effect was tested on the herd-test-date within milking system variance. A multiple comparison of means was performed for the main effect of milking system, and for interactions between milking system and days in milk, and milking system and parity using Bonferroni's correction ( $p < .05$ ).

## Results

### Descriptive statistics

Milk yield, FP, PP, CP and SCS averaged 30.2 kg/d, 3.78%, 3.38%, 2.64% and 2.84, respectively. Means of RCT,  $k_{20}$  and  $a_{30}$  were 22.9 min, 6.80 min, and 23.1 mm, and exhibited coefficients of variation of 26, 32 and 42%, respectively (data not shown). Saturated FA, UFA, MUFA and PUFA averaged 2.71, 1.18, 0.93 and 0.06 g/100 g milk, respectively. Among individual FA, C16:0 was the most important (1.17 g/100 g milk), followed by the sum of C18:1 (0.73 g/100 g milk), C14:0

(0.41 g/100 g milk) and C18:0 (0.34 g/100 g milk). Free FA averaged 0.57 mmol/100 g milk fat (data not shown), which is very close to the value (0.50 mmol/100 g milk fat) considered normal for fresh milk.

### Significance of fixed effects

The significance of fixed effects included in the statistical analysis is reported in Table 1. Stage of lactation was the most important factor ( $p < .001$ ) in explaining part of the variation of all studied traits, followed by parity, which was relevant for most variables, except for FP, SFA, UFA, C16:0, C18:0 and FFA. Milking system significantly affected ( $p < .05$ ) MY, pH, RCT, C16:0 and FFA. Among interaction effects, parity  $\times$  stage of lactation was the most important to explain the variability of analysed milk characteristics, except for fat, SFA, C14:0, C16:0 and FFA. Milking system  $\times$  stage of lactation effect had a strong influence ( $p < .05$ ) on MY, PP, CP, pH,  $a_{30}$  and FFA. Finally, milking system  $\times$  parity effect was important ( $p < .05$ ) for MY, PP, CP and SCS (Table 1).

Cows milked by AMS produced more milk (+1 kg/d;  $p < .05$ ) compared with those milked by CMP (Table 1). Milk from cows milked by AMS had greater but not significant SCS (3.17 vs. 3.03), slightly higher pH (6.64 vs. 6.63;  $p < .05$ ) and slightly longer RCT (23.9 vs.

22.7 min;  $p < .05$ ) than milk from CMP. Regarding FA composition, only C16:0 and FFA were significantly ( $p < .05$ ) different between AMS and CMP (Table 1). In particular, milk FFA content was greater for AMS than CMP (0.64 vs. 0.48 mmol/100 g milk fat;  $p < .05$ ). Milk yield across parities was greater for cows from AMS than CMP (Table 2) even if significance was obtained only for animals in third (32.4 vs. 31.3 kg/d;  $p < .05$ ) and fifth, and later lactations (31.7 vs. 29.1 kg/d;  $p < .05$ ).

The trend of MY, protein and  $a_{30}$  across lactation and milking system is depicted in Figures 1–3, respectively. Significant differences between AMS and CMP were detected until 170 days in milk, where cows milked by AMS yielded more milk than those milked

**Table 2.** Least squares means (standard errors within brackets) of milk yield, kg/d, for the interaction effect between milking system<sup>a</sup> and parity<sup>b</sup>.

Parity	MS	
	AMS	CMP
1	27.8 (0.4)	27.4 (0.4)
2	30.3 (0.4)	29.7 (0.4)
3	32.4 (0.4)*	31.3 (0.4)†
4	31.3 (0.5)	30.9 (0.5)
$\geq 5$	31.7 (0.5)*	29.1 (0.6)†

<sup>a</sup>MS: milking system; AMS: automatic milking system; CMP: conventional milking parlour.

<sup>b</sup>Least squares means with different symbols (\*,†) within a row are significantly different ( $p < .05$ ).

**Table 1.**  $p$ -Value<sup>a</sup> of fixed effects<sup>b</sup> included in the statistical analysis of milk yield, quality traits, coagulation properties and fatty acid composition, and least squares means (standard errors within parentheses) of the studied traits for milking system effect<sup>c</sup>.

