

## Alpha<sub>s1</sub>-casein, milk composition and coagulation properties of goat milk

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### Abstract

Amounts of alpha<sub>s1</sub>-casein ( $\alpha_{s1}$ -CN), protein, fat, SNF and total solids were measured in 125 goat milk samples. Coagulation time, coagulation rate and curd firmness were measured in 75 goat milk samples by dynamic mechanical analysis using a Bohlin VOR Rheometer. After adjustments were made for month, time of milk collection and animal age, it was confirmed that goat milk with high percent total solids, SNF, fat and protein coagulated faster (high coagulation rate) and formed a firmer curd than milk that had lower levels of milk components. Coagulation was delayed (long coagulation time) in milk with high protein but resulting curds were firmer than curds made from low protein milks. Amount of  $\alpha_{s1}$ -CN was positively correlated with milk components and coagulation time. Goat milk that lacked  $\alpha_{s1}$ -CN had lower percentages of milk components and poorer coagulation properties than milk that contained  $\alpha_{s1}$ -CN, suggesting that the presence of  $\alpha_{s1}$ -CN in milk should improve coagulation properties. However, percent total solids, SNF and protein were more highly correlated with coagulation properties than  $\alpha_{s1}$ -CN. Thus, measuring total solids, SNF or protein may be more practical in predicting cheese-making potential of goat milk than measuring  $\alpha_{s1}$ -CN, which is more tedious and expensive. Milk from Nubians and Nubian×Alpine crosses contained a higher amount of  $\alpha_{s1}$ -CN and other milk components, and exhibited higher coagulation rate and curd firmness than milk from Toggenburgs and Saanen×Alpine crosses. Selection of goats with high solids, particularly Nubians, is recommended if cheese-making is the objective. © 2000 Elsevier Science B.V. All rights reserved.

*Keywords:* Goat milk; Milk composition; Goat cheese; Coagulation; Alpha<sub>s1</sub>-casein

### 1. Introduction

Coagulation properties of milk are of interest because they influence cheese yield and quality (Storry and Ford, 1982a; Okigbo et al., 1985b; Osteren et al., 1997). Cheese-making conditions such as culture and enzyme type/concentration and incubation

temperature, as well as individual milk components, influence coagulation properties (Storry and Ford, 1982b; Okigbo et al., 1985b). Thus, any factor that influences milk composition also influences coagulation properties (Storry et al., 1983; Politis and Ng-Kwai-Hang, 1988; Aleandri et al., 1989). Ruminant milk composition is influenced by breed, age, stage of lactation, lactation number, month of sampling, nutrition, and environmental and genetic factors (Storry et al., 1983; Politis and Ng-Kwai-Hang, 1988; Aleandri et al., 1990; Clark, 1993). While not all of these factors may be known or can be controlled by breeders, proper selection of dairy goats on the basis of

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breed or of milk composition should enhance the cheese-making ability of goat milk.

Rennet-induced coagulation of milk proceeds in two phases. The first, or enzymatic phase, involves rapid proteolysis of approximately 90% of the kappa-casein ( $\kappa$ -casein) (Walstra et al., 1984). When  $\kappa$ -casein is hydrolyzed, the casein micelle becomes unstable and susceptible to precipitation by calcium. Aggregation of the casein micelles occurs during the second, or non-enzymatic phase (Garnot and Olson, 1982; Storry and Ford, 1982a; Walstra et al., 1984). Coagulation time (CT) is the point at which coagulation is first notable; the time when casein micelles have aggregated sufficiently to form visible flocs. Coagulation rate (CR) is a measure of how quickly the curd firms once coagulation has begun. Ideally, cheese-makers would like to minimize CT and maximize CR since both influence the processing time required for cheese production. Most importantly, cheese-makers want to maximize curd firmness (CF) since it is the primary coagulation property that influences cheese quality, yield, and economic returns. A firm curd improves cheese yield by encouraging retention of milk components (Bynum and Olson, 1982; Okigbo et al., 1985a,b; Marziali and Ng-Kwai-Hang, 1986; Politis and Ng-Kwai-Hang, 1988; Aleandri et al., 1990; Martin and Addeo, 1996).

Extensive research has been conducted to study the relationships between cow milk composition and coagulation properties (Jen and Ashworth, 1970; Schaar, 1984; Okigbo et al., 1985b; Marziali and Ng-Kwai-Hang, 1986; Pagnacco and Caroli, 1987; Politis and Ng-Kwai-Hang, 1988). However, this type of research on goat milk is more limited (Storry et al., 1983; Remeuf and Lenoir, 1986; Ambrosoli et al., 1988). Cow and goat milk researchers agree that high fat and protein, casein in particular, contribute to better coagulation properties and cheese yield. In addition, research has shown that when protein or casein levels are high in cow milk, CT tends to be short, CR fast, and CF high (Jen and Ashworth, 1970; Schaar, 1984; Okigbo et al., 1985b; Marziali and Ng-Kwai-Hang, 1986; Politis and Ng-Kwai-Hang, 1988). On the other hand, Ambrosoli et al. (1988), who studied goat milk, showed that when protein or casein levels were high, CR was fast, and CF was high, but CT was long.

Until the late 1980s, it was believed that goat milk lacked the protein alpha<sub>s1</sub>-casein ( $\alpha_{s1}$ -CN) (Jenness,

1980; Grosclaude et al., 1987; Brignon et al., 1989; Mora-Gutierrez et al., 1991).  $\alpha_{s1}$ -CN, the primary protein in cow milk, is a structural component of the casein micelle, and plays a functional role in cheese curd formation (Walstra et al., 1984). Cow and goat milk contain similar proportions of  $\kappa$ -CN (10–24%) and  $\alpha_{s2}$ -CN (5–19%). However, goat milk contains higher levels of beta-casein ( $\beta$ -CN: 42–64% versus 34–41%) and lower levels of  $\alpha_{s1}$ -CN (4–26% versus 36–40%) than cow milk (Walstra et al., 1984; Law and Tziboula, 1992). As  $\alpha_{s1}$ -CN participates in cheese curd formation, the impact of low  $\alpha_{s1}$ -CN in milk can be significant. One result of this difference is that cheese curds made from goat milk tend to be softer than those of cow milk even at similar casein levels (Storry et al., 1983; Remeuf and Lenoir, 1986; Ambrosoli et al., 1988; Risch, 1992). Optimization of curd firmness is important to cheese-makers to maximize cheese yield. In addition, shorter CT and higher CR would shorten the processing time required for cheese production.

