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Are cheese-making properties of dual purpose cattle impaired by highland grazing? A case study using Aosta Red Pied cows

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ABSTRACT

Summer transhumance is often practiced in mountainous farming systems. It includes moving dairy cows from lowland (LO) to highland (HI) pastures during summer. It is known that high genetic merit cows are susceptible to the HI conditions, but it is unclear if this also applies to more adapted, regional cow types. The present study aimed to investigate the effect of HI sojourn on cheese-making properties of Aosta Red Pied cows, a dual purpose cattle type. Milk coagulation properties were measured in the milk of 47 cows before and after transhumance. Sources of variation were investigated using linear mixed models, including parity, site, milking time, the interaction parity × site, milking time × site and milking time × parity. Cow was nested within site, and used as subject for repetition, and sampling date was included as repeated factor. Curd-firming time and curd firmness did not vary between LO and HI, whereas rennet coagulation time was prolonged in HI compared to LO in both primiparous (16.4 vs. 18.5 min) and multiparous cows (17.5 vs. 21.1 min, respectively). The percentage of non-coagulating samples was greater in HI (15.0%) compared to LO (8.5%). The lower milk reactivity to rennet addition in HI seems to be mostly related to the simultaneously increasing somatic cell score. Morning and evening milk were similar in coagulation properties. In conclusion, even indigenous dual purpose cows were affected by HI conditions and the experience the multiparous cows had with the transhumance was not helpful either.

Introduction

In European mountainous regions, dairy cows are often moved from lowland (LO) to highland (HI) pastures in late spring or early summer in transhumance systems established since centuries (Zendri et al. 2016). The maintenance of this activity is threatened by high labour load and low income. However, since recently, producing milk and cheeses on HI pastures is gaining attention again for various reasons. Firstly, for consumers these foods are attractive due to the presumed higher health value. This perception is based on their high proportions of n-3 fatty acids in the food lipids and on their great percentage of conjugated linoleic acids in total fat, compared to products from LO (Leiber et al. 2005a). Secondly, HI pastures products are characterised by specific sensory properties (Buchin et al. 1999) and are well suited to market by defining restricted geographical areas and typical

manufacturing as Protected Designation of Origin (PDO) (Buchin et al. 1999). Thirdly, public authorities and organisations are promoting agricultural use of HI pastures through several conservation actions in order to preserve marginal landscapes and biodiversity (Maretto and Cassandro 2014; Niero et al. 2016b).

However, in order to be able to profit from the added value of the dairy foods and thus help maintaining the management of these areas, premium prices have to be realised (Pretto et al. 2009; Zendri et al. 2016). One of the drawbacks of the HI dairy systems is the impairment of milk coagulation properties (MCP) reported from studies with specialised dairy cattle breeds, such as Holstein Friesian, Brown Swiss and Brown Italian (Leiber et al. 2005b, 2006; Bovolenta et al. 2008). The main reason explaining the longer rennet coagulation time (RCT) and the less favourable curd properties, measured as curd-firming time (k_{20})

and curd firmness (a_{30}), is the energy deficit these cows are experiencing at high altitude, harsh climatic conditions and steep slopes (Christen et al. 1996; Kreuzer et al. 1998). This results in a lower milk protein content (Leiber et al. 2005b) which reduces both cheese yield and milk coagulation performance, with the latter due to the lower density of the casein (CN) molecules in milk. It is yet unclear if the use of autochthonous or local dual purpose cattle breeds, which would fit better to extensive grazing systems, help to preserve biodiversity and have a lower impact on the environment (Visentin et al. 2015b), may better tolerate the harsh conditions due to their comparably lower milk yield (MY) and, possibly, due to an inherited adaptation.

This research question was investigated through the example of summer grazing systems using Aosta Red Pied (ARP) cows, an autochthonous dual purpose cattle breed, reared in northwestern Italy. Fontina cheese (FC), a PDO product, is manufactured from full-fat and unpasteurised ARP milk, from LO or HI pastures, within 2 h after every single morning and evening milking (Mazza et al. 2016). For this purpose, milk from 47 ARP cows was repeatedly sampled from May to July, including both LO to HI pastures.

Materials and methods

Experimental design and milk sample collection

Procedures of the experiment are excluded from the authorisation of the animal welfare committee. The herd of 47 ARP cows belonged to the Institut Agricole Régional (Aosta, Italy) and was subjected to the official milk recording system.

