



## Differences in Y-chromosome haplotype frequencies at the microgeographical level

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### Abstract

Five Y-chromosome short tandem repeat polymorphisms were studied in Cantabria, a small region in Northern Spain. DNA samples were obtained from male subjects from three different areas: a mostly urban area near the coast, with a mixed population, and two mountain areas that have traditionally had less opportunity for social interaction with neighbouring communities. Statistically significant differences were found in allele and haplotype frequencies between the areas studied, revealing the existence of a population structuration at this microgeographical level.

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### 1. Introduction

The analysis of Y-chromosome polymorphisms has been a subject of considerable interest in recent years. The absence of recombination makes them a very useful tool to characterize paternal lineages, which may be particularly useful in some paternity disputes, such as those of a male offspring with an absent alleged father. On the other hand, the effective population size of Y chromosomes is smaller than that of autosomes. Thus, Y-

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polymorphisms may be more likely to reveal differences among subpopulations due to genetic drift and other factors.

Studies of Y microsatellites have usually shown little variation among Western European countries [1]. However, the possibility of population stratification within certain regions with specific historical and demographic characteristics still remains. The clarification of this issue is of importance not only from the perspective of population genetic studies, but also for a correct interpretation in forensic cases.

## 2. Materials and methods

### 2.1. Population

We studied male subjects living in Cantabria, a region in Northern Spain with a population of 530 000. This region of 5000 km<sup>2</sup> is situated between the Cantabrian Sea and the Cantabrian Mountains, and has a flat, well-communicated, and densely populated coastal area. It includes the region capital and has about 400 000 inhabitants. On the other hand, Cantabria has a mountainous area with several valleys that have traditionally had difficult communications. Their inhabitants have had less opportunity for social and economic interaction with people from other areas. Among them are the Liébana and the Pas valleys, each with a population about 5000. The study group consisted of 181 subjects living either in the coastal area ( $n=97$ ) or in one of those valleys, the Liébana valley ( $n=43$ ) and the Pas valley ( $n=41$ ).

### 2.2. DNA analysis

The following loci were studied: DYS390, DYS19, DYS 389-1, DYS389-2, and DYS393. DNA was isolated from peripheral blood by the Qiagen method. A pentaplex PCR was carried out as described by Gusmao et al. [2]. The size of amplified fragments was determined in a ABI Prism 310 analyser. The differences in allelic frequencies of isolated loci and the corresponding haplotypes were estimated by an extension of Fisher's exact test based on a Markov chain with 10 000 possible tables, carried out with SPSS software. Coancestry coefficients and population distances were computed with TFPGA and Arlequin packages.

## 3. Results

There were significant differences in allele frequencies between the three subpopulations studied regarding DYS390 ( $p < 0.001$ ; Fig. 1), DYS389-1 ( $p = 0.043$ ), and DYS393 ( $p = 0.004$ ) systems. However, no statistically significant differences existed in DYS19 and DYS389-2 allelic frequencies.

As expected, strong allelic associations were found to exist between the loci studied. Among the 181 individuals genotyped, 79 different haplotypes were found. The two most common were present in 29 and 16 individuals, respectively (16% and 9% of the overall

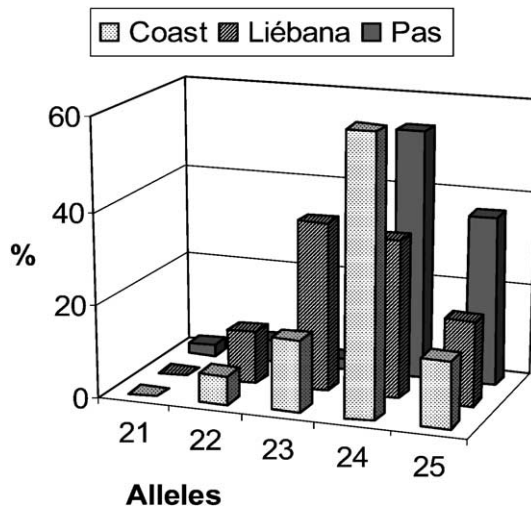


Fig. 1. Allele frequency distributions at the DYS390 locus in the three geographical areas studied.

population). The overall haplotype diversity was somewhat higher in the coastal area (0.95) than in the Liébana and Pas valleys (0.94 and 0.90, respectively). Significant differences in haplotype frequencies were also observed between the three subpopulations ( $p < 0.001$ ; Fig. 2). Pair-wise comparisons of haplotype frequencies revealed the following

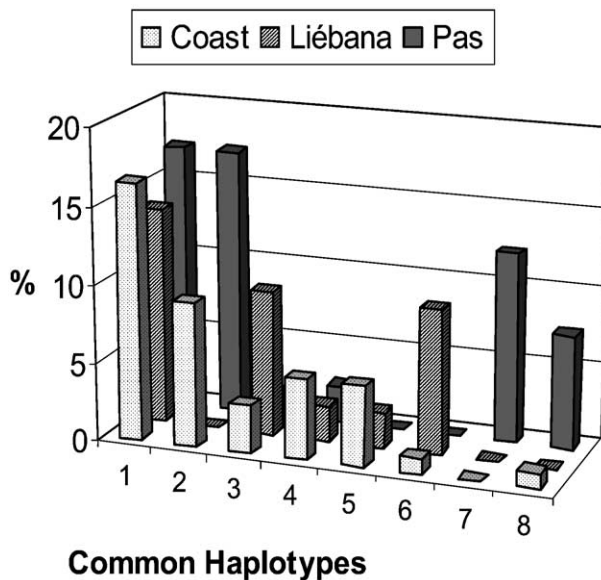


Fig. 2. Frequencies of the most common haplotypes.

*p*-values: Coast-Liébana, 0.002; Coast-Pas, 0.076; Liébana-Pas, <0.001. AMOVA analysis showed an overall *F*<sub>st</sub> value of 0.036 (95% confidence interval 0.006–0.065). Reynold's distances were 0.039, 0.013 and 0.081, respectively.

#### **4. Discussion**

A marked population stratification has been previously reported in some areas, frequently in the developing world, with communities with different languages and religion, as well as frequent consanguineous unions [3]. Albeit preliminary due to the small sample size, these results reveal that Y-chromosome microsatellites may also show significant population substructure at the microgeographical level in small European regions with distinctive demographic features and low social interaction with neighbouring communities, even in the absence of obvious ethnic or language differences. Although the overall *F*<sub>st</sub> value in this study was relatively small, the differences in some haplotype frequencies may need to be taken into consideration in certain trace–donor match analyses.

#### **Acknowledgements**

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