The potential for improving milk coagulation properties by selecting for high  $\alpha_{s1}$ -CN milk producers may be significant, but it is first necessary to establish whether  $\alpha_{s1}$ -CN is related to superior composition and coagulation properties. Previous research on milk from Saanen and Alpine goat breeds showed that  $\alpha_{s1}$ -CN is correlated with other milk components and coagulation properties (Ambrosoli et al., 1988). Samples were categorized as having a low or high amount of  $\alpha_{s1}$ -CN. Milks with high levels of  $\alpha_{s1}$ -CN had higher total solids, total protein, casein and phosphorus, and lower pH than milks with low levels of  $\alpha_{s1}$ -CN. Milks with high levels of  $\alpha_{s1}$ -CN also had long CT and formed firm curds. Ambrosoli et al. (1988) did not study milk from Nubians, Toggenburgs, Oberhaslis, or LaManchas but suggested that an unknown breed-linked factor may exist. Mora-Gutierrez et al. (1991), who examined milk from 25 Alpine and Nubian goats, did not see distinct breed differences in amount of  $\alpha_{s1}$ -CN, but saw great variability within breeds, suggesting genetic regulation of the protein.

To date, no studies have surveyed relationships between milk composition and coagulation properties of the six most common dairy goat breeds in USA and cross-breed combinations. The objective of this study was to observe how total solids, SNF, fat, protein,

casein,  $\alpha_{s1}$ -CN, and breed influence CT, CR and CF of goat milk.

## 2. Materials and methods

### 2.1. Milk collection

One hundred and twenty-five (125) milk samples were collected from 90 individual dairy goats. Seventy samples were collected between June–August 1994; 55 samples were collected in May, June, October and November 1996. Sampling was done during the regular morning (05.00 h) or evening (17.00 h) milking. All milk samples were collected from Side Hill Acres, Candor, NY; a herd composed of about 200 dairy goats representing Alpine, LaMancha, Nubian, Oberhasli, Saanen, Toggenburg breeds and some crosses. Herd feeding and health conditions were good. Goats were randomly selected for milk collection based on stanchion position. Each sample was collected mid-way through the milking process by removing inflations and hand-milking approximately 50 ml into individual Whirlpak<sup>®</sup> bags which were then placed on ice. Samples were transported to Cornell University where milk composition and coagulation properties were analyzed that afternoon (if morning collection) or refrigerated and analyzed the following morning (if previous evening collection).

### 2.2. Milk composition

Total solids were determined in duplicate by oven drying (AOAC, 1990c). Fat content was determined in duplicate using the Mojonnier ether extraction method (AOAC, 1990b). Solids-non-fat were calculated by subtraction. The Acid Orange 12 dye binding method (AOAC, 1990a), modified for use with a Bausch & Lomb Spectronic 20 (Rochester, NY) and a Bausch & Lomb short path length flow through cuvet (0.299 mm), was used to determine the true protein content of milk (personal communication, Udy Corp., 1991). The AO12 dye binding method for percent true protein was calibrated against the Kjeldahl Direct method for determination of percent true protein (AOAC, 1991; Clark, 1993). Casein content of 55 samples (milk collected in 1996 only) was determined by first precipitating the casein with acetic acid and

sodium acetate, then substituting the whey protein filtrate for milk in the modified AO12 dye binding method.

### 2.3. Coagulation properties

Dynamic mechanical analysis was chosen for analyzing the coagulation properties of goat milk because it continuously and non-destructively measures the sinusoidal elastic and viscous response of the curd during its formation and provides information on CR and strength over time (Bohlin et al., 1984). Due to equipment availability, coagulation properties of only 75 individual goat milk samples were analyzed in two sets of experiments, separated by 18 months. Based on Bohlin et al. (1984), CR, CF and CT were measured on goat milk samples using a bob (#100 004) and cup (#100 087) assembly unit and 1.064 g cm torsion bar in a Bohlin VOR Rheometer (Bohlin Reologi Incentive Group, Cranbury, NJ). An IBM PS/Value Point computer (IBM, Armonk, NY) controlled the system.

A goat cheese-making method was designed to mimic farmstead cheese-making (Le Jaouen, 1987), except that no pasteurization or heat treatment was used. As temperature, culture, and rennet affect coagulum development, treatment conditions were held constant and the same lots of culture or rennet were used within an experiment. The procedure used for analyzing coagulation properties of 45 samples tested in the summer of 1994 is as follows. First, 500  $\mu$ l (6 ml/kg) MD088 Ezal<sup>®</sup> lactic culture (Marschall products, Groupe Rhone-Poulenc, Dange Saint-Romain, France) were added to a 25 ml sample of goat milk maintained at 35°C. After 10 s, calf rennet (Chr. Hansen, Inc., Milwaukee, WI) diluted 1:10 (to obtain 0.1 ml/kg milk) was added to the milk and mixed by inversion for 15 s. After 30 s, 15 ml of the milk mixture was poured into the Bohlin cup, and the bob was lowered below the surface of the milk. An air-diverting funnel was secured around the bob and cup to minimize surface dehydration. At 120 s, oscillation was initiated. The Bohlin Rheometer was set at 1 Hz frequency and 1% amplitude. Measurements including complex modulus ( $G^*$ , Pa), storage modulus ( $G'$ , Pa), loss modulus ( $G''$ , Pa), phase change ( $\delta$ ), range (% deflection), and viscosity of the sample were made at 60 s intervals for 3900 s.