In May 2016, milk sampling started in the LO barn (Montfleury, Aosta, Italy, 580 m a.s.l.). Milking took place in the morning from 5.00 a.m. to 7.30 a.m. and in the evening from 4.00 p.m. to 6.30 p.m. All cows were sampled during two morning milkings. In addition, 14 cows out of this group, half of them were primiparous and half of them were multiparous, were sampled also at two evening milkings. The total amount of milk samples collected in LO was 122. In the barn animals were tethered during the night and during milking, while during the day they were grazing LO pastures. In the barn, animals were additionally fed with local hay offered at an amount of 10 to 12 kg per cow per day, and with 3 kg of concentrate per cow and per day during milking.

Milk from the same ARP cows was sampled in the highlands (Val di Rhêmes, Rhêmes Notre-Dame, Italy, from 1800 to 2100 m a.s.l.) between June and July 2016. At that site, milking was performed from 4.30 a.m. to 7.00 a.m. and from 3.30 p.m. to 6.00 p.m. At HI, cows had permanent and free access to pasture and water, using the strip grazing technique to provide them daily fresh grass. Milking was accomplished directly on the pasture with a mobile milking parlour (Eliar 4, Eli IAR, Institut Agricole Régional, 4 milking places). During milking, 2 kg of concentrate per cow per day was provided. The same 47 cows as in LO were sampled two times during the morning milkings and the same 14 cows were sampled additionally during one more morning milking and three evening milkings. The total amount of milk samples collected in HI was 150.

The LO barn and the HI mobile milking parlour were equipped with the same milking device (Afimilk Ltd, Kibbutz Afikim, Israel). The device recorded milk yield (MY) for each cow at each milking and sampled 50 mL of milk. Immediately afterwards 18 mg preservative tablet containing 8 mg Bronopol (2-bromo-2-nitro-propan-1,3-diol) and 0.30 mg Natamycin were added to inhibit bacteria, yeast and mould growth. Samples were straightaway stored at 4 °C and successively transferred, by refrigerated shipping, to the laboratory of the Breeders Association of Veneto Region (ARAV, Padova, Italy) for analysis of milk composition and MCP.

Analysis of milk composition and milk coagulation properties

Analyses of contents of fat, protein, CN, lactose and milk urea nitrogen (MUN) were carried out using a MilkoScan FT6000 (Foss Electric A/S, Hillerød, Denmark). Determination of somatic cell count (SCC) was accomplished through a Fossomatic (Foss Electric). Values of SCC were transformed to somatic cell score (SCS) as $3 + \log_2(SCC/100{,}000)$ to achieve normality and homogeneity of variances. The MCP was determined using the Formagraph (Foss Electric A/S Hillerød, Denmark) as lactodynamographic tool, following the method proposed by McMahon and Brown (1982). The coagulation properties measured were RCT (min) as the time from rennet addition to the beginning of coagulation, k_{20} (min) as the time from the gel development to a width of 20 mm of the bell-shaped graph created by the Formagraph and a_{30} (mm) as the graph width at 30 min after rennet addition. Samples that did not coagulate within 30 min were considered as non-coagulating.

Table 1. Composition of the grass from the lowland ($n = 4$) and the highland ($n = 4$) pastures.

Site	Lowland		Highland	
	Mean	SD	Mean	SD
Biomass, g dry matter/m^2	139	26	253	106
Chemical composition, g/kg dry matter				
Crude protein	202	12	149	20
Ether extract	31.90	2.00	30.40	2.90
Total ash	123	7	74	5
Neutral detergent fibre	354	29	439	39
Acid detergent fibre	237	28	312	11
Acid detergent lignin	41.10	10.90	65.00	4.80
Ca	12.70	1.00	8.40	1.60
P	4.12	0.27	2.50	0.43
Energy and protein per kg dry matter				
NE_L, MJ	6.32	0.10	6.05	0.59
PDIE, g	109	2	100	12
PDIN, g	135	8	96	17

NE_L: net energy for lactation; PDIE: absorbable protein at the duodenum according to supply with fermentable energy and rumen undegradable protein; PDIN: absorbable protein at the duodenum according to supply with rumen degradable protein and rumen undegradable protein; SD: standard deviation.

