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**Visentin, G., Penasa, M., Gottardo, P., Niero, G., Isaia, M.,
Cassandro, M., De Marchi, M.**

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Poljoprivredni fakultet u Osijeku, Poljoprivredni institut Osijek

Faculty of Agriculture in Osijek, Agricultural Institute Osijek

MILK COAGULATION PROPERTIES OF CATTLE BREEDS REARED IN ALPINE AREA

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SUMMARY

The aim of the present study was to apply mid-infrared spectroscopy prediction models developed for milk coagulation properties (MCP) to a spectral dataset of 123,240 records collected over a 2-year period in the Alpine area, and to investigate sources of variation of the predicted MCP. Mixed linear models included fixed effects of breed, month and year of sampling, days in milk, parity, and the interactions between the main effects. Random effects were herd nested within breed, cow nested within breed, and the residual. All fixed effects were significant ($P < 0.01$) in explaining the variation of MCP. In particular, milk clotting characteristics varied significantly among breeds, and local Alpine Grey breed exhibited the most favourable processing characteristics. Milk coagulation properties varied across lactation and were at their worst after the peak.

Key-words: *mid-infrared spectroscopy, milk quality, local breeds*

INTRODUCTION

European Union (EU) dairy products market is addressed in maximizing added value of milk, by processing it into different food categories. For instance, EU contributes to half of the total world cheese production. Therefore, an important challenge for the dairy industry is to segregate milk based on its processing quality. Milk coagulation properties (MCP) are indicators of milk processing characteristics and they include rennet coagulation time (RCT, min), curd-firming time (k_{20} , min), and curd firmness (a_{30} , mm). Large-scale monitoring of these traits is difficult due to expensive and time-consuming gold standard methods (e.g., Formagraph). Mid-infrared spectroscopy (MIRS) can overlap this issue and has been proposed as an effective tool to predict innovative milk quality traits at the population level, such as MCP (De Marchi et al., 2013, 2014). There is a paucity of studies that have investigated MCP variation among cattle breeds, including local populations. Therefore, aim of the present study was to investigate sources of variation of MIRS-predicted MCP in major cattle breeds in Bolzano Province, North-Eastern Italian Alps.

MATERIAL AND METHODS

Data collection

A total of 132,380 individual milk samples from 15,173 cows were collected between January 2012 and December 2013 in Bolzano Province, a mountainous area of the Italian Alps. Cow breeds included in the dataset were Holstein-Friesian (HF, 13%), Brown Swiss (BS, 36%), Simmental (SI, 30%), and Alpine Grey (AG, 21%). Cows were from 6 to 450 days-in-milk (DIM), and from 1st to 15th parity. Immediately after collection, samples were added with preservative (Bronysolv; ANA. LI.TIK Austria, Vienna, Austria) and processed in the laboratory of the South Tirol Dairy Association (Bolzano, Italy) according to recommendations by International Committee for Animal Recording. Each milk sample was analyzed using a MilkoScan FT6000 (Foss Electric A/S, Hillerød, Denmark) to determine protein (%), casein (%), fat (%), lactose (%), urea (mg/dL), and pH. Spectral information of milk quality traits, containing 1,060 transmittance data in the region between 900 and 5,000 cm^{-1} , were also retrieved from the South Tirol Dairy

M.Sc. Giulio Visentin (giulio.visentin@studenti.unipd.it), Assistant Prof. Mauro Penasa, M.Sc. Paolo Gottardo, M.Sc. Giovanni Niero, M.Sc. Martina Isaia, Prof. Dr. Martino Cassandro, Assoc. Prof. Massimo De Marchi - University of Padova, Department of Agronomy, Food, Natural Resources, Animals and Environment, Viale dell'Università 16, 35020 Legnaro (PD), Italy

Association. Somatic cell count (SCC) was assessed by Fossmatic (Foss Electric A/S, Hillerød, Denmark) and transformed to somatic cell score (SCS) through the formula $SCS = [3 + \log_2(SCC/100,000)]$.

Laboratory analysis and MIRS prediction models

A calibration dataset of 923 milk samples was used to build MIRS prediction models for RCT, k_{20} , and a_{30} . Reference values of MCP were determined by Formagraph (Foss Electric A/S, Hillerød, Denmark), and MIRS spectra were stored by Milkoscan FT6000 (Foss Electric A/S, Hillerød, Denmark). Full details about the reference method are available in De Marchi et al. (2013). Prediction models were built through partial least squares regression after uninformative variables elimination as recently implemented by Gottardo et al. (2015). Coefficient of determination (root mean square error) in validation was 0.55 (2.86 min), 0.58 (1.00 min), and 0.56 (8.43 mm) for RCT, k_{20} , and a_{30} , respectively.

