

## Y-CHROMOSOME VARIABILITY OF IN BRAZILIAN SHEEP BREEDS

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### INTRODUCTION

A thorough genetic and physical mapping of the y-specific region (SRY) is hampered by lack of recombination between this region and the X chromosome and by the complexity of the repetitive regions. Thus, mapping of the SRY region is only available for mice and humans. However, knowledge of this region is very important in livestock, given the disproportionately high gametic contribution of some males in the genetic constitution of some breeds. These y-specific markers are also useful for sex identification, paternity exclusion tests and evolutionary studies. The SRY region is poorly known in sheep and only one polymorphism has been identified yet (Meadows *et al.*, 2004). The objective of this work was to apply a fast typing methodology, by PCR-RFLP technique, of a Single Nucleotide Polymorphism (SNP) and to study its frequency in naturalized Brazilian sheep.

### MATERIAL AND METHODS

A total of 559 bp of the SRY promoter in Santa Inês, Bergamácia, Morada Nova, Rabo Largo, Somalis, Ile de France, Hampshire, Corriedale, Dorper, and Damara (10 individuals each) were sequenced. The SNP was identified by Meadows *et al.* (2004) as a G/A transition located upstream the SRY gene. The identity of the SNP was confirmed by alignment of the fragments with a GenBank sequence (AF026566), using MEGA v.3 (Kumar *et al.*, 2004). A restriction fragment analysis conducted with WebCutter (<http://rna.lundberg.gu.se/cutter2/>) indicated that endonuclease *DdeI* may be used for diagnosing this SNP.

SNP frequency was estimated from a sample size of 123 from the following groups: (1) naturalized Brazilian breeds Santa Inês (n=48), Morada Nova (n=3), Bergamácia (n=19), Rabo Largo (n=20); (2) woolen commercial breeds: Ile de France (n=11), Corriedale (n=2), Hampshire (n=13); and (3) recently introduced African breeds: Damara (n=3) and Dorper (n=4). Unavailability of rams determined unbalanced sample sizes. DNA was extracted from leukocytes using a non-organic protocol modified by Miller *et al.* (1988), and the upstream fragment of SRY gene was amplified by PCR using primers developed by Meadows *et al.* (2004). Aliquots of PCR product were digested with restriction enzyme *DdeI*, incubated at 37°C for two hours, and fragments were visualized with silver staining on 6% polyacrylamide gels. Twenty per cent of the samples were repeated as a control of results. Individuals were considered as mutants (SNP\_A) or wild type (SNP\_G), following the GenBank sequence AF026566.

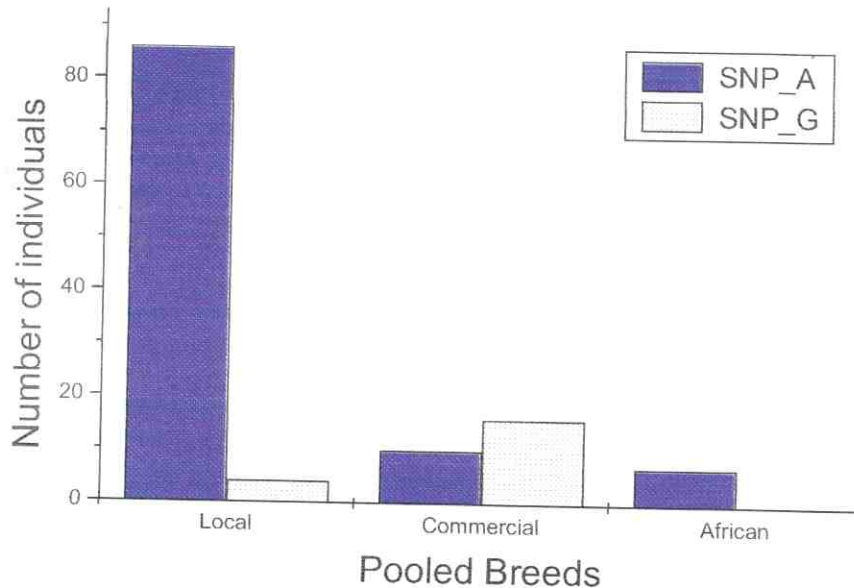
### RESULTS AND DISCUSSION

All sequenced samples confirmed the RFLP-PCR results. The mutant allele had high frequency among naturalized Brazilian and the African breeds (Table 1 and Figure 1). Although the wild type was more frequent among woolen commercial breeds (Figure 1), it was not fixed in these breeds. Some naturalized Brazilian breeds had only the mutant form (Table 1).

**Table 1. Frequency of the Y-chromosome SNP in sheep breeds in Brazil. N= samples size.**

Breed	N	Frequency SNP A	Frequency SNP G
Santa Inês	48	0.98	0.02
Bergamácia	19	0.89	0.11
Morada Nova	3	1.00	-
Rabo Largo	20	0.95	0.05
Ile de France	11	0.27	0.73
Hampshire	13	0.38	0.62
Corriedale	2	1.00	-
Damara	3	1.00	-
Dorper	4	1.00	-
Total samples/ Mean Proportion	123	0.80	0.20

Despite a reduced sample size, Meadows et al. (2004) characterized the prevalence of SNP\_G in the large European woolen breeds, whereas was more typical of more primitive breeds from Asia. Considering the high frequency of SNP\_G in the woolen commercial breeds, can be concluded that the results of this study are partially consistent with this overall pattern. These patterns await confirmation from more encompassing typing studies around the World.



**Figure 1. Frequency of the SRY-SNP alleles among three groups of sheep sampled in Brazil.**

#### CONCLUSION

The results demonstrate prevalence of the SNP\_A mutant allele in Brazilian naturalized breeds, suggesting a pattern similar to the more primitive, Asian breeds. We suggest that the woolen European breeds have had a rather recent influence in the country, a hypothesis that will demand the utilization of a greater number of markers and larger sample sizes.

#### REFERENCES

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