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Kappa-casein influence on yield and quality of cottage cheese

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***Abstract*— Genetic markers associated with the quality of milk and dairy products are gaining great importance in the selection of cattle today. The markers include the kappa-casein gene (CSN3). The studies of many authors provide information on the effect of alleles and kappa-casein genotypes on the technological properties of milk, the quality of cheese. Data on the effect of the gene on the quantity and quality of cottage cheese, however, is covered much less. The influence of genetic modifications of kappa-casein (AA, AB, BB) on the quantity and quality indicators of the cottage cheese samples made from milk of Ural type cows is studied in the research. According to the research results, the genotype of kappa-casein affects the yield and quality of cottage cheese. A higher amount of cottage cheese is produced from the milk of cows with B-allelic variants of the CSN3 gene, by 12.25 g - 24, 90 g higher compared to the AA variant. The yield of the cottage cheese produced from the milk of animals with genotypes AB and BB kappa - casein is higher by 1.19% -2.42%. According to the results of the organoleptic assessment, the best indicators were obtained from cottage cheese samples made from cows' milk containing kappa-casein allele B in their genotype B. They had an advantage in tasting quality, the taste and smell are pure, milky, the color is white or white with a cream shade, evenly distributed over the whole mass. Larger particles of milk protein are revealed in sample No. 6, gathered from the milk of animals with BB genotype. Samples of cottage cheese made from cow's milk with variants AB and BB kappa-casein contained 0.29-0.93% more protein.**

Keywords—kappa-casein, milk proteins, cottage cheese, cattle, quality parameters, yield of the cottage cheese.

I. INTRODUCTION

The quality of milk and the possibility of its use in production of dairy products depend on various factors, the influence of protein polymorphism is an important factor [1, 2, 3, 4].

Milk proteins are valuable components in biological terms, they contain all the essential amino acids. The main protein – casein is commonly considered during the production of cheese and cottage cheese [5].

Nowadays, modern methods of molecular genetics are widely used to identify genes responsible for the quality of milk and dairy products. The main trend of molecular genetics, which is based on the use of marker genes, is a polymorphism of genes that increase the productivity of animals. Using the method of polymerase chain reaction (PCR) with restriction analysis, made it possible to identify kappa-casein variants in animals without taking into account gender, age, or physiological state. The method is based on splitting the DNA molecule into fragments by restructuring and following by recognition of segments [6]. Breeding animals for protein in milk is a promising trend not only in Russia, but also abroad [5].

The kappa-casein gene appears to be one of the promising markers associated with better milk quality, higher milk protein content and fermented milk protein products (CSN3) [4, 7].

The kappa-casein gene has several alleles. Scientists have identified 7-10 alleles: A, B, C, E, F, G, H, I, X and A1, of which A and B are the most common, containing 2 amino acid substitutions. Options are listed in decreasing order of electrophoretic mobility. Both alleles are involved in the phenotype of heterozygotes, and the codominant type of inheritance of the trait has been identified. Allele A contains the amino acids threonine (Thr) and aspartate (Asp) at positions 136 'and 148', respectively. In option B, threonine (Thr-136) is replaced by isoleucine (Iso), and aspartate (As-148) by alanine (Ala). Amino acid sequences is caused by the point mutations presence in the kappa-casein gene. The chain length of the protein κappa-casein consists of 169 amino acids. Genetic variants are different from each other in chemical composition and are characterized by the number of amino acid residues. All polymorphic variants have the greatest number of amino acid residues of glutamic acid and the smallest amount of tryptophan. Kappa - casein is a phosphoglycoprotein, compared with other casein fractions contains carbohydrates, sialic acid and a small amount of phosphorus. The kappa-casein gene has a size of 13 thousand base pairs, consists of 5 exons with a length of 850 base pairs and 4 introns. The introns are long, including the second intron, which breaks the sequence that encodes the signal peptide. The third intron contains repeating sequences and polymorphic microsatellites. Kappa-casein is the only component of casein that is affected by rennet, which is necessary for the manufacture of cheese and curd mass. Kappa-casein is the only component of casein that is affected by rennet, which is necessary for the manufacture of cheese and curd mass.

Milk proteins have a high nutritional value, contain all essential and nonessential amino acids, therefore they are easily absorbed by human body. Milk proteins are more

breed-like and depend on the individual genetic characteristics of cattle. Therefore, the identification of protein milk markers (kappa-casein) in the animal genome contributes to the creation of a herd of cows with a high content of proteins in milk, necessary for the production of cheese and cottage cheese. The genotype of an animal according to polymorphic systems is a lifelong marker, independent of the state of the body and environmental conditions.