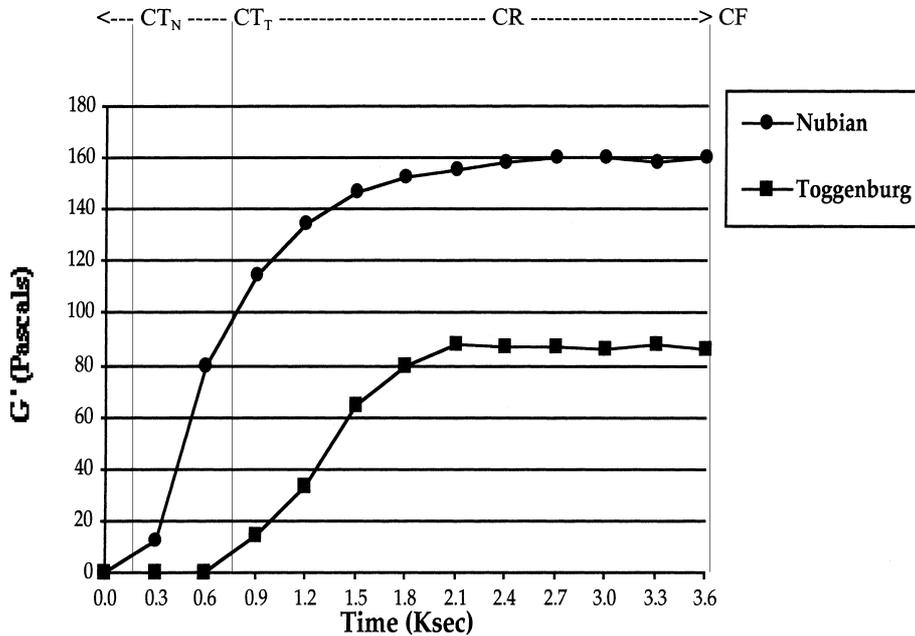


Fig. 1. Typical results of coagulating goat milk from dynamic mechanical analysis on the Bohlin VOR Rheometer, indicating time (ks) and  $G'$  (Pa). Coagulation time for Nubian ( $CT_N$ ) and Toggenburg ( $CT_T$ ) breeds, coagulation rate (CR), and curd firmness (CF) are noted above the graph.

Thirty goat milk samples were collected and analyzed in the fall of 1996. The analysis procedure was the same as that used in 1994, except that the samples were maintained at  $38^\circ\text{C}$  and coagulated with  $300\ \mu\text{l}$  culture and microbial rennet (Chr. Hansen, Inc., 100% strength chymosin from *Aspergillus niger*). These differences were controlled in the statistical analysis.

Fig. 1 shows typical coagulation results of Nubian and Toggenburg milk obtained from dynamic mechanical analysis (DMA) on the Bohlin Rheometer. Coagulation time is indicated for both Nubian ( $CT_N$ ) and Toggenburg ( $CT_T$ ) milks. CT was defined as the point when the range readings (data not shown) were greater than or equal to 0.50% (because the torque element was not sensitive enough to record results accurately below a range of 0.50%). Range measures the deflection ( $\pm 100\%$ ) of the measuring head (torque element), or the strength of the signal. The CT for the Nubian milk sample was 300 s, and for Toggenburg milk was 900 s.

Periods of time where CR and CF were measured are indicated above the figure. CR was defined as the slope of the logarithmic function of  $G'$  on time, between 300 and 3600 s. The CR of the Nubian milk

sample was 129 Pa/s and the Toggenburg milk sample was 105 Pa/s. Curd firmness was defined as the elastic properties, or  $G'$  value, measured 1 h (3600 s) after the start of oscillation. The CF measurement was 160 Pa for this Nubian milk sample and 85 Pa for the Toggenburg milk sample.

#### 2.4. Isolation and identification of $\alpha_{s1}$ -CN

Casein was isoelectrically precipitated from whole goat milk. First, skim milk was separated from whole milk by centrifugation in a Sorvall RC-5B (DuPont Co., Wilmington, DE) at  $2500\times g$  (4250 rpm) and  $30^\circ\text{C}$  for 20 min. The resulting skim milk was diluted with an equal volume of distilled water and 10% acetic acid. After mixing, 1 M sodium acetate was added, and the mixture was centrifuged at  $500\times g$  (1800 rpm). The casein pellicle was dispersed in distilled water and dissolved with 0.2 N NaOH. Samples were coagulated and washed a total of four times, ending with washed casein. Individual isoelectrically precipitated casein samples were frozen until reverse-phase high pressure liquid chromatography (RP-HPLC) was performed on the samples.

$\alpha_{s1}$ -CN was separated from the other caseins and identified using an RP-HPLC method adapted from Jaubert and Martin (1992). Lyophilized standards from whole goat casein were supplied by Mahe (INRA Laboratoire de Genetique Biochimique, Centre de Jouy-en-Josas, France).

Before analysis, the standard (10 ppm) or sample (100 ppm) was reduced with 10 mM dithiothreitol in a 40°C Versa-bath<sup>®</sup> (Fisher Scientific, Pittsburgh, PA) for 1 h, then diluted with the same volume of 0.10% trifluoroacetic acid and filtered through a 0.45  $\mu$ m Millipore (Bedford, MA) filter. Due to the fast degradation of trifluoroacetic acid, all isoelectrically precipitated casein and lyophilized standards were prepared (denatured, diluted, and filtered) within 2 h of the HPLC run to minimize ionic species degradation and baseline drift.

Mobile phase solvent A, 0.10% trifluoroacetic acid (Sigma Chemical Co., St. Louis, MO) in HPLC grade water ( $\sim$ pH 2.00) and solvent B, 0.096% trifluoroacetic acid in 80% acetonitrile ( $\sim$ pH 1.80) were prepared daily to minimize degradation of trifluoroacetic acid. Fresh solvents were dispensed into smaller flasks that were tempered to 40°C in the Versa-bath<sup>®</sup> and continuously flushed with high purity grade helium gas.

