Preliminary communication - Prethodno priopćenje UDK: 637.112.2

Relationship of somatic cell count and composition and coagulation properties of ewe's milk

doi: 10.15567/mljekarstvo.2015.0208

*Javier Caballero Villalobos[1], Ana I. Garzón Sigler[1], Bonastre Oliete[2], Ramón Arias Sánchez[2], Lorena Jiménez[2], Nieves Núñez Sánchez[1], Andrés L. Martínez Marín[1]**

[1]Departamento de Producción Animal, Universidad de Córdoba, Ctra. Madrid-Cádiz, km. 396, 14071 Córdoba, Spain

[2]Laboratorio de Lactología, Centro Regional de Selección y Reproducción Animal de la Junta de Comunidades de Castilla-La Mancha, Avenida del Vino, 10, 13300 Valdepeñas, Ciudad Real, Spain

Received - Prispjelo: 09.12.2014.

Accepted - Prihvaćeno: 02.03.2015.

Abstract

The relationship between somatic cell count (SCC) and raw milk composition and its coagulation properties measured at native or standardised pH values were investigated in Manchega ewes' milk. A total of 84 bulk tank milk samples from flocks included in the National Association of Manchega Sheep Breeders were used. According to their SCC, milk samples were divided into three terciles named low (562±138 cells/mL), medium (956±115 cells/mL) and high (1705±428 cells/mL) SCC groups. Within each SCC group, two pH treatments were applied before determining coagulation properties (rennet clotting time, curd firming time and curd firmness): no acidification of milk (coagulation at native pH) and acidification of milk at pH 6.5. Native milk pH significantly increased ($P<0.05$) as SCC rose. With respect to raw milk composition, fat contents were not affected ($P>0.05$) by SCC, protein content tended to be higher in the high SCC group ($P=0.05$) and lactose content was significantly lower ($P<0.05$) in that group. At native pH, the high SCC group had longer rennet clotting time, higher curd firming time and lower curd firmness after 30 min of rennet addition than the low and medium SCC groups ($P<0.05$). Standardising milk pH at 6.5 prior to rennet addition clearly cancelled out ($P<0.05$) the negative effects of high SCC on milk coagulation properties. In conclusion, despite the fact that acidification before renneting improved the coagulation properties of milk with high SCC, more research would be needed to determine the sensorial properties of cheese manufactured under such conditions.

Key words: sheep, somatic cell count, milk composition, milk clotting

Introduction

A major aspect of the quality of sheep milk is its capability to be transformed into high-quality cheeses and to produce high yields of cheese from each litre of milk (Bencini, 2002). The amount and quality of cheese that can be obtained mainly depends on the coagulation properties of the used milk (Bencini and Pulina, 1997) and its fat and casein contents (Politis and Ng-Kwai-Hang, 1988a). Coagulation properties are rennet clotting time, curd firming time, and curd firmness after 30 and 60 min of rennet addition. These parameters have been shown to be positively related to cheese yield (Ng-Kwai-Hang et al., 1989) and have been commonly used by researchers to assess milk processing performance (Ng-Kwai-Hang et al., 1989; Pirisi et al., 2000). Milk somatic cell count (SCC) is a widely used marker for both udder health and

*Corresponding author: e-mail: pa1martm@uco.es

milk quality. High SCC is either the consequence of an inflammatory process due to the presence of an intramammary infection or, under non-pathological conditions, the result of physiological processes such as advanced stage of lactation (Raynal-Ljutovac et al., 2007). Previous research indicate that high SCC in sheep milk alters milk pH and composition (Vivar-Quintana et al., 2006; Martí de Olives et al., 2013) as well as milk coagulation properties (Pirisi et al., 2000; Bianchi et al., 2004), which in turn reduces cheese yielding capacity of milk and cheese making efficiency (Politis and Ng-Kwai-Hang, 1988a, 1988b). On the other hand, pH before renneting affects milk coagulation properties (Balcones et al., 1996; Bencini, 2002). Since SCC is positively related to milk pH (Raynal-Ljutovac et al., 2007), lowering it would have positive effects on coagulation properties of milk with high SCC (Pirisi et al., 2000). These authors studied the effects of pH prior to renneting (native and standardised at 6.5) on milk coagulation properties, but they did not compare pH treatments within the SCC groups in their study.