Grass sampling and analysis

Two replicates of $10 \, cm \times 10 \, m$ strips were randomly chosen on two different pastures in LO and HI each resulting in four samples of fresh grass from both the site. Samples were weighed, then dried for 120 h at 60 °C and milled through a 5 mm screen. These pre-dried samples were weighed and analysed for contents of dry matter (DM), crude protein (CP), ether extract, total ash, neutral detergent fibre (NDF), acid detergent fibre (ADF), acid detergent lignin, Ca and P by NIRSystems 5000 (Foss Electric A/S, Hillerød, Denmark). From weights of fresh and pre-dried samples as well as DM content, the biomass per m^2 on the pastures was calculated. Contents of net energy for lactation (NE_L), PDIE and PDIN (for definitions see footnotes of Table 1) of the samples were estimated using the regressions underlying the calculation module of the official Swiss feeding recommendations for ruminants (Agroscope 2017).

Statistical analysis

By using the Shapiro–Wilk's test, it was first confirmed that all variables were normally distributed. Pearson's correlations between the traits were calculated by the CORR procedure of SAS (SAS Institute Inc., Cary, NC). Data from the 47 and 14 cows, respectively, were analysed by different models for analysis of variance using the MIXED procedure of SAS. In the first model, 188 samples from 47 cows and morning milkings only were included. Parity (primiparous and multiparous), site (LO and HI) and the interaction between parity and site were used as fixed effects. In the second model, 140 samples from the 14 cows with data from morning and evening milkings were used to assess milking time effect. Parity (primiparous and multiparous), site (LO and HI), milking time (morning and evening), and the interactions between milking time with site and parity were considered as fixed effects. The interaction between site and parity and the three-way interactions were excluded after determining that they were not significant. From the results obtained from Model 2 only means of milking time and site were presented, as parity effects were presented with the larger dataset in Model 1. In both the models, cow was nested within site, and used as subject for repetition, whereas date of sampling was included as repeated factor. Multiple comparisons among least square means were tested using Bonferroni's correction, and effects were considered significant at $p < .05$. Non-coagulating samples were considered in the statistical evaluation of milk yield and milk composition, whereas they were treated as missing values in case of RCT, k_{20} and a_{30}.

Results and discussion

Grass quality on the lowland and highland pastures

Table 1 shows grass composition from LO and HI pastures. The mean biomass of grass from the LO and HI was not different. This is due to the great variability (SD) of the available biomass of the grasslands from the HI. Indeed, the four LO pastures were more homogenous than the four HI pastures that represent the range over the whole summer grazing season. Contents of CP, Ca and P were higher by 36, 51 and 65% in the LO compared to the HI grass. The HI grass was more fibrous than the LO grass (24, 32 and 58% higher on average for contents of NDF, ADF and acid detergent lignin, respectively). The average NE_L content was quite similar for LO and HI grass, although it varied more in the HI grass. The PDIN content was higher by 41% for LO compared to HI grass.

Variation and correlations of milk quality and technological traits in Aosta Red Pied cows

In terms of milk yield and milk gross composition, results of the present study are comparable to those reported by Battaglini et al. (2009) and Renna et al. (2014), who studied milk yield and milk composition of Aosta cows. The contents of fat, protein and CN in the ARP milk were lower than those found in pure dairy breeds including Holstein–Friesian, Brown Swiss and Simmental cows (Penasa et al. 2014).

When compared with milk from other local and dual purpose cow breeds reared in the Italian alpine region, such as Alpine Grey, Rendena and Burlina, the milk from the ARP cows had greater protein content, a lower fat content and lower SCS (De Marchi et al. 2007; Niero et al. 2016b). Concerning MCP, among 236 rennet-treated samples, only 164 samples (about 70%) were able to reach k_{20} (Table 2). The ARP results were at the average of other Italian local breeds kept in alpine areas; they had a more favourable MCP than Burlina cows (Niero et al. 2016b) but less favourable technological traits than Alpine Gray and Rendena breeds (De Marchi et al. 2007).