Two Waters 501 HPLC pumps were controlled by a Waters Automated Gradient Controller (Waters division of Millipore, Milford, MA). A linear gradient was run from 42 to 58% solvent B in 42 min. The Waters 484 Tunable Absorbance Detector gain was set at 0.005 absorbance units full scale and the eluent absorbance was continuously monitored at 214 nm. A Hewlett-Packard 3392A integrator (Palo Alto, CA) recorded the results. Chromatography was run on a Phenomenex (Torrance, CA) reverse-phase C4 column (Atlantis 5 mm C4 300A, 4.6 mm $\times$ 25 cm) and guard column (Atlantis 5 mm C4 300A, 4.6 mm $\times$ 30 mm). A Rheodyne (Cotati, CA) model 7010 sample injector was used to deliver a 20  $\mu$ l sample onto the column. A Powerstat voltage control unit (Superior Electric Co., now carried by Warner Electric, Ann Arbor, MI), attached to heating tape (Glas-col Apparatus Co., Terre Haute, IN) insulated with flame resistant tape, was used to maintain the column at 40°C for optimal separation of peaks.

Retention times and peak areas of each casein peak of known standards and samples were entered into a

Microsoft Excel<sup>™</sup> (Redmond, WA) spreadsheet. Retention times were calculated for each peak, relative to  $\beta$ -CN. Zero, one, or two peaks could be present in the  $\alpha_{s1}$ -CN region. By the use of standards and comparison with previously published results for caprine caseins (Jaubert and Martin, 1992), the peaks were identified. Individual  $\alpha_{s1}$ -CN peak areas were added to obtain a value of total  $\alpha_{s1}$ -CN. The value was then compared to the sum of all casein peaks, total CN peak area, to obtain a value of  $\alpha_{s1}$ -CN relative to total CN for statistical analysis. Relative values of  $\alpha_{s1}$ -CN/total CN were also categorized into 'null', 'intermediate' and 'high'  $\alpha_{s1}$ -CN level groups for additional statistical analysis. Null samples were defined as those that lacked  $\alpha_{s1}$ -CN peaks, intermediate samples contained 0.01–0.24%  $\alpha_{s1}$ -CN relative to total CN, and high samples had 0.25% or more  $\alpha_{s1}$ -CN relative to total CN.

### 2.5. Statistical methodology

Pearson correlation coefficients were determined using SAS statistical software (SAS/STAT<sup>®</sup>, 1989). A variant of the Kruskal–Wallis non-parametric test for one-way ANOVA by ranks was used to detect differences between the six breeds and two cross-breeds, controlling for month of collection and linear and quadratic terms (Zar, 1996). As milk components exhibited a curvilinear trend over months, the quadratic month term was included in the analysis. A general linear model (GLM) was used to analyze 'null', 'intermediate' and 'high' group differences (MINITAB, 1991). Differences were considered significant when resultant *p*-values were less than 0.05.

## 3. Results and discussion

### 3.1. Separation of casein

Fig. 2 shows RP-HPLC chromatograms for (a) a standard that lacks  $\alpha_{s1}$ -CN and (b) a standard that contains  $\alpha_{s1}$ -CN. The bracketed region labeled I indicates  $\kappa$ -CN, II indicates  $\alpha_{s2}$ -CN, III indicates  $\alpha_{s1}$ -CN, and IV indicates  $\beta$ -CN. Elution order of caseins is supported by previous workers (Mora-Gutierrez et al., 1991; Jaubert and Martin, 1992). Double peaks in the  $\kappa$ -CN,  $\alpha_{s1}$ -CN, and  $\alpha_{s2}$ -CN regions of chromatograms

