

PHYLOGENETIC RELATIONSHIPS AMONG BRAZILIAN SHEEP BREEDS

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Summary

Partial mtDNA control region was sequenced (588bp) in order to investigate the phylogenetic relationships among nine Brazilian sheep breeds. A total of 18 haplotypes were identified from 34 samples. The results suggest introgression between commercial Corriedale and local adapted Crioula Lanada breeds. All other sheep sequences were closely related, and there was no apparent phylogenetic structure relative to breed sampled.

Keywords

Conservation genetics, genetic characterization, introgression, mtDNA, *Ovis aries*

Introduction

Conservation of genetic resources requires the characterization of available stock is important for preservation and management as well as the evaluation of phylogenetic origins of the genetic groups available. Independent studies [2,5] show that the mitochondrial genome of *Ovis aries* is divided in two large haplo-groups, one of European origin (Type B) and the other, probably, of Asiatic origin (Type A). The objective of this study was to verify phylogenetic relationships between naturalised sheep breeds in Brazil, as well as verify mitochondrial types in these breeds.

Material and Methods

The naturalised breeds used in this study included: Santa Inês, Bergamácia, Rabo Largo, Morada Nova, Somalis and Crioula Lanada. To support phylogenetic analyses the commercial breeds Ile de France, Corriedale and Hampshire were also included. DNA was extracted from blood samples using a non-organic protocol. The region selected for mtDNA analysis was the first half of the control region (RC) or D-loop (1.2 Kb). The PCR products (*Polymerase Chain Reaction*) obtained from primers described by [2,5], were purified and sequenced in an ABI Prism 310 automatic sequencer (Applied Biosystems). The obtained sequences were aligned and edited by the program *Bioedit* [1]. Genetic distances and phylogenetic relationships were obtained using the program MEGA v.3.0 [3]. A sequence of *Ovis aries* Type B (European) and *Ovis ammon* were added with their respective access numbers to GenBank: AF010406 and AJ251327 to complement the analyses.

Results and Discussion

In total, 588 bases of the 5' region of the sheep D-loop were analysed at least twice in each of the 34 individuals distributed over the nine breeds cited above. Heteroplasm was observed in the breeds with a variation of 3 to 5 repetitions in 75 base pairs. For standardisation, only fragments with four repetitions in 75 base pairs were sequenced. From 30 variable sites (all transitional), 18 haplotypes were distributed within the 34 individuals analysed (Table 1). All haplotypes were of the B type (European).

TABLE 1. Frequency and distribution of haplotypes identified in the control region (D-loop) of mtDNA in nine breeds of sheep in Brazil.

Breed	HAPLOTYPES																	Total		
	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	Q		R	
Bergamácia							1	1	2	1	1									6
Crioula Lanada	2	1																		3
Morada Nova										1	1	2								4
Rabo Largo									1	2				1	1					5
Santa Inês								1	1	1							3			5
Somalis									1	1						1			1	5
Corriedale	2																			2
Hampshire			1	1																2
Ile de France					1	1														2
Total	4	1	1	1	1	1	1	2	5	1	6	1	2	1	1	1	3	1		34

The presence of common haplotypes (I and K) suggests proximity between naturalised Brazilian breeds and the occurrence of introgression between these in the recent past. There was also clear introgression between the Corriedale wool breed and the naturalised Crioula Lanada, since these were the only breeds that shared haplotype A.

Mean nucleotide diversity in the sequences for all breeds used was 0.011 (± 0.003) while mean diversity between breeds was 0.004 (± 0.001). These results agree with those from earlier studies showing there is small genetic divergence between naturalised Brazilian breeds of sheep [4].

A phylogenetic tree was obtained from the distance matrix *-p* and the *Neighbour-joining* (NJ) algorithm followed by a bootstrap analysis with 1000 repetitions. Two main divisions were seen in the tree, one formed by the most common haplotypes and the other by the less common ones. Due to the close proximity between the breeds, it was not possible to identify a clear phylogenetic relationship between the breeds analysed. Nevertheless, the relationship between the less common haplotypes was similar to the grouping found with the same samples using RAPD and microsatellite markers. Future studies should include other breeds and the frequency of haplotypes using RFLP-PCR.

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