Trait <sup>d</sup>	$p$ -Value						RSD <sup>e</sup>	MS	
	MS	PAR	DIM	MS $\times$ PAR	MS $\times$ DIM	PAR $\times$ DIM		AMS	CMP
Milk yield, kg/d	.042	<.001	<.001	.005	<.001	<.001	4.83	30.7 (0.4)*	29.7 (0.4)†
Quality traits									
Fat, %	.696	.319	<.001	.640	.434	.054	0.61	3.78 (0.04)	3.76 (0.04)
Protein, %	.328	<.001	<.001	.007	<.001	<.001	0.25	3.39 (0.02)	3.36 (0.02)
Casein, %	.399	<.001	<.001	.019	<.001	<.001	0.19	2.64 (0.01)	2.62 (0.01)
Lactose, %	.994	<.001	<.001	.378	.184	<.001	0.13	4.78 (0.01)	4.78 (0.01)
SCS	.163	<.001	<.001	.033	.633	<.001	1.42	3.17 (0.07)	3.03 (0.08)
pH	.041	<.001	<.001	.068	<.001	<.001	0.04	6.64 (0.01)*	6.63 (0.01)†
Coagulation properties									
RCT, min	.044	<.001	<.001	.988	.262	<.001	3.49	23.9 (0.4)*	22.7 (0.4)†
$k_{20}$ , min	.820	<.001	<.001	.521	.459	<.001	1.04	6.90 (0.22)	6.83 (0.23)
$a_{30}$ , mm	.233	<.001	<.001	.255	<.001	<.001	6.10	21.6 (0.7)	22.7 (0.6)
Fatty acids, g/100 g milk									
SFA	.386	.290	<.001	.865	.065	.287	0.44	2.72 (0.03)	2.68 (0.03)
UFA	.530	.887	<.001	.634	.229	<.001	0.23	1.18 (0.02)	1.20 (0.02)
MUFA	.151	<.001	<.001	.678	.262	<.001	0.17	0.91 (0.01)	0.94 (0.01)
PUFA	.919	<.001	<.001	.553	.371	.007	0.02	0.06 (0.01)	0.06 (0.01)
C14:0	.305	<.001	<.001	.528	.218	.377	0.06	0.41 (0.01)	0.40 (0.01)
C16:0	.039	.226	<.001	.234	.297	.945	0.21	1.19 (0.02)*	1.14 (0.02)†
C18:0	.051	.984	<.001	.353	.580	.011	0.08	0.35 (0.01)	0.34 (0.01)
$\Sigma$ C18:1	.652	.001	<.001	.770	.474	.006	0.17	0.73 (0.01)	0.74 (0.01)
FFA, mmol/100 g milk fat	.008	.913	<.001	.753	.016	.119	0.21	0.64 (0.04)*	0.48 (0.04)†

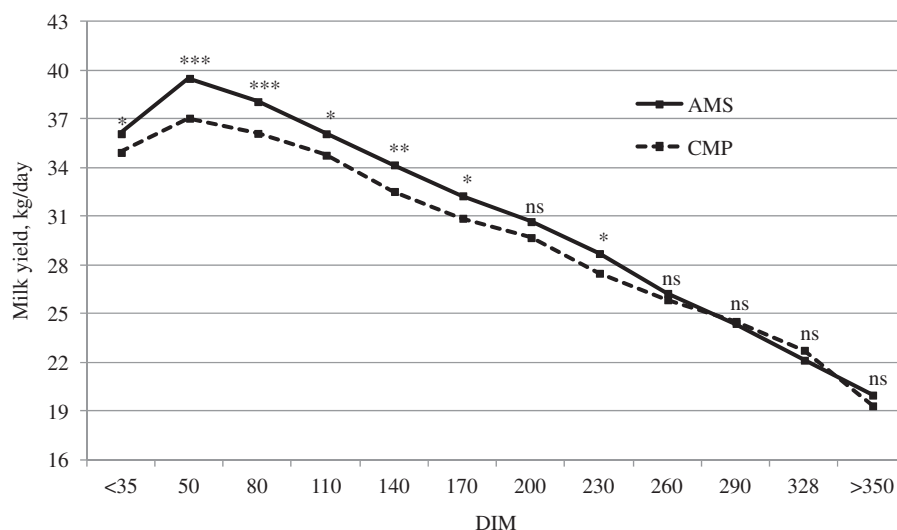
<sup>a</sup>Statistical significance was set to  $p < .05$ .

<sup>b</sup>MS: milking system; PAR: parity; DIM: stage of lactation. Significance of MS effect was tested on the herd-test-date within MS variance.