Pearson's correlation coefficients between traits of Milk composition, pH and MCP are presented in Table 3. Rennet coagulation time was only weakly correlated with milk protein and CN content and a_{30} also had only a moderate correlation with the same traits. This rather weak relationship might at least be partly explained by observations that the curd of the slow coagulating milks had not enough time to develop, thus providing only a limited description of the potential gel firmness. The three MCP traits were closely correlated. Rennet coagulation time had a medium and positive correlation with k_{20} and was strongly negatively correlated with a_{30}. Milk pH showed weak but significant correlations with RCT, k_{20} and a_{30} in a way that a lower milk pH was associated with more favourable MCP. This is consistent with the results of previous studies (Ikonen et al. 2004; Toffanin et al. 2015). Negative low correlations were found between RCT and milk protein content and between RCT and CN content which was unexpected because higher casein content may increase RCT due to the higher ratio of substrate to enzyme. Among the MCP traits measured, k_{20} showed the highest (negative) correlations with milk protein and CN contents, while correlations between a_{30} and milk protein and CN contents were slightly lower and positive. These relationships, different from those with contents of fat, lactose, MUN and SCS, were expected and in agreement with previous studies (Visentin et al. 2015a). Indeed, CN is the only milk constituent reacting to rennet addition and curd formation occurs because of the aggregation of para-CN micelles, which subsequently enclose other milk constituents (Visentin et al. 2015a).

Effect of highland grazing and parity on milk yield and composition

In HI, compared to LO grazing period, MY was lower in both primiparous and multiparous cows (Table 4). A small part of this effect could have resulted from the concomitantly progressing lactation. The finding of a depression in MY in HI compared to LO is in agreement with findings from a number of studies with pure dairy breed cows with their typically higher

Table 2. Descriptive statistics of production and milk-related traits.

Item	No. of samples[a]	Mean	SD	Minimum	Maximum
Production-related traits					
Days in milk, d	264	176	48	42	269
Parity, n	271	3.29	2.33	1.00	10.00
Milk yield, kg/d	197	15.50	4.60	5.60	27.80
Milk composition					
Fat, %	258	3.80	0.81	1.84	8.09
Protein, %	258	3.34	0.25	2.79	4.00
Casein, %	258	2.62	0.21	2.12	3.15
Lactose, %	258	4.74	0.20	4.06	5.12
Urea nitrogen, mg/dL	257	19.20	5.40	4.00	35.30
Somatic cell score	258	2.73	1.70	−1.64	7.79
Milk acidity (pH)	258	6.62	0.06	6.44	6.83
Milk coagulation traits					
RCT, min	236	18.80	4.90	4.50	29.00
k_{20}, min	164	5.16	1.44	2.45	9.15
a_{30}, mm	237	27.40	12.90	2.40	58.40

RCT: rennet coagulation time; k_{20}: curd-firming time; a_{30}: curd firmness 30 min after rennet addition; SD: standard deviation.
[a]Number of cows: 47.

Table 3. Pearson's correlation coefficients between milk composition, milk pH and milk coagulation traits.

Item	Protein	Casein	Lactose	MUN	SCS	pH	RCT	k_{20}	a_{30}
Fat	0.32***	0.29***	−0.16**	−0.33***	0.26***	−0.28***	0.00	−0.32***	0.11
Protein		0.98***	0.03	−0.06	0.20***	−0.06	−0.07*	−0.51***	0.36***
Casein			0.03	0.02	0.21***	−0.23***	−0.06*	−0.52***	0.35***
Lactose				0.37***	−0.50***	0.51***	−0.15*	−0.03	0.16*
MUN					−0.40***	0.26***	−0.08	−0.01	0.09*
SCS						−0.16*	0.22***	−0.12	−0.13*
pH							0.16*	0.27***	−0.17***
RCT								0.46***	−0.84***
k_{20}									−0.84***

MUN: milk urea nitrogen; SCS: somatic cell score; RCT: rennet coagulation time; k_{20}: curd-firming time; a_{30}: curd firmness 30 min after rennet addition.
*$p < .05$.
**$p < .01$.
***$p < .001$.

initial MY (Christen et al. 1996; Leiber et al. 2005b). Consistently with previous observations by Leiber et al. (2004, 2006), the higher milk fat content at the HI site could be explained by the increase in body fat mobilisation of the animals in order to cope with alpine conditions and by the higher content of fibre – the main nutrient transformed to milk fatty acids by *de novo* synthesis from acetate – of the HI compared to the LO pasture (Table 1) (Leiber et al. 2006). The ARP cows of the present study showed no depression in milk protein content which was repeatedly found when moving pure dairy breed cows to higher altitude pastures (Christen et al. 1996; Leiber et al. 2005b). Milk lactose content was lower on HI than on LO ($p < .001$), but only in the multiparous cows (interaction, $p < .001$). However, this decline was without practical relevance from its magnitude (Tiezzi et al. 2013). There was also a large decrease ($p < .001$) in MUN from LO to HI, which can be explained by excessive dietary CP content of the LO pasture compared to the HI pasture. Like described before (Berry et al. 2001; Leiber et al. 2006), there was an increase by HI grazing in SCS, suggesting an increased incidence of subclinical mastitis. This increase was more pronounced in the primiparous ($+200\%$) compared to the multiparous cows ($+82\%$) where the LO level was lower ($p < .05$) in the primiparous cows. Possible reasons include carry-over effects from disturbed milk let-down during transport and walking, a higher frequency of injuries of the mammary gland when climbing the steep slopes and a lower hygiene standard when milking on pasture especially during times of intensive precipitation (Berry et al. 2001).