The study of the polymorphism of the kappa-casein gene has advantages over traditional methods. It became possible to test the kappa-casein genotypes in lactating cows, bulls and young animals, to obtain amplification products in necessary quantities for more detailed studies. Currently, DNA polymorphism research methods are used for genotyping kappa-casein alleles. Scientists have developed several methods for genotyping the A- and B-alleles of the kappa-casein gene using restriction endonucleases [8].

For a number of years, Russia did not breed for maintaining the high frequency of the B allele kappa-casein in herds, which led to a decrease in its frequency in herds, in some cases to its complete absence. Currently, the situation has changed, with the use of DNA methods for this locus, an analysis of populations belonging to 11 cattle breeds has been carried out. The frequency of occurrence of alleles and kappa-casein genotypes can vary both between breeds and within the breed. They may have significant differences in various herds of the same breed, and in different groups within the same herd. Based on research by scientists, the genotyping of kappa-casein has an important role in animal breeding. Gene frequencies are more dependent on the genotype of a limited number of sires, which are used in farms for artificial insemination, and can vary significantly in the next generation. The frequency of allelic variants of the kappa-casein gene occurrence may depend on the breed and region of the enterprise. Most cattle breeds have a high frequency of allele A, and a low one - allele B, which is typical of black-and-white and Holstein cattle. Such changes are explained by different blood levels of animals according to the Holstein breed. In Jersey breed animals, on the contrary, a high frequency of allele B was detected (its share is 80%). Testing the A and B alleles is a relevant area, since cows milk containing the Kappa-casein B type allele has better heat resistance, a short coagulation time, faster coagulation, and lower costs for the production of cottage cheese and cheese compared to the A allele, the best quality of dairy products. Milk input for production of curd in animals with AA genotype is 8-10% higher. The European Association of Livestock Breeders consider the kappa casein option to be an economically important breeding criterion for dairy cattle breeds. In Germany, Denmark, Holland, selection for kappa-casein genotypes is included in cattle breeding programs. Due to the high practical significance of the kappa-casein index, in some Russian breeding organizations, this gene is indicated in ox catalogs. [9].

Researchers have proven that kappa-casein affects the technological properties of milk. The technological properties of milk characterize the nutritional value of dairy products, their yield and quality. The technological process in the production of protein milk products (cheese, curd) depends on the quality indicators of milk [8, 9, 10].

Cottage cheese refers to fermented protein foods made from whole, normalized or skimmed milk, fermented with leaven, followed by separation from the whey lump. Cottage cheese contains easily digestible and complete proteins, therefore, it has a high biological and nutritional value. There are 2 ways to make cottage cheese by the method of clot formation: acid-rennet (milk-clotting enzyme and lactic acid) and acid (lactic acid) [5, 8].

The yield and quality of cottage cheese depend on the organoleptic, physicochemical, technological, biological properties of milk [4].

The purpose of this study is to evaluate the yield and quality of cottage cheese made from the milk of cows with different kappa-casein genotypes.

II. METHODOLOGY AND MATERIALS

Cottage cheese was made from the milk of experimental Holstein cows in 2 breeding enterprises of the Sverdlovsk region (Experience 1 and Experience 2). Milk sampling for the production of cottage cheese was made during morning and evening milking and equaled 1 liter from the cow. A previously conducted study of animals using DNA technology at the Ural Veterinary Research Institute aimed to identify the kappa-casein genotypes. Based on the results of genotyping, animals were divided into 3 groups (AA, AB and BB genotypes), 3 samples of curd were obtained (in 2 experimental groups). Cottage cheese was produced by the acid method [2]. The quality of the curd samples was assessed by organoleptic and physicochemical methods. Acidity was determined by the titrimetric method using the indicator phenolphthalein (GOST standard 3624-92), MJ acid method (GOST standard 5867-90). The physicochemical indicators of the quality of the curd samples were evaluated on the basis of the Federal Law No. 88-FZ “Technical Regulations for Milk and Dairy Products [11].

III. YIELD AND QUALITY PARAMETERS OF COTTAGE CHEESE

A comparative analysis of mass (g), cottage cheese yield (%), milk input for the production of 1 kg of cottage cheese and quality indicators (organoleptic and physicochemical) of 6 cottage cheese samples was carried out. The results of cottage cheese sample production are demonstrated in Table 1.

