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A comparison of the predicted coagulation characteristics and composition of milk from multi-breed herds of Holstein-Friesian, Brown Swiss and Simmental cows

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ABSTRACT

The milk coagulation properties (MCP) and composition, as predicted by mid-infrared spectroscopy, were compared between Holstein-Friesian (HF), Brown Swiss (BS) and Simmental (SI) cows from mixed herds. Records ($n = 8524$) of rennet coagulation time (RCT, min) and curd firmness (a_{30}, mm) were analysed using a mixed linear model. Milk from BS coagulated earlier and showed a firmer curd than milk from HF and SI breeds. Rennet coagulation time was shortest in the first 90 d of lactation, and a_{30} was lowest at the beginning and end of lactation. Herd exerted a strong effect on MCP, as the differences between the best and the worst farm for RCT and a_{30} were 7.8 min and 13.1 mm, respectively. In conclusion, the BS breed produced milk more suitable for cheese production than that from SI and HF. Further research is required to understand how farm management can improve coagulation characteristics of milk.

1. Introduction

The dairy sector is of economic importance in Italy, and contributes about 10% to the agriculture gross domestic product (Cassandro, 2003). This milk is mainly processed into high-quality cheese, including Protected Designation of Origin products, and thus renneting properties are of great interest for the dairy industry; milk with high capacity to properly react to rennet and to produce a firm curd results in higher cheese yield (Bynum & Olson, 1982; Riddell-Lawrence & Hicks, 1989).

Milk coagulation properties (MCP) can be measured by several instruments as summarised by O'Callaghan, O'Donnell, and Payne (2002). Among mechanical tools, the Formagraph and the Computerised Renneting Meter have been the most used to determine MCP (e.g., Cassandro et al., 2008; Ikonen, Ahlfors, Kempe, Ojala, & Ruottinen, 1999); these devices produce a typical diagram as reported by Dal Zotto et al. (2008) and provide measures of rennet coagulation time (RCT), which is the interval, in minutes, between the addition of the clotting enzyme and the beginning of the coagulation, and curd firmness (a_{30}), which is the width, in millimetres, of the diagram 30 min after the addition of the coagulant. The main disadvantages of the afore-mentioned instruments are the limited number of samples processed per hour, the costs related to the analysis and to the skilled personnel involved. Therefore, mid-infrared spectroscopy (MIRS) has been suggested as rapid, non-invasive and cost-effective tool to routinely predict economically important traits such as fat, protein and casein contents, somatic cell score (SCS), and MCP (De Marchi et al., 2009; De Marchi, Penasa, Tiezzi, Toffanin, & Cassandro, 2012; De Marchi, Toffanin, Cassandro, & Penasa, 2013).

Rennet coagulation time and a_{30} are influenced by several factors such as titratable acidity (Formaggioni, Malacarne, Summer, Fossa, & Mariani, 2001), somatic cell count (SCC; Politis & Ng-Kwai-Hang, 1988), contents of protein, casein, calcium and phosphorus (Summer, Malacarne, Martuzzi, & Mariani, 2002), rennet concentration (Pretto et al., 2011; Van Hooydonk & Walstra, 1987), and sampling and laboratory procedures (Cipolat-Gotet, Cecchinato, De Marchi, Penasa, & Bittante, 2012; Dal Zotto et al., 2008; Pretto et al., 2011). Besides physical and chemical factors, and experimental procedures, additive genetic effects have been reported for MCP (e.g., Cassandro et al., 2008; Ikonen, Morri, Tyrisevä, Ruottinen, & Ojala, 2004; Tiezzi, Pretto, De Marchi, Penasa, & Cassandro, 2013), as well as a strong breed effect.

Most studies have measured MCP on a limited number of samples from single-breed herds using mechanical devices (e.g., De Marchi, Dal Zotto, Cassandro, & Bittante, 2007; Malacarne et al., 2006). The knowledge of the actual differences in MCP among breeds could be valuable; if certain breeds produce milk especially

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suitable for cheese production, then the segregation of milk could enhance the efficiency of the dairy industry and maximise the value of the raw material (Auldist, Mullins, O'Brien, O'Kennedy, & Guinee, 2002). To our knowledge, no studies have attempted to estimate breed differences for MCP predicted by MIRS in multi-breed herds with repeated records per cow. Therefore, the aim of this study was to compare three cattle breeds in mixed dairy herds for predicted MCP and composition traits.