Effect of highland grazing and parity on milk coagulation traits

Parity had only weak effects on milk coagulation traits. In addition, as could have been expected from the lack of adverse effect on milk protein and CN content, curd firmness (k_{20} and a_{30}) remained unaffected by moving cows from LO to HI (Table 4). Rennet coagulation time increased from LO to HI, in both primiparous and multiparous cows and concomitantly pH declined. A longer RCT of milk produced on HI pastures was also found by other authors (Leiber et al. 2006) who measured MCP with lactodynamographic tool. Different from that, Zendri et al. (2016) reported an improvement in MCP predicted from MIRS data in HI compared to LO sites. Leiber et al. (2006) attributed the increase in RCT mainly to the lower protein and CN content of HI milk compared to that of LO, which was not the case in the present study. Thus, the unfavourable effect of HI on RCT found in the present study could be partially due to the concomitantly higher SCS, because a high density of somatic cells is antagonistic in this respect (Politis and Ng-Kwai-Hang, 1988).

A sufficiently high Ca content of the milk is also important for a favourable MCP (Franzoi et al. 2017). The grass from LO pasture was richer in Ca than that from the HI pasture (Table 1), but cows always received a Ca containing mineral feed and Ca contents of the milk are difficult to influence by diet anyway as the cow tries to keep milk composition stable for their offspring (Gaucheron 2005). Milk Ca content was not measured in the present study. Overall, it is established that MCP are influenced also by other effects

Table 4. Least squares means of cows of different parity status at different sites on milk yield and milk-related traits.

Parity status	Primiparous		Multiparous			p Value		
Site	Lowland	Highland	Lowland	Highland	SEM	Parity	Site	Parity $\times$ site
No of cows	10	10	37	37				
No of samples	40	40	74	74				
Milk yield, kg/day	13.300[b,c]	10.800[c]	19.600[a]	14.200[b]	1.100	<.001	<.001	.018
Milk composition								
Fat, %	3.330[b]	4.190[a]	3.440[b]	3.990[a]	0.183	.749	<.001	.264
Protein, %	3.280	3.340	3.280	3.310	0.070	.842	.317	.702
Casein, %	2.570	2.650	2.540	2.580	0.060	.437	.101	.598
Lactose, %	4.870[a]	4.800[a]	4.810[a]	4.570[b]	0.041	<.001	<.001	<.001
Urea nitrogen, mg/dL	22.200[a]	15.500[b]	22.200[a]	16.100[b]	0.82	.629	<.001	.669
Somatic cell score	0.960[c]	2.880[a,b]	2.150[b,c]	3.910[a]	0.404	.008	<.001	.704
pH	6.630[a,b]	6.590[b]	6.640[a]	6.590[b]	0.013	.559	<.001	.526
Milk coagulation traits								
Rennet coagulation time, min	16.400[b]	18.500[a,b]	17.500[b]	21.100[a]	1.170	.083	<.001	.339
k_{20}, min	5.630	4.980	5.450	5.150	0.431	.985	.136	.567
a_{30}, mm	30.100	30.100	29.300	30.000	3.250	.125	.119	.134

k_{20}: curd-firming time; a_{30}: curd firmness 30 min after rennet addition.
[a–c]Least squares means with different superscripts within a row are significantly different ($p < .05$).

Table 5. Least squares means of milking time at different sites on milk yield and milk-related traits.

