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Milk coagulation properties and milk protein genetic variants of three cattle breeds/types in Sri Lanka

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Abstract

Milk coagulation is the primary step in the development of most dairy products. Raw milk from individual cows and different breeds exhibit distinct coagulation capacities. This variation is largely influenced by milk protein genetic variants. The aim of the present study was to evaluate differences in coagulation properties between milk obtained from three cattle breeds/types found in Sri Lanka. A total of 90 milk samples (400mL from each individual) were collected from two Sri Lankan cattle breeds/types (Thamankaduwa White/TW and Local/"*Batu*" cattle) and one European cattle breed (Friesian). Collected samples were subjected to enzymatic coagulation using commercial rennet source (Chymax®, Christian Hansen Standard, Denmark) and lactic acid bacteria (LAB) coagulation using a commercial starter culture(YFL 8 12,Christian Hansen Standard, Denmark) to determine milk coagulation properties. Different properties of milk coagulum such as yield, curd firmness, syneresis and rheological properties were evaluated. The biochemical composition (lactose, protein, fat, solid-non-fat) of milk samples were determined. Capillary Zone Electrophoresis (CZE) method was used to determine milk protein genetic variants. Experimental design was Nested Completely Randomized Design with three treatments. Milk coagulation time and curd firmness after enzymatic-coagulation were not significantly different ($p>0.05$) among the breeds. Coagulum yield was significantly higher ($p<0.05$) for the TW type than that of other breeds. Coagulum yield was negatively correlated with β-caseinA1 and α-lactalbumin in both enzymatic (-0.58) and LAB coagulation (-0.69). Coagulum yield was positively correlated ($p<0.05$) with β-casein B variant (0.70), protein (0.34) and lactose (0.36) contents. Meltability value was weakly and positively ($p<0.05$) correlated (0.34) with fat content of milk. Overall results indicate that there is a significant correlation between milk coagulation properties and milk protein genetic variants in three cattle breeds/types considered in the current study. TW type is the unique among studied breeds in terms of coagulation properties and milk protein genetic variants.

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1.Introduction

Through many years, dairy products have become a part of human diet. During past few years consumption of coagulated and fermented dairy products (cheese and yogurt) has been increased all over the world while fluid milk consumption has been decreased. Milk coagulation properties are important measures of the technological quality of milk[1]. Both enzymatic coagulation using milk clotting enzymes (such as rennet) and acid induced coagulation using lactic acid bacteria could be considered equally important to dairy industry. Raw milk from individual cows and different breeds exhibit distinct coagulation capacities and it affects the technological properties in cheese processing. General composition of cow and buffalo milk is an essential consideration to variation of milk coagulation. This variation is largely influenced by milk protein composition. Therefore, the effect of genetic polymorphism in the major milk protein on milk coagulation is also an important consideration. Also identification of genetic improvement of milk coagulation properties is crucial for selecting desired cattle breeds for manufacturing of variety of cheese and yogurt like dairy products[2].In general cattle breeds/types (Thamankaduwa White/TW, Common local/"*Batu*" and Friesian) contribute largely to the fluid milk production and dairy manufacturing in Sri Lanka. The primary objective of the present study was to identify enzymatic and lactic acid bacteria induced milk coagulation of these three cattle breeds/types in relation to milk composition and milk protein genetic variants.

2.Methodology

Fresh milk samples (400mL each) were collected from 90 individuals of cattle belongs to three cattle breeds/types. Cattle breeds/types were Friesian (n =30), TW cattle (n= 30) and Local ("*Batu*") cattle (n=30). Enzymatic coagulation of milk was determined using a method described by Berridge[3] and with commercial (Chymax®, Chr. Hansen's standard, Denmark) rennet solution. Milk samples were allowed to coagulate with commercial starter culture (YFL 812, Chr. Hansen standard, Denmark) in order to determine the lactic acid bacteria coagulation.

Different properties of coagulum such as milk coagulation time, yield of coagulum, curd firmness, syneresis and rheological properties (meltability value) were determined in both coagulation processes. To measure of curd firmness, 4465 Instron Universal Testing Machine (100, Royal Street, Canton, USA) was used. Meltability test was done by taking coagulum samples with aluminium borer and then allowed to melt at 106±1°C for 5 minutes. Ratios between unmelted and melted coagulum volumes were taken. Syneresis of milk coagulum samples were measured as per Wu *et al.*[4].

The biochemical composition (lactose, protein, fat, solid-non-fat) of milk samples was determined using "Lactoscan S" ultrasonic portable milk analyzer (Milkotronic Ltd., Bulgaria). As quality parameters somatic cell counts (SCC) were determined by using Delaval Cell Counter DCC (Delaval International AB, Tumba, Sweden).

Capillary Zone Electrophoresis (CZE) method was used to determine protein composition and its variation in milk samples. CZE analysis was carried out with a G-1600AX Capillary Electrophoresis system controlled by Chemstation software version A 10.02 (Agilent Technologies Co., SE-164 94, Kista, Sweden). Identification of the milk protein genetic variants and relative concentrations of milk protein genetic variants were done using standard electropherogrm.