2. Materials and methods

2.1. Data

The initial data consisted of 8960 individual milk samples collected between September 2011 and February 2012 from 39 multi-breed dairy herds of Veneto region (northeast Italy). Farms were enrolled in the official monthly test-day milk recording system. Days in milk were restricted to be between 5 and 600 d, and parity between 1 and 6. Moreover, samples that exceeded 4 standard deviations from the mean of each trait were discarded from the dataset. Following editing of the data as above, 8524 milk samples from 3055 cows were available for statistical analysis. The size of the herds ranged from 20 to 134 with a mean of 78 cows, and animals were from three cattle breeds: Holstein-Friesian (HF, $n = 1675$), Brown Swiss (BS, $n = 914$) and Simmental (SI, $n = 466$). Twenty-two farms were HF + BS, 13 were HF + SI, and 4 were HF + BS + SI. Breed contribution was calculated for each monthly herd-test-date as the percentage of cows controlled for each breed of the total cows of the farm. Contributions were averaged over the period considered to account for random fluctuations of the consistency of the breed within herd, and each herd had to guarantee that none of the afore-mentioned breeds accounted for more than 90% of total animals and that the least represented breed accounted for at least 5 cows controlled in each test-date.

Milk samples were analysed for fat, protein and casein contents, SCC, RCT and a_{30} in the laboratory of the Breeders Association of Veneto region (Padova, Italy) using Milko-Scan FT6000 (Foss Electric A/S, Hillerød, Denmark). Mid-infrared spectroscopy models used to routinely predict MCP were developed by De Marchi et al. (2009, 2012) and implemented on the Milko-Scan FT6000; the authors obtained coefficients of determination of cross-validation of 0.76 and 0.70 for RCT and a_{30}, respectively (De Marchi et al., 2012; Tiezzi et al., 2013). In addition to quality traits and MCP, information on daily milk yield of cows was available. Casein index was calculated as the ratio of casein to protein content and SCS was obtained via base-2 log-transformation of SCC as

$$SCS = 3 + \log_2(SCC/100,000) \quad (1)$$

2.2. Statistical analysis

Data were analysed using the MIXED procedure of SAS (SAS, 2008) according to the following linear model:

$$y_{ijklmno} = \mu + H_i + M_j + P_k + DIM_l + B_m + (P \times B)_{km} + (DIM \times B)_{lm} + cow_n(B_m) + \varepsilon_{ijklmno} \quad (2)$$

where $y_{ijklmno}$ is the dependent variable (daily milk yield, fat content, protein content, casein content, casein index, SCS, RCT or a_{30}); μ is the overall intercept of the model; H_i is the fixed effect of the ith herd ($i = 1$–39); M_j is the fixed effect of the jth month of sampling (j = September, October, November, December, January, February); P_k is the fixed effect of the kth parity of the cow (k = first, second, third, and fourth and later parities); DIM_l is the fixed effect of the lth class of stage of lactation of the cow ($l = 1$ to 12, the first being a class from 5 to 30 d, followed by classes of 30 d each, and the last being a class from 330 to 600 d); B_m is the fixed effect of the mth breed of the cow (m = Holstein-Friesian, Brown Swiss, Simmental); $(P \times B)_{km}$ is the fixed interaction effect between parity and breed; $(DIM \times B)_{lm}$ is the fixed interaction effect between DIM and breed; $cow_n(B_m)$ is the random effect of the nth cow ($n = 1$–3055) nested within the mth breed $N \sim (0, \sigma^2_{cow(B)})$; and ε is the random residual $N \sim (0, \sigma^2_\varepsilon)$. Significance of breed effect was tested on the cow within-breed variance. A multiple comparison of means was performed for the main effect of breed, using Bonferroni's test ($P < 0.05$).