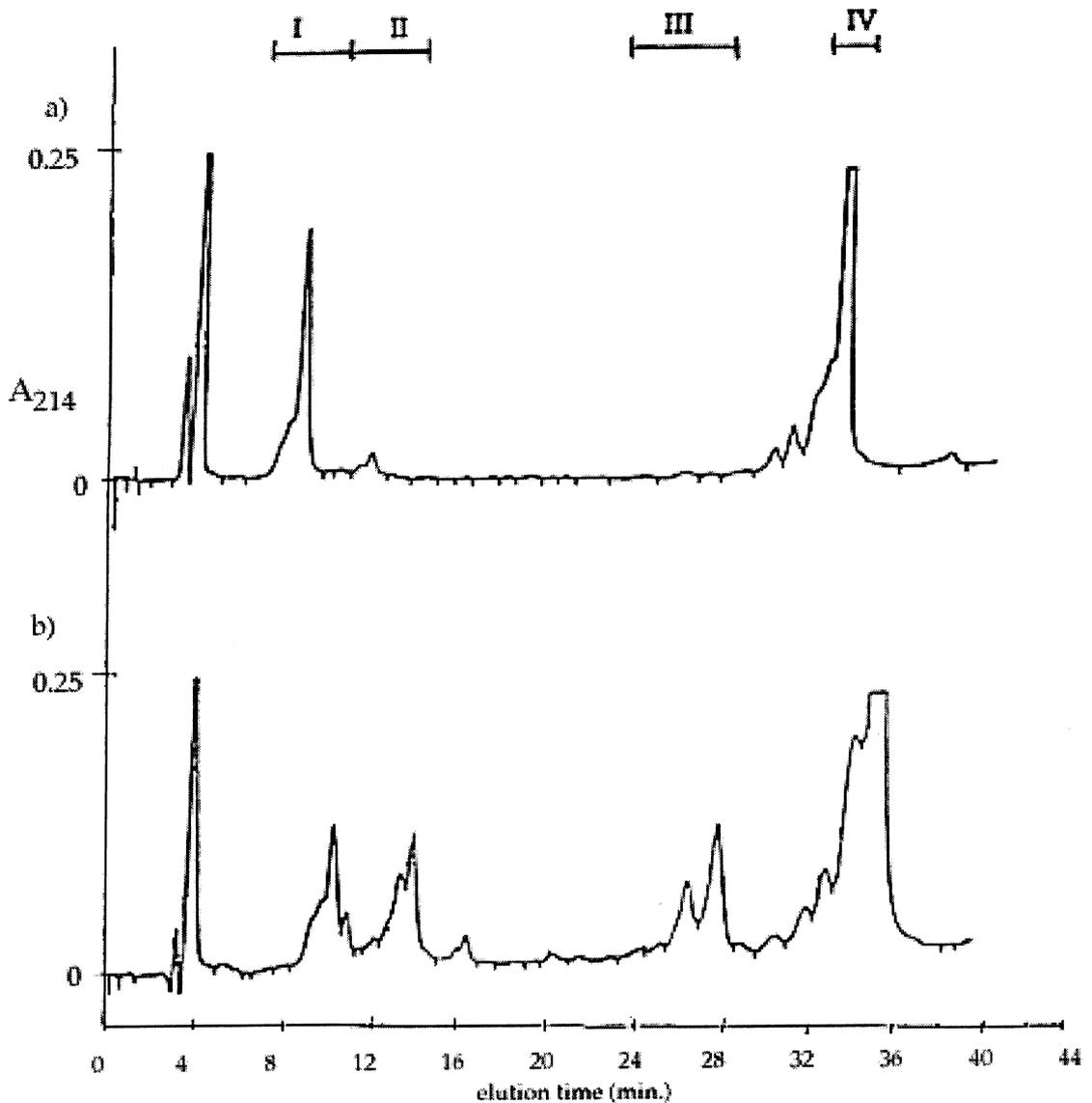


Fig. 2. Separation of goat milk caseins by RP-HPLC. Standards: (a) lacks  $\alpha_{s1}$ -CN and (b) contains  $\alpha_{s1}$ -CN (standards provided by INRA). Regions indicated: I —  $\kappa$ -CN, II —  $\alpha_{s2}$ -CN, III —  $\alpha_{s1}$ -CN and IV —  $\beta$ -CN.

indicate that some genetic variants of each protein can be separated by this method.

### 3.2. Relationships between milk composition and coagulation properties

After adjustments were made for month, time of milk collection and animal age, a number of

significant Pearson correlations ( $r$ -values) were found to exist between milk composition and coagulation properties (Table 1). Although repeated measures were taken on 35 goats (29 goats twice, five goats three times, and one goat four times), all measures were considered independent in the statistical analysis and the repeated measures did not affect the conclusions.

Table 1  
Pearson correlations between goat milk composition and coagulation properties<sup>a</sup>

	TS	SNF	F	TP	CN	$\alpha_{s1}$	CT	CR
SNF	0.64***							
F	0.93***	0.32***						
TP	0.53***	0.84***	0.25**					
CN	0.44**	0.76***	0.15	0.92***				
$\alpha_{s1}$	0.32***	0.33***	0.23*	0.33***	0.51***			
CT	0.15*	0.33**	0.02	0.40***	0.30	0.26*		
CR	0.59***	0.64***	0.44***	0.70***	0.48**	0.09	-0.02	
CF	0.50***	0.43***	0.44***	0.37**	0.31	0.17	0.33**	0.65***

<sup>a</sup> TS: total solids, SNF: solids-not-fat, F: fat, TP: protein, CN: casein,  $\alpha_{s1}$ :  $\alpha_{s1}$ -CN, CT: coagulation time, CR: coagulation rate and CF: curd firmness.

\*  $p < 0.05$ ; \*\*  $p < 0.01$ ; \*\*\*  $p < 0.001$ .

All milk components were positively correlated with amount of  $\alpha_{s1}$ -CN. Amount of  $\alpha_{s1}$ -CN was most highly correlated with percent casein, followed by SNF, protein, total solids, and fat. These findings are consistent with Ambrosoli et al. (1988), who reported that  $\alpha_{s1}$ -CN was positively correlated with total solids, total protein and casein.

During coagulation, casein and calcium phosphate bridge micelles together to form a network which entraps fat and other solids. It follows, then, that more available protein, primarily in the form of casein, should favor rapid formation of the casein network, entrapment of more solids into the curd, and a firmer curd. Our results support this statement. CR was positively correlated with percent total solids, SNF, fat, protein, casein, and CF. CF also showed significant positive correlations with percent total solids, SNF, fat and protein. The relationships of casein with CF and CT were not significant due to the low number of casein measures corresponding to coagulation property measures.

CT was positively correlated with percent total solids, SNF, protein,  $\alpha_{s1}$ -CN and CF. These relationships show that milk with high solids, particularly protein, formed curd more quickly (high CR) and firmly (high CF) than milk with low solids. These findings are supported by other authors who studied fewer animals or only a couple of breeds (Storry et al., 1983; Remeuf and Lenoir, 1986; Ambrosoli et al., 1988). Ambrosoli et al. (1988) studied the milk of 89 Alpine and Saanen goats that were distributed in four herds, and found that milk with high total solids, protein, casein, and phosphorus also had high CT,

CR and CF. Remeuf and Lenoir (1986) studied the milk of 60 Alpine and Saanen goats and also found that casein was positively correlated with CR and CF. Storry et al. (1983) looked at five samples of goat milk representing four different breeds and concluded that casein, fat, calcium, phosphorus, and magnesium were positively correlated with CF.