Phenotypic characterization

Statistical analysis was carried out using SAS software (ver. 9.3, SAS Institute Inc., Cary, NC, USA). Principal component analysis (PROC PRINCOMP) was performed on both the initial validation dataset and the calibration dataset, providing a new matrix of uncorrelated variables called principal components (PC) which explain, in a descending order, a portion of the total variance. This procedure aimed to select spectral observations in the validation dataset similar to those in the calibration dataset. This was achieved by selecting only spectral data in the validation dataset whose first and second PC (PC-1 and PC-2, respectively) were within the same range of PC-1 and PC-2 of spectral data in the calibration dataset. Therefore, 123,240 observations from 15,066 cows were retained for further statistical analysis, whereas the remaining 9,140 records were classified as outliers and discarded. Prediction models were subsequently applied to the edited validation dataset to predict RCT, k_{20} , and a_{30} . Sources of variation of MCP were investigated using PROC MIXED according to the following mixed linear model:

$$Y_{ijklmno} = \mu + B_i + M_j + Y_k + DIM_l + Parity_m + (B \times M)_{ij} + (B \times Y)_{ik} + (B \times DIM)_{il} + (B \times Parity)_{im} + (DIM \times Parity)_{lm} + H_n(B_i) + Cow_o(B_i) + e_{ijklmno}$$

where $Y_{ijklmno}$ is the dependant variable (MIRS-predicted RCT, k_{20} , or a_{30}), μ is the overall intercept of the model, B_i is the fixed effect of the i^{th} breed ($i = \text{HF, BS, SI, AG}$), M_j is the fixed effect of the j^{th} month of sampling ($j = 1$ to 12), Y_k is the fixed effect of the k^{th} year of sampling ($k = 2012, 2013$), DIM_l is the fixed effect of the l^{th} class of DIM ($l = 6$ to 30, 31 to 60, 61 to 90, 91 to 120, 121 to 150, 151 to 180, 181 to 210, 211 to 240, 241 to 270, 271 to 300, 301 to 330, 331 to 360, 361 to 390, 391 to 450 days), $Parity_m$ is the fixed effect of the m^{th}

parity ($m = 1$ to 5, with class 5 including cows from parity 5 to 15), $(B \times M)_{ij}$ is the fixed interaction effect between breed and month of sampling, $(B \times Y)_{ik}$ is the fixed interaction effect between breed and year of sampling, $(B \times DIM)_{il}$ is the fixed interaction effect between breed and DIM, $(B \times Parity)_{im}$ is the fixed interaction effect between breed and parity, $(DIM \times Parity)_{lm}$ is the fixed interaction effect between DIM and parity, $H_n(B_i)$ is the random effect of the n^{th} herd nested within the i^{th} breed $\sim N(0, s^2_{H(B)})$, $Cow_o(B_i)$ is the random effect of the o^{th} cow nested within the i^{th} breed $\sim N(0, s^2_{Cow(B)})$, and $e_{ijklmno}$ is the random residual $\sim N(0, \sigma^2_e)$. A multiple comparison of means was performed for breed effect using Bonferroni's test ($P < 0.05$).

RESULTS AND DISCUSSION

Means and variation

Descriptive statistics of milk quality traits and MCP included in calibration dataset are reported in Table 1. Coefficient of variation of MCP ranged from 24.8% (RCT) to 44.1% (a_{30}). These results are comparable with those (20.0 and 38.7%, respectively) of another large-scale research considering MIRS-predicted MCP on a multi-breed dataset (Penasa et al., 2014). Large data variation is desired when information is used to develop MIRS prediction models that will be subsequently applied for phenotyping at the population level (Visentin et al., 2015).

Table 1. Descriptive statistics¹ of milk samples included in calibration dataset (n=923)

Trait ²	Mean	SD	Minimum	Maximum	CV, %
Fat, %	4.09	0.72	1.71	9.19	17.6
Protein, %	3.61	0.45	2.34	5.50	12.6
Casein, %	2.82	0.36	1.71	4.38	12.9
SCS	2.80	1.86	-3.64	8.87	66.3
pH	6.65	0.07	5.87	6.92	1.1
RCT, min	18.57	4.61	4.30	29.00	24.8
k_{20} , min	5.20	1.63	2.00	13.15	31.3
a_{30} , mm	28.07	12.37	2.40	57.12	44.1

¹SD=standard deviation; CV=coefficient of variation; ²SCS=somatic cell score; RCT=rennet coagulation time; k_{20} =curd-firming time; a_{30} =curd firmness

Table 2. Least squares means (standard error) of milk coagulation properties across breeds

Trait ¹	Holstein-Friesian	Brown Swiss	Alpine Grey	Simmental
RCT, min	22.44 (0.15) ^a	22.15 (0.07) ^a	21.84 (0.09) ^b	21.80 (0.08) ^b
k_{20} , min	7.08 (0.05) ^a	5.75 (0.02) ^b	6.13 (0.03) ^c	6.21 (0.03) ^c
a_{30} , mm	14.82 (0.39) ^a	19.86 (0.19) ^b	19.71 (0.24) ^{bc}	18.90 (0.21) ^c

