Genetic Structure of Alpha S1 Casein in Slovak Pinzgau Cattle

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Abstract
The work was oriented to identification of α-s1 casein gene polymorphism and analysis of genotype structure in population of Slovak Pinzgau cattle. The material involved 39 cattle. Bovine genomic DNA was isolated by commercial kit NucleoSpin Blood (Macherey-Nagel) and ethanol precipitation and used in order to estimate α-s1 casein genotypes by means of PCR-RFLP method. The PCR products were digested with MaeIII restriction enzyme. In the population included in the study, homozygote genotype BB (39 animals) and heterozygote genotype BC (3 animals). Homozygote genotype CC has not been observed. In the total population of cattle homozygotes BB – 0.9231 were the most frequent, while BC – 0.0769 were the least frequent ones. This suggests a superiority of allele B – 0.9615.

Keywords: α-S1 casein, cattle, PCR-RFLP.

1. Introduction

The caseins are the major milks proteins of mammals. Their dual function for the suckling infant is to serve as a major source of amino acid, as well as to transport phosphate and calcium in sufficient amounts to support growth of bones [1]. Effects of milk protein polymorphism on milk production traits (milk, fat, and protein yield and also fat and protein percentage) have been investigated during the past decades and, in some cases, results are still conflicting [2, 3 ,4].
α-S1- casein (CSN1S1) is localised in bovine chromosome 6 [5, 6]. CSN1S1 genotype significantly influenced milk yield, fat yield, and protein yield with the highest yields obtained for the genotype BB. Protein percentage was influenced by CSN1S1, with the genotypes BC and BB, respectively, having the highest percentages. Significantly higher lactation cheese yields were estimated with CSN1S1 genotype BB [2].

2. Materials and methods

The material involved 39 cattle. Bovine genomic DNA was isolated by commercial kit NucleoSpin Blood (Macherey-Nagel) and used in order to estimate CSN1S1 genotypes by means of PCR-RFLP method.
DNA primers described by Koczan (1993) [7] were used to PCR amplification: forward primer 5´- TGC ATG TTC TCA TAA TAA CC - 3´ and reverse primer 5´ - GAA GAA GCA GCA AGC TGG - 3´.
The PCR reaction elaborated by Koczan (1993) [7] was modificated.
The reaction mixture in the total volume 25 µl containing 50 ng DNA, 0.5 U Taq polymerase (Fermentas), 1 mM MgCl2, 200 µM dNTP, 10 pM of each primer. The following amplification parameters were applied: 94°C for 3 minutes

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followed by 30 cycles: 94 °C for 30 seconds, 59 °C for 30 seconds, 72 °C for 30 seconds. The reaction was completed by the final synthesis: 72 °C for 10 minutes.
The PCR products of 310 bp were digested with 6 units of the MaeIII restriction enzyme (Roche). Restriction digestion fragments were loaded on 3 % agarose gel (Invitrogen) containing ethidium bromide and the gel were analyzed in the UV rays.

3. Results and discussion

In the population of Slovak Pinzgau cattle we detected two genotypes: homozygote genotype BB 36 animals and heterozygote genotype BC 3 animals. Homozygote genotype CC has not been observed.
Frequencies of genotypes and alleles determined in the total population (39 animals), are presented in Table 1. In the total population of cattle homozygotes BB – 0.9231 were the most frequent, while BC – 0.0769 were the least frequent ones. This suggests a superiority of allele B – 0.9615.
Genetic equilibrium of analysed population was evaluated on the base χ2-test. In the population included in the study non-significant diffrences in frequencies of genotypes were found. Frequencies of alleles in our population were similar.
Effectivity of allele's function of the gene CSN1S1 in cattle are presented in table 2.
Frequencies of alleles in our population were similar to those of CSN1S1 gene as reported by Sadik et al. (1972) [8]. Grosclaude et al. (1974) [9] reported slight superiority of allele B for Jersey, Guerusey, Normande, Reggiana. The higher frequency of the allele B in population of Pinzgau cattle reported by Beja-Pereira et al. (2003) [10]. Caroli et al., (2004) [11] present lower frequency of the allele C in population of Reggiana breed. Miluchová (2008) [12] observed superiority of allel B in Slovak Pinzgau population.

Table 1. Freqeuncy of genotypes and alleles of *CSN1S1*gene in the population of cattle

		Frequency of genotypes				Frequency of alleles		
	n				χ^2			p
		AA	AG	GG		A	G	
cattle	39	0.9231	0.0769	-	0.115	0.9615	0.0385	0.9443

Table 2. Effectivity of alleles CSN1S1 gene function in population Slovak Pinzgau cattle

	Heterozygosity		PIC	Ca	ENA	V%
	H_{obs}	H_{exp}				
cattle	0.0769	0.0740	0.0712	0.926	1.0799	7.59

4. Conclusions

It may be concluded that CSN1S1 is a polymorphic gene with a slight superiority of genotype BB and a superiority of allele B. Genetic structure examined in population of Slovak Pinzgau cattle remained within the range quoted in literature for other cattle breeds.

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