STUDY OF INDEL GENETIC MARKERS WITH FORENSIC INTEREST AND ANCESTRY INFORMATIVE POWER IN PALOP'S IMMIGRANT POPULATIONS IN LISBOA - PRELIMINARY RESULTS

Ana Inácio[1,2], Heloísa Afonso Costa[1,6], Cláudia Vieira da Silva[1,6], Teresa Ribeiro[1,6], Maria João Porto[1,6], Jorge Costa Santos[1,3,4,6], Gilberto Igrejas[2], António Amorim[1,4,5,6]

1 - Serviço de Genética e Biologia Forenses, Delegação do Sul, Instituto Nacional de Medicina Legal e Ciências Forenses, Lisboa, Portugal
2 - Departamento de Genética e Biotecnologia, Universidade de Trás-os-Montes e Alto Douro, Vila Real, Portugal
3 - Faculdade de Medicina, Universidade de Lisboa, Lisboa, Portugal
4 - Instituto Superior de Ciências da Saúde Egas Moniz, Monte da Caparica, Portugal
5 - Faculdade de Ciências, Universidade de Lisboa, Lisboa, Portugal
6 - CENCIFOR - Centro de Ciências Forenses, Coimbra, Portugal

Introduction

The increasing number of immigrants in Portugal is an unavoidable reality. According to Portugal Contemporary Base - PORDATA -, by the end of 2013, the foreign population living in Portugal was about 398.268, *i.e.*, about 4% of the total population. The total number of immigrants from Portuguese-speaking African countries (PALOP) was about 100.800, from which 20.000 were from Angola, 42.000 from Cabo Verde, 176.600 from Guiné-Bissau, 2.850 from Moçambique and 10.170 from São Tomé e Príncipe. From those 100.800, about 75.000 are part of Lisboa population.

The migratory phenomenon in Portugal can be one of the main factors for the genetic variability.

In the last years, a new class of autosomal insertion/deletion markers - InDel -, has gained interest in forensic genetics. They are characterized by the presence or absence of a specific sequence of nucleotides. Significative differences in allele frequencies of InDel markers, between different groups or populations, can be used as evolutionary and ancestry indicators.

Since there is few data for InDel markers of PALOP immigrants living in Lisboa, our aim is to characterize those groups of individuals by typing them with, at least, 30 markers and compare different groups of individuals/populations.

Material and Methods

Sample collection and DNA extraction

We studied 454 bloodstain samples, 258 from Angola, 124 from Guiné-Bissau and 72 from Moçambique, collected from immigrant individuals inhabitants of Lisboa metropolitan area, and undergoing forensic investigations in INMLCF. An interview was conducted in order to register personal data of the studied individuals, particularly the name, the age, the birth place, the individual ethnicity and his parental ethnicity. For each individual in study, peripheral blood samples were collected on Whatman® paper and DNA extraction was carried using Chelex®100 resin extraction method[1].

DNA typing

InDel typing was performed with Investigator® DIPplex PCR Amplification Kit. Allele definition was achieved in ABI 3130 Genetic Analyser (Applied Biosystems) by comparison to reference ladders provided by the manufacturer (Applied Biosystems).

Data analyses

Haplotype diversity and pairwise genetic distances were calculated using the Arlequin software ver. 3.5[2]. Phylogenetic comparasion between the three populations and others was accomplished with Arlequin software ver. 3.5 and with PHYLIP – Phylogeny Inference Package (version 3.2)[3].

Results

The haplotypes of individuals were obtained in a electropherogram in which we verify the panel of the 30 characteristic markers of this amplification kit, plus the amelogenin marker. An example of an electropherogram is presented in Figure 1.

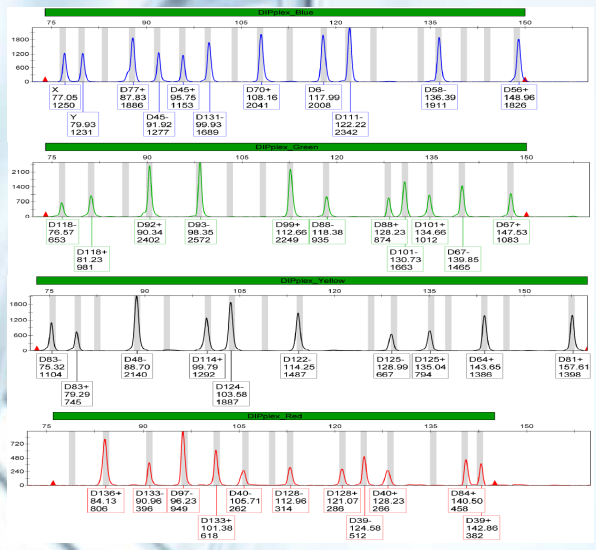

Figure 1 – Example of an electropherogram from an individual's haplotype with the various markers used in this study. For each marker, we can have an insertion and/or a deletion. If the individual just has one of those, we call him homozygotic for that marker; if he has both of them, that individual is heterozygotic for that marker.

Through the haplotypes obtained for the three populations in study, it was possible to verify the existence of genetic differences between populations. In figure 2 we presented a phylogenetic tree representative of the genetic distances between the studied populations.

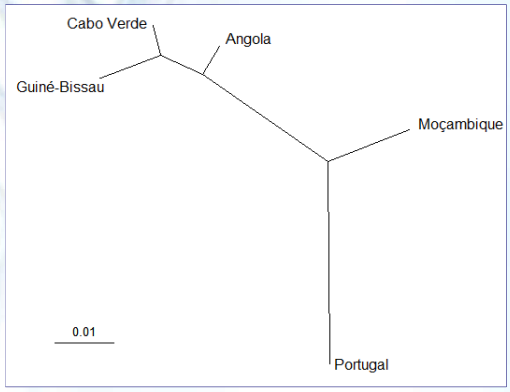

Figure 2 – Phylogenetic tree representing the genetic distances among populations under study.

Discussion/Conclusion

Through the results obtained we can confirm that studied populations show significant genetic distances between them and between them and the Portuguese population and so we can conclude that they introduce genetic variability in the Portuguese population.

References

[1] - P. Walsh, D. Metzger, R. Higuchi. Chelex 100 as a médium for simple extraction of DNA for PCR-based typing from forensic material. Biotechniques. 1991; 10(4):506-13.

[2] - L. Excoffier, G. Laval, S. Schneider. Arlequin (version 3.0): An integrated software package for population genetics data analysis. Evolutionary Bioinformatics Online. 2005 (1):47-50.

[3] - Felsenstein, J. PHYLIP - Phylogeny Inference Package (Version 3.2). Cladistics. 1989 (5): 164-166