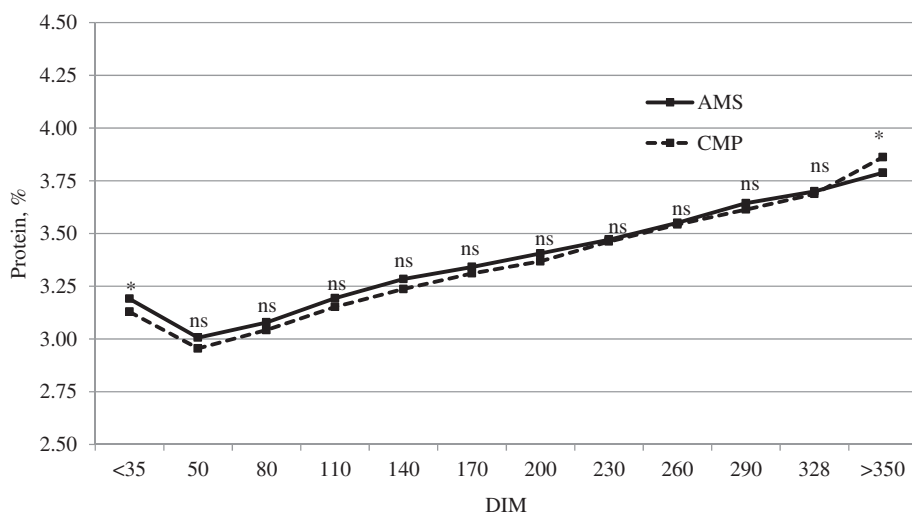
<sup>c</sup>AMS: automatic milking system; CMP: conventional milking parlour. Least squares means with different symbols (\*,†) within a row are significantly different ( $p < .05$ ).

<sup>d</sup>SCS: somatic cell score; RCT: rennet coagulation time;  $k_{20}$ : curd-firming time;  $a_{30}$ : curd firmness; SFA: saturated fatty acids; UFA: unsaturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids; FFA: free fatty acids.

<sup>e</sup>RSD: residual standard deviation.



**Figure 1.** Least squares means of milk yield for the interaction effect between milking system (AMS: automatic milking system; CMP: conventional milking parlour) and days in milk (DIM). Significance of the differences between least squares means of AMS and CMP within each class of DIM is reported above the solid line. ns, not significant; \* $p < .05$ ; \*\* $p < .01$ ; \*\*\* $p < .001$ .



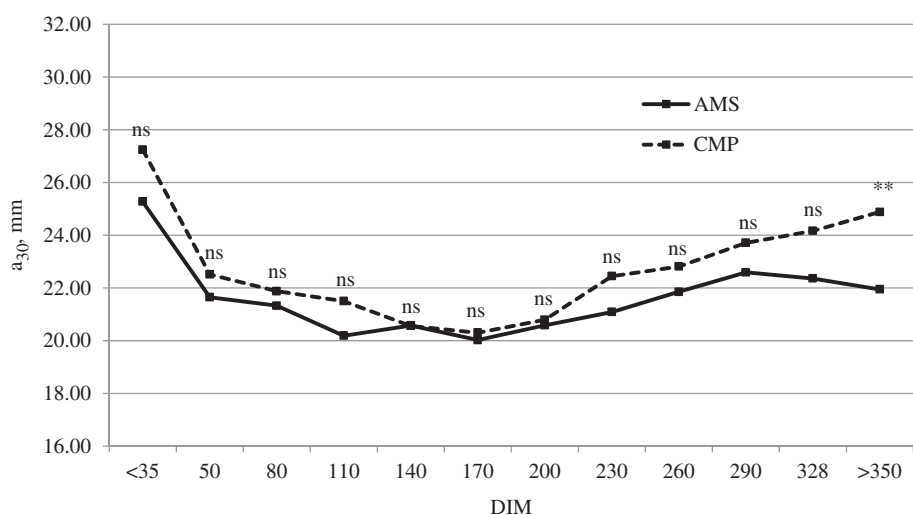
**Figure 2.** Least squares means of protein percentage for the interaction effect between milking system (AMS: automatic milking system; CMP: conventional milking parlour) and days in milk (DIM). Significance of the differences between least squares means of AMS and CMP within each class of DIM is reported above the solid line. ns, not significant; \* $p < .05$ .