Milk with high solids, particularly SNF and protein, began coagulating later (long CT) than milk with low solids, suggesting protein delays the onset of coagulation. These results are supported by Ambrosoli et al. (1988) who found that coagulation began earlier in goat milk with low casein than in goat milk with high casein. It is suggested that the elongation of CT at high protein levels may in part be due to the presence of higher amounts of  $\alpha_{s1}$ -CN and  $\alpha_{s2}$ -CN in the milk. These two protein fractions may delay curd formation by binding  $\text{Ca}^{2+}$  ions, making fewer available for binding after proteolysis of  $\kappa$ -casein by rennet (Ambrosoli et al., 1988). The positive correlation between CT and CF in the present study shows that milks that took a long time to begin coagulating also formed firmer curds. However, this does not imply that a long CT will always lead to high CF.

Although processors would like to minimize CT and maximize CR, CF is probably the most critical coagulation property because it influences cheese yield. Since total solids were most significantly positively correlated with CF, it may be profitable to supplement solids or concentrate goat milk prior to cheese-making if the goal is to enhance CR and CF.

When  $\alpha_{s1}$ -CN amount was categorized into 'null', 'intermediate' and 'high' level clusters and analyzed

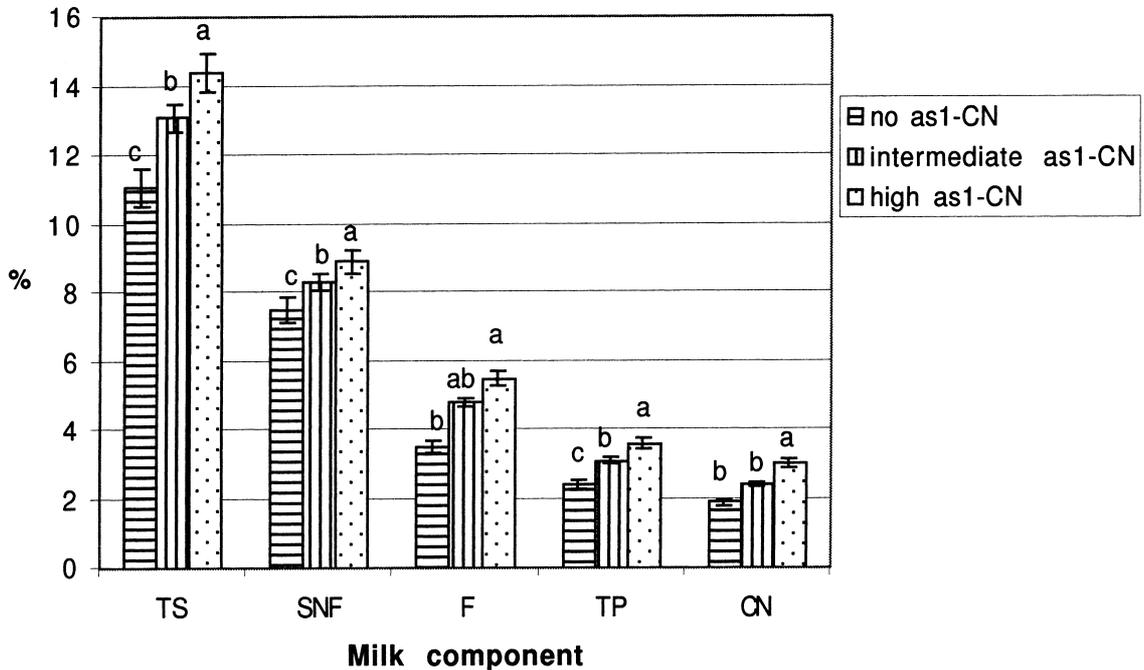


Fig. 3. The effect of null, intermediate and high levels of  $\alpha_{s1}$ -casein upon goat milk composition (TS, total solids; SNF, solids-not-fat; F, fat; TP, protein; CN, casein). Different letters above bars indicate that significant differences exist between the means of the same heading.

by GLM, some notable differences were found (Figs. 3 and 4). Goat milks containing higher  $\alpha_{s1}$ -CN had significantly greater levels of all milk components than milks without  $\alpha_{s1}$ -CN. In addition, milk that contained intermediate amounts of  $\alpha_{s1}$ -CN had significantly more total solids, SNF, protein and casein than null samples (Fig. 3). Samples with high  $\alpha_{s1}$ -CN also had significantly higher total solids, SNF and protein than samples containing intermediate amounts.

Average CR values of milks containing high amounts of  $\alpha_{s1}$ -CN were significantly higher than for milks lacking  $\alpha_{s1}$ -CN (Fig. 4). Study samples that lacked  $\alpha_{s1}$ -CN exhibited the shortest CT, low CR and low CF, but no significant differences in CT ( $p=0.08$ ) or CF ( $p=0.06$ ) were found due to the low number of null samples ( $n=4$ ) and variability in the coagulation properties. Ambrosoli et al. (1988) showed that high levels of  $\alpha_{s1}$ -CN were associated with long CT and high CF, but no conclusions were made regarding CR.

In the present study, total solids, SNF, protein and fat were highly correlated ( $p<0.01$ ) with CR and CF

(Table 1). Total solids and SNF indicated cheese-making suitability more effectively than  $\alpha_{s1}$ -CN itself. Therefore, it is not recommended that  $\alpha_{s1}$ -CN alone be measured for the purpose of predicting coagulation properties since the methodology is more time-consuming and expensive than traditional analytical methods used to determine milk composition.