¹RCT=rennet coagulation time; k_{20} =curd-firming time; a_{30} =curd firmness

Sources of variation for milk coagulation properties

Fixed effects included in the mixed linear model for MCP were highly significant ($P < 0.01$; data not shown). Cow breed was an important source of variation for milk coagulation characteristics, and least squares means for the breed effect are reported in Table 2. Dual-purpose cows (SI and AG) exhibited more favourable MCP than dairy breeds (HF and BS). These findings are consistent with another large-scale study on Italian dairy cattle breeds conducted by Penasa et al. (2014). Recently, Preto et al. (2013) demonstrated that a_{30} has a positive association with cheese yield. This is a crucial point of the present study, especially in countries highly specialized in cheese production, such as Italy. Indeed, although characterized by lower milk production compared to cosmopolitan breeds, milk of local AG cows can potentially result in greater cheese yield and this could lead to more profit for the dairy industry. Moreover, this can also contribute to the valorisation of local breeds, that nowadays are giving a service to

rural area populations (e.g., maintenance of the territory where they are reared and preservation of the local traditions) without recognition in any milk payment system. These results, anyway, need further investigation.

Least squares means of MCP across lactation are depicted in Figure 1. At the very beginning of lactation, MCP had the most favourable values for cheese processing, with low values of RCT and high values of a_{30} . The opposite trend of RCT and a_{30} is due to the strong and negative correlation between them (-0.87). The strong association between these two traits was previously reported by Cassandro et al. (2008) in Italian Holstein-Friesian dairy cows. Rennet coagulation time increased up to 180 DIM and decreased slightly thereafter, until the end of lactation. Curd firmness, on the other hand, exhibited less desired values immediately after the lactation peak, and then became more favourable for cheese-making after 6 months of lactation. None of the traits, anyway, reached the levels observed at the beginning of lactation. These findings are consistent with Ikonen et al. (2004) and Penasa et al. (2014).

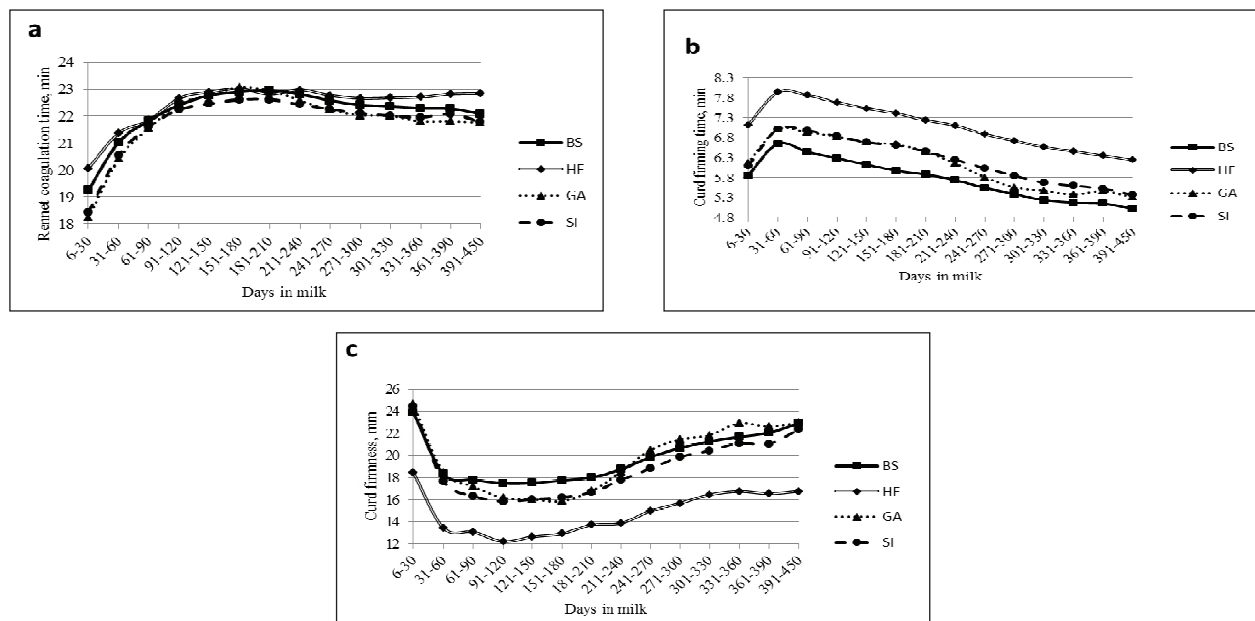


Figure 1. Least squares means of (a) rennet coagulation time, (b) curd-firming time, and (c) curd firmness across lactation.

CONCLUSION

This study provides for the first time a description of predicted MCP of AG cattle breed using individual samples and describing MCP variation in Alpine dairy system. The results demonstrated that MCP variation depended significantly on several environmental factors, including cow breed and DIM. Local AG cow had more favourable clotting characteristics compared to Holstein-Friesian breed, and this can be exploited as a strong point for its valorisation. Further research will estimate genetic parameters of MCP for these breeds.

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