with CMP. The most pronounced differences between the two milking systems were around the peak of lactation (Figure 1). Concerning protein percentage, no statistical differences between AMS and CMP were detected across days in milk, with the exception of the beginning and end of lactation where milk of cows milked with AMS had greater and lower protein percentage than milk of cows milked with CMP, respectively (Figure 2). Overall, cows milked by CMP produced milk with better  $a_{30}$  compared with AMS but significance was found only at the end of lactation (Figure 3). The difference between AMS and CMP for FFA across days in milk is presented in Figure 4. Cows milked with AMS produced milk with greater FFA

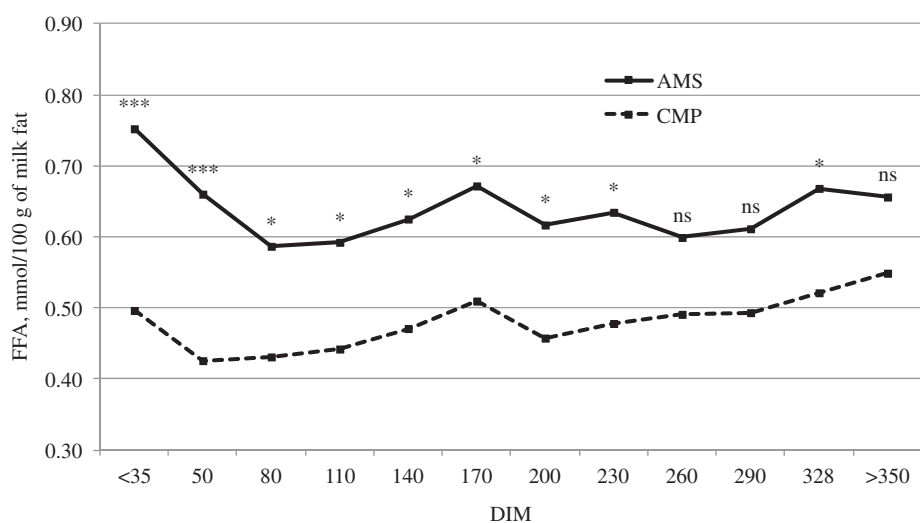
content across lactation. The greatest difference between AMS and CMP was detected within the first 80 days after calving, whereas the smallest differences were observed after 260 days in milk.

## Discussion

In North Italy about half of cows are reared in small-to-medium size herds (<50 cows). The potential economic benefit of the introduction of AMS in small-to-medium herds is an attractive perspective for farmers as discussed by Rotz et al. (2003) in a study which aimed at evaluating the interaction of AMS with farm size and animal production level. As previously



**Figure 3.** Least squares means of curd firmness ( $a_{30}$ ) for the interaction effect between milking system (AMS: automatic milking system; CMP: conventional milking parlour) and days in milk (DIM). Significance of the differences between least squares means of AMS and CMP within each class of DIM is reported above the dashed line. ns, not significant; \*\* $p < .01$ .



**Figure 4.** Least squares means of free fatty acids (FFA) for the interaction effect between milking system (AMS: automatic milking system; CMP: conventional milking parlour) and days in milk (DIM). Significance of the differences between least squares means of AMS and CMP within each class of DIM is reported above the solid line. ns, not significant; \* $p < .05$ ; \*\*\* $p < .001$ .

mentioned, the main advantages of AMS compared with CMP are the reduction of labour for milking and the increase of milk production.

### Effects of AMS on milk yield and quality traits

Cows from AMS produced 3.4% more milk per day than cows from CMP; this result may be the consequence of the direct relationship between milking frequency and milk secretion. Several authors demonstrated an increase of milk production (6 to 28%) with milking frequency (Hillerton et al. 1990; Knight et al. 1992; Klei et al. 1997; Stelwagen 2001; Løvendahl & Chagunda 2011). Traditional milk quality

traits were not affected by milking system, in agreement with the literature (Amos et al. 1985; Svennersten-Sjaunja et al. 2000; Abeni et al. 2005; Innocente & Biasutti 2013). Also, Klei et al. (1997) did not report any positive effect between two and three milkings per day for protein and fat percentages. Compared to CMP, the use of AMS increased MY in early lactation. Abeni et al. (2005) and Svennersten-Sjaunja and Pettersson (2008) reported significant interaction effects between milking system and stage of lactation from 91 to 180 days in milk, supporting findings from the present research. Concerning PP and CP, no information about the effect of AMS on these traits across lactation are available in the literature



for comparison. Concerning milk SCC, the effect of introducing AMS in the dairy herd is quite contradictory among studies; our results indicated no difference in SCS for cows milked with AMS and CMP, although they were numerically higher in AMS milk. An increase of milk SCC after the introduction of AMS was observed by several authors in bulk milk (Klungel et al. 2000; Rasmussen et al. 2001; Kruijper et al. 2002; de Koning et al. 2003).