Milk of the Manchega sheep breed is used to make Manchego cheese, which is one of the most important and well-known sheep's milk cheese in Spain and abroad. Manchego is a high-fat, hard, ripened cheese, manufactured at a factory level under controlled conditions using raw or pasteurized milk and with starter addition (Gonzalez-Viñas et al., 2001). Therefore, the objective of this study was to determine the effects of SCC on composition and clotting parameters, measured at native and standardised pH values, of Manchega ewe milk.

Materials and methods

A total of 84 bulk tank milk samples from flocks included in the National Association of Manchega Sheep Breeders were used. Milk samples were stored at 4 ºC until analysis within 3 days after collection. For each sample, pH (Crison Basic 20 pH-meter; Crison Instruments, Barcelona, Spain), SCC (Fossomatic; Foss Electric, Hillerød, Denmark), and fat, total protein, lactose and total solid contents (Milko Scan; Foss Electric, Hillerød, Denmark) were determined. According to their SCC, milk samples were divided into three terciles (i.e. 28 samples each group) named low (562±138 cells/mL), medium (956±115 cells/mL) and high (1705±428 cells/mL) SCC groups. Within each SCC group, two pH treatments were applied before determining coagulation properties: no acidification of milk (coagulation at native pH) and acidification of milk at pH 6.5 by adding a lactic acid solution (10 %) before renneting.

Coagulation properties were measured by using a Formagraph (Foss Electric, Hillerød, Denmark). The testing time of the analysis was set up at 60 min to investigate if milk that did not form a curd within the conventional threshold of 30 min showed coagulation aptitude after this time (De Marchi et al., 2012). The measured traits were rennet clotting time (named r, it is the interval in min from the addition of the rennet to the beginning of coagulation), curd firming time (named k20, it is the interval in min from the beginning of coagulation to the moment the width of the graph achieves a separation of 20 mm), and curd firmness (named A30 or A60, it is the width of the graph measured in mm after 30 or 60 min of rennet addition). For the samples that did not form a curd after 60 min, curd firmness values were arbitrarily assumed as 1 mm. Also, in samples that did not curdle after 60 min, coagulation time and curd firming time were attributed an arbitrary value of 60 min.

All data were analysed using the GLM procedure of SAS 9.1 (SAS Institute Inc., Cary, NC). The statistical analysis of milk pH and composition data included SCC group as fixed effect in the model. The statistical analysis of milk coagulation properties included SCC group, pH and their interaction as fixed effects. Tukey's test was used to compare least squares means. Statistical significance was declared at $P<0.05$.

Results and discussion

Composition and pH values of bulk tank milk of Manchega ewes are shown in Table 1. The fact that native milk pH significantly increased ($P<0.05$) as SCC rose is commonly reported in the literature (Raynal-Ljutovac et al., 2007). This effect is attributed to increased permeability of the mammary epithelium, which can lead to the transfer of components from blood to milk, including citrates, bicarbonates, and Na and Cl ions. Higher levels of citrate and bicarbonate may be responsible for elevated pH levels (Harmon, 1994; Kitchen, 1981).