Milking time	Morning		Evening			p Value		
Site	Lowland	Highland	Lowland	Highland	SEM	Time	Site	Time × Site
No of cows	7	7	7	7				
No of samples	35	35	35	35				
Milk yield, kg/milking	8.720[a]	6.490[b]	6.960[b]	6.350[b]	0.341	<.001	<.001	<.001
Milk composition								
Fat, %	3.310[b]	3.820[a]	4.090[a]	3.850[a]	0.169	<.001	.276	.002
Protein, %	3.340[b]	3.420[b]	3.570[a]	3.390[b]	0.050	<.001	.150	<.001
Casein, %	2.610[c]	2.720[a,b]	2.780[a]	2.680[b,c]	0.043	<.001	.927	<.001
Lactose, %	4.860[a]	4.790[a,b]	4.830[a,b]	4.760[b]	0.041	.079	.019	.929
Urea nitrogen, mg/dL	22.200[a,b]	19.500[b]	23.700[a]	19.700[b]	0.868	.155	<.001	.261
Somatic cell score	1.320[c]	2.780[a]	2.070[b]	2.620[a,b]	0.305	.022	<.001	<.001
pH	6.640	6.640	6.650	6.650	0.012	.083	.909	.212
Milk coagulation traits								
Rennet coagulation time, min	17.700	19.400	18.300	19.800	1.220	.458	.070	.869
k_{20}, min	5.330	4.950	4.670	5.450	0.389	.705	.363	.008
a_{30}, mm	28.500	28.700	29.200	26.200	3.49	.643	.525	.430

k_{20}: curd-firming time; a_{30}: curd firmness 30 min after rennet addition.
[a–c]Least squares means with different superscripts within a row are significantly different ($p < .05$).

that have not been taken into account, such as laboratory procedures, detailed milk protein and mineral composition (Niero et al. 2016a; Visentin et al. 2016) and genetics of cows (Ikonen et al. 2004). Nevertheless, since the cows used in the present study were the same on LO and HI, the latter factor was not contributing in the present study.

Differences between morning vs. evening milk

The effect of daytime of milking was investigated as it has a specific relevance since Fontina cheese has to be manufactured within 2 h after every single respective milking, as stated in the official procedural disciplinary manual (Giannino et al. 2009). Due to the longer nocturnal milking interval (13 h) than that during daytime (11 h), the milk amount was higher ($p < .001$) in the morning than in the evening, but mainly in LO (interaction, $p < .001$) (Table 5). There were also interactions ($p < .01$ to .001) between milking time and site in contents of fat, protein and CN as well as SCS resulted affected by milking daytime. In LO, contents of fat, protein and CN were higher ($p < .05$) in the evening milk, whereas this difference disappeared in HI. The same was true for SCS. One likely reason for that was mainly a dilution effect in LO where morning and evening milk amount clearly differed. The different milk composition did not result in differences between morning and evening milk in pH, RCT, k_{20} and a_{30}. There was a milking time × site interaction ($p < .001$) in k_{20} which decreased in the evening in LO and increased in the evening in HI.

Non-coagulating samples

In the present study, 13.2% of milk samples did not coagulate within 30 min from rennet addition. This proportion was close to 12.9% reported by Toffanin

et al. (2015), who studied MCP through lactodynamographic tool reference analyses in milk from Holstein–Friesian cows kept in an intensive system. Conversely, the proportion found in the present study was considerably greater than the 2.5% reported by Penasa et al. (2014) for milk from Holstein–Friesian, Brown Swiss and Simmental cows reared in multibreed herds. Actually, it is quite difficult to compare the results, because Penasa et al. (2014) predicted MCP from MIRS spectra in the time-span from 5 to 30 min and considering samples that showed values out of this range as non-coagulating (NC) milk. In the present study, the proportion of NC milk samples increased from 8.5% at LO to 15.0% at HI. On average, the NC milk samples had slightly lower contents of fat, protein and CN (3.69, 3.24 and 2.53% vs. 3.90, 3.45 and 2.72%, respectively), but considerably greater SCS (3.35 vs. 2.42) compared with means obtained by the coagulating samples. In the lowland, the NC samples showed a similar fat, protein and CN content (data not shown), but considerably greater SCS (3.24) when compared with mean obtained for coagulating LO milk samples (2.33). Finally, also the highland NC samples had a similar protein and CN content (data not shown), but again a greater SCS (3.60) with respect to the average value obtained for coagulating milk samples of HI (2.50). Overall, these findings suggest that SCS plays a major role in impairing the coagulation aptitude of cow's milk. This relationship was recently confirmed with the help of a study of by Summer et al. (2015) when relating SCS in bulk milk and milk coagulation properties.