### 3.3. Relationships among breed, milk composition, and coagulation properties

Since milk components were positively correlated with CR and CF, it would follow that milk of breeds with high solids milk should have good coagulation properties. This was found to be the case. Significant differences in milk composition were found between each of the six goat breeds and two cross-breed combinations tested (Table 2). Nubian and Nubian  $\times$  Alpine crosses produced milk that was high in all milk components. Milk from the Nubian breed was found to have the highest percent total solids, SNF, fat and protein (supported by USDA (1998) production breed

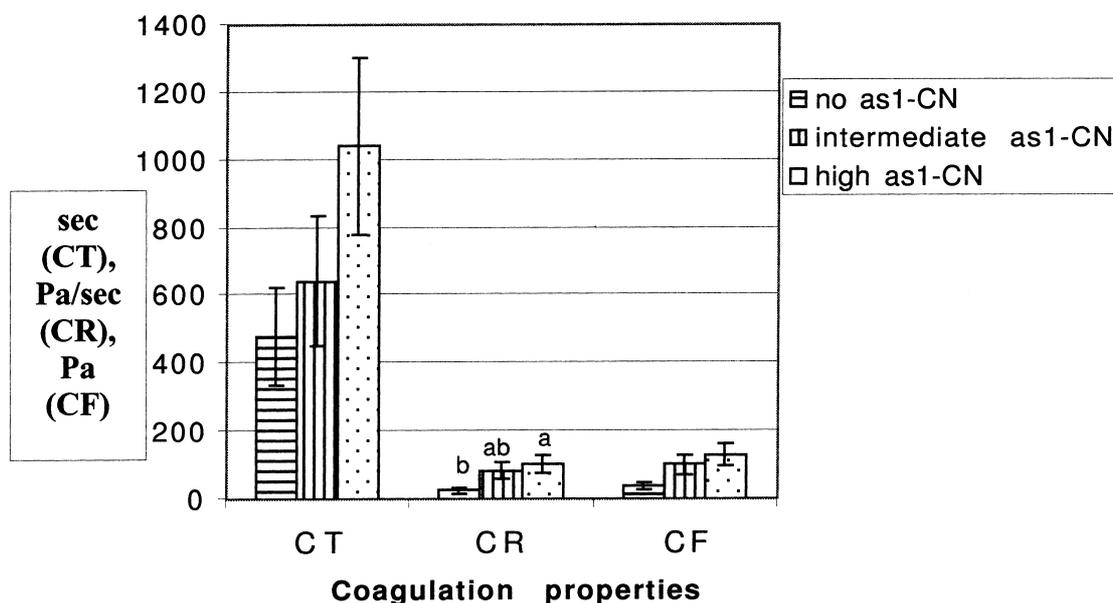


Fig. 4. The effect of null, intermediate and high levels of  $\alpha_{s1}$ -casein upon goat milk coagulation properties (CT, coagulation time; CR, coagulation rate; CF, curd firmness). Different letters above bars indicate that significant differences exist between the means of the same heading.

averages); and Nubian  $\times$  Alpine crosses had milk with the highest average percent  $\alpha_{s1}$ -CN. Milk from Nubians, LaManchas, Saanens, and Nubian  $\times$  Alpine crosses had higher amounts of  $\alpha_{s1}$ -CN than milk of Alpine, Oberhasli, Toggenburg and Saanen  $\times$  Alpine crosses. The Swiss breeds (Saanen  $\times$  Alpine crosses and Toggenburgs, in particular) typically produced milk with

lower percentages of components than the non-Swiss breeds (Nubian and LaMancha). Our findings are supported by Mora-Gutierrez et al. (1991) who, after cluster analysis of data, found a higher incidence of Alpine goats producing low  $\alpha_{s1}$ -CN than Nubians.

Significant differences were found between the average coagulation properties of each goat breed

Table 2  
Least square (LS) means (and S.E.) of milk composition (%) of goats representing different breeds

Breed	<i>n</i> <sup>c</sup>	Total solids	SNF	Fat	Protein	Casein	$\alpha_{s1}$ -CN
Nubian	6	16.02 <sup>a</sup> (0.70)	9.01 <sup>a</sup> (0.22)	7.02 <sup>a</sup> (0.62)	3.59 <sup>a</sup> (0.16)	2.77 <sup>a</sup> (0.18)	0.24 <sup>a</sup> (0.11)
N $\times$ A <sup>f</sup>	8	13.98 <sup>a,b,c</sup> (0.61)	8.60 <sup>a,b</sup> (0.19)	5.38 <sup>a,b</sup> (0.54)	3.52 <sup>a</sup> (0.14)	<sup>g</sup>	0.28 <sup>a</sup> (0.13)
LaMancha	30	13.67 <sup>a,b</sup> (0.31)	8.72 <sup>a,b</sup> (0.10)	4.95 <sup>b</sup> (0.28)	3.34 <sup>a,b</sup> (0.07)	2.70 <sup>a</sup> (0.09)	0.25 <sup>a</sup> (0.19)
Saanen	13	12.98 <sup>b,c,e</sup> (0.51)	8.34 <sup>b,c</sup> (0.16)	4.64 <sup>b,c</sup> (0.45)	3.03 <sup>b,c</sup> (0.11)	2.48 <sup>a,b,c</sup> (0.10)	0.23 <sup>a</sup> (0.17)
Alpine	41	12.93 <sup>c,d,e</sup> (0.26)	8.14 <sup>d</sup> (0.08)	4.79 <sup>b,c</sup> (0.23)	3.02 <sup>c,d</sup> (0.06)	2.43 <sup>a,b,c,d</sup> (0.09)	0.10 <sup>b</sup> (0.09)
Oberhasli	3	11.70 <sup>d,e</sup> (1.00)	8.40 <sup>b,c</sup> (0.32)	3.29 <sup>c</sup> (0.88)	3.35 <sup>a,b,c</sup> (0.23)	2.44 <sup>a,b,c,d</sup> (0.22)	0.06 <sup>b</sup> (0.04)
Toggenburg	11	11.83 <sup>e</sup> (0.66)	7.56 <sup>e</sup> (0.21)	4.27 <sup>b,c</sup> (0.59)	2.76 <sup>d</sup> (0.12)	2.28 <sup>d</sup> (0.10)	0.09 <sup>b</sup> (0.04)
S $\times$ A <sup>f</sup>	6	12.06 <sup>e</sup> (0.69)	7.86 <sup>d,e</sup> (0.22)	4.20 <sup>b,c</sup> (0.62)	2.76 <sup>d</sup> (0.16)	2.16 <sup>c,d</sup> (0.22)	0.07 <sup>b</sup> (0.11)

<sup>a,b,c,d</sup> Significant differences exist between LS means in the same column with different superscripts (Kruskal–Wallis non-parametric test for one-way ANOVA by ranks,  $p < 0.05$ ).

