The casein genes in goat breeds from different Continents: analysis by Polymerase Chain Reaction – Single Strand Conformation Polymorphism (PCR-SSCP)

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ABSTRACT: A screening of casein gene variability was carried out by Polymerase Chain Reaction – Single Strand Conformation Polymorphism in 8 goat breeds from Sudan (Nubian goat), Turkey (Angora Goat Lalahan Tiftic, Angora Goat Yerkoy, Hair goat) and India (Jammu, Maharashtra, Rajasthan, South Goat). A total of 16 different alleles or groups of alleles were found, showing conspicuous differences among breeds. The allele frequencies were submitted to cluster analysis in order to highlight differences between breeds, also including data from Red Sokoto, West African Dwarf Nigeria, West African Dwarf Cameroon, and Borno Goat. The tree obtained from the cluster analysis showed two main lineages. The West African goat clustered together, the Indian and Turkish breeds were in the other group. Nubian goat was found in an intermediate position.

Key words: Milk, Casein, Genes, Goat, Variability.

INTRODUCTION – A high variability characterises the goat genes *CSN1S1*, *CSN2*, *CSN1S2*, and *CSN3* respectively coding for α_{s1}-casein, β-casein, α, and κ-casein (Caroli *et al.*, 2006). Such extensive genetic variation strongly affects milk composition traits (Martin *et al.*, 2002). Recent studies analysed simultaneously great part of the variation of the goat casein gene complex in different breeds (Sacchi *et al.*, 2005; Caroli *et al.*, 2006; Caroli *et al.*, 2007). This requires several techniques at the DNA level to identify the great number of polymorphisms described. Polymerase Chain Reaction – Single Strand Conformation Polymorphism (PCR-SSCP) is a cheap molecular tool for DNA typing allowing the simultaneous detection of several alleles. The aim of this work was to increase the knowledge about the casein gene structure in the goat species and to compare different breeds from all over the world. Preliminary data on the PCR-SSCP analyses of the casein genes in goat breeds from different Continents are presented.

MATERIAL AND METHODS – Table 1 provides information about the breeds analysed. Even if some breeds are represented by a rather low number of individuals, they contribute to the knowledge on both goat species and casein complex evolution. DNA was extracted from blood/tissue by standard methods and submitted to the PCR-SSCP analyses shown in Table 2. The allele frequencies were submitted to CLUSTER procedure followed by TREE

procedure (SAS, 1989) in order to highlight the differences between breeds. Data from Red Sokoto (RS), West African Dwarf Nigeria (WADN), West African Dwarf Cameroon (WADC), and Borno Goat (BG) were also included, considering only the allele groups typed in the present work.

Table 1.   Information about the goat breeds analysed.

Breed	Abbreviation	Origin	N of samples
Jammu	JA	India	14
Maharashtra	MA	India	9
Rajasthan	RA	India	25
South Goat	SG	India	29
Angora Goat Lalahan Tiftic	AGLT	Turkey	23
Angora Goat Yerkoy	AGY	Turkey	20
Hair Goat	HG	Turkey	48
Nubian	NU	Sudan	25

Table 2.   PCR-SSCP analyses of casein genes.

Gene	Alleles o groups of alleles identifiable	Method reference
CSN1S1	A/0[1], B/E, B', F, N	Caroli *et al.,* 2007
CSN2	A, C, E, 0	Chessa *et al.,* 2005
CSN1S2	A, B, C, E	Chiatti *et al.,* 2007
CSN3	A, B/B', B'', C, C', E, D/I/K/L, G, H, J, M	Prinzenberg *et al.,* 2005

Table 3.   Allele frequencies in the different breeds. Blanks: allele frequency = 0.000.

