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Derivation of multivariate indices of milk composition, coagulation properties, and individual cheese yield in dairy sheep

M. G. Manca,* J. Serdino,* G. Gaspa,* P. Urgeghe,* I. Ibba,† M. Contu,† P. Fresi,‡ and N. P. P. Macciotta*[1]
*Dipartimento di Agraria, Università di Sassari, 07100 Sassari, Italy
†Associazione Regionale Allevatori della Sardegna, 09128 Cagliari, Italy
‡Associazione Nazionale della Pastorizia, 00161 Rome, Italy

ABSTRACT

Milk composition and its technological properties are traits of interest for the dairy sheep industry because almost all milk produced is processed into cheese. However, several variables define milk technological properties and a complex correlation pattern exists among them. In the present work, we measured milk composition, coagulation properties, and individual cheese yields in a sample of 991 Sarda breed ewes in 47 flocks. The work aimed to study the correlation pattern among measured variables and to obtain new synthetic indicators of milk composition and cheese-making properties. Multivariate factor analysis was carried out on individual measures of milk coagulation parameters; cheese yield; fat, protein, and lactose percentages; somatic cell score; casein percentage; NaCl content; pH; and freezing point. Four factors that were able to explain about 76% of the original variance were extracted. They were clearly interpretable: the first was associated with composition and cheese yield, the second with udder health status, the third with coagulation, and the fourth with curd characteristics. Factor scores were then analyzed by using a mixed linear model that included the fixed effect of parity, lambing month, and lactation stage, and the random effect of flock-test date. The patterns of factor scores along lactation stages were coherent with their technical meaning. A relevant effect of flock-test date was detected, especially on the 2 factors related to milk coagulation properties. Results of the present study suggest the existence of a simpler latent structure that regulates relationships between variables defining milk composition and coagulation properties in sheep. Heritability estimates for the 4 extracted factors were from low to moderate, suggesting potential use of these new variables as breeding goals.

Key words: factor analysis, sheep milk, coagulation properties, cheese yield

INTRODUCTION

Dairy sheep farming is widespread in many regions of Europe, especially in the Mediterranean area, where about 63% of the world's sheep milk is produced (FAOSTAT, 2014). The most important producers are Greece (699,500 t/yr), Romania (651,912 t/yr), Spain (552,517 t/yr), and Italy (406,177 t/yr; FAOSTAT, 2014). The Italian dairy sheep stock consists of about 5,500,000 animals (FAO, 2014), 60% of which are of the Sarda breed. The average gross milk production of the Sarda dairy industry is about 330,000 t/yr (Osservatorio Regionale per l'Agricoltura, 2012). All milk is destined to cheese production. In particular, 3 Protected Designation of Origin cheeses from Sarda sheep milk are currently manufactured in Italy: Pecorino Romano, which is largely exported to the United States (about 10,000 t/yr; ISMEA, 2013), Pecorino Sardo, and Fiore Sardo.

The breeding program of the Sarda breed includes 220,268 ewes (ICAR, 2014). Current selection goals are lactation milk yield and scrapie resistance. Fat and protein percentages and udder morphology are measured routinely only on first-lactation ewes, mainly to reduce costs of phenotype recording. However, there is interest in milk quality, especially on novel phenotypes related to nutritional characteristics, such as milk fatty acid composition (Carta et al., 2009) and cheese-making suitability.

Several variables contribute to the definition of technological properties of milk, and different phenotypes have been proposed to reliably characterize its cheese-making ability. Micro-manufacturing experiments have been suggested as a tool to quantify individual cheese yield in sheep (Othmane et al., 2002a). This variable is of great interest even though its use in routine phenotype recording programs is problematic. Milk coagulation properties (**MCP**) are assessed measures of milk cheese-making aptitude (Bittante et al., 2012; Pretto

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[1]Corresponding author: macciott@uniss.it

et al., 2013). They have been suggested as breeding goals for dairy cattle (Ikonen et al., 2004; Bittante et al., 2012; Chessa et al., 2013). Coagulation properties are usually defined by 3 variables: rennet coagulation time (**RCT**, min), curd firming time ($\mathbf{k_{20}}$, min), and curd firmness ($\mathbf{a_{30}}$, mm). They are measured directly by mechanical and optical techniques (Bittante, 2011). Moreover, they can be estimated by mid-infrared reflectance spectroscopy (MIRS) of fresh milk by using appropriate calibration algorithms (Dal Zotto et al., 2008; Bittante, 2011).