<sup>c</sup> *n* denotes casein of only one N  $\times$  A cross tested.

<sup>f</sup> Cross-breeds — N  $\times$  A: Nubian  $\times$  Alpine and S  $\times$  A: Saanen  $\times$  Alpine.

<sup>g</sup> Insufficient number of samples for statistical analysis.

Table 3  
Least square (LS) means (and S.E.) of coagulation property measurements on goat milks representing different breeds and crosses

Breed	<i>n</i>	Coagulation time (s)	Coagulation rate (Pa/s)	Curd firmness (Pa)
Nubian	4	531 <sup>a,b</sup> (300)	128 <sup>a</sup> (23)	134 <sup>a,b</sup> (36)
N×A <sup>c</sup>	4	536 <sup>a,b</sup> (301)	145 <sup>a</sup> (23)	178 <sup>a</sup> (36)
LaMancha	13	964 <sup>a</sup> (154)	61 <sup>c,d</sup> (11)	101 <sup>a,b,c</sup> (19)
Saanen	8	456 <sup>a,b</sup> (221)	89 <sup>a,b,c,d</sup> (26)	120 <sup>a,b,c</sup> (27)
Alpine	27	729 <sup>b</sup> (119)	81 <sup>b,c,d</sup> (9)	96 <sup>c,d</sup> (14)
Oberhasli	3	346 <sup>a,b</sup> (346)	88 <sup>a,b,c</sup> (26)	84 <sup>a,b,c,d</sup> (42)
Toggenburg	4	829 <sup>a,b</sup> (316)	68 <sup>a,b,c,d</sup> (24)	66 <sup>c,d</sup> (38)
S×A <sup>c</sup>	4	658 <sup>a,b</sup> (312)	46 <sup>d</sup> (23)	52 <sup>d</sup> (38)

<sup>a,b,c,d</sup> Significant differences exist between LS means in the same column with different superscripts (Kruskal–Wallis non-parametric test for one-way ANOVA by ranks,  $p < 0.05$ ).

<sup>c</sup> Cross-breeds — N×A: Nubian×Alpine and S×A: Saanen×Alpine.

and cross-breed combination tested (Table 3). Milk from Nubians and Nubian×Alpine crosses formed a curd quickly (high CR) and firmly (high CF). Milk from Toggenburgs and LaManchas had long CT and low CR. Milk from Saanen×Alpine crosses had short CT and low CR values. The breeds in this study that produced milk containing a high amount of  $\alpha_{s1}$ -CN, namely Nubian and Nubian×Alpine crosses, exhibited the most favorable combination of milk composition and coagulation properties. High variability in coagulation property data (particularly CT) resulted in few significant differences among breeds. Nonetheless, these findings suggest that Nubian ancestry in a cross combination may improve milk composition and coagulation properties.

Ambrosoli et al. (1988) found that the milk of the Alpine breed showed better coagulation properties (short CT, high CR and high CF) than the Saanen breed even though no significant differences in milk composition were found. In the present study milk from Saanens exhibited significantly higher SNF and  $\alpha_{s1}$ -CN than milk from Alpines, but coagulation property differences (shorter CT, higher CR and higher CF) were not significant. It is possible that differences in milk composition and coagulation properties were due to genetic factors beyond breed differences alone, namely genetic variants, which warrants further study.

Nubian and Nubian×Alpine milk contained more  $\alpha_{s1}$ -CN than the other breeds;  $\alpha_{s1}$ -CN was associated with better milk composition; and better milk composition was related to better coagulation properties. Therefore, inclusion of Nubian goats in a herd may

increase the incidence of higher levels of  $\alpha_{s1}$ -CN in the milk supply and improve milk composition and coagulation properties. One might think it worthwhile for breeders to select animals on the basis of amount of  $\alpha_{s1}$ -CN in milk if their goal is to make cheese. However, we found non-significant correlations between  $\alpha_{s1}$ -CN and CR and CF. As percent total solids, SNF and protein are good indicators of coagulation properties, these components, rather than  $\alpha_{s1}$ -CN amount, may be adequate for indicating cheese-making ability of goat milk. In addition, HPLC requires a greater investment of time, resources, and money than traditional milk composition analyses.

#### 4. Conclusions

Our results indicate that goat milk containing high total solids, SNF, protein and fat coagulates quickly and forms a firmer curd than milk containing low levels of these components. It is of interest to cheesemakers to increase coagulation rate and curd firmness because both are economic factors. Therefore, selection for goats that produce high solids is recommended for those who are interested in cheese-making.

The RP-HPLC method used in this study was able to separate caseins, and enabled the quantification of  $\alpha_{s1}$ -CN relative to total casein in goat milk.  $\alpha_{s1}$ -CN was highly correlated with individual milk components and coagulation time. Goat milks that lacked  $\alpha_{s1}$ -CN had lower percent total solids, SNF, fat, protein and casein and lower coagulation rate than milks

that contained high amounts of  $\alpha_{s1}$ -CN. Milks that lacked  $\alpha_{s1}$ -CN tended to have lower values for coagulation time and curd firmness, but differences were not significant. Milk from Nubians, Nubian  $\times$  Alpine crosses LaManchas, and Saanens contained higher  $\alpha_{s1}$ -CN than milk from Alpines, Oberhaslis, Toggenburgs and Saanen  $\times$  Alpine crosses. Since milk from Nubian and Nubian  $\times$  Alpine crosses exhibited a more favorable combination of milk composition and coagulation properties than the other breeds, inclusion of the Nubian breed in a herd may be beneficial if cheese-making is the objective.

Percent total solids, SNF, and protein were better predictors of coagulation properties than  $\alpha_{s1}$ -CN. Thus, measuring for  $\alpha_{s1}$ -CN, which is time consuming and expensive, is not highly recommended for goat breeders or cheese-makers.

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