| Allele	JA	MA	RA	SG	AGLT	AGY	HG	NU
CSN1S1*A/0[1]	0.208	0.438	0.591	0.679	0.795	0.632	0.656	0.042
CSN1S1*B/E	0.792	0.563	0.409	0.321	0.205	0.342	0.323	0.604
CSN1S1*F							0.010	0.354
CSN1S1*B'						0.026	0.010	
CSN2*A	0.450	0.667	0.250	0.167	0.095	0.300	0.240	0.523
CSN2*C	0.550	0.333	0.750	0.833	0.905	0.700	0.760	0.477
CSN1S2*A	0.636	0.313	0.500	0.429	0.643	0.650	0.479	0.826
CSN1S2*B		0.063	0.038	0.375				
CSN1S2*C	0.364	0.625	0.462	0.161	0.357	0.350	0.510	0.174
CSN1S2*E				0.036			0.010	
CSN3*A	0.286	0.400	0.533	0.182	0.174	0.083	0.218	0.045
CSN3*B/B'	0.357	0.200	0.300	0.523	0.652	0.722	0.359	0.909
CSN3*C'							0.026	
CSN3*D	0.286	0.400	0.133	0.273	0.152	0.194	0.295	0.023
CSN3*G					0.022		0.103	0.023
CSN3*M	0.071		0.033	0.023				

RESULTS AND CONCLUSIONS – A total of 16 different alleles or group of alleles showing conspicuous differences among breeds were found (Table 3). The breed abbreviations are reported in Table 1.

As to the calcium-sensitive caseins, some main observations can be drawn. The faint *CSN1S1*F* allele occurred only in Hair Goat at a very low frequency, and in Nubian goat at a frequency higher than 0.35, whereas it was a rare allele in other Africa breeds (Caroli *et al.*, 2007). Only two alleles (*A* and *C*) were found at *CSN2* gene. The *CSN2*C* allele was predominant on *CSN2*A* in all breeds except Maharashtra and Nubian. The *CSN1S2*A* prevailed in all breeds except Maharashtra and Hair Goat. *CSN1S2*B* occurred at a rather high frequency in South Goat.

Six alleles were found at *CSN3,* among which *CSN3*B/B'* was predominant in all breeds except Maharashtra and Rajasthan. Nubian was almost monomorphic for this allele. *CSN3*D* was found in all breeds, with frequencies ranging from 0.023 (Nubian) to 0.4 (Maharashtra).

The tree obtained from CLUSTER procedure (Figure 1) resulted in two main lineages. The West African goat breeds clustered together (on the right side of the figure), whereas the Indian and Turkish breeds were in the other group (on the left side). The Nubian goat was in an intermediate position. The Indian and Turkish breeds were differently separated within the latter group where two main sub-clusters occurred. Jammu and Maharashtra were clearly separated from the other breeds. In the other sub-cluster South Goat was closer to Angora Goat Lalahan Tiftic and Angora Goat Yerkoy, and Rajasthan was closer to Hair Goat. Even if a higher number of DNA analyses, currently in progress, is needed to identify all the possible variation within the casein complex, the PCR-SSCP methodology employed was a useful tool to characterize the casein variation of the goat breeds analysed, allowing to discriminate two main clusters among them.

Figure 1.   Tree obtained from CLUSTER procedure on the basis of casein gene frequencies in the different breeds.
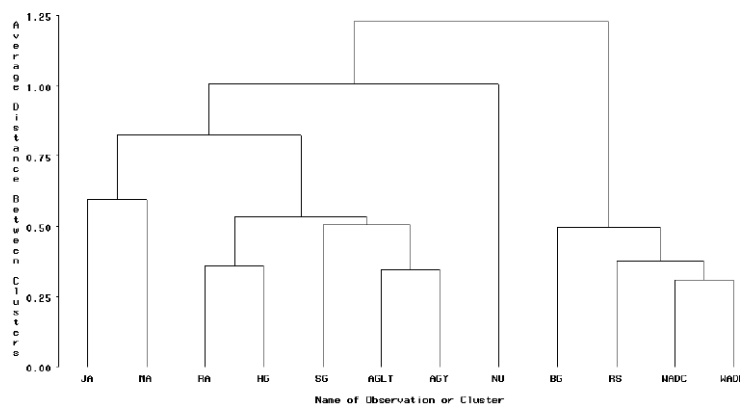
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