Conclusions

In the present study, Aosta Red Pied cows were used as a model to investigate the effect of mountainous

farming and grazing on milk coagulation properties of dual purpose cattle. The highland sojourn indeed compromised these properties in terms of rennet coagulation time, while rennet firming and firmness remained unaffected by the transhumance. The latter was different from effects found in the more susceptible specialised dairy breed cows. It was interesting to note that Aosta Red Pied cows familiar with the transhumance system and the specific alpine pasture areas from previous years (multiparous cows) had no advantage over the primiparous cows. A very undesired phenomenon of the highland conditions was that, along with a substantial increase in somatic cell score, the percentage of non-coagulating milk samples were considerably increased compared to the lowland conditions. This indicates that also the adapted cow types such as the Aosta Red Pied cattle need special care under highland conditions, where milking hygienic standards are more difficult to maintain and cows are more exposed to situations resulting in mammary gland injuries. As the results show, this not only is important for maintaining cow's health but also to produce raw milk with favourable cheese-making properties. Finally, the results show that morning and evening milk appear similarly suitable for cheese-making.

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Disclosure statement

No potential conflicts of interest were reported by the authors.

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References

Agroscope. 2017. Feeding Recommendations and Nutrient Tables for Ruminants; [accessed 2017 Feb 15]. https://www.agroscope.admin.ch/Agroscope/en/home/services/support-services/feedstuffs/feed-recommendations-for-ruminants.html.

Battaglini LM, Renna M, Garda A, Lussiana C, Malfatto V, Mimosi A, Bianchi M. 2009. Comparing milk yield, chemical properties and somatic cell count from organic and conventional mountain farming systems. Ital J Anim Sci. 8:384–386.

Berry NR, Büeler T, Jewell PL, Sutter F, Kreuzer M. 2001. The effect of supplementary feeding on composition and renneting properties of milk from cows rotationally grazed at high altitude. Milchwissenschaft. 56:123–126.

Bovolenta S, Sacca E, Corazzin M, Gasperi F, Biasioli F, Ventura W. 2008. Effects of stocking density and supplement level on milk production and cheese characteristics in Brown cows grazing on mountain pasture. J Dairy Res. 75:357–364.

Buchin S, Martin B, Dupont D, Bornard A, Achilleos C. 1999. Influence of the composition of Alpine highland pasture on the chemical, rheological and sensory properties of cheese. J Dairy Res. 66:579–588.

Christen RE, Kunz PL, Langhans W, Leuenberger H, Sutter F, Kreuzer M. 1996. Productivity, requirements and efficiency of feed and nitrogen utilization of grass-fed early lactating cows exposed to high Alpine conditions. J Anim Physiol Anim Nutr. 76:22–35.

De Marchi M, Dal Zotto R, Cassandro M, Bittante G. 2007. Milk coagulation ability of five dairy cattle breeds. J Dairy Sci. 90:3986–3992.

Franzoi M, Niero G, Penasa M, Cassandro M, De Marchi M. 2017. Development and validation of a new method for the quantification of soluble and micellar calcium, magnesium, and potassium in milk. J Dairy Sci. 101:1–6.

Gaucheron F. 2005. The minerals of milk. Reprod Nutr Developm. 45:473–483.

Giannino ML, Marzotto M, Dellaglio F, Feligini M. 2009. Study of microbial diversity in raw milk and fresh curd used for Fontina cheese production by culture-independent methods. Int J Food Microbiol. 130:188–195.

Ikonen T, Morri S, Tyrisevä AM, Ruottinen O, Ojala M. 2004. Genetic and phenotypic correlations between milk coagulation properties, milk production traits, somatic cell count, casein content, and pH of milk. J Dairy Sci. 87:458–467.

Kreuzer M, Langhans W, Sutter F, Christen RE, Leuenberger H, Kunz PL. 1998. Metabolic response of early-lactating cows exposed to transport and high altitude grazing conditions. Anim Sci. 67:237–248.

Leiber F, Kreuzer M, Jörg B, Leuenberger H, Wettstein HR. 2004. Contribution of altitude and alpine origin of forage to the influence of alpine sojourn of cows on intake, nitrogen conversion, metabolic stress and milk synthesis. Anim Sci. 78:451–466.

Leiber F, Kreuzer M, Nigg D, Wettstein HR, Scheeder MRL. 2005a. A study on the causes for the elevated n-3 fatty acids in cows' milk of alpine origin. Lipids. 40:191–202.