3. Results and discussion

3.1. Descriptive statistics and significance of fixed effects

Means of milk yield, fat content, protein content, casein content, casein index and SCS were 27.7 kg d^{-1}, 3.97%, 3.57%, 2.82%, 79.1% and 3.11, respectively (Table 1), with large variability among traits: the coefficient of variation ranged from 2.65% (casein index) to 62.4% (SCS). Rennet coagulation time and a_{30} averaged 20.2 min and 23.5 mm, respectively, and exhibited coefficients of variation of 20.0 and 38.7% (Table 1). Herd, month of sampling, DIM and breed effects were highly significant ($P < 0.001$) in explaining the variability of the traits studied (Table 2). Parity was significant ($P < 0.001$) for all traits except for fat content ($P = 0.28$). The interaction effect DIM × breed was significant in explaining the variation for fat, protein, and casein percentages, SCS and MCP ($P < 0.05$). Finally, the interaction effect parity × breed was important only for SCS ($P < 0.001$; Table 2).

3.2. Mid-infrared spectroscopy and noncoagulating milk

Milk coagulation properties were predicted using models developed by De Marchi et al. (2009, 2012) and currently implemented in several milk quality laboratories of Italy. The models are suitable for predicting RCT in the range from 5 to 30 min and samples that show values out of this range are classified as noncoagulating (NC). Noncoagulation of milk has a multifactorial origin as it is correlated with pH, SCC, mastitis, κ-casein concentration and genotypes, stage of lactation, breed of cow, and additive genetic effects. Recently, De Marchi et al. (2013) reported that there is no specific spectral information that distinguishes NC from coagulating samples. However, the same authors found that the most accurate prediction models were developed for RCT, curd-firming time and a_{30}, whereas curd firmness 60 min after enzyme addition could not be accurately predicted. Based on these results, De Marchi et al. (2013) concluded that MIRS might be proposed in payment systems to reward or penalise milk according to MCP.

Table 1
Descriptive statistics of milk yield, composition and coagulation traits.

Trait[a]	Mean	CV (%)[b]	Minimum	Maximum
Milk yield (kg d^{-1})	27.7	33.2	4.00	64.3
Fat (%)	3.97	21.7	1.10	10.1
Protein (%)	3.57	12.6	2.39	5.44
Casein (%)	2.82	13.5	1.77	4.37
Casein index (%)	79.1	2.65	69.0	88.8
SCS	3.11	62.4	−1.64	10.4
RCT (min)	20.2	20.0	2.26	29.9
a_{30} (mm)	23.5	38.7	0.04	58.7

[a] SCS, somatic cell score; RCT, rennet coagulation time; a_{30}, curd firmness 30 min after rennet addition.
[b] CV, coefficient of variation.

Table 2
F-value and significance of fixed effects included in the analysis for milk yield, composition and coagulation traits.[a]

Trait[b]	Herd	Month	Parity (P)	DIM	Breed (B)[c]	P × B	DIM × B	RSD
Milk yield (kg d^{-1})	45.42***	45.47***	62.79***	210.96***	109.25***	0.27	1.44	4.304
Fat (%)	16.34***	35.52***	1.29	40.41***	96.81***	1.78	2.17**	0.649
Protein (%)	8.58***	124.28***	30.02***	351.22***	236.41***	1.14	4.23***	0.210
Casein (%)	8.84***	29.20***	37.39***	339.04***	237.07***	1.24	4.05***	0.176
Casein index (%)	24.88***	2366.77***	125.82***	27.52***	48.16***	0.93	0.89	1.039
SCS	10.41***	9.52***	38.55***	18.80***	13.77***	4.41***	1.76*	1.248
RCT (min)	17.86***	121.37***	9.83***	118.82***	77.66***	1.29	2.17**	2.603
a_{30} (mm)	10.41***	9.08***	25.91***	50.50***	141.54***	1.80	2.38***	5.746

[a] Statistical significance is given as: $*P < 0.05$; $**P < 0.01$; $***P < 0.001$.
[b] Abbreviations are: SCS, somatic cell score; RCT, rennet coagulation time; a_{30}, curd firmness 30 min after rennet addition; DIM, days in milk; RSD, residual standard deviation.
[c] Breed effect was tested on the cow within-breed variance.