Experimental design was Nested Completely Randomized Design (Nested CRD) with three treatments (Breeds/types). All the data were obtained from milk coagulation properties and proximate analyses were statistically analyzed using Generalized Linear Model (GLM) procedure of the SAS program (Version 9.1, SAS Institute Inc., 2000) and Pearson's correlation analyses were done using SPSS software (version 16.0).

3. Results, Discussion, Conclusion and Recommendations

Milk solid-non-fat levels were significantly higher ($p<0.05$) in TW (9.45%) and Local cattle (9.44%) than that of Friesian cattle (8.87%) while there was no significant difference in SCC in milk among three breeds/types ($p>0.05$). Rennet coagulation time and curd firmness after enzymatic coagulation were not significantly different among breeds/types. In LAB coagulation, highest curd firmness was showed in TW (885.15Pa) while lowest value was showed in Friesian breed (720.27Pa). In addition, highest coagulum yield of enzymatic coagulation process was observed in TW (83.76%) than Local (62.09%) and Friesian (66.52%). Lowest syneresis values were observed in local breed than other two cattle in both coagulation processes ($P<0.05$). Highest meltability ratio was observed in TW (5.77) and lowest ratio was observed in Friesian breed (1.01).TW cattle has the highest total casein (87.12%), κ-casein while low total casein level was observed in Local breed (84.59%) and Friesian breed (84.27%). Moreover, β-casein B variant was only present in TW cattle (Table1). Milk protein is one of the main factors which affect on the variation of milk coagulation properties among different cattle breeds[4]. In this study, correlations between milk coagulation properties, milk protein genetic variants and biochemical composition could be obtained (Table1). High level of κ-casein influenced on yield level, because, high κ-casein results smaller casein micelles and has significant improvement of coagulation properties. Therefore, curd get firmer and capable to retain larger amount of substances, hence increasing yield. It has been shown that β-casein B genetic variant could also increase the yield of cheese[5]. According to results of current study, milk coagulum yield was increased with the level of β-casein B ($P<0.05$) and κ-casein level (Table 1).

Table1. Milk protein genetic variants, biochemical compositions and correlations of milk coagulation properties milk protein genetic variants and biochemical compositions among three cattle breeds/types

	β-CNB	β-CN A1	β- CN A2	αs-CN	κ-CN	α-la	β-Lg	Protein	Fat	Lactose
Milk protein genetic variants (%) and biochemical compositions (%)										
TW	3.03	0.00	26.15	38.82	19.12	0.77	8.59	3.48	3.84	4.92
Local	0.00	0.49	33.96	38.92	11.22	1.23	9.72	3.47	3.31	4.91
Friesian	0.00	14.96	22.87	38.75	7.69	2.96	10.05	3.27	3.08	4.59
Correlation in enzymatic coagulation										
MCT/min	0.17	-0.03	0.08	-0.25	0.03	-0.22	-0.54*	0.13	-0.22	0.12
Firmness Pa	0.07	-0.18	-0.11	0.38	0.47	-0.49	-0.43	-0.19	0.32*	-0.17
Yield %	0.70**	-0.58*	0.17	0.10	0.51	-0.58*	-0.42	0.25	0.21	0.26*
Syneresis %	0.14	0.41	-0.39	-0.21	-0.05	0.48	0.39	0.19	-0.23	0.18
Meltability	0.33	-0.37	0.11	-0.17	0.36	-0.39	-0.20	0.19	0.34*	0.22
Correlation in LAB coagulation										
Yield %	0.23	-0.68**	0.34	0.09	0.48	-0.68**	-0.27	0.34**	0.22	0.36**
Firmness Pa	0.47	-0.48	0.12	-0.07	0.51	-0.29	-0.29	0.19	0.01	0.19
Syneresis %	0.10	0.09	0.08	-0.03	-0.32	0.44	-0.08	-0.10	-0.39**	-0.19

*Correlation was significant at 0.05 **Correlation was significant at 0.01

CN-Casein, lg-Lactoglobulin, MCT-milk coagulation properties

In milk coagulation process, whey protein is normally removed from milk coagulum; hence as major protein source casein is remained. Therefore, whey protein should have negative correlation with milk coagulum yield. According to current results, there was negative moderate correlation ($p<0.05$) between major whey protein (α-lactalbumin/α-la) and coagulum yield in both coagulum processes (Table 1).Normally when the fat content increases, number of bonds that are occupied with fat globules increases and makes impedance to whey drainage. Therefore, in this study, significant ($p<0.01$) negative correlation between milk fat level and syneresis percentage could be observed only in LAB coagulation. There was a positive correlation ($p=0.05$) between coagulum yield and both milk protein and lactose contents in LAB coagulation and significant positive correlation ($p<0.05$) between milk fat level and meltability value in enzymatic coagulation (Table1). Therefore; these results support the conclusion that there is a difference in coagulation properties of milk among the three cattle breeds/types with the correlation between milk coagulation properties and genetic variants of milk protein. TW type shows highest coagulum yield, highest meltability value and curd firmness than that of other two cattle breeds. In relation to milk protein composition TW shows highest level of total casein, κ-casein and β-casein B variants. Hence, TW cattle could be considered as a cattle type with better milk coagulation properties and with the unique milk protein genetic variants compared to that of other breeds considered in the current study.

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