TABLE I. MANUFACTURING CRAFT FROM MILK OF COWS GENOTYPED BY CAPPA-CASEIN

Sample	**Genotype**	**Mass, g**	**Yield of cottage cheese, %**	**Milk input per 1 kg of cottage cheese, kg**
Test 1				
1	AA	180,15	17, 51	5, 71
2	AB	200,34	19, 47	5,14
3	BB	192,40	18, 70	5,35
Test 2				
4	AA	158,20	15,39	6,50
5	AB	170,53	16,59	6,03
6	BB	183,10	17,81	5,61

Studies have shown that in the two experimental groups, the highest yield of cottage cheese (19, 47% and 17, 81%) and lower milk consumption per 1 kg of cottage cheese (5.14

kg and 5.61 kg) were obtained from the milk of cows containing in the genome Kappa casein B allele. In group 1, the output of cottage cheese made from the milk of animals with the AB genotype kappa-casein was 1.96% and 0.77% higher, the consumption of milk was 0.57 kg and 0.21 kg lower compared to AA and BB genotypes. In the experimental group 2, the highest yield of curd (by 1.22% and 2.42%) and the smallest consumption of milk for the production of 1 kg of curd (by 0.42 kg and 0.89 kg) were detected in cows with the BB genotype.

An expert group was formed to study the quality of curd samples according to organoleptic indicators. The results of sensory evaluation are presented in Table 2.

TABLE II. RESULTS OF SAMPLES' ORGANOLEPTIC TESTS

Sample	**Genotype**	**Organoleptic parameters**		
		Appearance and consistency	***Taste and smell***	***Color***
Test 1				
1	AA	Soft, smeary	The taste is bitter, sour milk smell	White, with a cream shade, not evenly distributed over the mass
2	AB	Soft, presence of milk protein particles	Pure, Fermented Milk	White
3	BB	Soft, presence of milk protein particles	Pure, Fermented Milk	White with cream tint
Test 2				
4	AA	Loose, smearing	Pure, Fermented Milk	White with cream tint
5	AB	Soft, presence of milk protein particles	Pure, Fermented Milk	White
6	BB	Soft, presence of milk protein particles	Pure, Fermented Milk	White

According to the table, all samples with the exception of No. 1 had good taste qualities: the taste is pure, fermented milk-liked, there were no additional tastes and smells. Sample No. 1 had a bitter aftertaste. The consistency of the best were samples №2, №3, №5, №6. Samples No. 1 and No. 4 had a soft, smearing consistency. The color of the samples is white or white with a cream shade. Sample cottage cheese No. 1 had a white with a cream tinge color that was not evenly spread throughout the mass. The quality of grain was the best in sample number 6, it had a larger grain.

Physicochemical parameters (protein, fat, acidity, moisture content) 6 samples of cottage cheese (Table 3).

TABLE III. PHYSICAL AND CHEMICAL INDICATORS OF QUALITY OF THE COTTAGE CHEESE

Sample	**Genotype**	**Indicator**			
		Protein,%	***Fat ,%***	***Acidity, °T, no more than***	***Moisture, %***
Test 1					
1	AA	12,02	8,3	184	74,9
2	AB	12,95	8,02	179	74,1
3	BB	12,31	8,06	180	74,8
Test 2					
4	AA	11,10	7,05	175	75,6
5	AB	12,0	8,10	178	75,3
6	BB	12,0	7,35	181	75,1

In group 1, the protein content in the sample of cottage cheese No. 2, made from the milk of cows with the AV kappa-casein genotype, is 0.64% and 0.93% higher than other samples. In experimental group 2 samples of cottage cheese No. 5 and No. 6, obtained from the milk of animals with genotypes AB and BB, kappa-casein contains 0.9% more protein compared to the AA genotype. In samples of cottage cheese No. 1 and No. 5, produced from the milk of cows with options AA and AV kappa-casein, a higher fat content is observed, in group 1 - by 0.24% and 0.28% higher; in group 2 - by 1.05% and 0.75%, respectively. The highest moisture content was observed in cottage cheese samples No. 1 and No. 4 (AA genotype) - 74.9% and 75.6%.

IV. CONCLUSIONS AND RECOMMENDATIONS

The results of the study prove that genetic variants of the kappa-casein gene influence the quality and quantity of cottage cheese. The highest quality samples of cottage cheese were obtained from the milk of cows with genotypes AB and BB kappa-casein. Therefore, in order to increase the yield of protein fermented milk products and improve their quality indicators, it is necessary to carry out the genotyping of animals by the kappa-casein gene and identify carriers of the allele B

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