HLA-DQA1 POLYMORPHISM IN A PORTUGUESE POPULATION

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INTRODUCTION

The polymerase chain reaction (PCR) is a powerful and sensitive techique which offers new tools for forensic analysis both in identification of stains and in paternity investigation.

By use of PCR amplification and reserve dot-blot procedure the polymorphism at the HLA-DQA1 locus revealed six major allelic types - DQA1.1, DQA1.2, DQA1.3, DQA2, DQA3 and DQA4 (1).

We present data on HLA-DQA1 allele frequencies and distribution of the different genotypes at this locus in a sample of a Portuguese population, mainly from the Southern part of the country.

MATERIAL AND METHODS

Blood samples were obtained from 106 unrelated individuals collected for paternity cases presented to the Institute of Legal Medicine in Lisbon.

The individuals of the investigated samples can be divided according to their genetic background into five geographical zones of Portugal: 57% from the South, 10% from the Center, 16% from the North, 11% from Madeira and 6% from Açores Islands.

DNA was extracted from bloodstains with Sigma chelating resin, using the method described by Barros et al. (2).

Amplification by PCR was performed during 32 cycles on approximatly 0.1µg of human genomic DNA using a DNA thermal cycler (denaturation at 94ºC for 1 min, annealing at 60ºC for 30s and extension at 72ºC for 30s).

The HLA-DQA1 alleles were typed by the reverse dot-blot procedure as described in the Amplitype HLA-DQ Alpha Forensic DNA Amplification and typing Kit (Perkin Elmer).

RESULTS

The distribution of HLA-DQA1 genotypes and allele frequencies for the Portuguese population studied are shown in Table 1. The distribution of genotypes indicate that there is no deviation from Hardy-Weinberg equilibrium, as shown by the χ^2 test ($\chi^2 = 12.21$; d.f. = 15; $0.50 < p < 0.70$).

Table 1. Distribution of HLA-DQA1 Genotypes and Allele Frequencies in the Portuguese Population

Genotype	Observed		Expected	
	n	%	n	%
1.1-1.1	3	2.83	4.57	4.31
1.1-1.2	4	3.77	4.98	4.70
1.1-1.3	4	3.77	4.98	4.70
1.1-2	7	6.61	7.47	7.05
1.1-3	3	2.83	3.94	3.72
1.1-4	20	18.87	13.49	12.73
1.2-1.2	2	1.89	1.36	1.28
1.2-1.3	1	0.94	2.72	2.56
1.2-2	5	4.72	4.08	3.85
1.2-3	2	1.89	2.15	2.03
1.2-4	8	7.55	7.36	6.94
1.3-1.3	0	0.00	1.36	1.28
1.3-2	6	5.66	4.08	3.85
1.3-3	4	3.77	2.15	2.03
1.3-4	9	8.49	7.36	6.94
2-2	3	2.83	3.06	2.88
2-3	4	3.77	3.23	3.04
2-4	8	7.55	11.04	10.41
3-3	1	0.94	0.85	0.80
3-4	4	3.77	5.83	5.50
4-4	8	7.55	9.06	9.40
TOTAL	106	100.00	106.02	100.00

HLA DQA1.1= 0.2075	
HLA DQA1.2= 0.1132	
HLA DQA1.3= 0.1132	χ^2=12.21
HLA DQA 2 = 0.1698	df= 15
HLA DQA 3 = 0.0896	$0.50 < p < 0.70$
HLA DQA 4 = 0.3066	CE= 0.58

In the Portuguese population the HLA-DQA1.1-4 genotype is the most frequent (18.87%), followed by HLA-DQA1.2-4, DQA1.3-4, DQA2-4 and DQA4-4 genotypes (7.55%-8.49%). In our sample, we have not observed any HLA-DQA1.3-1.3 genotype.

The allele frequency analysis reveals that the most common HLA-DQA1 alleles in the Portuguese population are: HLA-DQA4 allele and HLA-DQA1.1 allele with frequencies, respectively, of 0.3066 and 0.2075.

In our sample, we have obtained, a power of discrimination of 0.9210 and an allelic diversity of 0.8042, which is in good agreement with other populations studied at the some locus (5).

We have introduced the HLA-DQA1 polymorphism study in 51 paternity testing cases. The 54,5% of exclusions obtained, corresponding to 12 incompatibilities in 22 cases of non-paternity, is in good agreement with the theoretical a priori chance exclusion value (C.E. = 0.58).

DISCUSSION

The studied sample of the Portuguese population is composed of individuals whose genetic background is from several geographical zones of Portugal - North, Center and South, and also Madeira and Açores Islands, with higher incidence (57% of individuals) from the South.

When compared to other Caucasian data, our allele frequencies show some differences in the distribution of HLA-DQA1.1 and HLA-DQA1.3 alleles, which are more frequent in our population than in Barcelona (3), Italian (4) or US Caucasian populations(5).

Our study also shows that data on HLA-DQA2 and HLA-DQA3 allele frequencies, in the Portuguese population, are very similar to the Italian ones.

We can infere that HLA-DQA1 polymorphism exibits differences among several ethnic groups.

Due to the power of exclusion of 0.58 and the power of discrimination of 0.92 we can assign the HLA-DQA1 system as a good genetic marker in the Portuguese population both for paternity investigation and for individual identification in forensic analysis.

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