To our knowledge, no information is available in the scientific literature to explain the interaction between milking system and parity. Our results demonstrated that cows of parity 5 or greater produced 2.6 kg/d more milk in AMS than CMP, and it can be hypothesised that cows from AMS and so milked more than two times per day maintain a better productive and health status respect to cows milked in CMP. The possibility for the cows to choose the time of milking could preserve the health of the udder, especially in high-productive animals.

### **Effects of AMS on milk pH and coagulation properties**

Milk from herds equipped with AMS had slightly worse technological features compared with milk from herds equipped with CMP; indeed, pH (6.64 vs. 6.63;  $p < .05$ ) and RCT (23.9 vs. 22.7 min;  $p < .05$ ) were greater in AMS than CMP. Milk acidity, and in particular pH, affects the rate of hydrolysis of k-casein by chymosin; this relationship confirms our results and was reported also by Bastian et al. (1991) who studied the effect of plasmin activity on clotting time and pH, and by several phenotypic and genetic studies carried out in Italian HF farms (De Marchi et al. 2013; Tiezzi et al. 2013; Penasa et al. 2014). Innocente and Biasutti (2013) did not detect significant differences between AMS and CMP for MCP determined by Formagraph in bulk milk samples collected in 14 farms and O'Brien et al. (2002) did not report significant effects on MCP by increasing milking frequency. The statistical analysis revealed that  $a_{30}$  was significantly affected by the interaction effect between milking system and stage of lactation.

### **Effects of AMS on milk fatty acid composition**

Overall, no significant differences between milk from AMS and CMP existed for FA composition, except for C16:0 and FFA contents, which were greater in AMS than CMP. These results agree with findings of Svennersten-Sjaunja et al. (2002) who investigated the effect of increasing milking frequency on milk FA

profile and of Knight et al. (1992) who explored the variation of acetyl-CoA carboxylase and FA synthetase in udder half milked 4 times daily compared with the other udder half milked twice daily. In contrast, Wiking et al. (2006) reported a significant reduction of PUFA by increasing milking frequency. Greater content of FFA in milk from AMS confirms previous studies conducted at farm and udder level (Wiking et al. 2006). The difference between AMS and CMP for FFA across lactation seems to be related to MY; in fact, greater contents in FFA for cows milked by AMS with respect to CMP were depicted in the first three months of lactation where cows produced more milk per day. Rasmussen et al. (2006) demonstrated that air intake in AMS did not have the largest influence on FFA at the cow or herd level and confirmed that MY at individual milking influences the FFA concentration. Abeni et al. (2005), analysing individual samples of HF cows, reported that milk fat lipolysis was affected by milking system and showed that FFA content was greater in milk fat from AMS than CMP. Free FA derive from the degradation of milk fat into glycerol and FFA through lipolysis reaction. Cooling and mechanical treatments of milk can break the membrane of the fat globule, resulting in an increase of FFA. The increasing of FFA is firstly related to increasing milking frequency or short milking intervals (Klei et al. 1997; Justesen & Rasmussen 2000; Klungel et al. 2000; Wiktorsson et al. 2000; de Koning & Rodenburg 2004; Slaghuis et al. 2004; Wiking et al. 2006). Another explanation for the greater FFA content of milk from cows milked more than 2 times daily is the fat globule size; shorter milking intervals are associated with larger fat globules which are more susceptible to lipolysis than smaller ones (Wiking et al. 2003; Abeni et al. 2005).

### **Conclusions**

The present study investigated the effect of milking system on milk coagulation traits and fatty acid composition of HF cows. Milking system had an effect on MY and FFA content. On average, cows milked by AMS produced more milk and with greater FFA content than cows milked by CMP. Traditional milk quality traits were not affected by milking system and MCP differed marginally between AMS and CMP. Results from the present study suggest that AMS can be adopted without detrimental effects on milk features, especially on milk coagulation characteristics. An economic analysis is required before recommending this system to producers in specialised cheese production areas.

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## Disclosure statement

The authors declare that there is no conflict of interest associated with this paper. The authors alone are responsible for the content and writing of this article.

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