In the present study, only 2.5% of samples did not exhibit rennet coagulation within 30 min from the beginning of the analysis. This percentage is much lower than that (9.7%) reported by Cassandro et al. (2008) in an Italian study. However, Cassandro et al. (2008) used data from Holstein-Friesian cows only, whereas the present study included also Brown Swiss and Simmental breeds. Moreover, it is quite difficult to compare results from the present research with those of Cassandro et al. (2008), since they collected milk samples in 2003 and measured MCP using a mechanical instrument instead of MIRS.

3.3. *Effect of breed*

Table 3 shows the least squares means (LSM) of milk yield, quality traits and MCP for BS, HF and SI breeds. As expected, HF produced significantly (27.5 kg d^{-1}; $P < 0.05$) more milk than BS (24.1 kg d^{-1}) and SI (23.5 kg d^{-1}) cows, and BS exhibited much better milk composition compared with the other two breeds, especially HF ($P < 0.05$): fat, protein and casein contents, and casein index were 4.25 and 3.83%, 3.76 and 3.47%, 2.99 and 2.74%, and 79.3 and 78.8% for BS and HF, respectively. The composition of milk from SI cows was intermediate between that of BS and HF breeds ($P < 0.05$). These findings are consistent with previous studies which reported the superiority of HF for milk yield compared with other breeds (Cecchinato et al., 2011; Tyrisevä, Vahlsten, Ruottinen, & Ojala, 2004) and its inferiority for quality traits (Auldist et al., 2002; Auldist, Johnston, White, Fitzsimons, & Boland, 2004; De Marchi et al., 2007; Tyrisevä et al., 2004). Somatic cell score was notably lower for SI (2.97; $P < 0.05$) than BS (3.15) and HF (3.43) breeds, confirming the findings of De Marchi et al. (2007).

Milk from BS cows coagulated 1.90 and 1.10 min earlier, and exhibited a firmer curd by 6.00 and 3.20 mm, than milk from HF and SI, respectively (Table 3). Only a few studies have compared coagulation traits from cows of different breeds in mixed herds or under similar conditions (Auldist et al., 2002, 2004; De Marchi, Bittante, Dal Zotto, Dalvit, & Cassandro, 2008; Pomiès, Martin, Chilliard, Pradel, & Rémond, 2007; Tyrisevä et al., 2004). Pomiès et al. (2007) assessed MCP of Montbéliarde and HF cows; milk samples from the two breeds had very similar RCT (11.7 versus 11.8 min, respectively), whereas a_{30} was much higher for milk of the former breed (44.8 versus. 34.4 mm, respectively). These results are consistent with those from our research, which reported a difference of −0.80 min for RCT and 2.8 mm for a_{30} between SI and HF. De Marchi et al. (2008) investigated MCP of individual milk samples from HF and BS cows in mixed herds, which were also comparable in terms of size, management and feeding practices; BS performed much better than HF for RCT (−1.90 min) and a_{30} (4.40 mm), in agreement with the present results. Auldist et al. (2002) assessed rennet coagulation characteristics of Irish Friesian, HF, Montbèliarde and Normande cows, and concluded that milk from the latter two breeds coagulated more quickly and exhibited a firmer curd than milk from Irish Friesian and HF. The coagulation time was shorter and the curd was firmer in milk from HF than Finnish Ayrshire cows in the study of Tyrisevä et al. (2004) conducted in Finland, while Auldist et al. (2004) reported similar performance between Friesian and Jersey cows in New Zealand. Using bulk milk from single-breed herds, De Marchi et al. (2007) compared MCP of BS, HF and SI cows; RCT was approximately 2 min shorter for BS and SI compared with HF, and curd was much firmer for BS than the other 2 breeds, particularly HF.

Table 3
Least squares means (with standard errors) of milk yield, composition and coagulation traits of cows of different breeds.[a]

Trait	BS	HF	SI
Milk yield (kg d^{-1})	24.1 (0.25)[a]	27.5 (0.18)[b]	23.5 (0.33)[a]
Fat (%)	4.25 (0.03)[a]	3.83 (0.02)[b]	4.05 (0.04)[c]
Protein (%)	3.76 (0.01)[a]	3.47 (0.01)[b]	3.64 (0.02)[c]
Casein (%)	2.99 (0.01)[a]	2.74 (0.01)[b]	2.88 (0.01)[c]
Casein index (%)	79.3 (0.04)[a]	78.8 (0.03)[b]	79.1 (0.06)[c]
SCS	3.15 (0.07)[a]	3.43 (0.05)[b]	2.97 (0.09)[a]
RCT (min)	19.1 (0.13)[a]	21.0 (0.09)[b]	20.2 (0.18)[c]
a_{30} (mm)	26.8 (0.31)[a]	20.8 (0.22)[b]	23.6 (0.42)[c]