Table 1. Effects of somatic cell count (SCC, mean ± standard deviation in cells/mL) on the composition of bulk tank milk of Manchega ewes

	SCC group			
Traits	Low (562±138) n = 28	Medium (956±115) n = 28	High (1705±428) n = 28	S.E.M
Native pH	6.57 [c]	6.65 [b]	6.81 [a]	0.013
Fat, %	8.37	8.31	8.30	0.104
Total protein, %	6.25	6.21	6.41	0.051
Lactose, %	4.55 [a]	4.58 [a]	4.41 [b]	0.024
Total solids, %	20.01	19.93	19.99	0.141

S.E.M. Standard error of the mean
[a,b,c]Means without a common superscript are statistically different at $P<0.05$

Table 2. Effects of somatic cell count (SCC, mean ± standard deviation in cells/mL) and pH prior to renneting on the coagulation properties of bulk tank milk of Manchega ewes

	SCC group						
	Low (562±138) n = 28		Medium (956±115) n = 28		High (1705±428) n = 28		
Traits[1]	N	L	N	L	N	L	S.E.M.
r, min	28.13 [b]	18.76 [c]	28.91 [b]	20.79 [c]	35.65 [a]	22.78 [bc]	0.760
k20, min	3.96 [b]	3.88 [b]	4.07 [b]	5.19 [ab]	11.62 [a]	4.07 [b]	0.714
A30, m m	15.25 [b]	37.80 [a]	15.09 [b]	35.42 [a]	6.01 [c]	28.90 [a]	1.458
A60, mm	42.43 [ab]	49.00 [a]	36.14 [b]	47.86 [a]	35.77 [b]	47.08 [a]	0.796

N = native pH, L = pH standardised at 6.5, S.E.M. Standard error of the mean
a,b,c - Means without a common superscript are statistically different at $P<0.05$
[1]r - rennet clotting time, k20 - curd firming time, A30 and A60 - curd firmness after 30 and 60 min of rennet addition, respectively

With respect to raw milk composition, fat contents were not affected ($P>0.05$) by SCC. Protein content tended to be higher in the high SCC group ($P=0.05$) while lactose content was lower ($P<0.05$) in that group. Our results are complementary with most previous published research on sheep milk, which indicate that fat content does not change, protein content either increases or does not change, and lactose content decreases with increasing SCC (Albenzio et al., 2004; Nudda et al., 2003; Pirisi et al., 2000; Vivar-Quintana et al., 2006). The negative effect of high SCC on the milk lactose content has been related to a decreased synthesis capacity of the mammary gland due to the damage of epithelial tissue, but also because of a lesser availability of its precursor, glucose, due to competition for energy between secretor cells and those with phagocyte functions (Martí de Olives et al., 2013). On the other hand, the absence of clear changes in protein content could be due to a reduced protein synthesis by the udder and an increment of proteins coming from the bloodstream, both having opposite effects (Vivar-Quintana et al., 2006). Moreover, Martí de Olives et al. (2013) did not observe differences in casein content, but found lower casein to protein ratio due to high SCC in milk of Manchega ewes.

At native pH, the high SCC group had longer rennet clotting time, higher curd firming time and lower curd firmness after 30 min of rennet addition than the low and medium SCC groups ($P<0.05$, Table 2). Curd firmness after 60 min was slightly lower, but not statistically different, in the medium and high SCC groups compared with the low SCC group. Standardising milk pH at 6.5 decreased ($P<0.05$) rennet clotting time and stepped up ($P<0.05$) curd firmness at 30 min in all SCC groups. It also decreased ($P<0.05$) curd firming time in the high SCC group and increased curd firmness at 60 min in the medium and high SCC groups. Albenzio et al. (2004), Bianchi et al. (2004) and Pirisi et al. (2000) also reported poorer coagulation properties of ovine milk as SCC increased. The negative relationships between SCC and milk coagulation properties could to be related to increased casein breakdown due to a higher plasmin activity in the milk with high SCC (Albenzio et al., 2005; Bianchi et al., 2004; Leitner et al., 2004). In addition, the high SCC could negatively affect both the first phase of rennet coagulation due to elevated pH, as the optimum pH of chymosin activity is in the acidic range (Kumar et al., 2010), and the aggregation of paracasein micelle due to a lower Ca^{2+} activity, as suggested by Leitner et al. (2004). On the other hand, Ng-Kwai-Hang et al. (1989) observed that a shorter clotting time, a faster rate of firming, and a harder curd resulted in a higher cheese yield and efficiency due to lower losses of milk fat and protein in whey. Moreover, Pirisi et al. (2000) and Albenzio et al. (2004) reported a greater loss of protein in whey when ewe milk with high SCC was used for cheese manufacture. Therefore, higher yields of cheese might be expected in the milk with high SCC renneted at standardised pH compared with milk renneted at native pH.