Although most of literature on MCP deals with dairy cattle, some studies have been carried out on dairy sheep. In particular, effects of fat and protein contents, temperature, pH (Park, 2007), SCC (Nudda et al., 2001), milk protein polymorphism (Piredda et al., 1993), and parity (Pazzola et al., 2014) on MCP have been investigated. Moreover, environmental factors such as stage of lactation (Jaramillo et al., 2008), flock-test date (Pazzola et al., 2014), and lambing season (Martini et al., 2008) have been found to affect sheep milk renneting properties.

Some authors have pointed out that a complex correlation pattern exists between traits that contribute to define the technological properties of milk, resulting in difficulties in the elucidation of causal relationships among variables (Ikonen et al., 2004; Cecchinato et al., 2011; Bland et al., 2015). In the specific case of MCP, the high degree of correlation that exists among RCT, k_{20}, and a_{30} represents a limitation for the use of these variables in the interpretation of the milk coagulation pattern (Bittante et al., 2015).

Multivariate statistics offers a set of methodologies for studying and dissecting complex correlation patterns. In particular, factor analysis has been used to study the correlation structure among milk compositional variables and MCP in Italian Brown Swiss cattle, allowing for the extraction of latent factors that were interpreted as indicators of milk composition, coagulation, acidity, and mammary gland health (Macciotta et al., 2012). Todaro et al. (2001) used factor analysis to study milk composition of ewes of the Valle del Belice breed and found 3 latent factors from 14 original variables, related to cheese yield, mastitis infection, and aptitude of milk for cheese making. In a further work on Girgentana goats, the same authors extracted 3 factors related to coagulation time, milk yield, and curd firmness (Todaro et al., 2005). Factor analysis was also used by Abilleira et al. (2010) to study relationships between milk composition, coagulation properties, and season of production in Latxa sheep.

The aim of the present work was to study the relationship pattern between milk composition, MCP, and experimental cheese yield in Sarda ewes by using multivariate factor analysis. This approach was expected to generate new variables with potential technical meaning that could be used for management and breeding purposes.

MATERIALS AND METHODS

Animals and Milk Sampling

The study was carried out on 991 Sarda ewes in 47 flocks (about 18 ewes per herd) located in the 4 historical provinces of Sardinia, Italy. All the ewes involved in the experiment were officially recorded in the herd book of Sarda breed. The individual milk samples (1 per animal from the morning milking) were collected between April and July 2014. Preservative was added to all milk samples, stored at room temperature, and processed 24 h after collection. Chemical composition of milk (fat percentage, protein percentage, lactose percentage, pH, urea, NaCl) and cryoscopy index were determined by Fourier transform mid-infrared (FT-MIR) spectroscopy equipment (MilkoScan, Foss Electric, Hillerød, Denmark). Calibration algorithms were developed according to FIL-IDF rules (ISO 9622:2013; IDF, 2013). Somatic cell count values (Fossomatic, Foss Electric) were also measured and converted to SCS by logarithm transformation as proposed by Ali and Shook (1980).

Analysis of MCP

The MCP were measured using a Formagraph (Foss Electric). Briefly, 10 mL of each individual sample was heated to 35°C before the addition of 200 μL of rennet solution (Hansen Naturen 215, with 80 ± 5% chymosin and 20 ± 5% pepsin, Pacovis Amrein AG, Bern, Switzerland) diluted to 0.8% in distilled water, resulting in a final dosage of 0.034 international milk clotting units (**IMCU**)/mL. This analysis ended within 30 min after rennet addition and produced a diagram as reported by Bittante et al. (2012). The following coagulation traits were recorded: RCT (which describes the time in minutes from the addition of the enzyme to the beginning of the coagulation process), k_{20} (which describes the time in minutes needed to obtain a curd firmness of 20 mm), and a_{30} (which describes the width in millimeters of the diagram at 30 min after the rennet addition).

Individual Laboratory Cheese Yield Analysis

The individual laboratory cheese yield (**ILCY**) was measured according to the method proposed by Othmane et al. (2002a) with some modifications. Raw milk samples were heated to 40°C and gently mixed by