[a] Least squares means with different superscript letters are significantly different according to Bonferroni's test ($P < 0.05$). Abbreviations are: BS, Brown Swiss; HF, Holstein-Friesian; SI, Simmental; SCS, somatic cell score; RCT, rennet coagulation time; a_{30}, curd firmness 30 min after rennet addition.

3.4. *Herd effect*

Coagulation characteristics of milk varied considerably among farms; LSM of the best and the worst herds differed by 7.8 min for RCT and 13.1 mm for a_{30} (data not shown), suggesting that management practices and feeding had a notable impact on MCP and thus that it is possible to produce milk with better ability to coagulate by improving the farm environmental conditions. The relationship between the aptitude of milk to coagulate and farm characteristics needs specific research to collect management and feeding information from the herds.

Few studies have investigated the effect of herd on MCP. In contrast to our findings, Ikonen et al. (2004), Tyrisevä et al. (2004) and Vallas et al. (2010) reported that MCP were not strongly affected by herd environment, whereas Ikonen et al. (1999) estimated a significant influence of the farms on the variation of RCT. Vallas et al. (2010) reported that less than 5% of the total variation of MCP was explained by herd differences, whereas Tyrisevä et al. (2004) estimated slightly higher proportions of variation explained by herd (6–9%). The frequent feeding (more than twice a day) of concentrates was associated with good MCP in the work of

Tyrisevä et al. (2004), and a reduction of the occurrence of poorly coagulating samples. Other authors reported that udder health of the cows is an important management factor affecting MCP, and mastitis has a detrimental effect on milk coagulation ability (Bergère & Lenoir, 2000). Finally, Macheboeuf, Coulon, and D'Hour (1993) and Malossini, Bovolenta, Poras, Dalla Rosa, and Ventura (1996) found that the energy level of the diet was correlated with MCP.

3.5. Interaction effects

Fig. 1 depicts the LSM of MCP for the different breeds across DIM. Rennet coagulation time showed the highest values between calving and 90 d of lactation. Brown Swiss cows performed better than other breeds across DIM, particularly from 60 d onward, whereas HF performed worse than BS and SI. Curd firmness exhibited the best values at the beginning and in the last 3 months of lactation. As for RCT, BS performed better than SI and especially HF across DIM. The DIM effect was an important source of variation for RCT and a_{30} ($P < 0.001$; Table 2), and the trend for MCP during the first weeks of lactation was consistent with that reported in the literature (Cipolat-Gotet et al., 2012; Ikonen et al., 2004; Ostersen, Foldager, & Hermansen, 1997; Tyrisevä et al., 2004). The good coagulation characteristics at the beginning of the lactation could be related to the low values of milk pH during this stage (Ikonen et al., 2004; Mariani, Pecorari, & Fossa, 1982; Tyrisevä et al., 2004). Moreover, the proportion of non-coagulating milk samples has been observed to increase across DIM (Mariani et al., 1982; Okigbo, Richardson, Brown, & Ernstrom, 1985).

Fig. 2 shows the LSM of MCP for the different breeds across parities. Rennet coagulation time and a_{30} were better in primiparous than multiparous animals. Simmental cows were intermediate between BS and HF in first lactation, whereas they were much more close to RCT and a_{30} of HF breed in third and later parities. Parity had a strong effect on RCT and a_{30} ($P < 0.001$), which deteriorated across lactations. In the literature, reports on the effect of parity on MCP are contradictory. Similarly to our findings, Tyrisevä, Ikonen, and Ojala (2003) observed better values of MCP in first lactation cows, whereas Ikonen et al. (2004) reported lower values of curd firmness for primiparous than multiparous cows. Finally, Ikonen et al. (1999) and Tyrisevä et al. (2004) reported that lactation number had no influence on MCP.