Cheese producers can adjust pH value of milk to achieve the desired acidity by varying the percentage of inoculum of starter cultures (Bencini and Pulina, 1997) or by direct addition of food-grade acid (Lucey and Kelly, 1994). However, despite the fact that direct acid addition improved milk coagulation properties in ovine milk with high SCC in the present work, it should be noted that some authors have reported off-flavours and texture defects after ripening in cheese manufactured from high SCC ovine milk (Jaeggi et al., 2003; Revilla et al., 2007).

Conclusions

High SCC decreased milk lactose content and worsened clotting parameters (rennet clotting time, curd firming time and curd firmness). Standardising milk pH at 6.5 prior to rennet addition clearly cancelled out the negative effects of high SCC on coagulation properties. However, further studies should be performed to determine the sensorial properties of cheese manufactured under such conditions.

Acknowledgments

This work was financed by the PII10-0003 project of the Viceconsejería de Ciencia y Tecnología (Junta de Comunidades de Castilla-La Mancha, Spain). It was part of a research line continued in the RTA2011-00057-C02 project of the Instituto Nacional de Investigación y Tecnología Agraria y Alimentaria (Ministerio de Economía y Competitividad, Madrid, Spain).

Odnos broja somatskih stanica i sastav i koagulacijska svojstva ovčjeg mlijeka

Sažetak

Cilj ovog rada bio je istražiti odnos između broja somatskih stanica, sastava sirovog mlijeka, i koagulacijskih svojstava sirovog mlijeka kod prirodne i standardizirane pH vrijednosti mlijeka mančego ovce. Analizirano je 84 uzoraka skupnog mlijeka stada Državnog udruženja uzgajivača mančego ovce. Prema broju somatskih stanica/mL (BSS/mL), uzorci mlijeka podijeljeni su u 3 skupine - niska (562±138 BSS/mL), srednja (956±115 BSS/mL) i visoka (1705±428 BSS/mL). U okviru svake skupine, pH je mjeren dva puta prije određivanja koagulacijskih osobina (vrijeme zgrušavanja sirilom, vrijeme učvršćivanja gruša i tvrdoća gruša): bez zakiseljavanja mlijeka (koagulacija kod prirodne pH vrijednosti)

i zakiseljavanje kod pH vrijednosti 6,5. pH vrijednost mlijeka signifikantno je povećana (P<0,05) povećanjem BSS. BSS nije utjecao (P>0,05) na udjel masti, dok je udjel proteina bio veći u grupi s visokim BSS (P=0,05), a udjel laktoze bio je signifikantno niži (P<0,05) u toj skupini. Kod prirodne pH vrijednosti mlijeka, visoki BSS utjecao je na duže vrijeme zgrušavanja mlijeka sirilom, sporije učvršćivanje gruša i na manju čvrstoću gruša nakon 30 min od dodatka sirila, u odnosu na skupinu u kojoj je BSS bio nizak i srednji (P<0,05). Standardizacija pH mlijeka na 6,5 prije dodavanja sirila anulirala je (P<0,05) negativan utjecaj visokog BSS na koagulacijska svojstva mlijeka. Može se zaključiti da su, unatoč činjenici kako je acidifikacija mlijeka prije zgrušavanja poboljšala koagulacijske osobine mlijeka s visokim BSS, daljnja istraživanja potrebna kako bi se utvrdila senzorska svojstva sira proizvedenog takvim postupkom.

Ključne riječi: ovce, broj somatskih stanica, sastav mlijeka, zgrušavanje mlijeka

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