Leiber F, Nigg D, Kunz C, Scheeder MR, Wettstein HR, Kreuzer M. 2005b. Protein composition, plasmin activity and cheesemaking properties of cows' milk produced at two altitudes from hay of lowland and high-alpine origins. J Dairy Res. 72:65–74.

Leiber F, Kreuzer M, Leuenberger H, Wettstein HR. 2006. Contribution of diet type and pasture conditions to the influence of high altitude grazing on intake, performance and composition and renneting properties of the milk of cows. Anim Res. 55:37–53.

Maretto F, Cassandro M. 2014. Temporal variation in genetic diversity and population structure of Burlina cattle breed. Ital J Anim Sci. 13:322–329.

Mazza S, Guzzo N, Sartori C, Mantovani R. 2016. Genetic correlations between type and test-day milk yield in small dual-purpose cattle populations: the Aosta Red Pied breed as a case study. J Dairy Sci. 99:8127–8136.

McMahon DJ, Brown RJ. 1982. Evaluation of Formagraph for comparing rennet solution. J Dairy Sci. 65:1639–1642.

Niero G, Penasa M, Gottardo P, Cassandro M, De Marchi M. 2016a. *Short communication:* selecting the most informative mid-infrared spectra wavenumbers to improve the accuracy of prediction models for detailed milk protein content. J Dairy Sci. 99:1853–1858.

Niero G, Visentin G, Ton S, De Marchi M, Penasa M, Cassandro M. 2016b. Phenotypic characterisation of milk technological traits, protein fractions, and major mineral and fatty acid composition of Burlina cattle breed. Ital J Anim Sci. 15:576–583.

Penasa M, Tiezzi F, Sturaro A, Cassandro M, De Marchi M. 2014. A comparison of the predicted coagulation characteristics and composition of milk from multi-breed herds of Holstein-Friesian, Brown Swiss and Simmental cows. Int Dairy J. 35:6–10.

Politis I, Ng-Kwai-Hang KF. 1988. Effects of somatic cell counts and milk composition on the coagulating properties of milk. J Dairy Sci. 71:1740–1746.

Pretto D, De Marchi M, Dalvit C, Cassandro M. 2009. Comparing profitability of Burlina and Holstein Friesian cattle breeds. Ital J Anim Sci. 8(Supp 3):65–67.

Renna M, Cornale P, Lussiana C, Battaglini LM, Turille G, Mimosi A. 2014. Milk yield, gross composition and fatty acid profile of dual-purpose Aosta Red Pied cows fed separate concentrate-forage versus total mixed ration. Anim Sci J. 85:37–45.

Summer A, Franceschi P, Formaggioni P, Malacarne M. 2015. Influence of milk somatic cell content on Parmigiano-Reggiano cheese yield. J. Dairy Res. 82:222–227.

Tiezzi F, Pretto D, De Marchi M, Penasa M, Cassandro M. 2013. Heritability and repeatability of milk coagulation properties predicted by mid-infrared spectroscopy during routine data recording, and their relationships with milk yield and quality traits. Animal. 7:1592–1599.

Toffanin V, De Marchi M, Lopez-Villalobos N, Cassandro M. 2015. Effectiveness of mid-infrared spectroscopy for prediction of the contents of calcium and phosphorus, and titratable acidity of milk and their relationship with milk quality and coagulation properties. Int Dairy J. 41:68–73.

Visentin G, McDermott A, McParland S, Berry DP, Kenny OA, Brodkorb A, Fenelon MA, De Marchi M. 2015a. Prediction of bovine milk technological traits from mid-infrared spectroscopy analysis in dairy cows. J Dairy Sci. 98:6620–6629.

Visentin G, Penasa M, Gottardo P, Niero G, Isaia M, Cassandro M, De Marchi M. 2015b. Milk coagulation properties of cattle breeds reared in Alpine area. Poljoprivreda. 21:237–240.

Visentin G, Penasa M, Gottardo P, Cassandro M, De Marchi M. 2016. Predictive ability of mid-infrared spectroscopy for major mineral composition and coagulation traits of bovine milk by using the uninformative variable selection algorithm. J Dairy Sci. 99:8137–8145.

Zendri F, Ramanzin M, Cipolat-Gotet C, Sturaro E. 2016. Variation of milk coagulation properties, cheese yield, and nutrients recovery in curd of cows of different breeds before, during and after transhumance to highland summer pastures. J Dairy Res. 84:39–48.