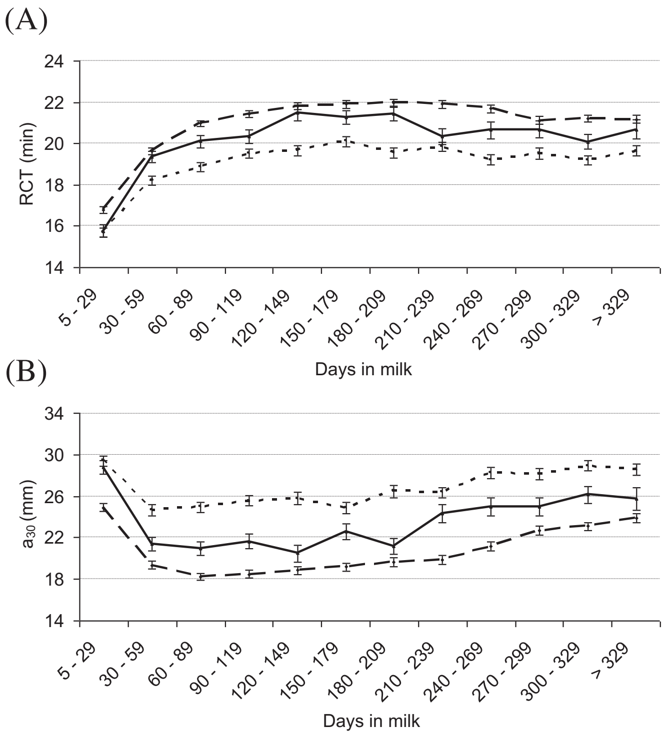

Fig. 1. Least squares means (with standard errors) of (A) rennet coagulation time (RCT, min) and (B) curd firmness 30 min after rennet addition (a_{30}, mm) of cows of different days in milk (DIM) and breeds (BS, Brown Swiss; HF, Holstein-Friesian; SI, Simmental).

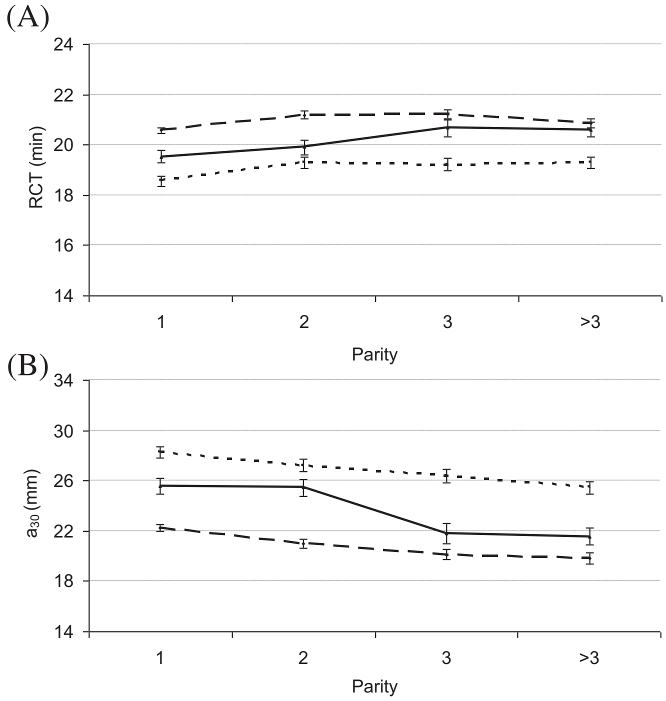

Fig. 2. Least squares means (with standard errors) of (A) rennet coagulation time (RCT, min) and (B) curd firmness 30 min after rennet addition (a_{30}, mm) of cows of different parities and breeds (BS, Brown Swiss; HF, Holstein-Friesian; SI, Simmental).

4. Conclusion

Milk quality and MCP largely differed among cattle breeds in mixed herds. Brown Swiss produced less milk than HF cows but performed much better in terms of MCP and quality traits. Simmental was intermediate between HF and BS for all milk characteristics, and exhibited the lowest value of SCS. Stage of lactation and parity were also important for MCP, and an appreciable variation for these effects has been observed across breeds. Further research is needed to investigate the effects of farm characteristics (e.g., feeding strategies) on the variation of MCP to identify technical solutions which could help farmers to improve renneting properties.

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